Making the invisible visible: monitoring levels of gaseous carbon dioxide in the field and classroom

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Use of a gaseous carbon dioxide sensor and data-logger can make real to students the impact of their lifestyle choices on climate change

Most people have fantasised about what it would be like to be invisible to others. This article concerns the opposite – making an otherwise invisible gas, carbon dioxide, visible (at least in a quantitative sense). This is now something that can be achieved at a realistic cost in the school classroom, using the gaseous CO_2 sensor produced by the USA-based company Vernier (distributed in the UK by Instruments Direct (Services) Ltd – see Suppliers).

There is growing consensus amongst scientists that the world is going through a period of rapid climatic change, driven at least partly by anthropogenic production of greenhouse gases such as CO_2 (Maslin, 2004). The complexity of the issues involved means it is difficult to maintain students' interest, partly because they cannot see the *immediate* relevance of climate change to their own lives, and partly because they cannot see what impact their lifestyle choices (ecological footprint) might have on the global picture. Historically, climate change has always had a powerful effect on human affairs, as brilliantly described by Brian Fagan (Fagan, 2004), and the recent catastrophic effects of hurricane *Katrina* in the southern USA demonstrate that modern humanity is even more at risk, despite technological advances. Global warming is a key topic in most UK 14–18 biology syllabuses, and the new *Salters-Nuffield AS Biology* course (for 16–18 year-olds) includes a significant section on plants and climate change (Salters, 2005) where students are asked evaluate evidence.

Most school students will have been introduced to the critical role of photosynthesising plants in removing CO_2 from the atmosphere as part of the carbon cycle, and releasing oxygen as a waste gas. Even though the importance of learning these cycles is emphasised, it is normal for students to be expected to take the details on trust. Standing outside the classroom, in a green environment, it is simply not possible to actually *see* the carbon dioxide being taken up by the leaves of the plants, or released by the decomposers in the soil. But now there is technology that allows us to monitor what is going on, and even calculate some of the figures involved.

ABSTRACT

This article introduces the use of a gaseous carbon dioxide sensor and data-logger for a variety of purposes in the school laboratory, including measurements of respiratory and photosynthetic rates of animals, plants and microbes. Suggestions are made for taking measurements in the field, as well as demonstrations and project work in the school laboratory. A simple equation for converting rates of change of gaseous CO_2 concentration into energy or glucose equivalents is suggested.

Fantasy series of lessons on the carbon cycle

This series of lessons could be used at any level, and would emphasise the personal importance of photosynthesis, and of responsible citizenship in relation to ecological footprints, to each individual student.

Everybody fears suffocation: rebreathing air into an empty bag is only possible for a moment – then we are tearing the bag from our face and gulping fresh air. Imagine a lesson that begins with a class entering a laboratory. They notice the windows are closed, even though the day might be warm (if plants

BOX 1 Calculations based on rates of change in CO_2 concentration

If the rate of CO_2 uptake or production is known, then it can provide a means for calculating the equivalent number of glucose molecules that this amount of CO_2 represents, either being made during photosynthesis or broken down during respiration. If this assumption is made, then it is possible to estimate the amounts of energy being used in photosynthesis or released in respiration as below.

- Select an area of the graph, and select the rate button to get the rate of CO_2 change figure for this region. The software provides units for the rate of CO_2 change in ppm/second.

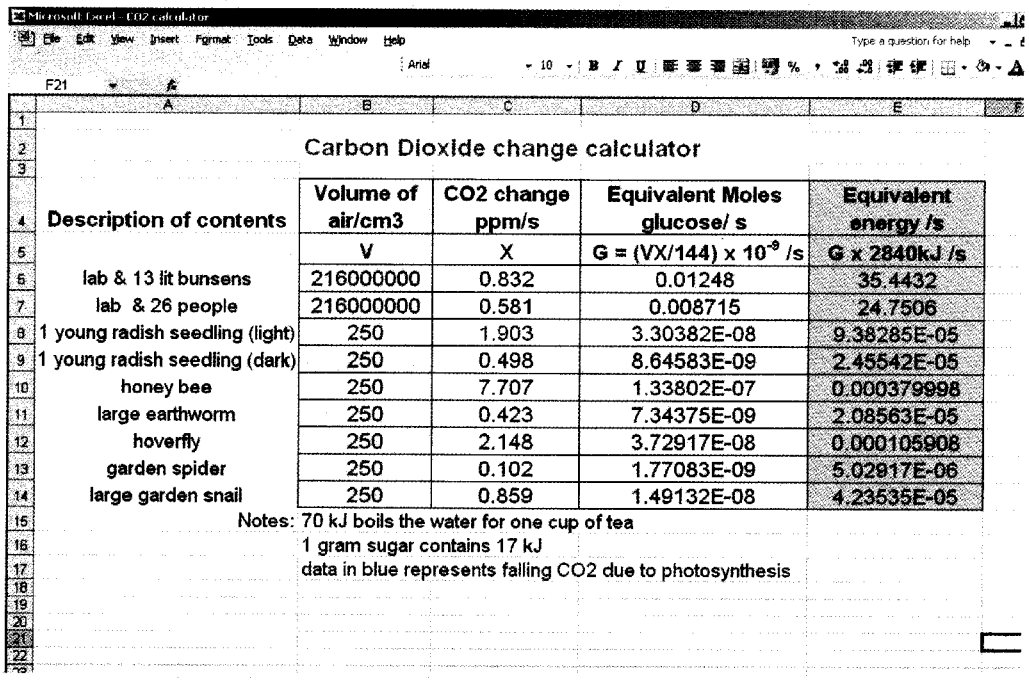
- This rate of change in CO_2 would be different for different-sized containers, so the next step is to take account of the volume of gas in the container being used, in units of cm^3:

If we assume that six CO_2 molecules are (used in photosynthesis to form/are released by the respiration of) a glucose molecule, then the **number of moles of glucose being formed per second = (rate of change (ppm per second) x volume of container (cm^3) / 144) x 10^{-9}**

- The complete aerobic respiration of glucose is worth 2840 kJ mol^{-1}. So the **rate of release of energy (or uptake of energy in photosynthesis) by the organism in kJ per second = (moles of glucose per second) x 2840 kJ.**

For many purposes, it would be a good idea to have an *Excel* worksheet set up so that simply entering the volume of the container, and the ppm/second figure, leads to the automatic calculation of glucose and energy equivalents (see Figure 1).

Naturally this figure can be adjusted to provide a rate in kJ per minute, per hour, or per day. To allow fair comparisons between different samples, the figure should also be related to the mass of biological material involved, to provide a rate as units of energy/unit mass/unit time.



Carbon Dioxide change calculator

Description of contents	Volume of air/cm3	CO2 change ppm/s	Equivalent Moles glucose/ s	Equivalent energy /s
	V	X	G = (VX/144) x 10^{-9} /s	G x 2840kJ /s
lab & 13 lit bunsens	216000000	0.832	0.01248	35.4432
lab & 26 people	216000000	0.581	0.008715	24.7506
1 young radish seedling (light)	250	1.903	3.30382E-08	9.38285E-05
1 young radish seedling (dark)	250	0.498	8.64583E-09	2.45542E-05
honey bee	250	7.707	1.33802E-07	0.000379998
large earthworm	250	0.423	7.34375E-09	2.08563E-05
hoverfly	250	2.148	3.72917E-08	0.000105908
garden spider	250	0.102	1.77083E-09	5.02917E-06
large garden snail	250	0.859	1.49132E-08	4.23535E-05

Notes: 70 kJ boils the water for one cup of tea
1 gram sugar contains 17 kJ
data in blue represents falling CO2 due to photosynthesis

Figure 1 Screen shot of *Excel* spreadsheet for converting carbon dioxide figures.

are present, they should be covered with black plastic bin-liners). On a screen there is a display provided by a data-projector linked to the CO_2 probe and data-logger, showing a scrolling graph of the CO_2 level in the room plotted against time. Once the door is closed and the class settled down, they notice the rising level of CO_2 on the display.

The mixing of CO_2, expelled on the breath of the teacher and students, with the air in the room will cause the graph to rise, and the gradient of the graph will provide a rate. This gradient can be instantly found using a tool button on the graph screen (see instructions on the use of *LoggerPro3* software later), giving the rate of change in parts per million (ppm) per second. This rather academic figure can be converted by a simple calculation to equivalent units of glucose or energy (see Box 1 and Figure 1).

For even more drama, the Bunsen burners in the laboratory could be lit for a 10-minute period; the change in the gradient of the graph will soon be apparent (a nice parallel with fossil fuel combustion and the composition of the global atmosphere). When the Bunsens are turned off and windows opened to allow fresh air to circulate in the room and to vent the CO_2, the change in the atmosphere can be followed on the scrolling graph (see Figure 2). Incidentally, building regulations recommend the CO_2 levels in rooms are kept below 1000 ppm; however, it is commonplace for stuffy teaching rooms to have levels as high as 4000 ppm! (Coley and Beistner, 2002).

The CO_2 probe is then placed in a boiling tube with a freshly detached plant leaf, covered by a lightproof sleeve (lightproof card or aluminium foil) (Figure 3). The first surprise for many students is to see that the leaf in the dark is doing what we do – creating CO_2 rather than taking it up; this provides an opportunity to recognise that respiration is a universal feature of living organisms. Then the sleeve is removed and a bright light shone on the tube; within a couple of minutes the CO_2 levels begin to fall (thunderous applause from the class!).

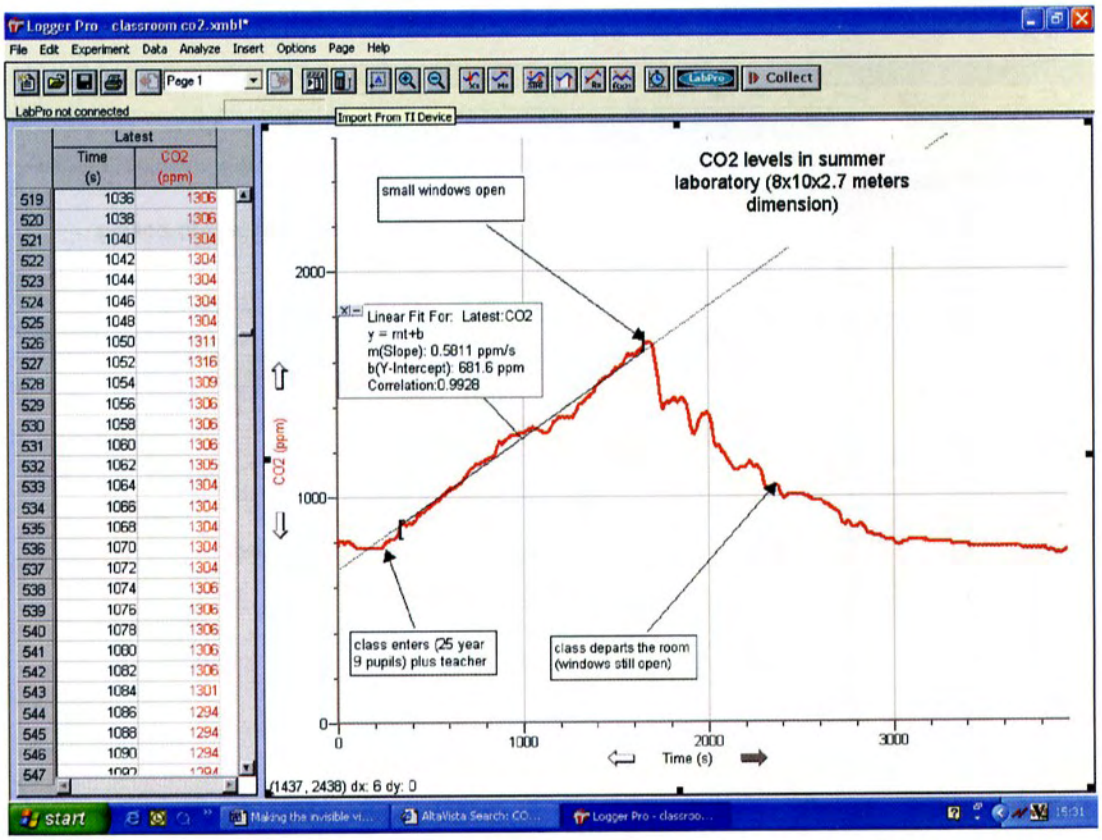


Figure 2 Screen shot showing the effects of a class of students on the composition of the atmosphere of a room.

Figure 3 A boiling tube and probe, with a folded plant leaf inside.

The advantage of using a single leaf in such a small container is that the change in carbon input/output will be seen in a couple of minutes (Figure 4).

The lesson finishes with an attempt to estimate the number of such illuminated leaves that would be needed to stabilise the CO_2 levels in the lab, by balancing the CO_2 output of the class with photosynthetic uptake of CO_2 by plant leaves. Guidance with the calculations is given in Boxes 1 and 2.

Future lessons might involve going into the garden or field to measure the real inputs/outputs of CO_2 by plant leaves, the soil and small invertebrates. The rest of this article describes the probe and a range of ways in which the CO_2 probe can be adapted for these purposes. Further suggestions and details can be found on the SAPS website (see websites).

Principle behind the CO_2 probe

The sensor depends on the fact that gaseous CO_2 intercepts pulses of infra-red radiation produced by a filament at the end of the sensor probe; the more CO_2 there is in the air, the less radiation reaches the infra-red sensor (Figure 5). This, of course, is the basis for the greenhouse-gas effect, whereby the loss of heat from Earth by radiation to outer space is limited by greenhouse gases such as CO_2 and methane. So discussing the function of the probe can actually form a useful basis for understanding the greenhouse gas effect.

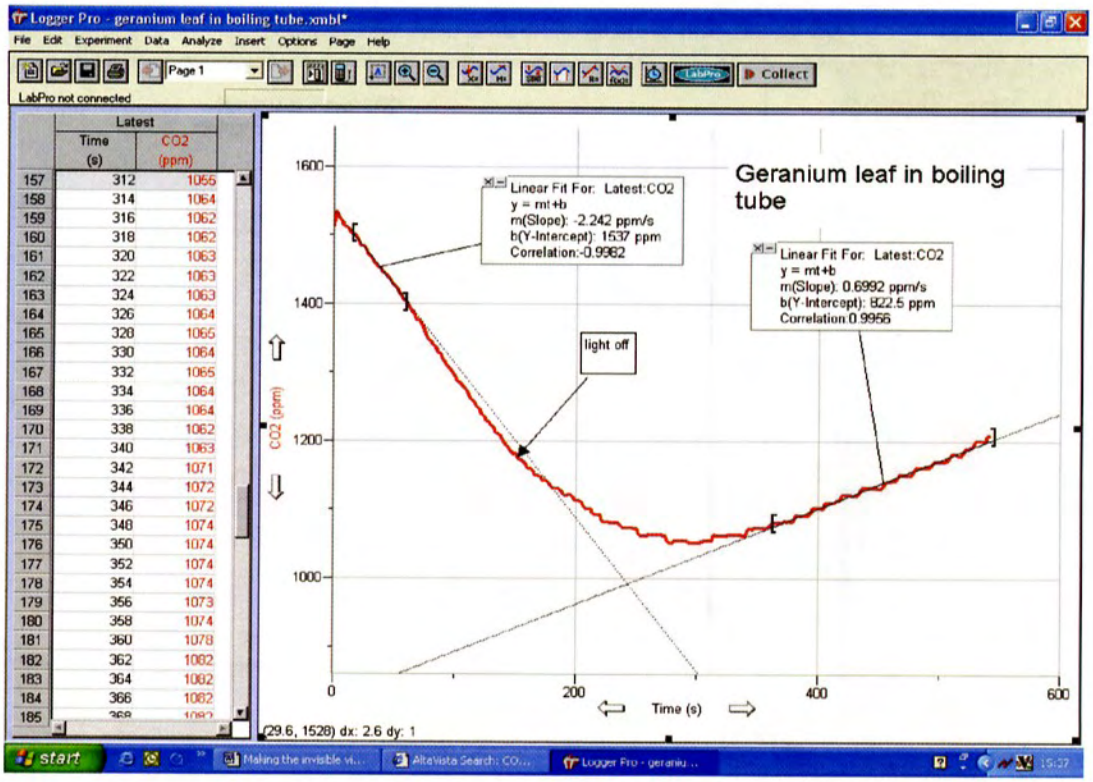


Figure 4 Screen shot showing the effect of turning off the light source to a geranium leaf in a boiling tube, on the concentration of CO_2 in the tube.

BOX 2 Calculating the need for plant leaves to reprocess an individual's CO_2, and replenish the oxygen consumed in respiration

- Find the volume of the laboratory by multiplying its length, breadth and height in metres, and then multiplying the answer by 10^6 to give its volume in cm^3.

- Assume that the gradient of the CO_2 increase in the laboratory, with the doors and windows closed (and plants covered), is largely due to the respiration of the students in the room, and use the formula below to calculate the *number of moles of glucose being respired per second by the students*:
(rate of change (ppm per second) x volume of container (cm^3) / 144) x 10^{-9}

- Divide this figure by the number of students in the room, to find out the number of *moles of glucose per second being respired by each student*.

- Assume that the leaf in the boiling tube is in a volume of 50 cm^3 of air and employ the formula above to calculate the *number of moles of glucose per second being made in the tube when the leaf is photosynthesising in the light*. This is the *net productivity** of the leaf, due to the fact that the leaf has to make the glucose to pay off its respiratory needs, before it actually makes a 'net profit' of extra glucose for use in growth, etc. This net productivity is what is needed to reprocess back to O_2 the CO_2 released on an individual person's breath.

- Divide the figure for the CO_2 output of an individual student by the figure for the CO_2 uptake by an illuminated plant leaf, and the answer gives *the number of illuminated leaves needed to reprocess the exhaled carbon dioxide of an individual student*.

A sobering thought for students is that plant leaves are only naturally illuminated for half of the 24 hour day/night cycle, and that climatic conditions, seasons, disease and many other factors will reduce further the capacity of a leaf to help us breath in this way! In addition, the draughtiness of the room will mean that the values for class respiration will be significantly less than the textbook values.

* To find the *gross productivity* of the leaf, employ the formula above to calculate the number of *moles of glucose per second being respired by the plant leaf in darkness* (because this is still happening when the leaf is in the light). The gross productivity of the leaf is the sum of this figure plus the net productivity figure.

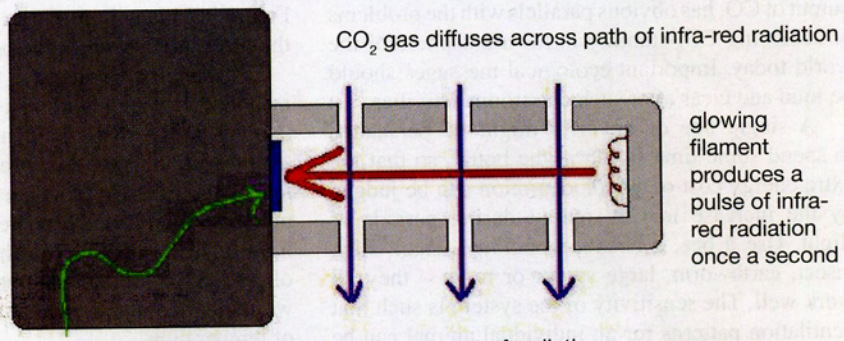


Figure 5 Principles upon which the CO_2 sensor depends.

Advantages of using a gaseous CO_2 sensor

What are the advantages of using a gaseous CO_2 sensor, as opposed to a gaseous or aqueous O_2 sensor? Gaseous diffusion is thousands of times faster than diffusion in water, allowing results in *minutes rather than hours* (allowing time for replications and modifications). There is much *less CO_2 as a fraction of the atmosphere*, so changes are more obvious, again allowing results to be obtained in minutes rather than hours (and by using small container volumes, only small amounts of biological materials are needed). No difficult experimental concepts are required to understand the fate of the CO_2 in photosynthesis – *what you see is what you get*. The analogy between the principle on which the probe works and global warming can be explored. Used with a laptop, the Vernier software allows *dramatic real-time plotting and annotation of data* whilst it is being collected, as well as sophisticated analysis tools. The data-logger can also be used on its own and the data downloaded later, or used with a Texas TI graphical calculator to provide a live display.

Methods for employing the CO_2 probe

Probe in a 250 cm^3 bottle (supplied with kit)

This can be used with either animal or plant material, or both animal and plant material, to show the evolution and uptake of CO_2 (Figure 6). If both sorts of material are present, it might be possible to achieve a balance (by varying the size or species of a leaf, or the intensity of light provided). Placing animals and plants together and achieving a balance of input and output of CO_2 has obvious parallels with the problems of achieving CO_2 stability in the atmosphere of the world today. Important ecological messages should be loud and clear after such classroom activities.

A single bee or hoverfly might be persuaded to spend some time flying in the bottle, so that the extra energy cost of such locomotion can be judged by the increase in CO_2 output during periods of flight. Use a bee, a beetle, woodlouse, snail, stick insect, earthworm, large spider or moth – they all work well. The sensitivity of the system is such that ventilation patterns for an individual animal can be distinguished; for example, a bee and an earthworm seem to exchange gas with their environment continuously, whereas a housefly seems to breath in a more cyclic manner, closing its spiracles between ventilation movements. A garden spider shows the same cyclical ventilation pattern. A bumblebee can

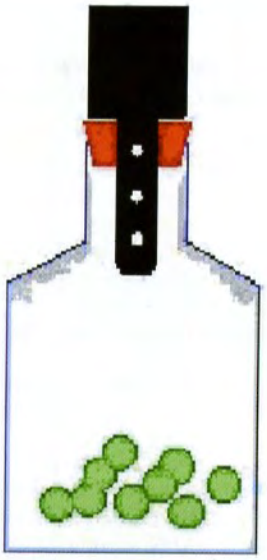


Figure 6 The 250 cm^3 bottle supplied with the sensor.

be used to demonstrate that after being fed with a drop of sugar solution, its metabolic rate increases. These observations would enliven any lesson dealing with the process of gas exchange between animals and their environment.

A significant point to note is that the orientation of the bottle should be kept the same when recordings are made, as CO_2 will concentrate slightly in the lower regions of the vessel.

Probe in a boiling tube

A substitute rubber bung must be created to allow the probe to be used in a boiling tube, as illustrated by Figure 7. This will provide a gas-proof seal between the probe and the air in the boiling tube.

The small air volume in the tube means that the response time will be as fast as possible. Very small animals are perhaps best studied in such a small chamber to increase the speed of result collection; a leaf can be rolled to fit inside the tube (Figure 3). The tubes can easily be illuminated to study the effects of light intensity or wavelength (wrap coloured sheets of cellophane around the tube) on photosynthesis, or warmed or cooled in a water bath to study the effects of temperature.

Small samples of soil can be placed in the tube, in order to measure the overall respiratory activity of the decomposers present – a major way in which carbon is recycled to the atmosphere. These soil samples can be modified by the addition of solutions of water, acids, lime, pesticides, fertilisers, and so

Figure 7 Dimensions for CO_2 sensor adaptor for a boiling tube – made by boring out the centre of a rubber bung.

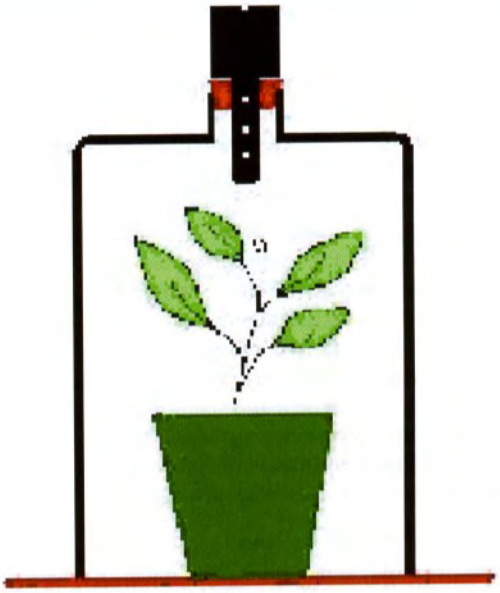


Figure 8 Sensor used with a bell jar on a glass sheet.

on, in order to assess how these affect the respiratory activities of the soil microbes.

Alternatively, a few cm^3 of a solution containing microbes, such as soil water, pond water, yeast cells in a sugar solution, non-sterilised milk, or bacteria growing in nutrient broth, seems to produce CO_2 at a steady rate that reflects the total populations of microbes.

Probe in a bell jar

For larger organisms, such as whole potted plants, a bell jar placed on a glass sheet should provide a useful container (Figure 8). With an air volume of several dm^3, the response time for the set-up will be slower than that for the smaller vessels. If soil microbes are to be kept out of the equation, then the plant pot of soil should be wrapped in plastic. This will give students a new appreciation of the value of potted plants on windowsills.

Probe in a plastic bottomless bottle

A plastic drinks bottle with the base removed and the probe fitted in the neck is useful for studying the carbon budget of patches of ground vegetation, and underlying soil, by simply pressing the bottom of the bottle onto the ground (Figure 9). The base needs to be pressed firmly onto the soil in order to create a seal. This has been used to compare sunlit patches of a lawn, with patches in the shade, and with bare

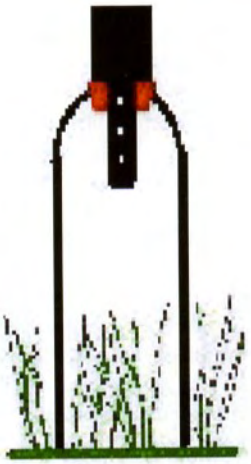


Figure 9 Sensor used with a bottomless plastic bottle.

soil. A brightly lit piece of a lawn will be a net importer of CO_2, whereas a part of the lawn in the shade may well be a net exporter, because the rate of photosynthesis in the grass leaves is not sufficient to exceed the respiration of the grass plants, their roots and the soil organisms.

Probe in a plastic bottomless bottle with plastic bag

A plastic bottle as above with a plastic bag sealed to its base can be used to isolate individual leaves or small branches on taller above-ground vegetation in order to study the carbon budget of the leaves still attached to the plant in the field (Figure 10). Larger and smaller clear plastic bottles can be adapted by cutting the necks in the appropriate place. This gives a fairly consistent volume of air surrounding the leaf. The plastic bag can be gathered up like a skirt, and stuck with tape, to form a reasonably CO_2-proof seal on the branch. Alternatively, home-made frames of fixed volumes could be made, using sheets of plastic and tape. Direct collection of CO_2 data from living plants in the field will make the whole concept of the carbon cycle much more real to students.

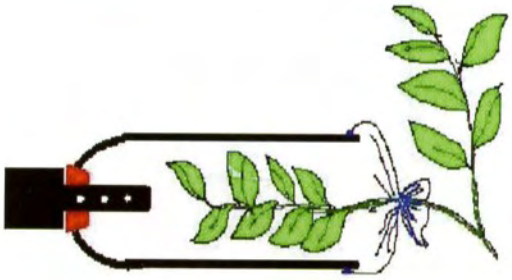


Figure 10 Plastic bottle and bag used to isolate a branch.

Probe in a metal sleeve

This enables the collection of CO_2 produced by the respiration of soil organisms. A simple steel cylinder (30 mm diameter by 150 mm long) accepts the probe and bung (Figure 11). The metal sleeve can be driven, say, 10 mm into the soil, thus providing a finite soil area for CO_2 sampling, before the probe is attached. Within a few minutes a trace of the CO_2 output of the soil will be obtained. This tool is particularly robust and can be easily cleaned for re-use.

An alternative is to collect soil samples, and carry out the measurements in boiling tubes, back in the laboratory – this would have the advantage of allowing varying amounts of water, pH buffers, and so on, to be added in order to assess the effects of soil moisture, pH, etc., on respiratory rates of decomposers. The respiratory rates of different kinds of soils and types of leaf litter, or, by excavating a hole, different soil horizons, can be compared.

A large proportion of the world's organic carbon is present in the soil as organic detritus, and the respiration of soil detritivores and decomposers releases a significant proportion of CO_2 back into the atmosphere. The warming of polar regions, including regions of permafrost where much of the world's peat is currently locked up in the ice, could lead to an accelerating greenhouse effect as the trapped carbon is respired. There is recent evidence to suggest that the UK soil is currently losing almost 1 per cent of its carbon each year, due to climatic change (Bellamy *et al.*, 2005), which presumably has been added to the atmosphere pool. Experimental studies on soil and peat respiration are currently highly relevant to this problem.

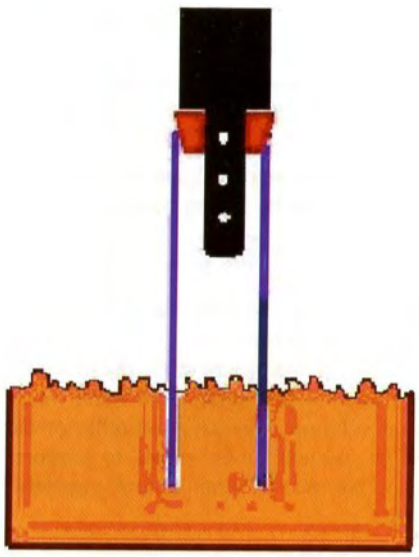


Figure 11 Sensor used with a metal cylinder to create a soil auger.

Using the CO_2 sensor, data-logger and software

A very strong reason for using the Vernier data-logging system is the exceptional quality of the *LabPro3* software that comes with it, and comes with a site and student licence. The following instructions give an idea of the use of the software with the CO_2 sensor.

Make sure all cables between sensor, data-logger and computer are attached, and that the data-logger is powered up. Then, when you start the *LapPro3* software running, it will recognise which probe is attached and will provide you with a blank graph already bearing the correct axes (time/seconds on

BOX 3 Suggested investigations using the CO_2 sensor

Effects of the following on leaf photosynthesis:

- CO_2 concentration
- temperature
- light intensity
- light wavelength
- daily rhythms
- stomatal density
- light/dark leaf adaptations
- herbivory
- action of herbicides
- C4 and CAM versus C3 plant leaves

Effects of the following on teaching room gaseous CO_2 concentrations:

- air-flow through the room
- number of people in the room
- biomass of people in the room
- physical activity in the room (compare a day when everyone is doing test, with another day)
- size of the room
- combustion sources of CO_2 (Bunsen burners)
- presence of plants

Isolate the different parts of a shrub in the field to study the carbon budget:

- in different aspects (shade-adapted leaves versus light-adapted leaves)
- at different times of the day
- in different weathers
- at different times of the year

Isolate the carbon budget of plant roots and associated soil, from the above-ground carbon budget (with a potted plant) to study investments in shoots/roots:

- when grown at different angles
- when mechanically stimulated/stressed

Effects of the following on respiration of small invertebrates:

- temperature
- activity (walking/flying)
- food sources
- stage of development
- species

Effects of the following on soil respiration:

- moisture content
- percentage organic matter
- temperature
- pH
- litter type
- mineral content
- mineral skeleton
- supplements of nutrients
- living plants
- organic and inorganic fertilisers
- pesticides
- salinity
- acid rain
- soil invertebrate population
- microbial populations
- season
- weather

the *x*-axis, and CO_2 level/ppm on the *y*-axis). At this stage, even before you collect any data, there will be a glow at the tip of the probe, about once every second, as it begins to register CO_2 levels.

To start collecting data, simply select the *Collect* button on the tool-bar at the top of the screen. The graph should immediately show data being collected. You can stop collecting at any time by clicking on the *Stop collecting* button. It is worth bearing in mind that there is a warm-up time of approximately 90 seconds before the data makes much sense. The usual default time for collecting is 300 seconds, but if you want to extend this time (which can be done whilst data is being collected), click on *Experiment* and then select *Extend collection* from the drop-down menu. If you wish to extend further, just keep selecting the same drop-down option. The scrolling columns on the left of the screen will show the numerical values of the readings as they are being taken.

You can enhance your view of the data as it is being collected by stretching or shrinking either or both of the *x* and *y* axes of the graph. To do this, move the cursor to just outside the border of the axis you wish to change (it will show a sort of squiggly line), and then right-click and drag away from or towards the origin of the axis to respectively stretch or shrink the axis of the graph. Don't worry if you have lost the scrolling data, you can move to where you want on the graph by clicking on the direction arrows found either side of the title for each axis. In this way you will be able to enlarge any portion of the developing graph to increase its dramatic impact.

Another very useful feature of the software is that labels, titles and other features can be added to the graph – even when data is still being collected. The most immediately useful analysis feature is the gradient function, which will get the software to calculate the slope of a best-fit line between any two points on the graph; and you can identify different parts of the graph for separate analysis. To do this simply highlight the portion of the graph you wish to study by clicking and dragging the cursor so that the area of interest is highlighted, and then click the *Linear Fit (R=)* button on the toolbar at the top of the page. A box will appear on the page, which indicates the slope as well as providing a correlation coefficient for the data in the region of interest. This box can be dragged to wherever it seems most appropriate on the page (if data are still being collected, the calculation will not be completed until the data collection stops).

If you select the *Stop collecting* data button, then you can save the file if you so wish. When you select *Start collecting* data again, the program will ask you if you want to erase the current data, or append the new data to the previous set. Normally you will want to start with a 'clean sheet', but occasionally you may wish to leave the previous trace on-screen and run the new trace over the same area of the graph; in this case you should select *Experiment* and then *Store latest run*. Then you can start to collect a new set of data, with the previous trace on screen in the background.

More investigations

Box 3 lists some suggestions for using the CO_2 sensor and data-logger for investigations both in and outside the classroom. These suggestions go well beyond the scope of fieldwork, but should give some idea of the range of uses to which the CO_2 probe might be applied.

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Useful websites

SAPS: www.saps.org.uk

Topical and up-to-date news on climate change: http://www.guardian.co.uk/climatechange/

Up-to-date commentary site on climate science by working climate scientists for the interested public and journalists: http://www.realclimate.org/

Hadley Centre for Climate Prediction and Research (part of the Met Office), provides a focus in the UK for scientific issues associated with climate change: http://www.met-office.gov.uk/research/hadleycentre/index.html

National air-quality archive allows students to see up-to-date data on precise hourly fluctuations in many major air pollutants in their own regions of the UK; excellent resource for data-analysis activities, allows urban and rural differences to be investigated: http://www.airquality.co.uk/archive/index.php

Simple animated carbon cycle for KS 3 and 4: http://www.purchon.com/ecology/carbon.htm

Concept map on the carbon cycle for KS 4+: http://www.schoolnet.edu.mo/general/biology/temp/cmap/carbon.html

Simple ecological footprint calculator: http://www.steppingforward.org.uk/calc/

More sophisticated footprint calculator: http://www.earthday.net/footprint/index.asp#

Suppliers

Instruments Direct (Services) Ltd, Unit 10, The Courtyard, Stenson Road, Coalville LE67 4JP Tel 01530 832 500. Website: www.inds.co.uk

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