Supporting Information

Colourful chemistry of anthocyanins: A tutorial review of applications of anthocyanins in school and university teaching

Iain A. Smellie,*[a] Isobel Everest[b] and Iain L. J. Patterson[a]

[a]EaStCHEM, School of Chemistry, University of St Andrews, North Haugh, St Andrews, KY16 9ST, UK.
[b]Bedford School, De Parys Ave, Bedford MK40 2TU, UK.

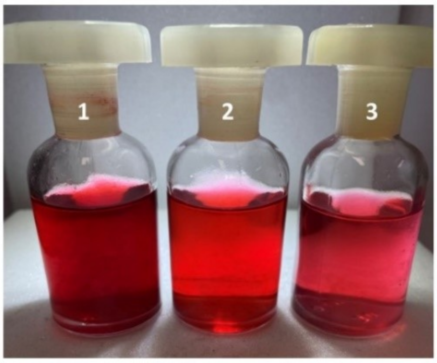
Supporting information for the example activities: Contents

A Red cabbage indicator experiment inspired by James Watt

Red cabbage indicator is usually made by soaking chopped red cabbage in hot water, and then filtering off the vegetable matter. This process is easy to do and the resulting indicator works well. However, samples stored for more than a day or so acquire a disagreeable odour. Watt overcame this by soaking chopped red cabbage in dilute sulfuric acid solutions, the resulting red extracts can be stored for over 12 months (in a cool location, not exposed to direct sunlight) without significant decomposition being detected.

- **Sulfuric acid (CAS 7664-93-9). This will be diluted further with water to make a 0.4 M solution.**
- **Calcium carbonate (CAS 471-34-1)**
- **Sodium hydrogen carbonate (CAS 144-55-8)**
- **Filter funnel**
- **Filter paper**
- **Chopped red cabbage**
- **Assorted beakers**

To prepare the extract, a red cabbage is finely chopped and soaked in dilute solutions of sulfuric acid (the cabbage is placed in a beaker and immersed in dilute acid). We have found that extracts made using 0.4 M sulfuric acid are stable enough for long term storage (Figure S1). The cabbage should be chopped roughly then left steeping overnight in twice its mass of 0.4 M sulfuric acid (22 mL 96% H_2SO_4 per litre), or can be heated for an hour or two if it is needed sooner. Allow to cool if it has been heated, then strain through a plastic sieve. A typical red cabbage usually will give 2-3 litres of strongly coloured extract. The residual acidic cabbage may be left to steep in a bucket of water then neutralised using sodium hydrogen carbonate prior to disposal. Diluted with 1 to 3 volumes of water, the extract solutions work well for the demonstration described below.



1 = Red cabbage extract diluted 1:1 with water, 0.2 M H_2SO_4
2 = Red cabbage extract diluted 1:3 with water, 0.1 M H_2SO_4
3 = Red cabbage extract diluted 1:7 with water, 0.05 M H_2SO_4

Figure S1: Solutions of red cabbage extract prepared from dilute sulfuric acid.

Demonstration procedure

$$CaCO_3 + H_2SO_4 + H_2O \rightarrow CaSO_4.2H_2O + CO_2$$

and

$$\text{flavylium}^+ \underset{+H^+}{\overset{-H^+}{\rightleftharpoons}} \text{quinoidal} \underset{+H^+}{\overset{-H^+}{\rightleftharpoons}} \text{quinoidal}^-$$

20-30 mL of red cabbage extract (in dilute sulfuric acid) is poured into a beaker. Calcium carbonate is then added and mixed until the solution turns purple/blue. The reaction produces carbon dioxide gas and the extract contains natural saponins, so the solid should be added gradually to control frothing. Calcium sulfate will precipitate during the reaction, and is readily removed by filtering through paper or simply decanting the solution if desired (see Figure S2). The neutralised solution decomposes overnight at room temperature. Watt mentioned that ammonia or the “fixed” (i.e. permanent rather than volatile) alkalis can also be used, though care must be taken not to add an excess otherwise the mixture turns green and becomes useless. Even the “chalk or whiting” of the time could turn the extract green. Modern $CaCO_3$ doesn’t seem to have the same effect. We have left the neutralised extract in contact with excess $CaCO_3$ for hours and the solution remained blue. The green colour is due to a mixture of blue and yellow components. Likely due to its aqueous insolubility $CaCO_3$ (in our hands) doesn’t produce the yellow material so is ideal for obtaining the blue stage, which can be missed when using strong alkalis or ammonia. If neutralising the extract to use in pH experiments, it may be best to stop at the purple stage. In any case, due to its insolubility, it is hard to overshoot when neutralising with $CaCO_3$.

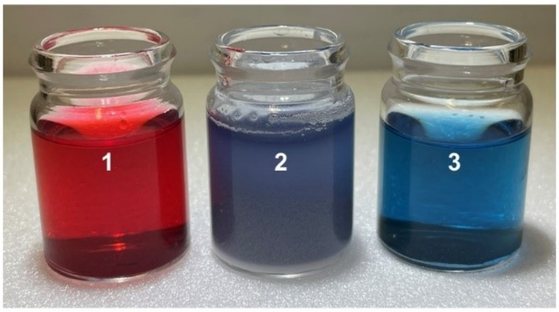


Figure S2: Red cabbage extract demonstration. Red cabbage in dilute sulfuric acid (1), red cabbage extract treated with calcium carbonate (2) and red cabbage extract treated with calcium carbonate after filtration (3).

Safety considerations

A sharp knife will be required to chop the cabbage, care should be taken to avoid injury.

The sulfuric acid extracts are dilute, but should be considered as corrosive to skin and eyes, it is recommended that safety glasses and a lab coat are worn. Take care when using concentrated sulfuric acid, always pour a slow stream of acid into rapidly stirred water.

The Safety Data Sheets (SDS) From the manufacturers of chemicals should be consulted prior to making a risk assessment. The chemical supplier for this activity was Merck Life Science UK Limited (Dorset UK), the key points from the associated safety data sheets are shown in Table 1. Educators in UK schools may wish to access chemical safety information provided by the Consortium of Local Education Authorities for the Provision of Science Services (CLEAPSS**, https://science.cleapss.org.uk/).**

Table 1: Safety data for chemicals required for preparation of James Watt's red cabbage indicator.

Substance	**CAS Number**	**Hazard Codes**	**Warnings**	**GHS Pictogram**
0.4 M Sulfuric acid[1]	**CAS 7664-93-9**	**H290**	**May be corrosive to metals**	
Calcium carbonate[2]	**CAS 471-34-1**	**No hazard code provided**	**No hazard statements provided**	**No pictogram provided**
Sodium hydrogen[2] carbonate	**CAS 144-55-8**	**No hazard code provided**	**No hazard statements provided**	**No pictogram provided**

1. **5-15% Solutions are considered eye and skin irritants (0.4 M H_2SO_4 is <5%).**
2. **Calcium carbonate and sodium hydrogen carbonate are** not considered to be hazardous substances according to Regulation (EC) No. 1272/2008.

Waste disposal

The aqueous waste from the demonstration should be diluted with water and presents a low hazard.

Calcium sulfate (CAS 10101-41-4) is recovered by filtration, this material is not considered to be a hazardous substance according to Regulation (EC) No. 1272/2008.

Aqueous and solid waste should be collected in appropriate containers for proper disposal (in line with local legislation).

Colourful pH and metal ion binding experiments for droplet art

To prepare the droplet experiments, the following chemicals and materials are required:

- 1 M Hydrochloric acid (CAS 7647-01-0)
- 1 M Ammonium acetate solution (CAS 631-61-8)
- Saturated sodium hydrogen carbonate solution (CAS 144-55-8)
- 1 M or 0.5 M Sodium hydroxide (CAS 1310-73-2)
- 0.1 M Potassium alum solution (CAS 7784-24-9)
- Raspberry or blackberry ("brambles") extracts
- Flower extract
- Black bean extract (food waste)
- Clear plastic sheet (acetate film or clear polypropylene A4 document holders
- Small pipettes or small "eye dropper" style bottles

Demonstration procedures

Raspberry/blackberry extract

The berry extract is best prepared by freezing a cupful (~200 g) of berries. The berries are then soaked in hot water for 30-40 minutes and gently mashed with a spoon or fork. The fruit mixture is filtered through a piece of cheesecloth or old a kitchen cloth into a bowl, glass jug or beaker. The fruit pulp retained in the cloth can be soaked again for a further 30-40 minutes if desired and filtered. The fruit juice will be strongly red in colour and usually has a pH in the range 3-4. Any juice that is not used in the demonstration can be stored in a freezer for at least 12 months for use in future.

Flower extracts

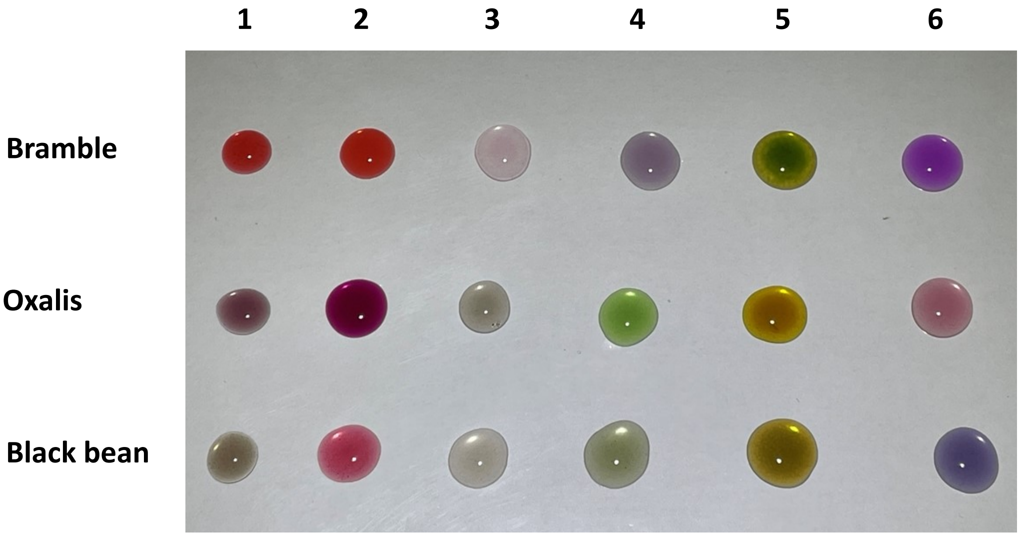
(a) Flower extracts can be obtained in most cases by crushing petals in 50 mL of hot water for 5 minutes. The volume of water or number of petals can be adjusted to get a sufficient concentration of pigment to be clearly visible, this is highly dependent on the type of flower.

(b) An alternative method is to mix flower petals in water then heat the mixture in a microwave. Microwaving is safest as a series of short bursts, e.g. 20 seconds at a time, so progress can be checked and evaporated water can be replaced if necessary. The beaker or flask should contain a liquid level no greater than 20%, this is in case the liquid becomes superheated. The glass will likely get very hot so careful handling is required.

Black bean extract

Black beans (*Phaseolus vulgaris*) are readily available from grocery stores, the instructions prior to cooking usually say the beans should be soaked in cold water. The soaking water turns a pale blue/purple colour over a period of a few hours, this liquid contains anthocyanins and it will change colour in response to pH changes. The beans are removed by draining off the liquid through a colander or sieve, and the liquid can be stored for several months in a freezer if required. The beans are not wasted, they are boiled after soaking and can be eaten or stored.

Droplets of the chosen extracts are positioned on the plastic sheet, then a drop of the chosen reagent can be added and any colour change observed. Alternatively, a small amount of the chosen indicator and the reagent can be premixed, then a droplet placed on the plastic sheet. Both methods allow good colour comparisons to be made (see Figure S3).



1 = Extract + water
2 = Extract + 1 M HCl
3 = Extract + sat. $CH_3CO_2NH_4$
4 = Extract + sat. $NaHCO_3$
5 = Extract + 1 M NaOH
6 = Extract + 5% $KAl(SO_4)_2$

Figure S3: Example droplet reactions (pH and effect of Al^{3+} ions).

Safety considerations

Small quantities of the solutions are used, but should be considered as corrosive to skin and eyes. It is also possible to use more dilute HCl and NaOH solutions if required. It is recommended that safety glasses and a lab coat are worn.

The Safety Data Sheets (SDS) From the manufacturer of chemicals should be consulted prior to making a risk assessment. The chemical supplier for this activity was Merck Life Science UK Limited (Dorset UK), the key points from the associated safety data sheets are shown in Table 2. Educators in UK schools may wish to access chemical safety information provided by the Consortium of Local Education Authorities for the Provision of Science Services (CLEAPSS**, https://science.cleapss.org.uk/).**

Table 2 Safety Data for chemicals required for metal binding/droplet art experiments.

Substance	CAS Number	Hazard Code	Warning	GHS Pictogram
1 M Hydrochloric acid[1]	**CAS** 7647-01-0	**H290**	**May be corrosive to metals**	
1 M Ammonium acetate solution[2]	**CAS 631-61-8**	**No hazard code provided**	**No hazard statements provided**	**No pictogram provided**
Sodium hydrogen carbonate[2]	**CAS 144-55-8**	**No hazard code provided**	**No hazard statements provided**	**No pictogram provided**
1 M Sodium hydroxide[3]	**CAS** 1310-73-2	**H290** **H314**	**May be corrosive to metals** **Causes severe skin burns and eye damage**	
0.1 M Potassium alum solution[2]	CAS 7784-24-9	**No hazard code provided**	**No hazard statements provided**	**No pictogram provided**

1. **10-25% Solutions are considered eye and skin irritants (1 M HCl is <4%).**
2. **Ammonium acetate, sodium hydrogen carbonate and potassium alum are** not considered to be hazardous substances according to Regulation (EC) No. 1272/2008.
3. **Solutions with a concentration <2% are considered eye and skin irritants. Solutions with concentrations >2% are considered corrosive to skin and eyes. 1 M solutions can be diluted if desired.**

Particular care should be taken when picking and extracting flowers since several plants contain toxic compounds. gloves should be worn to avoid skin contact and ingestion of flowers and seeds should be avoided. Examples of known hazardous flowers are provided below, please note that this is not an exhaustive list.

- Aconite “Monkshood” (*Aconitum napellus*)
- Autumn crocus (*Colchicum autumnale*)
- Begonia (*Begonia × semperflorens-cultorum*)
- Blue lobelia (*Lobelia erinus*)
- Clematis (*Clematis viticella*)
- Daffodil (*Narcissus pseudonarcissus*)
- Foxglove (*Digitalis purpurea*)
- Red Geranium (*Pelargonium × hortorum*)
- Hydrangea (*Hydrangea macrophylla*)
- Larkspur (*Delphinium elatum*)
- Morning glory (*Ipomoea purpurea*)
- Oleander (*Nerium oleander*)
- Petunia (*Petunia × atkinsiana*)
- Purple Iris (*Iris versicolor*)
- Red Azalea (*Azalea japonica*)
- Rhododendron (*Rhododendron ponticum*)
- Sweet pea (*Lathyrus odoratus*)
- Wisteria (*Wisteria floribunda*)

Waste disposal

The aqueous waste from the demonstration should be diluted with water and presents a low hazard.

Aqueous waste should be collected in appropriate containers for proper disposal (in line with local legislation).

Residual fruit pulp, used flower petals and other plant tissue should be disposed of as domestic garden waste or composted.

Detection of tin(II) ions in fruit juices

To prepare the demonstration, the following chemicals and materials are required:

- **Tin(II) chloride dihydrate (CAS 10025-69-1)**
- **Glycerol (CAS 56-81-5)**
- **Plastic tray or small beakers**
- **Raspberry or blackberry extract (see page S3)**
- **Fresh pineapple juice**
- **Pineapple juice from a carton**
- **Canned pineapple (chunks, rings or crushed)**

Fresh pineapple juice, pineapple juice in a carton and canned pineapple can be purchased from grocery stores or supermarkets.

If desired, tin(II) chloride can be used as a source of $Sn^{2+}$for control experiments. Tin(II) chloride dihydrate is a colourless solid and is readily available from commercial chemical suppliers. We have found that making a solution of tin(II) chloride in glycerol is more convenient than preparing aqueous solutions. The glycerol mixture does not require the addition of hydrochloric acid and can be stored for years rather than weeks.[1]

Due to its viscosity glycerol can be easier to measure out by mass rather than by volume. The tin solution is prepared by adding the required mass of tin(II) chloride to a measured quantity of glycerol (2 g $SnCl_2.2H_2O$ per 100 g or 80 mL of glycerol). The resulting viscous mixture is heated to 140 °C with occasional stirring until the solution becomes homogeneous. The cooled liquid is stored in a stoppered brown glass bottle (the liquid is easier to pour when slightly warm). The solution is approximately 0.1 M Sn^{2+}. While not essential, the solution can be diluted to 33-50% with water immediately before use to make it easier to mix with the plant extracts, however the diluted (now mostly aqueous) solution has poor keeping qualities.

1. A. Henriksen, *Analyst*, 1963, **88**, 898-899

Demonstration procedure

1. **10-15 mL of pineapple juice is transferred to a suitable container (such as a beaker) and mixed with 5 mL of berry extract. If tin ions are detected, a purple/violet colour will be observed).**
2. **Control samples of the berry extracts can also be prepared, for example, Figure S4 shows comparisons with bramble extract. The top row shows acidified bramble extract and tin(II) chloride mixed with blackberry extract. The latter shows the expected colour from interaction of tin ions with the anthocyanins in the blackberry extract (see Figure S4).**

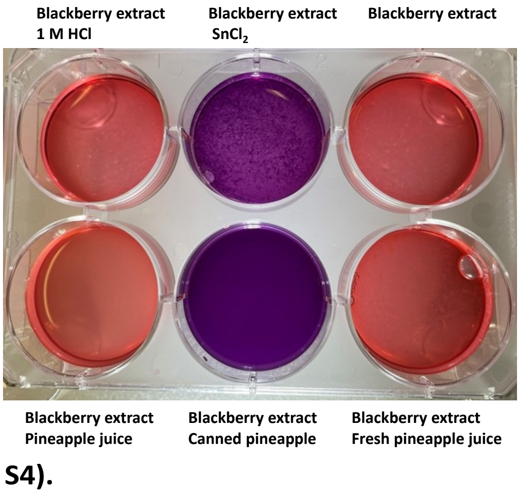


Figure S4: Example tests of pineapple juices with blackberry extracts.

Safety considerations

The hydrochloric acid solutions are dilute, but should be considered as corrosive to skin and eyes, it is recommended that safety glasses and a lab coat are worn.

Tin(II) chloride is corrosive on contact with skin and eyes.

The Safety Data Sheets (SDS) From the manufacturer of chemicals should be consulted prior to making a risk assessment. The chemical supplier for this activity was Merck Life Science UK Limited (Dorset UK), the key points from the associated safety data sheets are shown in Table 3. Educators in UK schools may wish to access chemical safety information provided by the Consortium of Local Education Authorities for the Provision of Science Services (CLEAPSS, **https://science.cleapss.org.uk/).**

Table 3 Safety Data for chemicals required for detection of tin(II) ions in fruit juices.

Substance	CAS Number	Hazard Codes	Warnings	GHS Pictogram
Tin(II) chloride dihydrate[1]	**CAS 100025-69-1**	**H290, H302+H332 H314 H317 H335 H373 H412**	**May be corrosive to metals Harmful if swallowed or if inhaled Causes severe skin burns and eye damage May cause an allergic skin reaction May cause respiratory irritation May cause damage to organs Harmful to aquatic life with long lasting effects**	
Glycerol[2]	**CAS 56-81-5**	**No hazard code provided**	**No hazard statements provided**	**No pictogram provided**

1. **Refers to the solid precursor, the solution used in the experiment is 0.1 M in glycerol.**
2. **Glycerol is** not considered to be a hazardous substance according to Regulation (EC) No. 1272/2008.

Waste disposal

The aqueous waste from the demonstrations with commercially available pineapple juice, fresh pineapple or canned pineapple should be diluted with water and present a low hazard.

The aqueous waste from control experiments with tin(II) chloride should be diluted with water prior to disposal as low hazard aqueous waste. The allowable limits for the tin(II) content of food products varies between countries, however values <150 ppm are typical.[2,3] The tin concentration of the aqueous waste solution from control experiments using tin(II) chloride can be adjusted to <100 ppm **by adding 25 mL of water to the test solution.**

Aqueous waste should be collected in appropriate containers for proper disposal (in line with local legislation).

Residual fruit juice or pineapple should be disposed of as domestic garden waste or composted.

2. S. Blunden and T. Wallace, *Food and Chem. Toxicol.*, 2003, **41**, 1651-1662.

3. C. C. Seow, Z. Abdul Rahman and N. A. Abdul Aziz, *Food Chem.*, 1984, **14**, 125-134.

A simple and quick demonstration of wine "pinking"

To prepare the wine pinking demonstration, the following chemicals and materials are required:

- **Sodium metabisulfite (CAS 7681-57-4)**
- **5 M Sulfuric acid (CAS 7664-93-9) (278 mL conc. H_2SO_4 per litre of solution)**
- **Sodium acetate trihydrate (CAS 6131-90-4) or anhydrous (CAS 127-09-3) (optional)**
- **Raspberry or blackberry extract (see page S3, red begonia extract also works very well)**
- **2 Wine glasses**
- **Assorted beakers**
- **Old wine bottle (optional)**

Demonstration procedure

1. Transfer 500 mL of distilled water to an empty wine bottle or beaker.

2. Note what volume of the berry or flower extract is required to tint the water to the desired shade of rose. Tap water may alter the colour. Discard the diluted solution.

3. To the required volume of berry or flower extract, add a solution of sodium metabisulfite in water (approximately 10%) dropwise until the colour almost disappears (be patient between additions and take care not to add too much or it is unlikely that the final step will work). A more convincing "white wine" is produced if a little colour remains. Note: The diluted berry or flower extract can be decolourised or nearly decolourised directly, though much more bisulfite will be required and an appreciable odour of sulfur dioxide will become evident upon acidification in step 7. If you don't have much extract, try step 2 on a small scale then scale up accordingly for the demonstration. Significantly less bisulfite is required to decolourise or nearly decolourise the neat extract, then consquently there's very little odour of sulfur dioxide when acidified.

4. Transfer the bleached extract to the empty wine bottle or beaker then add 500 mL of distilled water.

5. Place approximately 0.5 mL of 5 M sulfuric acid in the bottom of a wine glass without splashing the sides. This quantity will be invisible to the audience.

6. Pour 100-150 mL of the decolourised berry or flower solution into a clean wine glass (this can be described as "white wine" to the audience, see Figure S5).

7. Pour 100-150 mL of the decolourised berry mixture into the wine glass containing sulfuric acid. The liquid in the glass should become pink/pale red (this can be described as "rosé wine" to the audience). At low pH, the flavylium ion is favoured and "sulfite" is present as aqueous sulfur dioxide rather than bisulfite.

8. The "rosé" wine can be converted to "water" by adding all at once 30 mL of 1 M sodium acetate solution (slow addition doesn't create enough turbulence to mix the solutions effectively). This lowers the acidity from pH 1.7 to pH 5 where bisulfite predominates and the colourless flavene sulfonate can form. In addition, at this pH, the preferred form of anthocyanins is the colourless carbinol. Given the rapidity of the decolouristion, bisulfite addition is the more likely route.

Figure S5: Wine pinking demonstration. (Left to Right). Decolourised berry extract (treated with sodium metabisulfite) and decolourised berry extract acidified with dilute sulfuric acid.

Safety considerations

5 M sulfuric acid should be considered as corrosive to skin and eyes, it is recommended that safety glasses and a lab coat are worn. Take care when using concentrated sulfuric acid, always pour a slow stream of acid into rapidly stirred water.

When using familiar beverage containers and drinking vessels for demonstrations, take particular care than nobody can unwittingly consume liquid from them.

When the decolourised berry extract is added to acid, the bisulfite present will react to form a small quantity of sulfur dioxide. This demonstration must be conducted in a well-ventilated area. If the demonstrator has asthma, it is recommended they complete a more detailed risk assessment, sulfur dioxide and sulfites can trigger asthma attacks.

The Safety Data Sheets (SDS) From the manufacturer of chemicals should be consulted prior to making a risk assessment. The chemical supplier for this activity was Merck Life Science UK Limited (Dorset UK), the key points from the associated safety data sheets are shown in Table 4. Educators in UK schools may wish to access chemical safety information provided by the Consortium of Local Education Authorities for the Provision of Science Services (CLEAPSS**, https://science.cleapss.org.uk/).**

Table 4 Safety Data for chemicals required for a demonstration of wine "pinking".

Substance	CAS Number	Hazard Statements	Warning	GHS Pictogram
5 M Sulfuric acid[1]	**CAS 7664-93-9**	**H290** **H314**	**May be corrosive to metals** **Causes severe burns and eye damage**	
Sodium metabisulfite	CAS 7681-57-4	**H302** **H318**	**Harmful if swallowed** **Causes serious eye damage**	
Sodium acetate[2] trihydrate	CAS 6131-90-4	**No hazard code provided**	**No hazard statements provided**	**No pictogram provided**

1. **Solutions >15% are considered corrosive to eyes and skin (5 M H_2SO_4 is ~40%).**
2. **Sodium acetate trihydrate is** not considered to be a hazardous substance according to Regulation (EC) No. 1272/2008.

Waste disposal

The aqueous waste from the demonstration should be diluted with water and presents a low hazard.

Aqueous waste should be collected in appropriate containers for proper disposal (in line with local legislation).

Residual fruit pulp, used flower petals and other plant tissue should be disposed of as domestic garden waste or composted.

Anthocyanins and the Shinoda test

The Shinoda test is often used to test plant extracts from plant material. A sample of plant tissue is usually extracted in ethanol, mixed with dilute hydrochloric acid, and then magnesium metal added. If flavonoids are present a red, orange or violet colour is often observed. In the example below, commercially available rutin is used.

Magnesium ribbon or turnings (CAS 7439-95-4)
2 M hydrochloric acid (CAS 7647-01-0)
Ethanol (CAS 64-17-5)
Rutin (CAS 153-18-4)
Boiling tube

Demonstration procedure

1. **5 mg of rutin is dissolved in 3 mL of ethanol (warming in hot water dissolves the solid faster).**
2. **5 mL of 2 M hydrochloric acid is added to the solution of rutin.**
3. **Cut a 4 cm long piece of magnesium ribbon, fold it in half, and add the metal to the test solution.**
4. **As the reaction with the metal and acid proceeds, the solution will become warm, hydrogen gas will evolve, and the colour of the test solution should become red/orange (Figure S6).**

If pure rutin is not available, a commercial rutin supplement tablet can be used, choose one with the fewest excipients. A tablet containing 600 mg of "rutin" likely contains 600 mg of plant material with approximately 40 mg actual rutin. Boil a rutin tablet in 25 mL ethanol for a minute or two, allow to cool, then decant or filter through a small cotton wool plug to remove coarse grainy material. The solution doesn't have to be perfectly clear. Mix with 20 mL of water then leave to stand overnight if possible (not essential, but a lot of the murkiness settles out overnight). Pour 3 mL of the solution into a boiling tube, add 5 mL of 2 M hydrochloric acid, mix then drop in a 4 cm length of magnesium ribbon folded in half. Alternatively, 0.07 g of magnesium turnings can be used, or add a small chunk of sawn up Mg pencil sharpener or Mg rod (available from online suppliers). Larger chunks can be re-used since there's only enough acid to dissolve 0.1 g of Mg. If buying a magnesium pencil sharpener, make sure it is genuinely magnesium, most are aluminium.

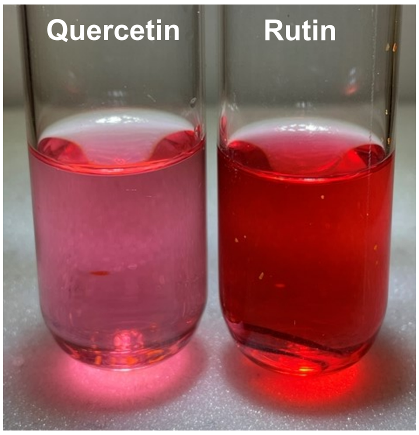


Figure S6: Example Shinoda test solutions.

Safety considerations

2 M hydrochloric acid should be considered as corrosive to skin and eyes, it is recommended that safety glasses and a lab coat are worn.

The reaction will release hydrogen gas, this is readily ignited, so the demonstration must be attempted in a well ventilated area and away from sources of ignition.

The boiling tube may become hot as the reaction proceeds, some of the ethanol may be become vapour, this should also be considered a fire hazard.

The Safety Data Sheets (SDS) From the manufacturer of chemicals should be consulted prior to making a risk assessment. The chemical supplier for this activity was Merck Life Science UK Limited (Dorset UK), the key points from the associated safety data sheets are shown in Table 5. Educators in UK schools may wish to access chemical safety information provided by the Consortium of Local Education Authorities for the Provision of Science Services (CLEAPSS, **https://science.cleapss.org.uk/).**

Table 5 Safety Data for chemicals required for a Shinoda test.

Substance	CAS Number	Hazard Statements	Warning	GHS Pictogram
Magnesium ribbon or turnings	**CAS 7439-95-4**	**H228** **H260**	**Flammable solid** **In contact with water releases flammable gases which may ignite spontaneously**	
2 M Hydrochloric acid[1]	**CAS** 7647-01-0	**H290**	**May be corrosive to metals**	
Ethanol[2]	CAS 64-17-5	**H225** **H319**	**Highly flammable liquid and vapour** **Causes serious eye irritation**	
Rutin[3]	CAS 153-18-4	**No hazard code provided**	**No hazard statements provided**	**No pictogram provided**

1. **10-25% Solutions are considered eye and skin irritants (1 M HCl is ~8%).**
2. **Solutions >50% are considered eye irritants.**
3. **Rutin is** not considered to be a hazardous substance according to Regulation (EC) No. 1272/2008.

Waste disposal

The aqueous waste from the demonstration should be diluted with water and presents a low hazard.

Unless being recovered for future tests, any residual magnesium metal should be allowed to fully react to form soluble magnesium salts (not classed as hazardous substances according to Regulation (EC) No. 1272/2008).

Aqueous waste should be collected in appropriate containers for proper disposal (in line with local legislation).