14–16 years

Leaf chromatography

ROYAL SOCIETY OF CHEMISTRY

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Introduction

Most leaves are green due to the chemical **chlorophyll**. This substance is important in photosynthesis (the process by which plants make their food).

In this experiment, you will investigate the different pigments present in a leaf, from chlorophyll to carotenes, using a technique called **paper chromatography**.

Introduction

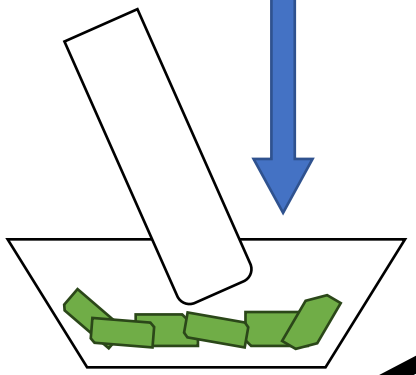
Chromatography has many real-world applications. These include testing the purity of foods and medicines, identifying the use of performance enhancing drugs during sports competitions, and testing for traces of explosives and accelerants from fires.

Learning objectives

1. Carry out paper chromatography to find the pigments present in leaves and calculate their R_f values.
2. Identify stationary and mobile phases in paper chromatography.
3. Describe what the chromatogram can tell us about substances.
4. Explain why water is not always used as the solvent in chromatography.

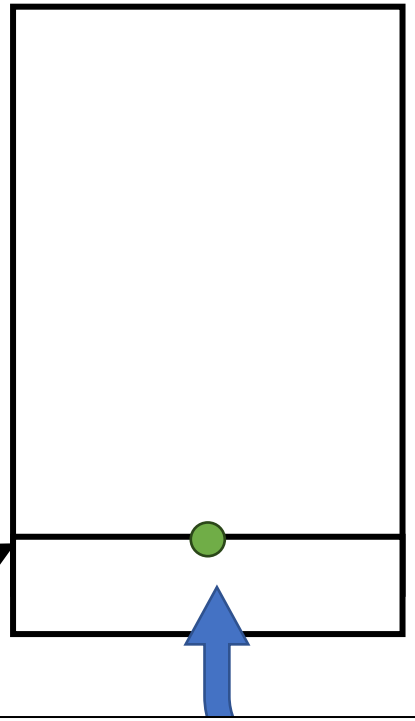
❶Cut-up leaves ❑

❷Add:
- leaves ❑
- sand ❑
- six drops of propanone ❑



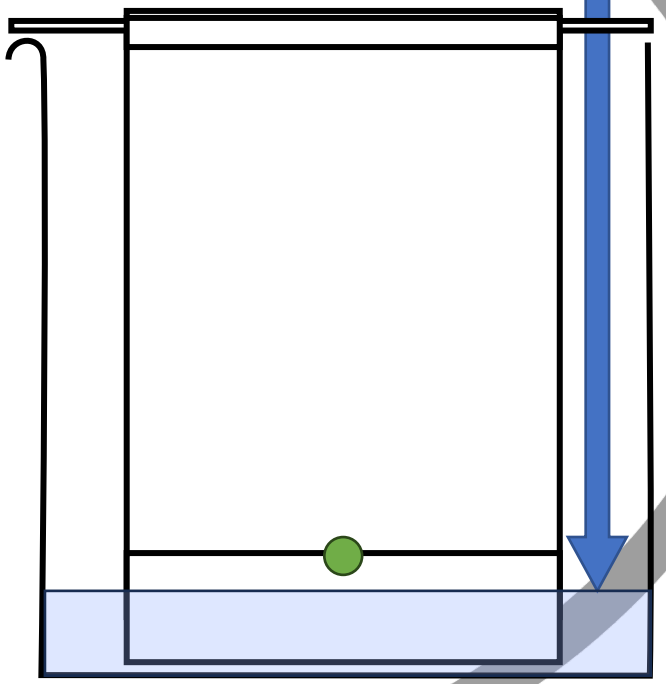
❸Grind for **three minutes** ❑

❹ Draw a line ❑



❺ Spot of leaf extract. Let dry. Repeat **five** times ❑ ❑ ❑ ❑ ❑

❼ Secure chromatography paper ❑



❽ Max 10 cm^3 of propanone. ❑

❾ Remove the chromatogram ❑

❿ Mark solvent front. ❑

Nuffield practical collection
Leaf chromatography
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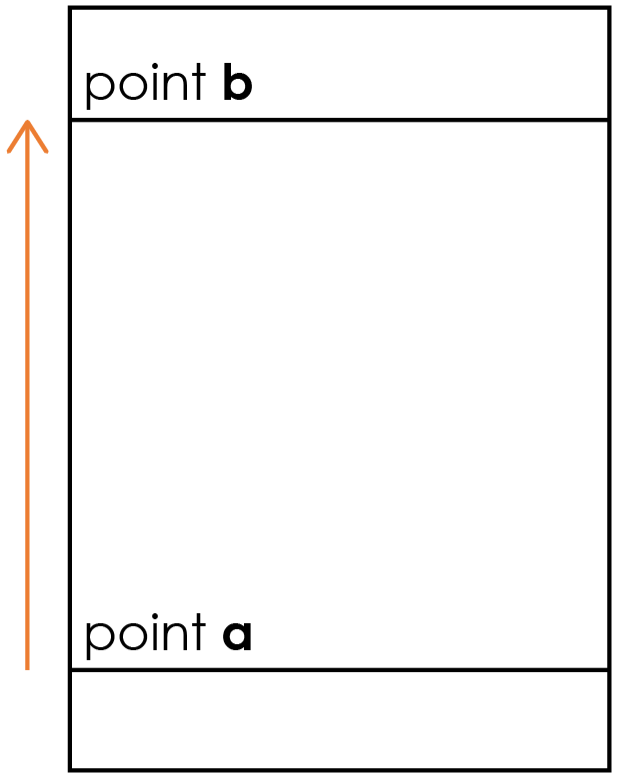
Results



Distance travelled by sample = _____ cm

(= distance between point **a** and the **centre of the spot**)

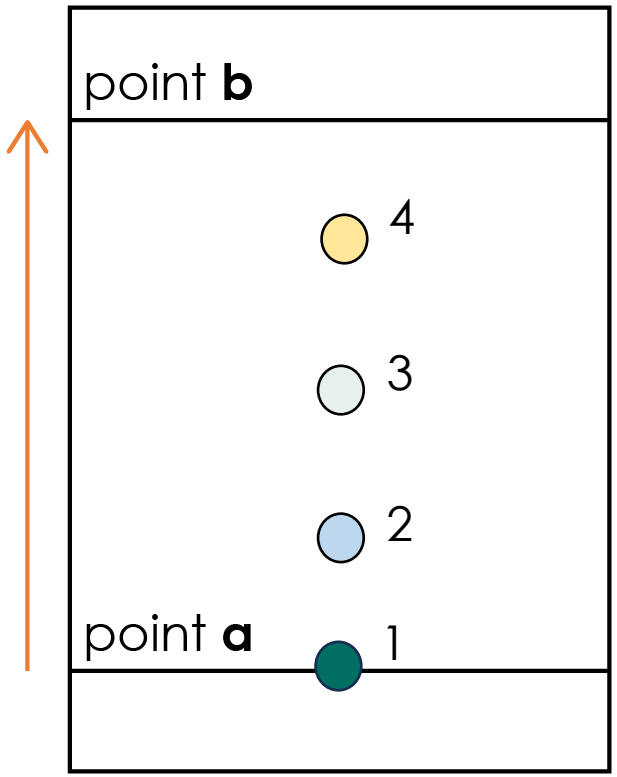
If there are multiple sample spots, calculate the distance for each spot separately

Spots on leaf sample	Distance travelled (cm)		R_f
	By spot	By solvent	

Distance travelled by solvent = _____ cm
(= distance between point **a** and point **b**)

$$R_f = \frac{\text{distance moved by spot}}{\text{distance moved by solvent}}$$

Example Results



Distance travelled by sample = _____ cm

(= distance between point **a** and the **centre of the spot**).

If there are multiple sample spots, calculate the distance for each spot separately.

Spots on leaf sample	Distance travelled (cm)		R_f
	By spot	By solvent	
1	0.0	5.0	0.00
2	1.6	5.0	
3	2.5	5.0	
4	4.5	5.0	

Distance travelled by solvent = **5.0** cm

(= distance between point **a** and point **b**)

$$R_f = \frac{\text{distance moved by spot}}{\text{distance moved by solvent}}$$

Questions

1. State the stationary and mobile phase in this chromatography experiment.
2. Explain why water was not used as the solvent in this chromatography experiment.
3. Explain why the markings on the chromatography paper must be drawn in pencil (and not in pen).
4. State and explain which of the chemicals (using the spot numbers) within the leaf pigment was the most soluble in the solvent.
5. Using your chromatogram, is the pigment found within the dye a **pure substance** or a **mixture**? Explain how you can tell.
6. Look at the example data provided by your teacher.

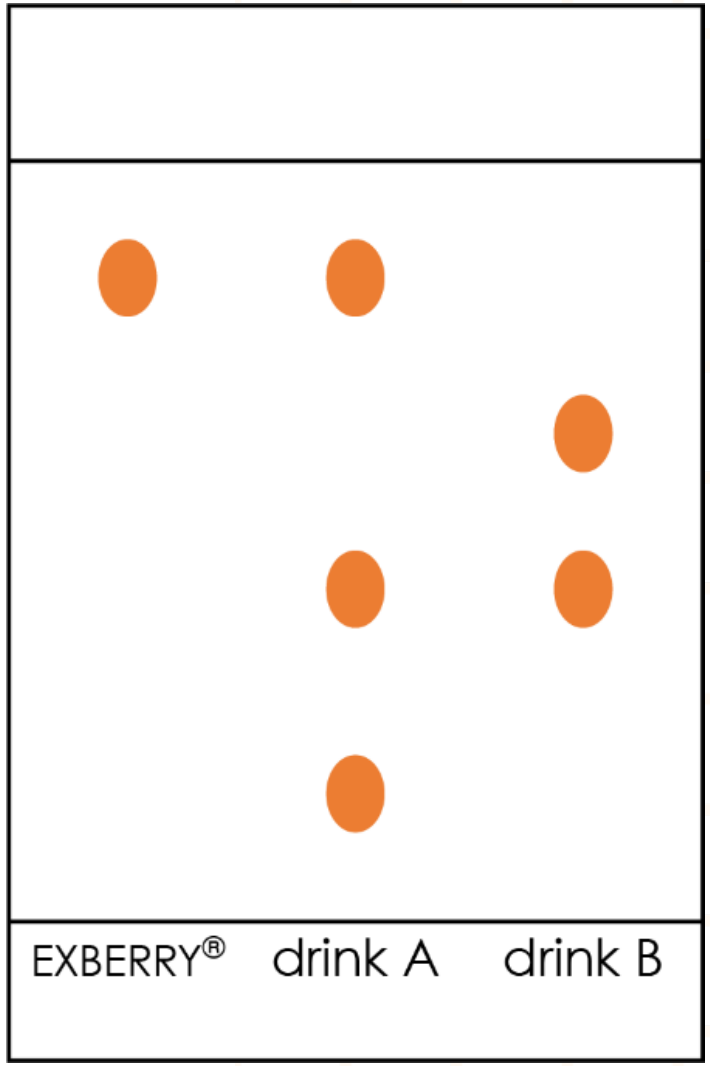
   a) Why did spot 1 not travel up the chromatography paper?
   b) Why did spots 2, 3 and 4 travel different distances up the chromatography paper?

Unscaffolded questions

Questions

7. Carotenes are natural pigments produced by plants, algae and some bacteria, fungi and archaea. EXBERRY®, a brand of plant-based carotene food colouring extracted from saltwater algae *Dunaliella salina*, is used in a number of food and drink items. A student wanted to investigate whether this food colouring is used in two different soft drinks, A and B. The results are shown on the chromatogram.

Use the chromatogram to draw **two** conclusions about the colourings used in drinks A and B.



Unscaffolded questions

[Carotene Food Coloring | EXBERRY®]

Questions

1. State the stationary (the phase which **does not move**) and mobile phase (the phase that **moves**).
2. Explain why water was **not** used as the solvent in this chromatography experiment. You should refer to **solubility** in your answer.
3. Explain why the markings on the chromatography paper must be drawn in pencil (and not in pen). You should refer to **solubility** in your answer.
4. Which of your spots travelled the greatest distance during the chromatography experiment? Is this the **most** or **least** soluble pigment?
5. Using your chromatogram, is the pigment found within the dye a **pure substance** or a **mixture**? Explain how you can tell using the number of spots produced by each pigment.

Scaffolded questions

Questions

6. Look at the example data provided by your teacher.

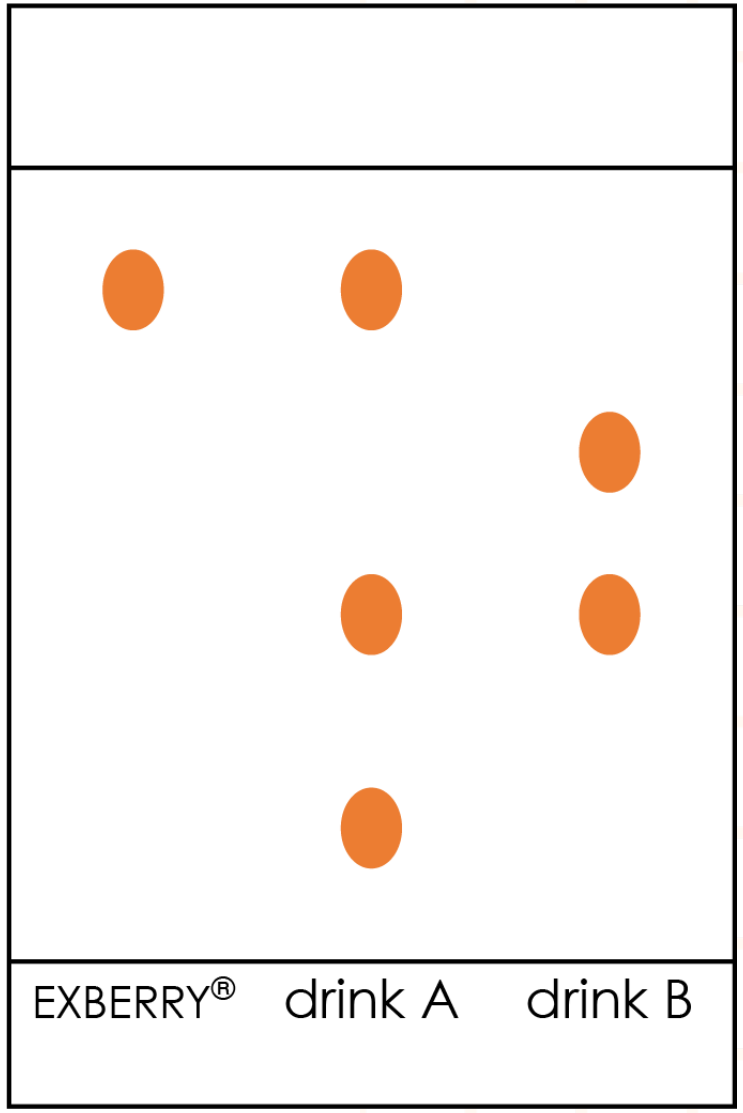
a) Why did spot 1 not travel up the chromatography paper? You should refer to **solubility** in your answer.

b) Why did spots 2, 3 and 4 travel different distances up the chromatography paper? You should refer to **solubility** in the solvent and attraction towards the chromatography paper in your answer.

Scaffolded questions

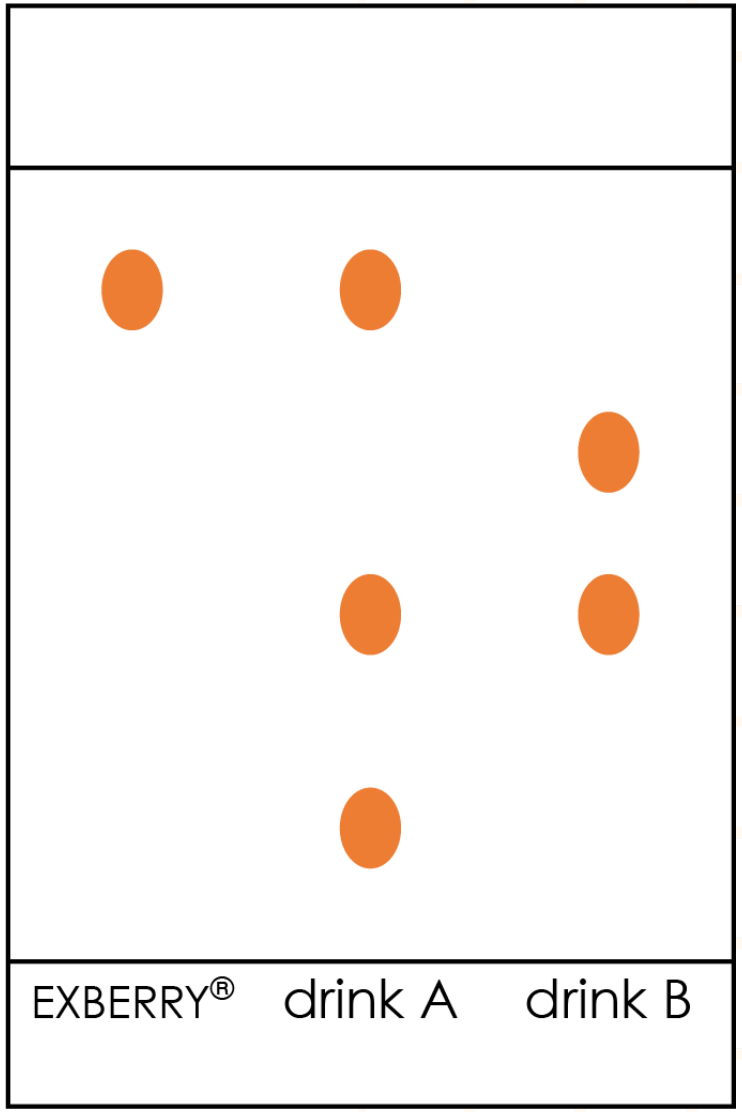
Questions

7. Carotenes are natural pigments produced by plants, algae and some bacteria, fungi and archaea. EXBERRY®, a brand of plant-based carotene food colouring extracted from saltwater algae *Dunaliella salina*, is used in a number of food and drink items. A student wanted to investigate whether this food colouring is used in two different soft drinks, A and B. The results are shown on the chromatogram.



Questions

a) Which of the two drinks, A or B, contains EXBERRY® colourings?
b) Which of the two drinks, A or B, contains the most colourings?
c) Do drinks A and B contain any common ingredients? Explain how you can tell using the chromatogram.
d) Which of the colourings was most soluble in the solvent used?



Scaffolded questions

Answers

1. Stationary phase = chromatography paper
   Mobile phase = propanone
2. Water is not used in this experiment as the dyes are **not soluble** in water, and would therefore not move up the chromatography paper.
3. Pencil must be used for all markings as it is **insoluble** in the solvent. Pen is soluble, and would also travel up the chromatography paper and interfere with results.
4. View alongside student results.
   Students should identify the pigment which has travelled the greatest distance – this is the **most soluble** pigment.
5. View alongside student results.
   If **more than one spot** is produced, the pigment is a mixture. Pure substances produce only one spot during chromatography.

Answers

6.

a) Spot 1 did not travel up the chromatography paper because it was insoluble in the solvent used.

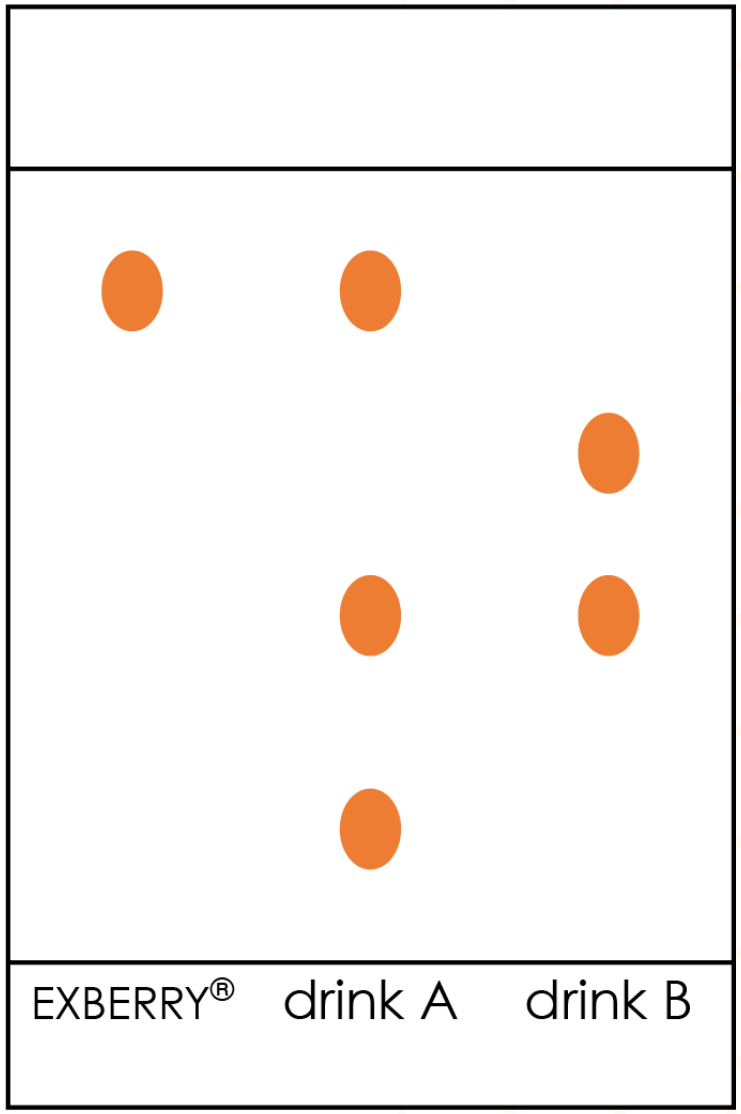
b) Spots 2, 3 and 4 travel different distances because they have different solubilities in the solvent and different attractions for the paper.
Because Spot 4 travelled the furthest, it has the highest solubility and the lowest attraction to the paper.
Spot 2 has the lowest solubility (apart from spot 1, which is insoluble) and the highest attraction to the paper.

Answers

7. Unscaffolded

Any two conclusions from:

- Drink A contains EXBERRY® but Drink B does not.
- Drink A contains three different colourings, one of which is EXBERRY®.
- Drink B contains two different colourings.
- EXBERRY® was the most soluble colouring.
- Drinks A and B contain only one common ingredient.



Answers

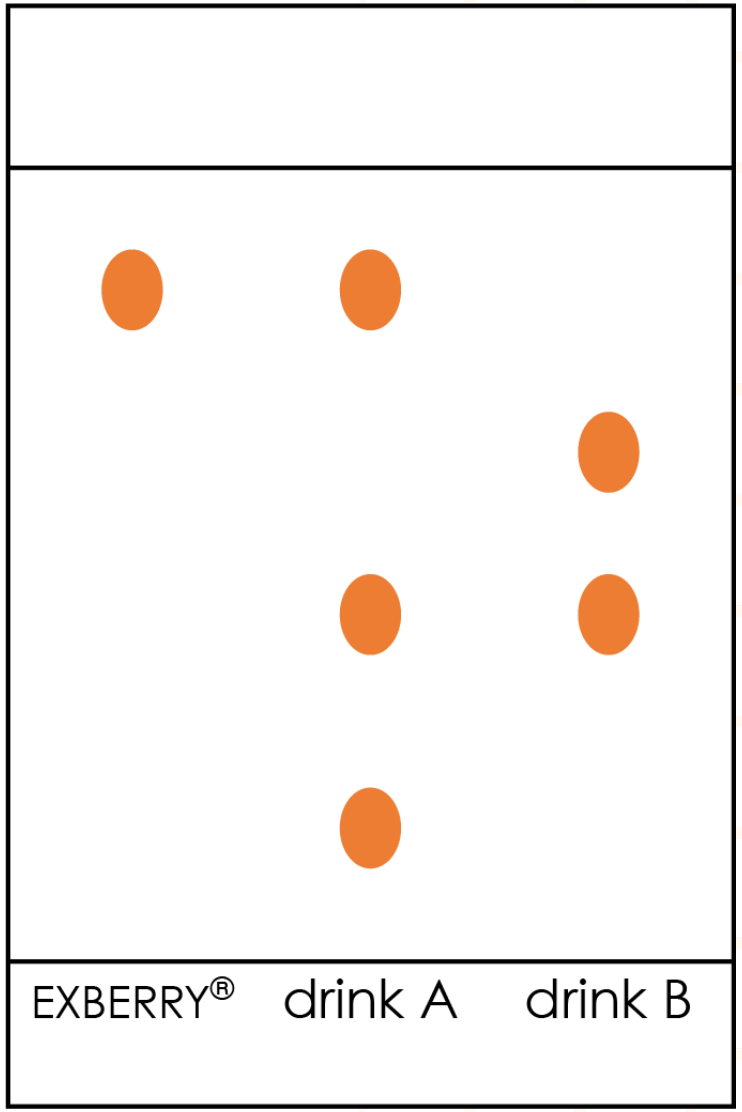
7. Scaffolded

a) Only Drink A contains EXBERRY® colouring.

b) Drink A contains the most colourings – three spots are produced.

c) Drinks A and B contain only one common ingredient. This is **not** EXBERRY®. They produced a spot with a common R_f value.

d) EXBERRY® was the most soluble colouring in the solvent.



Acknowledgements

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