What role does CO_2 play in root location by a soil insect?

Nicholas J. Hix[1], Xiaoxian Zhang[2], Peter J. Gregory[1,3] & Scott N. Johnson[1,3]

[1]School of Human & Environmental Sciences, Department of Soil Science, University of Reading RG6 6DW, UK.

[2]SIMBIOS Centre, University of Abertay, Dundee DD1 1HG, UK. [3]Scottish Crop Research Institute, Invergowrie, Dundee DD2 5DA, UK.

Scottish Crop Research Institute

The respiratory emission of CO_2 from roots is frequently proposed as an attractant that allows soil-dwelling insects to locate host plant roots, but this has recently become less certain for three reasons:
(a) CO_2 is emitted from many sources other than roots,
(b) spatially perceptible gradients from roots do not always persist because of high densities of roots in the upper layers and
(c) because soil contains more CO_2 than the atmosphere, vertical gradients are stronger than horizontal gradients from roots, which may also confound spatial orientation.

We investigated the role of CO_2 in root location by soil-dwelling neonatal larvae of the clover root weevil, *Sitona lepidus*.
Our experiments investigated:
1. the CO_2 concentrations in the vicinity of white clover roots,
2. whether larvae were attracted to point emissions of CO_2, and
3. whether trajectories (or movement paths) of larvae changed at various CO_2 concentrations.

Experiments

1. Rhizochambers were used to investigate CO_2 concentrations in the vicinity of white clover roots. Measurements were taken using a membrane-inlet mass-spectrometer (Hiden HPR-40).
2. Arenas (Fig. 1) were used to investigate the attraction of larvae to point emissions of CO_2 at different flow rates (5, 8, 16 and 32 ml hr^{-1}) for 30 individual larvae. The position of the larva was recorded at 10 sec intervals for 15 mins using an image analysis unit that incorporated a tracking algorithm.
3. Experiment 3 was identical to the above, except that CO_2 concentrations remained constant throughout the experiment (0, 380, 1000 and 2500 ppm). Larval trajectories were quantified using fractal analysis (see box 1).

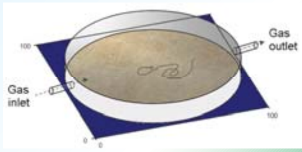

Fig. 1. *Arenas to investigate larval movement in experiments 2 and 3 (inlet/outlet sealed in experiment 2 which used constant CO_2 concentrations).*

Box 1: The Fractal Dimension (*D*)

The fractal dimension, *D*, of the trajectory (or movement path) is defined as:

$$L(\varepsilon) = k\varepsilon^{1-D} \qquad D \geq 1,$$

where $L(\varepsilon)$ is the total trajectory length and *k* is a positive constant. *D* = 1 is a linear trajectory, *D* values closer to 2 (the maximum) reflect increasingly tortuous trajectories.
- $D \rightarrow 1$ = movement **between** resource patches;
- $D \rightarrow 2$ = movement **within** a resource patch.

Results

Experiment 1

Mean CO_2 concentrations within 2 cm from white clover roots = 965 ppm

Experiment 2

There were no statistically significant differences in the numbers of individual larvae attracted to the control and to emissions of CO_2 for flow rates of 5ml hr^{-1} (P = 0.19), 8 ml hr^{-1} (P = 0.99), 16 ml hr^{-1} (P = 0.42) or 32 ml hr^{-1} (P = 0.42). Fishers exact *P* test used for analysis.

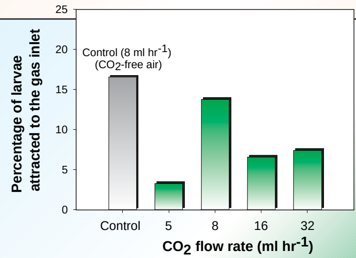

Fig. 2 *Percentage of larvae attracted to point emissions of CO_2.*

Experiment 3

Fractal dimension (*D*) values for larvae moving in different CO_2 conditions were significantly higher for larvae in the 1000 ppm arena ($F_{3,102}$ = 4.08, P = 0.009) than for other CO_2 conditions. This reflects more intensive searching of a smaller area at 1000 ppm.

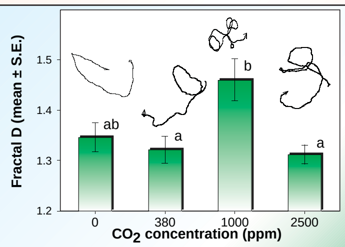

Fig. 3 *Mean fractal D values of larvae under different CO_2 conditions and examples of movement paths.*

Conclusions

- CO_2 is unlikely to act as an attractant for *S. lepidus* larvae for host location
- Searching intensified when larvae experienced CO_2 conditions similar to those around host plant roots (1000 ppm).
- CO_2 could therefore act as a 'search trigger' which indicates the potential presence of roots and initiates more intensive searches for more specific host location cues.

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