## Techniques

# An Environmental Cell to Control Simultaneously the Matric Potential and Gas Quality in Soil

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#### ABSTRACT

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A method is described for simultaneous control of matric potential, gas quality, and temperature in thin (2.5 cm) layers of soil. The apparatus permits easy sampling of the soil and soil biotica. The matric potential can be controlled between 0 bars (saturation) and approximately -1 bar; gas quality can be varied to include a single pure gas (e.g.,  $100\% O_2$ ) or any defined mixture of gases. The system uses low gas pressures ( $\leq 1$  bar) to control the matric potential and to circulate gas, thus preventing stagnation of the soil atmosphere. Once equilibrated, the cell can be maintained for

weeks without drying of the soil. Matric potentials can be adjusted while the system is operative, thereby allowing studies with soil under controlled wetting or drying. Repeatable soil moisture characteristic curves can be achieved and gas infiltration in emptied soil pores occurs within minutes. In addition to control of matric potential and gas quality in the soil, concurrent temperature control and illumination of the soil surface are possible by placing the cells in controlled environmental chambers.

Studies on the influence of water potential on the activities of plant pathogens and other microorganisms in soil are often limited by the lack of control of gas quality, most notably oxygen and carbon dioxide (1). Water and gas occupy the same pore space of soil and any change in water content or redistribution of water dictates a concurrent change in soil aeration (2). The greatest problem for researchers arises in wet soil where very small changes in matric potential can cause large changes in the pore space available for gas. As pointed out by Griffin (2), soil oxygen status rather than water potential is probably the relevant parameter determining behavior of microorganisms in wet soil. However, although techniques are available for maintaining soil water potentials down to about -1 bar, e.g., the pressure plate apparatus (3), few permit the simultaneous control of the soil atmosphere. Two systems have been described for simultaneous atmosphere and water content control (5,6), but neither allows for easy sampling of soil or biota in the soil, nor is it possible with the existing systems to either wet or drain a soil to the desired water content. The use of gas species under positive pressure will allow soil biologists to examine simultaneously the effects of aeration and of soil moisture and to determine the effects of these physical parameters on biological activity. This paper describes such a technique, including the means to sample the soil, for observations on the behavior of soil microorganisms.

## MATERIALS AND METHODS

Experimental cell. The cell used was a modified Tempe Pressure Cell (Fig. 1) (Soil Moisture Equipment Corp., Santa Barbara, CA). A water inlet port was added to the base of the cell and located at the edge of the water reservoir space under the porous plate. A gas outlet port was drilled in the top of the cell at a distance midway between the gas inlet port and the cell wall. Both the gas inlet and outlet ports were fitted with hose bibs. In addition, a gas sampling port with a replaceable rubber septum was placed in the cell wall as close as possible to the tension plate; this permitted

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sampling of the soil atmosphere with a gas-tight syringe. The syringe needle was modified to allow gas samples to be taken from a hole located in the side of the needle stem, as opposed to the distal end, to avoid clogging of the needle.

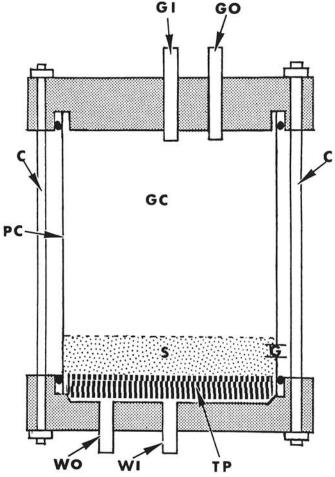


Fig. 1. Environmental cell. Clamp (C); gas sampling port (G); gas chamber (GC); gas inlet (GI); gas outlet (GO); pressure chamber wall (PC); soil (S); water inlet (WI); water outlet (WO); and porous plate (TP).

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Water system. A reservoir of sterilized, degassed water was used to continually wet the lower surface of the porous plate in the environmental cell (Fig. 2). The water was maintained at a pressure head of +0.5 cm above the center of the porous plates because it provided a convenient flow of 0.5 ml per minute per cell. All porous plates were positioned at the same height relative to the water reservoir. A single reservoir (10 L) supplied water to all cells for about 3 days. Water was allowed to flow through the space under the tension plate and discarded. This system permitted soil to be wetted and prevented soil from drying out during incubation at a constant matric potential.

Gas system. Pressurized gas was used to control the matric potential in the soil. Any gas or mixture of gases can be used. The pressurized gas was passed through two regulators (Fig. 2); the first reduced the tank pressure to 0-10 MPa and the second regulated the pressure from 0-100 KPa. This system reduced the flow rate and allowed conservation of the gas(es). One tank of gas was used to operate a number of cells. Each cell was maintained at a uniform pressure by a manifold (Small Parts Inc., Miami, FL), which split the single gas source equally. Gas passing into the cells was exhausted through the gas outlet ports, collected by a second manifold, and conducted to a mercury manometer (100 cm × 5 cm) constructed of Plexiglas (Fig. 2). The exhausted gas was forced down into the mercury via a glass tube held rigidly in place. The top of the manometer was plugged with porous foams, which prevented mercury from splashing out, yet allowed gas to escape. The manometer was operated in a fume hood. The matric potential was adjusted by lowering or raising the depth to which the glass rod extended into the mercury.

**Operation.** The porous plates were soaked in sterilized, degassed water for 48 hr and then placed in the modified Tempe Cell. The pressure in the water reservoir was adjusted to permit a constant flow (0.5 ml/min) beneath the porous plate. All air was removed from the space beneath the porous plate before adding soil to the cell. The maximum soil depth was 2.5 cm. The soil was allowed to wet to saturation (12 hr for a silt loam). The experimental cell was then tightly sealed, and the matric potential in the soil was adjusted by positioning the glass rod at the desired depth in the mercury manometer and by supplying sufficient gas to cause bubbling in the mercury. The matric potential  $(\psi)$  could be calculated by measuring the depth X the rod extended into the mercury (Fig. 2).

$$\psi \text{ bars} = X \text{cm} / 76.6 \text{ cm Hg.b} \tag{1}$$

Test material. Biological samples could be incorporated into the soil before placement of the soil in the cells, or buried in the cell while adding the test soil. The experimental cell permits a determination of the combined effects of water potential and oxygen diffusion on saprophytic growth of microorganisms in controlled but not stagnant environments. Test material can be sampled repeatedly from the same cell, or from one entire cell in a multi-cell system without disturbing the remaining cells, depending on the test material. Sampling requires that pressure be released and the cell opened long enough to collect a sample. The sampling process requires only minutes to complete, thus this process will have little effect on the soil water potential during the course of an experiment.

Soil moisture and gas infiltration determinations. A silt loam soil from Lind, WA, was used for characterization in the experimental chamber. The soil was air-dried, sieved through a 2-mm-mesh screen, and autoclaved twice for 2 hr (1.6 kg pressure/160 C). The prepared soil was placed in the cells to a depth of 2.5 cm and leveled by gently agitating the cell. The soil was then allowed to wet until saturated.

Time for saturation was determined by sampling the soil after 12, 18, 24, 36, 48, and 72 hr. At the prescribed times, three soil samples (1-2 gm) were taken from each of four cells and dried at 110 C for 48 hr to determine the water content. Water contents also were determined at several pressures for soil being wetted and dried. For adjustment of the matric potential during the draining

of soil pores, the soil was initially saturated, then drained by increasing the gas pressure in stepwise increments every 72 hr. For soil being wetted, gas pressure was decreased on the same schedule after that which had initially been saturated was dried to -0.95 bar. Three samples from each of four cells were taken 24, 48, and 72 hr following a pressure adjustment. Using both sets of data describing the soil water characteristics, the magnitude of hysteresis for the soil can be described.

Pure oxygen was used as the pressurized gas to determine when gas infiltrated soil pores drained of water. Saturated soil was drained to 25, 35, 45, and 50 —mbar. Forty-eight hours after adjustment of the gas pressures, gas samples  $(1-10\,\mu\text{l})$  were taken using a gas-tight syringe inserted into the soil,  $(2\,\text{cm})$  beyond the gas sampling port (Fig. 1). The samples were analyzed for nitrogen, oxygen, and carbon dioxide using a column  $(0.6\,\text{cm}\times1.8\,\text{m})$  of molecular sieve 5A (Applied Science Products, Fairfield, CA) and a hot-wire detector at 150 ma. A small amount of air was entrapped in "saturated" soil. All gas measurements were performed by gas chromatography using a helium carrier at 30 ml/min at a temperature of 50 C. Four cells were used in this experiment (Fig. 3).

## RESULTS AND DISCUSSION

Saturation of silt loam (2.5 cm thick) before the adjustment of the matric potential required 12 hr. A layer I cm thick required about 4-6 hr. Equilibration times after adjustment of the matric potential will vary with the soil used and the thickness of the soil layer. The modified pressure plate apparatus (3) requires similar equilibration periods. Soil moisture in silt loam (measured gravimetrically) 24 hr after a change in the pressure potential, was significantly different than the moisture content in samples taken after 48 and 72 hr, indicating that water potential equilibration for longer than 24 hr was required (Fig. 3). There was no statistically significant difference in water content between samples taken 48 and 72 hr after pressure potentials were adjusted. Soil water potentials in each of the four replicated cells were similar. It is important to realize that in this system as with any other, a thorough knowledge of the soil moisture characteristics for a soil is essential before valid interpretation may be made concerning the effects of the water potential on the activity of the test material.

One advantage of pressurized gas is the ease in obtaining uniform matric potentials more negative than (-0.3 bar. Another is that any gas species can be used. The vertical height required for

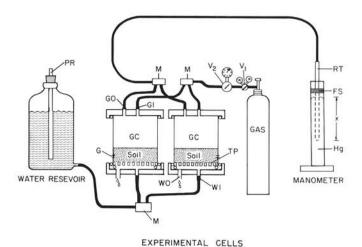


Fig. 2. Diagram of environmental cell system for the simultaneous control of matric potentials and soil aeration. In the manometer: foam stopper (FS); rigid tube (RT); and mercury (Hg); the depth RT is immersed into the Hg (X). In or connected to the experimental cells: gas chamber (GC); gas inlet (GI); gas outlet (GO); gas sampling port (G); manifold (M); porous plate (TP); water inlet (WI); water outlet (WO); and V<sub>2</sub> and V<sub>1</sub>, pressure regulator values. The water reservoir is maintained at ambient pressure by an open tube PR.

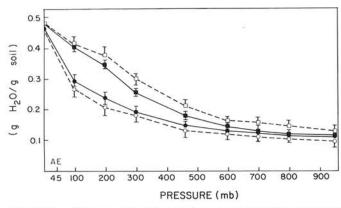


Fig. 3. Soil moisture characteristics and air-entry value of a Shano silt loam in environmental cells. Squares represent adjustments from saturation downward (drying curve) and circles from dry soil upward to saturation (wetting curve). Water contents at 24 hr (open symbol) and 48 hr (solid symbol) after adjustments to different pressures (mbar). Values are averages of eight replicates and standard deviations are indicated. The potential at which air entered (AE) the soil was determined by using oxygen as the pressurized gas in the system.

this system is slightly less than 1 m. For example, a pressure of -1 bar required a column of mercury 76.6 cm. By comparison, a hanging water column requires 3 m and 10 m to produce matric potentials of -0.3 bar and -1 bar, respectively. However, in the environmental cell system, the nature of the porous plate limits the matric potential obtainable. The porous plate used in this system had an air-entry value (AE) of about 1 bar; thus matric potentials were limited to -1 bar. The air-entry value is that pressure that will force water from sufficient pores, thereby allowing air to pass through the porous plate. All porous materials have an AE. Two ceramic porous plates are available: high flow (0.5 bar AE); and low flow (1 bar AE). By using the high flow plate, equilibration time could be reduced.

Gas effects can also be examined at numerous matric potentials that are more negative than the air-entry value of the soil used. For example, an anaerobic environment established with nitrogen could be used to determine the effects of water content on the growth or survival of anaerobic organisms at matric potentials not usually associated with anaerobiosis. In addition, the O<sub>2</sub> and CO<sub>2</sub> concentrations in the gas can be varied while maintaining a constant matric potential or vice versa. Using this approach, one

could test Griffin's (2) idea that O<sub>2</sub> and not water potential is the limiting factor of microbial activity in wet soil.

Another advantage of the system is the ease of maintaining constant conditions for extended periods of time. Hanging water columns must be sealed at the top to reduce evaporation, but this creates a stagnant atmosphere both in and above the soil and precludes long-term experiments. In the environmental cell, gas is flowing above the soil, preventing stagnation, and water is flowing beneath the tension plate, preventing soil dessication.

Gas movement into a soil as it drains occurs only when the air-entry value of the soil is exceeded by the pressure applied to the cell (Fig. 3). This limitation is inherent with all soils. Air-entry values will depend on the soil pore geometry (4). Again, each soil should be characterized to determine the matric potential at which gas infiltration starts. This measurement can be made readily by using a gas other than air and sampling for it through the gas sampling port as pressure is increased in saturated soil. In the silt loam soil it was determined that gas would enter the soil pores to a depth of 1.5 cm from the surface only after matric potential was decreased to -0.045 bar. At a greater potential energy, the water in soil pores prevented a significant amount of gas from entering the soil profile (Fig. 3).

This technique will not be useful for very short-term studies (e.g., < 12 hr) because soil water equilibrium will take about 24-48 hr, depending on the texture and thickness of the soil layer. Long-term studies, for which this system may be most useful, include survival of pathogens in crop residue, breakdown of organic matter in soil, microbial parasitism, and other soil-pathogen interface studies of interest to plant pathologists and soil microbiologists.

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