Science notes

Cabomba – an exocharmic plant!

Anne Adams, Gordon Moore, Alison Rutherford, Fiona Stewart, Kath Crawford and Paul Beaumont

Some years ago the phrase 'exocharmic reactions' was introduced into the literature to describe chemical reactions and demonstrations which fascinate, allure or delight the observer (Ramette, 1980). Despite their undoubted value in supporting curriculum delivery, collections of demonstrations for use in biology teaching are not widely available. We hope that this short contribution might encourage colleagues to consider sharing their favourite demonstrations with the wider teaching community.

In a recent publication, one of us (Crawford, 2005) explored how *Cabomba* might be used as a reliable alternative to *Elodea* and we wish to extend these observations in the experiments described. While the basic methodology that we have used in the experiments is not new, we strongly believe that we present here an experimental set-up that provides a stimulating way of engaging students in discussion and leads to greater understanding of plant respiration and the effect of light intensity on the rate of photosynthesis.

Materials and methods

Cabomba is available from most tropical fish suppliers as well as online (see, for example, the Blades Biological and Urmston Aquatics websites). We have found that the best way to maintain *Cabomba* is to put it in a constantly aerated tank (just use a simple aquarium pump), filled with tap water and located near a window, together with some *Elodea* (ratio 6 : 1 of *Cabomba* to *Elodea*). Under these conditions, the plants survive well together for months. In our experience, failure to add *Elodea* in the proportion suggested leads to the breakdown of the *Cabomba* over a period of one to two weeks.

The light source used in these experiments was a 35 W fluorescent tube (product number 56427) purchased from Focus DIY. No attempt to quantify light intensity falling on the sample was made. Qualitatively similar results can be obtained with a variety of light sources likely to be available in the school laboratory. Lamps that lead to significant changes in the temperature of the solutions are best avoided.

A stock solution of hydrogencarbonate indicator was prepared as follows:

1. Cresol red (0.10 g) [**caution: HARMFUL** if ingested in quantity or inhaled as dust] and thymol blue (0.20 g) [**caution: HARMFUL** if ingested in quantity or inhaled as dust] were dissolved in ethanol (20 cm^3) [**caution:** ethanol is **HIGHLY FLAMMABLE** and an **IRRITANT**].
2. Sodium hydrogencarbonate (0.85 g) was dissolved in freshly boiled, cooled distilled water (approximately 200 cm^3) and combined with the ethanolic solution of cresol red/thymol blue and made up to 1.0 dm^3 with distilled water.

Safety note: None of the experiments here present significant health and safety risks provided standard laboratory practice is observed. Students should be advised to wash their hands after handling plant material.

For routine use, 100 cm^3 of the stock solution was diluted to 1.0 dm^3 with freshly boiled, cooled distilled water.

Filters were obtained from Lee Filters. Detailed absorption and transmission characteristics of the filters are provided on both the Lee Filters and the SAPS websites (see *Websites*).

Results and discussion

The use of hydrogencarbonate indicator to monitor CO_2 levels in aqueous solutions is well established (for example, Nuffield, 1966; Eldridge, 2007). Figure 1 shows how the colour of the indicator solution changes over the pH range 6.8 to 9.2. Since CO_2 is an acidic gas, solutions high in CO_2 will have a lower pH than those which are low in CO_2.

Given that the process of photosynthesis consumes CO_2, one might predict that if some *Cabomba* were introduced into a solution of hydrogencarbonate indicator and irradiated for a sufficient period of time (4 hours in our experiments), then one would see a colour change in the indicator from its starting orange to purple as CO_2 is removed from solution. As can be seen from Figure 2, that is indeed what is observed. So, photosynthesis has taken place with a consequent reduction in the concentration of CO_2, causing the colour change.

The time required for observable colour changes will vary, with the principal factors being the light intensity and the spectral output of the lamp. Figure 3 shows a slightly different experimental arrangement. In this case we set up two measuring cylinders each containing a single strand of *Cabomba* and hydrogencarbonate indicator. Before illumination, the pH of these solutions was approximately 7.4 (orange). Both cylinders were placed in front of the lamp for 4 hours of illumination, but cylinder A was

Figure 2 Effect of irradiation on *Cabomba*/hydrogencarbonate indicator mixtures: (A) before irradiation (pH measured to be 7.4); (B) after 4 hours of irradiation (pH measured to be 8.3)

Figure 1 Hydrogencarbonate indicator solutions in the range pH 6.8 (pale yellow) to pH 9.2 (purple); pH increases in increments of 0.4 units

covered in black paper during this period. Cylinder B in Figure 3 has turned a purple colour for exactly the same reasons as described for Figure 2. Cylinder A in Figure 3 also shows a colour change from its starting point, but in this case there has been a lowering of pH indicating a rise in CO_2 concentration. This is an interesting observation, which can stimulate much discussion. The explanation for this observation is that the *Cabomba* in cylinder A is not photosynthesising but is producing CO_2 through the process of respiration and this gives rise to the colour change.

Students might be asked to predict whether respiration occurs in plants that are undergoing photosynthesis. The experimental set-up shown in Figure 4 shows this to be the case. We took a single strand of *Cabomba* and in two places along its length placed discs cut from the base of a Styrofoam™ cup. The diameter of the disc was such that it was the same size as the internal diameter of the measuring cylinder. A small hole (diameter 2–3 mm) was cut in the centre of the polystyrene disc and a further cut made from the centre of the disc to its outer edge. This allows the disc to be placed around the stems of the strands of *Cabomba* and thereby act as a collar. The *Cabomba* and discs were placed in a measuring cylinder containing hydrogencarbonate indicator (pH 7.4). One-third of the measuring cylinder was covered in black paper, one-third was covered with a filter which allows 50% of the light to be transmitted and the remaining third was left uncovered. The purpose of the discs is to reduce diffusion of hydrogencarbonate indicator between

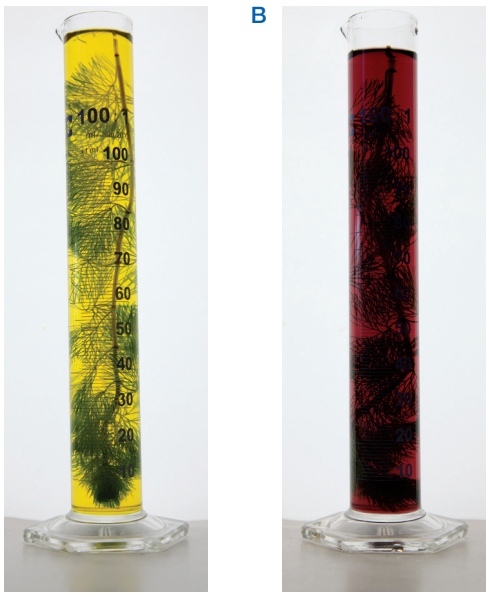

Figure 3 Effect of irradiation on *Cabomba*/hydrogencarbonate mixtures: (A) after 4 hours of irradiation during which the cylinder was covered in black paper (pH measured to be 6.8); (B) after 4 hours of irradiation (pH measured to be 8.3)

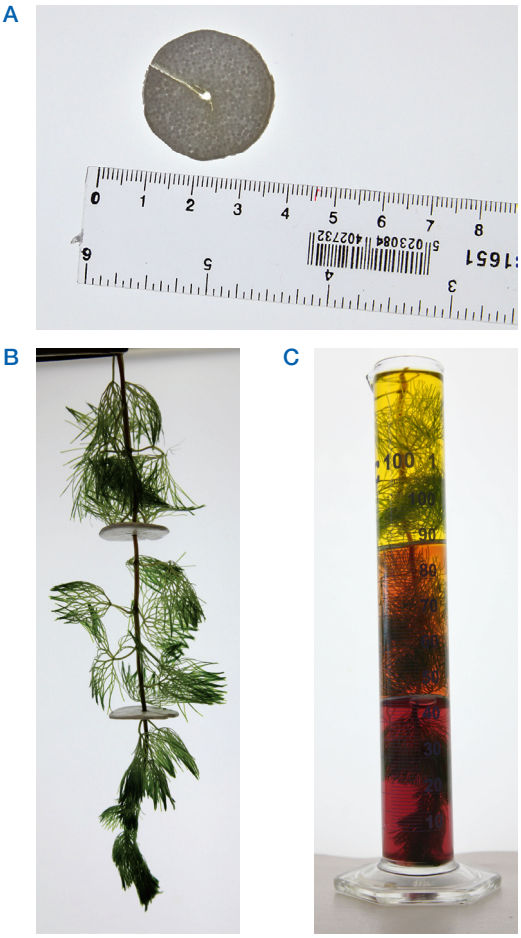

Figure 4 (A) Styrofoam™ fashioned to fit inside a 100 cm^3 measuring cylinder; (B) strand of *Cabomba* showing two collars in place; (C) effect of 4 hours of irradiation on *Cabomba*/hydrogencarbonate mixtures: before irradiation the pH was measured to be 7.4; the upper portion of the cylinder was covered with black paper during irradiation; the middle portion of the cylinder was covered with 50% neutral-density filter during irradiation; the lower portion of the cylinder was kept uncovered during irradiation

the three regions. The measuring cylinder was then placed in front of the lamp and irradiated for 4 hours. The resultant colour changes are shown in Figure 4. One could of course ask students to predict what they might see before removing the filters to reveal the colour changes that have taken place. In our experience, removal of the filters from the measuring cylinder in Figure 4 gives rise to one of those perhaps infrequent, yet indulgent, moments in teaching when your students say 'wow!' or some such similar remark as they note the variety of colours present in the cylinder.

It can be seen in this experiment that both respiration and photosynthesis occur at the same time in the same plant. The bottom one-third, receiving 100% of the light, goes purple (pH has increased to 8.3), implying CO_2 uptake and hence photosynthesis. The middle one-third, receiving 50% of the light, also shows a colour change, which implies net CO_2 uptake (pH has changed to 7.6) and hence photosynthesis, albeit at a lower rate. The colour change in the top one-third, receiving no light, shows a reduction in pH (pH has reduced to 6.8), which in turn can be explained by the fact that only respiration is occurring. From the observations shown in Figure 4 we might additionally conclude:

- photosynthesis does not take place in the dark;
- when plants are illuminated, the rate of photosynthesis is greater than the rate of respiration at the light levels used.

A subsequent experiment in which black paper is placed over the bottom one-third of the measuring cylinder shown in Figure 4, with the top one-third uncovered, followed by a period of illumination, leads to a reversal of colour change. In theory this reversal process can be repeated many times, but in practice diffusion through the pores and gaps in the polystyrene disc eventually make the colour changes less clear.

We believe that the experiments described can be used to show that both photosynthesis and respiration can occur simultaneously in a single plant, and be described as exocharmic.

Acknowledgements

Aspects of this work have been supported by the Scottish Government through funding received by SSERC as part of the Supporting Scottish Science Education through CPD project. Additional financial support from the Gatsby Charitable Foundation is gratefully acknowledged. The authors would like to thank Ian Birrell from SSERC for his help with photography.

Anne Adams, one of the authors of this manuscript, died on 23 February 2011. Anne was the most caring and delightful of colleagues and we shall miss her terribly. While we all have responsibility for the contents of this manuscript, it would not have been possible without her contribution.

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Websites

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Urmston Aquatics: www.urmstonaquatics.com.

Kath Crawford is the deputy director (projects) and **Paul Beaumont** is a senior advisor (science) at SSERC (2 Pitreavie Court, Dunfermline KY11 8UB); email: Paul.Beaumont@sserc.org.uk. **Anne Adams**, until her death, was a biology technician at SSERC. **Gordon Moore** is a science technician at Mearns Castle High School, **Alison Rutherford** is the principal teacher (biology) at Viewforth High School, Kirkcaldy, and **Fiona Stewart** is the principal teacher (biology) at Perth Grammar School.