

Testing milk

Hygiene and cleanliness are extremely important in the production of chocolate. In fact, it is the law that preparing, processing and packing any food are done hygienically. Strict hygiene standards are laid down for the premises, machinery and workers.

Biochemical Engineers help to design, test and apply ways of meeting these hygiene standards. The aim is to make sure that no food becomes contaminated by bacteria.

Bacteria (or other micro-organisms, such as moulds) affect the quality of food and may even lead to serious illness. They can contaminate food at any stage of production, from preparing ingredients, through processing to packing.

Imagine that it is your job to test the quality of milk before it is used to make chocolate.

your task

You are going to investigate a method of measuring the bacterial quality of milk. Read the instructions completely before beginning.

what you will need

- resazurin solution
- 1 cm³ and 10 cm³ graduated syringe or pipette and safety filler
- 4 x sterile test tubes and bungs
- test tube rack
- 4 x milk samples
- water bath at 36 °C
- Universal indicator strips
- eye protection

safety

Once the milk and resazurin solution are sealed in the test tubes, DO NOT remove the bungs. Your teacher will tell you how to tidy away the test tubes. Wear eye protection.



what you need to do

Your teacher will give you four milk samples of different ages, labelled A, B, C and D.

1. Use Universal indicator strips to find the pH of each milk sample. Record the results in a table.
2. Place the test tubes in the test tube rack. Label them A to D.
3. Measure 1 cm³ of resazurin solution into each test tube, using a syringe or pipette. (Do not pipette by mouth – use a safety filler.)
4. Measure 10 cm³ of each milk sample (A-D) into the correct test tube (A-D). Use a clean syringe or pipette for each.
5. Immediately place bungs in the test tubes. Turn the tubes upside down, then back again, three times to mix the contents.
6. If you are sharing a water bath, label your tubes, so you know which are yours.
7. Put the test tubes in the water bath and leave for half an hour.
8. After half an hour, check the colour of the resazurin in each tube.

DO NOT remove the bungs. Your teacher will tell you how to tidy away the test tubes.

results

Record your observations in a table like this, using the key to help:

milk sample	pH value	colour of solution	quality of milk
A			
B			
C			
D			

Key:	
colour of solution	quality of milk
blue (no colour change)	excellent
light blue	very good
purple	good
purple-pink	fair
light pink	poor
pink	bad
white	very bad

Place the milk samples in order, from highest to lowest quality. Does this match the pH values?

Which milk sample do you think is the oldest?

questions

- Why do you think there were differences in the quality of the milk samples?
- Suggest why bacteria might affect the pH value of the milk.
- What other factors could affect the quality of milk?
- What could be the consequences of using low quality milk?
- Share your results with the rest of the class. How do they compare?
- The test tubes were sterile, and the resazurin solution was freshly prepared using boiled water. Why are these steps important?
- What improvements, if any, could you make to your investigation?
- What further investigations could you carry out?

extension

Use secondary sources (books and Internet) to find out about hygiene in food production. What procedures are used to prevent and monitor for unwanted bacteria?

engineers

- There are four engineers shown on the poster. What other engineers do you think might be needed to help make chocolate products and other sweets?
- As well as helping to make chocolate products, where else might you find a biochemical engineer? Try to think of at least three areas of work.

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Curriculum links

England and Wales (Key Stage 3 Science Programme of Study)	
key concepts	1.1b, 1.2a
key processes	2.1a-c, 2.2a and b, 2.3a
range and content	3.3c
curriculum opportunities	4a-c
Northern Ireland (Science Statutory Requirements)	
knowledge, understanding and skills	develop: enquiry skills; critical thinking; practical skills research information learn about: organisms and health (microorganisms)
objective 1 – develop as individuals	mutual understanding: team work
objective 3 – as contributors to the economy/environment	identify skills used in: dairy and food industries economic importance and science behind: milk quality
Scotland (SQA Science Outcomes)	
third level	SCN 317P

Introducing the activity

You may wish to start by discussing what we mean by 'hygiene', and what the pupils already know about micro-organisms (or microbes). They should, at least, be able to establish that micro-organisms (microbes) are:

- small (only seen under a microscope)
- living
- germs – bacteria and viruses
- 'good' and 'bad'

Discuss how 'bad' micro-organisms (microbes) can be killed or their growth slowed down. Talk in particular about hygiene and food.

Ask the pupils to think about what steps there may be in the production of chocolate, including the collection of ingredients, such as cocoa beans and milk. This does not need to be too in-depth or accurate, but enough to establish that there are many steps and processes during which strict hygiene is essential.

You must also establish that, although hygiene standards and practices are very strict, tests and checks are still carried out to ensure the effectiveness of these measures.

Milk contains nutrients that make it a very good medium for the growth of many microbes. This is why dairy produce has to be processed carefully and has a limited shelf life. It deteriorates fast if storage conditions are not strictly controlled during delivery and, if strict hygiene procedures are not followed, bacteria would grow in the milk. Food manufacturers use the resazurin test to check incoming batches of milk before use.

Explain that pupils are going to investigate the resazurin method of measuring the bacterial quality of milk: a method which is widely used in the dairy industry.

The practical activity

It is suggested that this activity is carried out in small groups, although you may want the pupils to complete any research individually. It is your decision as to how much of the activity you need to discuss with the pupils before they are confident, and you are confident in them, of carrying out the experiment correctly and safely.

Each group should be given four milk samples of different ages, labelled A, B, C and D. The milk samples should be prepared in advance. Try using fresh milk, fresh milk after 24 hours at room temperature, fresh milk after 48 hours at room temperature (all within use-by date), and fresh milk just past its use-by date. Since pupils have to place the milk samples in order of age, it's best if you label them randomly.

By testing the bacterial content of each sample they should be able to work out which is the oldest. The pH should also indicate age – it should reduce due to bacteria converting lactose (milk sugar) into lactic acid.

For comparison, and to investigate the effect of exposure to the air, some groups could use milk that has been opened, part-used and then left for the times above. Others could use samples left unopened at room temperature.

Resazurin is a dye, which acts as a redox indicator. As microbial activity uses up available oxygen, the dye is reduced and gradually changes from blue to pink to clear. The rate of colour change, or the extent of change in a given time, is a semi-quantitative measure of the microbial activity. The quality of milk can be ascertained using the following table:

colour of solution	quality of milk
blue (no colour change)	excellent
light blue	very good
purple	good
purple-pink	fair
light pink	poor
pink	bad
white	very bad

Safety note: All glassware and bungs should be sterile. In other words, not contaminated with bacteria. Make sure pupils do not remove the bungs at the end of the experiment. Standard procedures for the safe disposal of incubated test tubes should be followed (i.e. autoclave). It is recommended that protective clothing and eye protection are worn.

Equipment

(Per group)

- 5 cm³ freshly prepared resazurin solution [**to make resazurin solution:** wearing eye protection, pour 50 cm³ distilled water into a conical flask; heat over a Bunsen flame to boiling point; remove from heat and add resazurin tablet (IRRITANT SOLID); put bung in conical flask and swirl gently until tablet has completely dissolved]
Resazurin tablets are available from education suppliers, such as Phillip Harris and Scientific and Chemical.
- 1 cm³ and 10 cm³ graduated syringe or pipette
- 4 x stoppered sterile test tubes
- method of labelling test tubes
- test tube rack
- 4 x milk samples
- water bath at 36 °C
- 4 x Universal indicator strips (pH 4-10, rather than Full Range indicator, pH 1-13)
- eye protection

Possible extension activities

- Compare the bacterial content of different types of milk (skimmed, semi-skimmed, UHT, etc.), or of milk stored in different conditions.
- Investigate whether UHT milk keeps any better than fresh milk after opening; and why or why not.
- Use secondary sources to find out about growing bacteria to make products such as yoghurt.