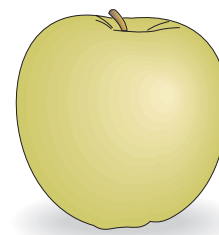


more juice from apples

Enzymatic juice extraction from apples was introduced 30 years ago and today some 5 million tons of apples are processed into juice annually throughout the world. Try this investigation with one or more different enzyme preparations, or different types of fruit.



To enhance the yield of fruit juice from apple pulp.

Preparation

The apple pulp can be prepared beforehand or bought as tinned purée. Some tinned purée contains starch however, and may not yield much juice — or it may require the addition of other enzymes (e.g., *amylase*).

Timing

This activity takes about 50 minutes.

Materials and equipment

Needed by each person or group

- Apple or tinned apple purée, 100 g
- Pectinase, e.g., Novozymes *Pectinex*[®], 1 cm³. *Dilute with an equal volume of distilled water immediately before use.*
- OPTIONAL: Cellulase enzyme, e.g., Novozymes *Celluclast*[®], 1 cm³. *Dilute with an equal volume of distilled water immediately before use.*
- OPTIONAL (if starch-containing purée is used): Amylase, e.g., Novozymes *Termamyf*[®] or *AMG*, 1 cm³. *Dilute with an equal volume of distilled water before use (See page 15 for activity graphs).*
- Coffee filter papers, 2
- Cling film
- Knife (if pre-pulped apple is not used)
- Glass stirring rods or plastic spoons, 2
- 1 cm³ syringes, 2 (for measuring out enzyme and water)
- Filter funnels, 2
- 100 cm³ measuring cylinders, 2
- 100 cm³ beakers, 2
- Water bath or incubator, set to 40 °C
- Stopclock
- Access to a balance

Procedure

1. Chop one medium-sized apple into small pieces, roughly 5 mm x 5 mm x 5 mm. Alternatively, tinned apple purée may be used.
2. Weigh half of the apple into one beaker and half into another (put about 50 g of apple in each).
3. Add 2 cm³ of diluted pectinase enzyme to one of the beakers, and 2 cm³ of water to the other. OPTIONAL: Other enzymes may be used too, either by themselves or in combination.
4. Stir the beakers' contents with a clean glass rod or spoon.
5. Cover the beakers with cling film, then incubate them in a water bath or incubator at 40 °C for 15–20 minutes.

6. Filter the juice from the apple pieces or purée, using coffee filter papers in funnels placed in measuring cylinders.
7. Record the volume of juice obtained from both lots of apple pulp at 5-minute intervals, and plot a graph of the results.


Safety

Do not consume the fruit juice

Juice prepared in this way must *not* be consumed. The proportion of enzyme used is far greater than that employed in commercial production, and it has not been handled aseptically. **Please refer to the general enzyme safety guidelines on page 11.**

Care should also be taken when handling knives.

Further activities


1. Compare the yield of juice from different varieties of apples (or other fruits or vegetables).
2. Investigate the effects of enzyme dosage and incubation temperature on juice yield.
3. Compare the yield of juice from pulp which has or has not undergone pre-oxidation (oxidation before pressing prevents certain enzyme inhibitors from working, see page 6).
4. Does the addition of cellulase to the purée increase the yield of juice? Do pectinase and cellulase in combination further enhance yields? 

commercial juice production

Where enzymes are used in commercial production, juice is extracted as follows:

- After they have been crushed, apples are usually left for 20–30 minutes so that enzyme inhibitors in the pulp are oxidised.
- The pulp is then heated to 30 °C before pectinases are added (this compares with a temperature of 50–60 °C which is needed if enzymes are not used). Typically, 130 cm³ of enzyme are added for every ton of apples.
- Enzyme treatment takes anything from 15 minutes to 2 hours, depending upon the exact nature of the enzyme, the dosage rate, the reaction temperature and the variety of apple used. Some varieties, like *Golden Delicious*, are very difficult to break down. During incubation, the pectinases degrade soluble pectin in the pulp, making the juice flow more freely.
- Next, the apples are pressed. Yields of juice may be increased by up to 20% by enzyme treatment, depending upon the age and variety of apple used and whether pre-oxidation is employed. Pectinase treatment is particularly effective with mature apples and those from cold storage. Significant increases in yield are not usually achieved from fresh, early season fruit.

resources

Smash more apple into juice! (1999)
BioTimes March 1999, No. 1, p. 8–9.
 www.biotimes.com/biotimes9901/main.htm



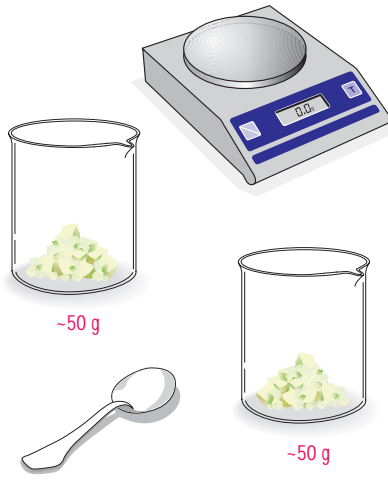
1 Roughly chop an apple into small pieces about 5 mm x 5 mm x 5 mm.



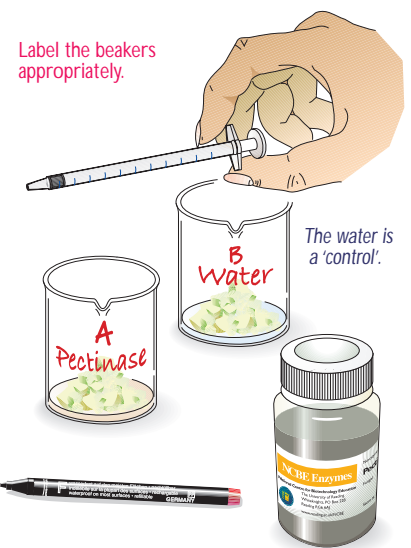
Tinned apple purée can be used instead if you are short of time.

Note: Some tinned apple purée contains starch. Make sure that you read the label.

2 Weigh equal amounts of chopped apple or purée into each of two beakers.



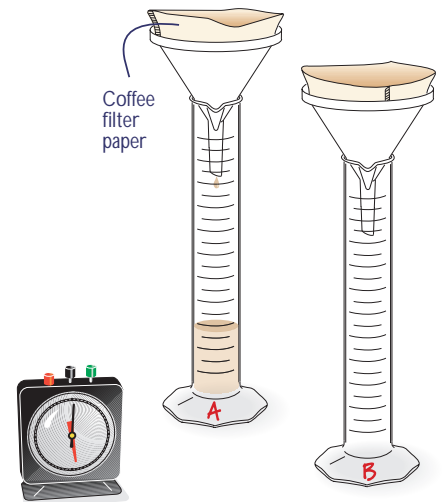
3 Add 2 cm³ of diluted pectinase enzyme to one beaker. Add 2 cm³ of distilled water to the other.



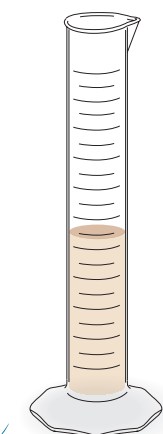
4 Stir the enzyme solution or water into each batch of fruit. Cover the beakers with cling film to stop the liquid evaporating.

5 Put both beakers in a water bath or incubator set at 40 °C for 15–20 minutes.
During this time, the enzyme will break down the fruit, helping to release the juice.

6 Filter the juice from each apple pulp preparation into a measuring cylinder.
Record the volume of juice collected at 5-minute intervals.

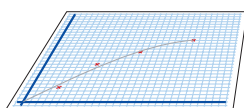


7 Plot the data as a graph, showing the total volume of juice (cm³) produced by each treatment against the time (hours).

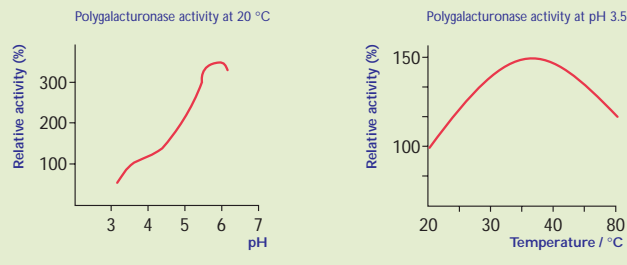


With this method, you could investigate the effect(s) of:

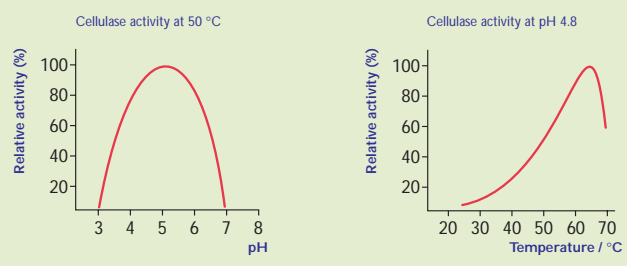
- enzyme concentration;
- the variety of apple used;
- the duration of incubation;
- the temperature of incubation;
- the effect(s) of other enzymes such as cellulase or amylase (either by themselves in combination with pectinase);
- the age of the fruit and the conditions in which it is stored before juice is extracted.



Pectinase (mixture of enzymes)



Cellulase (mixture of enzymes)



These graphs show the activity of two commercial preparations of pectinase and cellulase (Pectinex™ and Celluclast®). Both are mixtures of enzymes — the main enzyme in the pectinase preparation is polygalacturonase.

The data may help you to plan further investigations of your own.

Note: In practice, these graphs can only be used as a guide, as the enzyme activity will vary according to many factors, in addition to pH and temperature, including:

- the exact substrate used;
- the reaction time;
- the presence of inhibitors.

Data from Novozymes A/S.