

## Introduction

Many pupils are familiar with the technique of paper chromatography in its various forms either with inks or food dyes, but few are aware of column chromatography. The idea for trying out this experiment came about as the result of a conversation with an Edinburgh chemistry teacher (thanks Fiona).

This method uses a length of soda glass tubing approximately 20 cm long, or a glass Pasteur pipette as the column. Both columns give very good separation.

The stationary phase is starch and the mobile phase is water. The column is easily packed with a slurry of starch powder in distilled water in a few minutes.

This technique could be used to support learning outcomes **SCN 2-16a** - *I have participated in practical activities to separate simple mixtures of substances and can relate my findings to my everyday experience* and **SCN 3-16a** - *I can differentiate between pure substances and mixtures in common use and can select appropriate physical methods for separating mixtures into their components.*

The ability to show separate dyes in separate containers can show how this technique can recover the constituent parts – a task which is a little more difficult with paper chromatography.

## Chemicals

- soluble starch powder
- distilled water
- dry fine sand
- food colouring (ensure there is a mixture of colours in it as indicated by different E numbers)

## Equipment

- clamp stand, bosshead and clamp
- syringe, 10 cm<sup>3</sup>
- glass tubing, inside diameter 5 mm, approx. 20 cm long
- plastic tubing approximately 20 cm long of internal diameter to fit the syringe and glass tubing
- spatula
- beakers, 100 cm<sup>3</sup>, x3
- pasteur pipettes, x3
- stirring rod
- cotton wool

## Preparing the column

Prepare a slurry of starch in one of the beakers by adding approximately 4 cm<sup>3</sup> of distilled water to 3 g of the starch powder.

Insert a loose plug of cotton wool in one end of the column. This should be tight enough to hold the stationary medium in the tube but loose enough to allow liquid to pass through it easily.

Clamp the column vertically in the clamp stand and place a beaker under it.

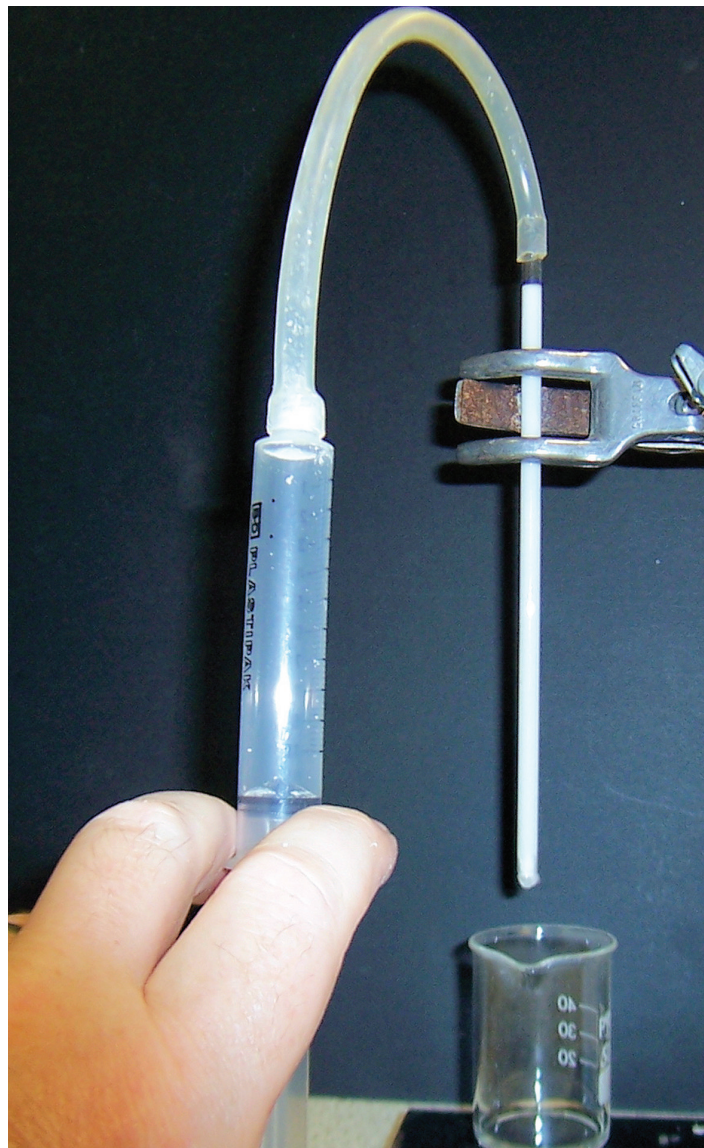


Figure 1 - Packing the column

Using a pipette, carefully fill the column with the slurry until it is 1 cm from the top.

Attach the short plastic tubing to the end of the syringe and ensure the plunger is withdrawn.

Attach the other end of the tubing to the top of the column and slowly and gently push in the plunger. The increase in pressure in the column will help to pack the starch and you will see drops of water drain into the beaker. Hold the plunger in for a few minutes until the air pressure in the column reduces. If you release the plunger too quickly the air rushing back into the syringe from the column can disrupt the packing of the stationary starch phase which can lead to uneven separation. Repeat the pressurising 2-3 times more until the column is packed but still damp.

You may also have to add more slurry until the starch is about 2-3 cm from the top when finished. (Figure 1)

### Method

The syringe and plastic tubing is used to introduce air into the column to aid the separation of any dyes in the food colouring. Because pressure is used in this way, the colours separate very quickly within minutes and the technique can be called *Flash Chromatography*. Since the system is not sealed, the increase in pressure is never great enough to cause the glass to crack.

Use the second pipette to introduce one drop of the food colouring to the column. Try to pour this carefully down the side of the glass so as not to disturb the starch.

With the plunger withdrawn and the plastic tubing attached to the column top, gradually press in the plunger until you feel some resistance and the dye is forced into the stationary phase. Note: It is not necessary to force the plunger completely in.

Hold the plunger in for a few minutes. When the pressure on the plunger is released, it should remain more or less in position.

Add a few millimetres of the sand to the top of the column.

Remove the tubing from the column and fill it to the top with distilled water (the eluent). Again, try to trickle this down the side of the glass onto the sand so as not to disturb the layer.

Reattach the tubing and pressurise as before until the water has been forced into the stationary phase.

Repeat with further additions of water as required.

We used Super Cook black food colouring from Tesco® which according to the label comprised three separate dyes:

- E102 – (Tartrazine, a yellow coloured dye),
- E122 – (Carmoisine, a reddish coloured dye) and
- E143 – (Green S).

If desired, the addition of more eluent and the pressurisation can be repeated until the colours have been forced through the entire column (Figures 3a & 3b)

After discarding any water which has been forced through, continued application of pressure enables the coloured dyes to be collected in separate small sample tubes. The addition of a few cm<sup>3</sup> of distilled water to these makes the colours more obvious.

If a few cm<sup>3</sup> of each of the dyes are recombined in another small sample tube, the original black colour is reformed. (Figure 4)

The technique can be repeated with other coloured food dyes such as green and blue, which are a mixture of other colours.

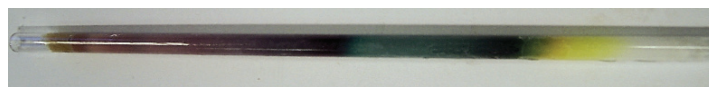


Figure 2 – The dye started to separate into three distinct bands of yellow, green and red/purple

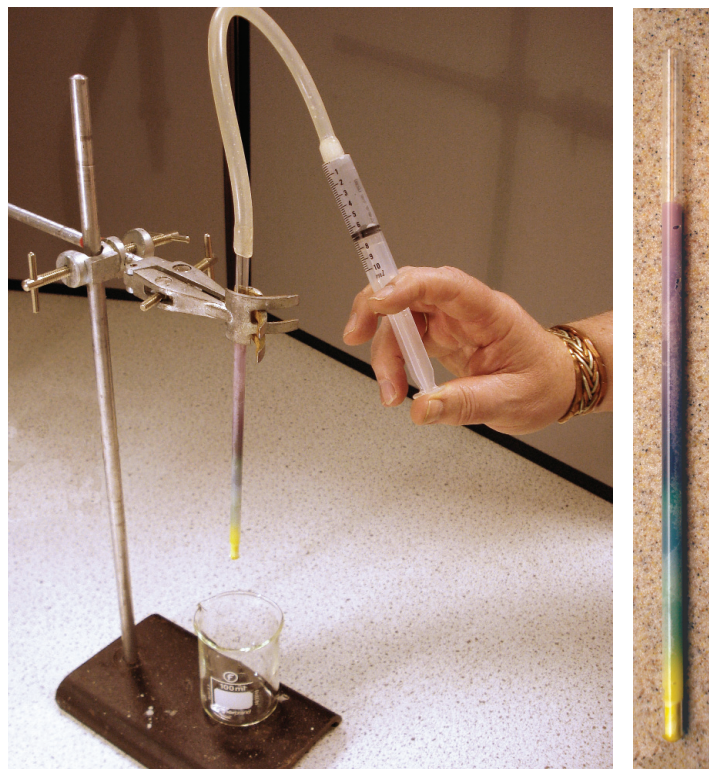


Figure 3a & 3b

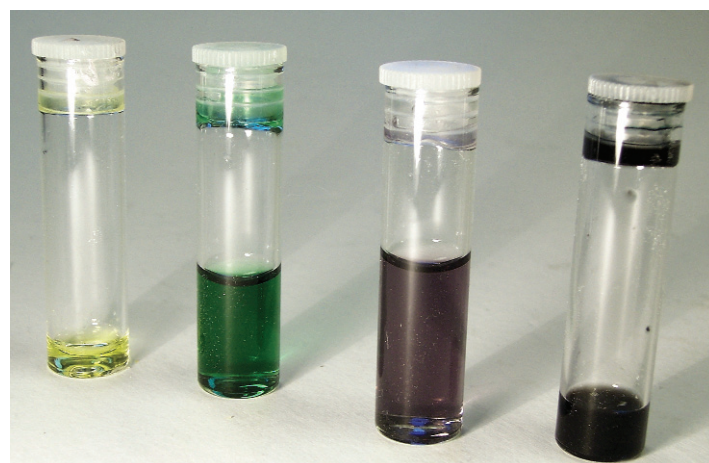


Figure 4  
 yellow      green      red/purple      black