Student activity: Extracting your own DNA

Materials

- Micropipettes or graduated transfer pipettes If you don't have micropipettes, you can use calibrated / graduated disposable plastic transfer pipettes. On these pipettes the 'stalk' is graduated, allowing volumes of less than 1 ml to be transferred with sufficient accuracy for this experiment.
- Disposable culture loops or buccal swabs
- Small Falcon tube or a test tube with a bung or cap Falcon tubes are calibrated test tubes with screw caps. If you don't have them, just use normal test tubes.
- Water bath at 40°C (optional)
- Disinfectant solution
- Lysis solution
- Proteinase K solution
- Sodium acetate solution
- Cold ethanol or isopropyl (rubbing) alcohol (keep in the freezer until required)

Procedure

1. Place 1 ml lysis solution in your Falcon tube or test tube.



- 2. Vigorously scrape a loop or swab around the inside of your cheeks and across your tongue.
- 3. Place the loop or swab in the lysis buffer and mix it around to dislodge your cells.



- 4. Put your loop or swab in the disinfectant.
- 5. Repeat steps 2-4 twice more to ensure you get plenty of cells. **Use a new loop or swab each time**.
- 6. Add 20 μ l (or 1 drop if you are using transfer pipettes) of proteinase K to your tube.

7. Cap the tube and invert it a couple of times to mix.



- 8. Incubate the mixture in the water bath or at room temperature for 10 min.
- 9. Add 100 μ l sodium acetate.





- 10. Cap your tube and shake well to mix.
- 11. Add 3 ml cold ethanol.
- 12. Cap the tube and invert it very slowly to mix.
- 13. Your DNA should appear as a whitish stringy precipitate.

Teaching activity

Safety note:

The solutions can irritate eyes and skin, so wear a lab coat, safety glasses and gloves. Saliva can carry diseases; only handle your own loops or swabs and put used items in the disinfectant.

Disposal: liquids can be poured down the sink with plenty of water. Used loops or swabs can be placed in normal waste after disinfecting for 15 minutes.

Questions for discussion

- What does 'lysis' mean? How does this help extract the DNA?
- The lysis buffer contains a detergent called SDS. Using your knowledge of cell structure, what do you think the detergent does?
- Inside cells, DNA is found tightly coiled up and bound to a variety of proteins.
 Which step helps to release the DNA from the proteins?
- What does the last step tell you about the solubility of DNA in both salty water and ethanol?
- How could you confirm that the white precipitate really is DNA?

Extension activities

- Compare this method of extracting DNA with the simpler methods using frozen peas (Madden, 2006) or kiwi fruit^{w2}. How do they differ? Which one works best? Can you explain why? Can you find out which method is closest to the method that professional geneticists use?
- Simply extracting someone's DNA is not enough to tell if they have a predisposition for obesity. What other tests would have to be done? Find out more about the techniques used in genetic research.
- In many countries, parents who carry serious genetic conditions like cystic fibrosis or haemophilia can opt for pre-implantation genetic diagnostics to avoid having children that carry the disease. Do you think this procedure should be available for parents who have a genetic predisposition for obesity? Do your classmates agree with you?

Sourcing and preparing the reagents

Lysis solution (50 ml)

- 1. Prepare tris-buffered saline (TBS) according to manufacturer's instructions or standard recipe.
 - TBS can be purchased as a ready-made solution, in tablet form or made up from scratch^{w4}.

Safety note: a ready-made sodium dodecyl sulphate (SDS) solution is recommended as powdered SDS is harmful if inhaled. If powdered SDS is used, the teacher should prepare the solution, wearing a mask and using the fume hood.

See also the general Science in School safety note on page 57.

2. If using ready-made 10 % SDS solution, add 5 ml SDS to 45 ml TBS.

If using powdered SDS, dissolve 0.5 g in 50 ml TBS.

3. Store in the fridge until required.

3 M Sodium acetate solution (for 50 ml)

- 1. Dissolve 12.3 g anhydrous sodium acetate in 50 ml distilled water.
- 2. Add dilute HCl to adjust to pH 5.2.
- 3. Store in the fridge until required.

Proteinase K (100 µg / ml)

- 1. Dissolve 1 mg proteinase K in 10 ml tris-buffered saline.
- 2. Only a very small amount of the enzyme is required, so you might want to make up a smaller volume if you have a sufficiently accurate balance. Simply adjust the quantities accordingly.
- 3. Store in the freezer until required.

Disinfectant solution

BACKGROUND

Suitable disinfectants include 0.015 M sodium hypochlorite solution, 1 % Virkon[®] solution or 5 % domestic bleach. After soaking for at least 15 min, the loops can be transferred to a plastic bag (wear gloves) and disposed of with normal waste. To help you find the necessary reagents, a list of product num-

bers for Sigma-Aldrich is given in table 2. However, you will also be able to source them from other suppliers.

Reagent	Sigma-Aldrich product number
Tris buffered saline (TBS)	Tablets: TS030 or 94158
Sodium dodecyl sulphate (SDS)	Powdered: L3771 10% solution: 71736
Sodium acetate	S2889
Proteinase K	P6556

 Table 2: Sigma-Aldrich product numbers for the reagents used in this protocol