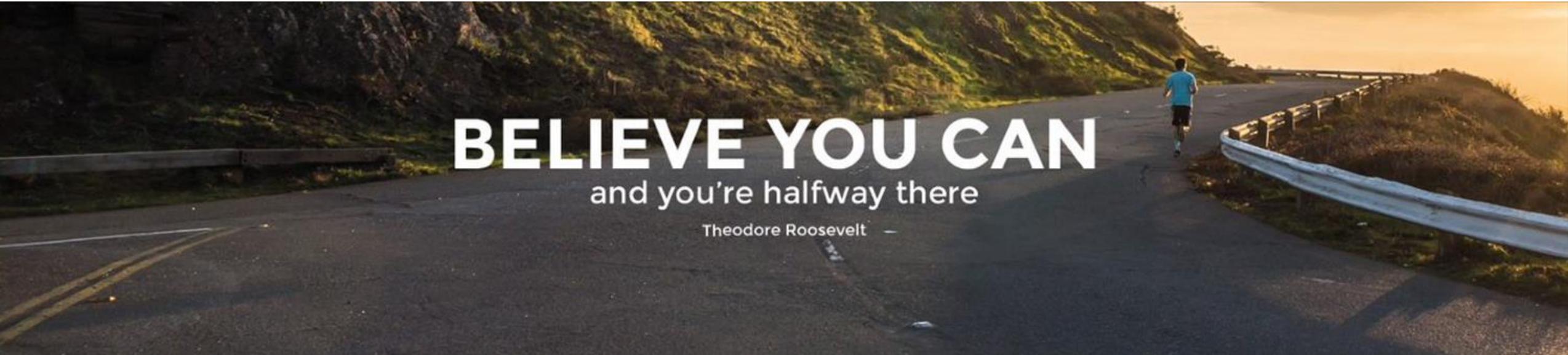


# EB Education Revision Guide



## How to work with Enzymes: Part 2

# Enzymes

## Breaking down molecules

Digestive enzymes break down large insoluble molecules into smaller, soluble molecules.

This needs to happen so that the molecules can pass through the walls of the small intestines into the blood.

In plants energy is stored in the form of starch, a carbohydrate. Enzymes in the plant will break down the starch into sugars when the plant needs energy. The plant will then respire using the sugar.

## Which enzymes?

**Carbohydrases** break down carbohydrates into simple sugars like glucose.

**Amylase** is a carbohydrase which breaks down starch.

**Proteases** break down proteins into **amino acids**.

**Lipases** break down lipids into **glycerol** and **fatty acids**.

**TOP TIP:**

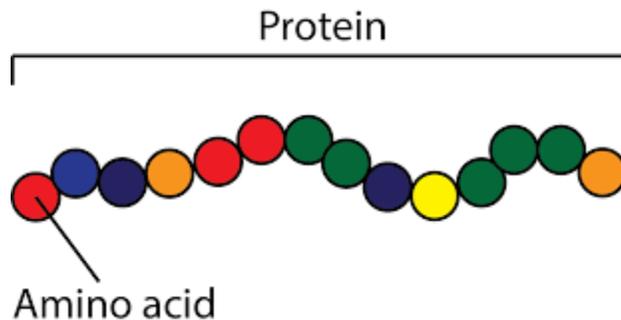
Fatty acids will lower the pH

# Enzymes

## Building molecules

Enzymes are used to combine smaller molecules to make larger molecules.

Carbohydrates, proteins and lipids are all made from smaller molecules and the joining of them is catalysed by enzymes.



Carbohydrates are made by joining simple sugars together. **Glycogen synthase** is an enzyme that joins together glucose molecules to make **glycogen**.

Glycogen stores energy in animals.

Proteins are made by joining amino acids together.

Lipids are made by joining fatty acids and glycerol together – many enzymes are involved in this.

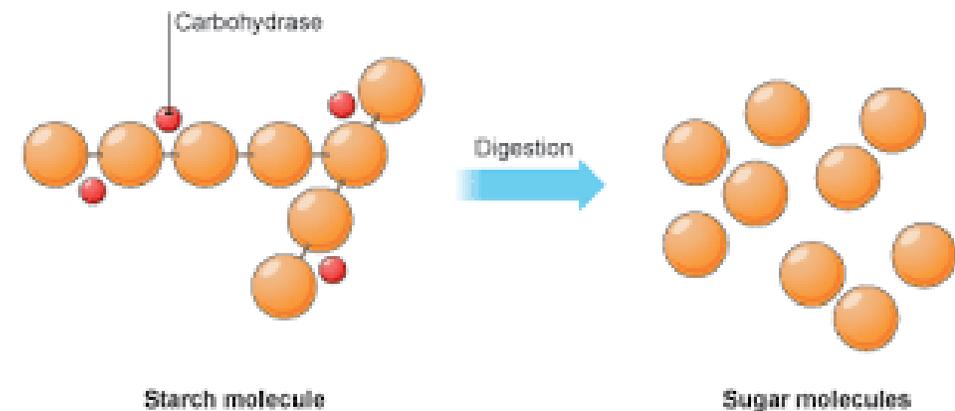
# Investigating enzymes

## Investigating the effect of pH on the enzyme amylase

The enzyme amylase controls the breakdown of starch in our digestive system.

We can simulate digestion using solutions of starch and amylase in test tubes. We can also determine the optimum conditions required.

The presence or absence of starch can be determined using iodine solution. If starch is present it will change from brown/orange to blue/black. In this experiment, we can measure how long the amylase takes to break down the starch at different pHs.



# Required Practical

## Method

- Use the syringe to place 2 cm<sup>3</sup> of amylase solution into a test tube.
- Using another syringe, add 1 cm<sup>3</sup> of pH solution to the test tube.
- Place the test tube into a water bath set at 30°C and leave for 5 minutes.
- Add a drop of iodine solution into each dimple of a spotting tile.
- After 5 minutes, use another syringe to add 2 cm<sup>3</sup> of starch to the test tube in the water bath and mix using a plastic pipette. Start a stopwatch.
- After 30 seconds, remove a drop of the solution and add it to the first drop of iodine on your spotting tile. (Hint – the iodine solution should turn blue–black.)
- Wait another 30 seconds. Then remove a second drop of the mixture to add to the next drop of iodine.
- Repeat until the iodine solution and the amylase/buffer/starch mixture remains orange. (Hint – this is the point at which the amylase has fully digested the starch.)
- Record the time taken for the amylase to fully digest the starch. (Hint – count how many iodine drops you have used.) Multiply the number of drops by 30, as each drop equals 30 seconds of reaction time.
- Repeat the whole procedure with a different pH buffer.





# Your turn:

2 Phenolphthalein is an indicator. It is pink in alkaline solutions and turns colourless as the pH decreases.

It can be used to measure the activity of the enzyme lipase on the breakdown of lipids.

Samples of milk containing phenolphthalein were incubated with lipase at different temperatures.

The time taken for the phenolphthalein to turn colourless was recorded and used to calculate the rate of enzyme activity.

Figure 10 shows these results.

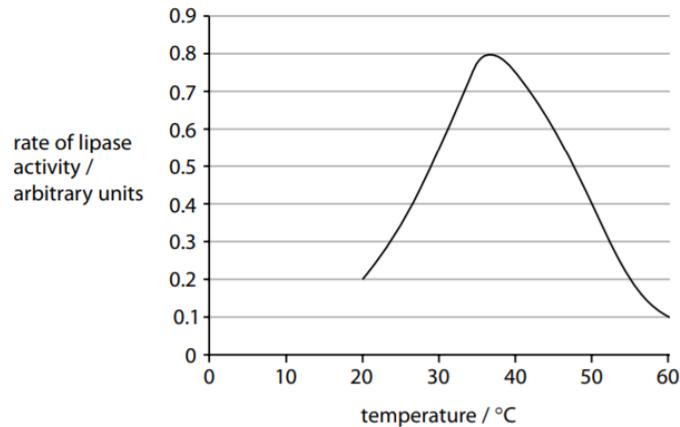


Figure 10

(a) (i) Explain why phenolphthalein turns colourless when lipase breaks down the lipids in milk.

(2)

(ii) Describe the effect of temperature on the activity of lipase, as shown in Figure 10.

(2)

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(iii) Explain why the activity of lipase changes above a temperature of 40°C.

(2)

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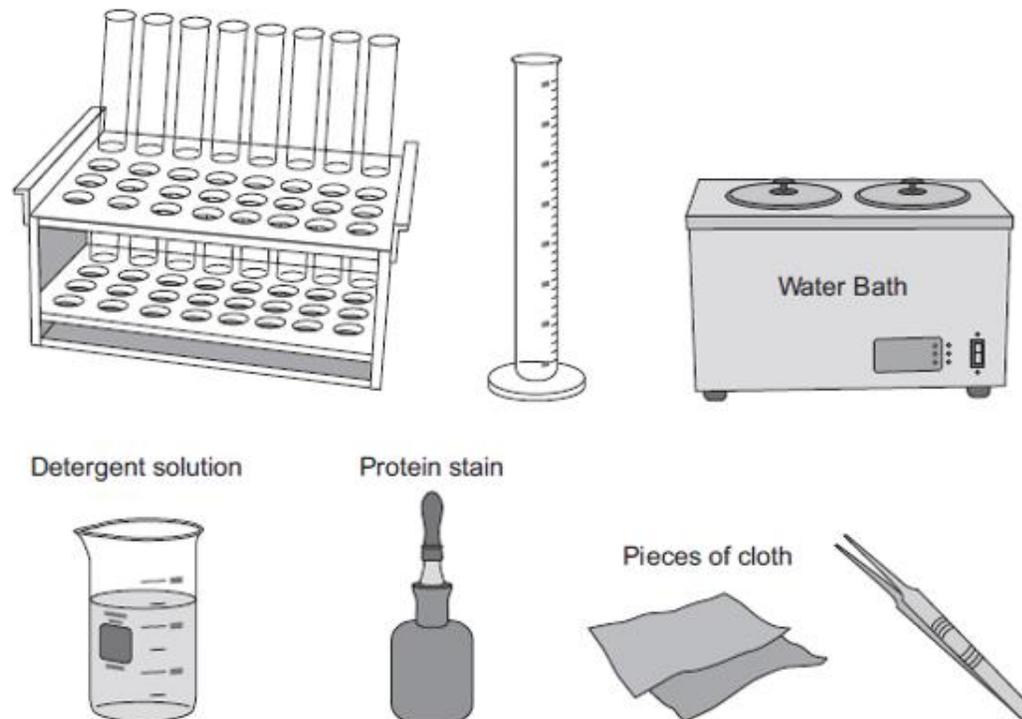
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# Your turn:

- Biological detergents contain protease enzymes.

(a) The drawings show some apparatus and materials.



Describe how you would use the apparatus and materials shown in the drawings to find the best temperature for removing stains from clothing.

You should include how you would make the investigation a fair test.

# Answers:

- 2 Phenolphthalein is an indicator. It is pink in alkaline solutions and turns colourless as the pH decreases.

It can be used to measure the activity of the enzyme lipase on the breakdown of lipids.

Samples of milk containing phenolphthalein were incubated with lipase at different temperatures.

The time taken for the phenolphthalein to turn colourless was recorded and used to calculate the rate of enzyme activity.

Figure 10 shows these results.

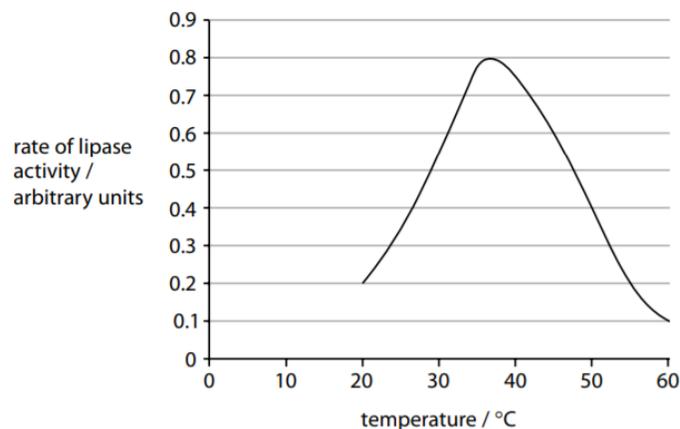


Figure 10

- (a) (i) Explain why phenolphthalein turns colourless when lipase breaks down the lipids in milk.

Fatty acids are formed when lipids are broken down by lipase. Fatty acids are acidic so the pH decreases.

- (ii) Describe the effect of temperature on the activity of lipase, as shown in Figure 10.

(2)

As the temperature increases from 20°C to 37 °C the rate of lipase activity increases. The rate of lipase activity is optimal at 37°C. Above 37°C the rate of lipase activity decreases.

- (iii) Explain why the activity of lipase changes above a temperature of 40°C.

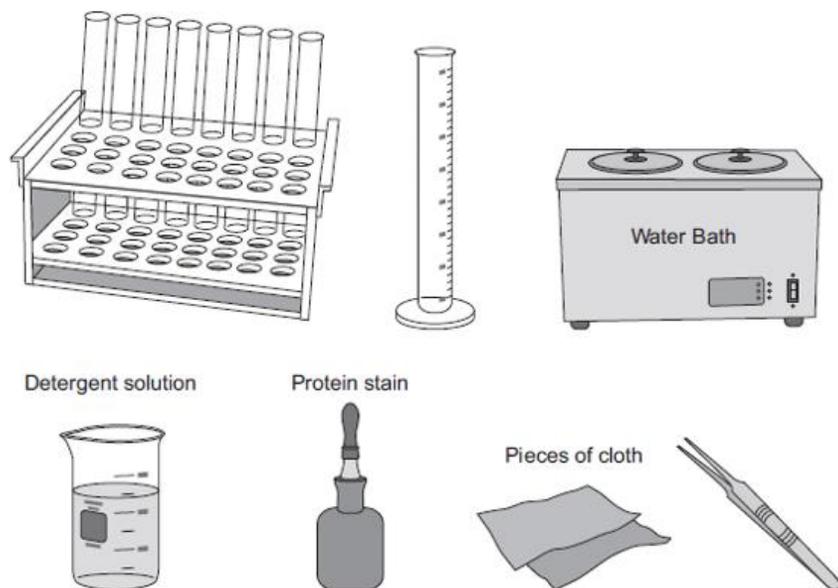
(2)

An increase above 40°C causes the active site of the enzyme to change shape. The enzyme becomes denatured and stops functioning.

# Answers:

Biological detergents contain protease enzymes.

(a) The drawings show some apparatus and materials.



Describe how you would use the apparatus and materials shown in the drawings to find the best temperature for removing stains from clothing.

You should include how you would make the investigation a fair test.

- measuring cylinder used to measure equal volumes of detergent solution
- use of dropping bottle to apply same number of drops / amount of stain to each piece of cloth
- include stainless cloth as control
- use of forceps to transfer cloths
- use of test tubes as containers for detergent solution + stained cloth
- use water bath to provide a range of temperatures
- cloths left in detergent solution at each temperature
- for same length of time or measure time taken to remove stain
- repetition
- assessing the stain removal

For more help and resources, or  
to work with us as a tutor, please  
contact us

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