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6. From milk to curds and whey – which enzyme?

Time

1 h.
Extension 1 h.

Curriculum links

Use of enzymes in cheese making. Catalytic action of enzyme.

Group size

2–3.

Materials and equipment

Materials per group

- 40 cm^3 of whole pasteurised milk ('silver top')
- 1 cm^3 of rennet essence (of bovine origin, containing both chymosin and pepsin)
- 1 cm^3 of 'Maxiren' (pure chymosin) from genetically engineered yeast
- 1 cm^3 of 'Rennilase' (microbial protease) from a fungus
- 1 cm^3 of deionised water.

For the Extension

Making a sweet syrup from the whey

- 2 cm^3 of lactase enzyme (Novo 'Lactozym®')
- 8 cm^3 of a 2% (w/v) sodium alginate solution (Note: sodium alginate is not readily soluble and requires both warm water and stirring to dissolve)
- 100 cm^3 of a 1.5% (w/v) calcium chloride solution, Diastix™ (available from pharmacies) or other glucose detector.

Maxiren, Rennilase and Novo Lactozym® are available from the National Centre for Biotechnology Education, Department of Microbiology, University of Reading, Whiteknights, PO Box 228, Reading RG6 2AJ. (Telephone 01734 873743.)

Equipment per group

- 10 cm^3 plastic syringes without needles
- 4 boiling tubes or sample bottles
- 4 glass rods
- thermometer

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- stop-watch
- access to water bath maintained at 37°C
- Safety glasses

For the extension

- Small piece (about 1 cm^2) of nylon gauze *eg* net curtain
- 10 cm^3 plastic syringe (without needle)
- 10 cm length of 4 mm diameter aquarium airline tubing to fit syringe
- aquarium airline tap or adjustable laboratory tubing clip (Hoffman clip)
- retort stand, boss and clamp
- 2 small beakers (*ca* 100 cm^3) or disposable plastic cups
- tea strainer.

Safety

Safety guidelines are given in the Teacher's notes of the National Dairy Council Publication.[1] (General guidance will be found in *Microbiology – An HMI guide for schools and non-advanced further education.* London: HMSO, 1985.)

Eye protection must be worn. Unnecessary contact with the enzyme or inhalation of dust from dried-up enzyme spills should be avoided. In case of spillage or contact with the eyes, rinse by flushing with water.

Risk assessment

A risk assessment must be carried out for this activity.

Commentary

This investigation is based on an problem designed by Dean Madden and John Scholar of the National Centre for Biotechnology Education for a practical workshop at the Natural History Museum, London, in May 1992.[1] On this occasion the students worked in groups of three and each group was given a box containing all the necessary apparatus. The practical details are adapted from a National Dairy Council Publication.[2] If the extension is completed this problem will fit well with 'Any glucose?'.

Procedure

The aim of this experiment is to compare the milk-clotting abilities of the three enzymes. The students should be able to design the experiment and appreciate that they may need to warm the milk to 37°C.

The formation of a precipitate may then be observed within 5 – 15 minutes. It may be seen adhering to the sides of the tubes when they are gently rocked from side to side, or clinging to a glass rod dipped into the liquid.

The activities of the three enzymes should be found to vary markedly.

Extension

You can extend the problem by investigating the activity of the enzymes at different temperatures and at different pH values.

Making a sweet syrup from the whey

Whey contains lactose which can be split up by the enzyme lactase to the simpler

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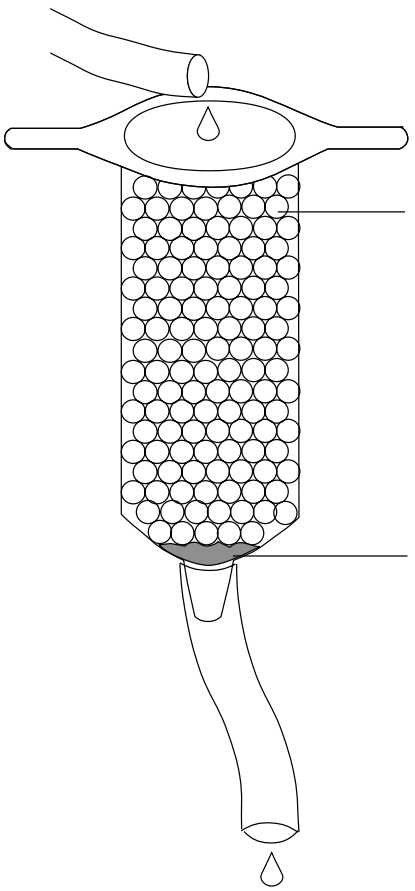
sugars glucose and galactose. These sugars are sweet and are the basis of a syrup that is used widely in the food industry. An interesting extension is therefore to ask the students to make this sweet syrup, containing glucose, from the whey by using an immobilised enzyme column (see below).

Practical details

The enzyme lactase converts the whey to a sweet syrup and this enzyme can be immobilised by trapping it in calcium alginate beads. The beads are packed into a small column, over which the whey is passed. Instructions for setting up the column and treating the whey are given below.

Immobilising the lactase enzyme and packing the column[3]

1. Draw up 2 cm^3 of enzyme in a syringe and transfer to a small beaker.
2. Using the same syringe, transfer 8 cm^3 of sodium alginate solution to the beaker and mix.
3. Draw the mixed solution into the syringe.
4. Pressing the plunger gently, add the mixture to the calcium chloride solution drop by drop.
5. Strain the beads.
6. Rinse with distilled water.
7. Pack the beads into the column.



Note: Put a small piece of gauze at the bottom of the syringe to stop the beads from blocking it.

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Esso

8. Treat the whey by pouring it through the column. The syrup coming out of the syringe should be found to contain glucose.

Tips

The tip of the syringe must not be allowed to come into contact with the calcium chloride solution. The immobilised enzyme beads should be allowed to harden for a few minutes before being separated from the liquid with a tea-strainer.

References

1. D. Madden, *National centre for biotechnology education Newsletter*, p12, Spring 1992.
2. *Dairy Biotechnology*, National Dairy Council 5/7 John Princes Street, London W1M 0AP.
3. D. Madden, *National centre for biotechnology education Newsletter*, p22, Spring 1992.

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Dean Madden and John Schollar of the National Centre for Biotechnology gave advice on the development of this activity and the experimental procedures described were developed in their laboratories.

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▼ Find out which of the three enzymes provided – rennet, genetically engineered chymosin and a fungal enzyme – is the best for coagulating milk to make curds and whey.

Traditionally rennet essence, obtained from the stomachs of calves or adult cows, is used for this process but biotechnology has made other enzymes available. The curds are used to make cheese, which is an enormous sector of the food industry. The whey can be treated to produce a sweet syrup that is used in preparing many different sorts of food.