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# Trasor, a transit time portable system for measuring soil respiration

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## Abstract

Magliulo V., Renella G., 1997. TRASOR. A Transit Time Portable System For Measuring Soil Respiration. *Comput. Electron. Agric.* A portable instrument (TRASOR: TRANSIT-time SOil Respirometer) dedicated to the measurement of the soil respiration in situ was assembled using a commercial infrared gas analyzer and a data logger. A closed system, TRASOR has a principle of operation resembling that of transit time leaf porometers, widely used in the 80s and early 90s. Transients may be monitored in real time in a graphical form, allowing for immediate detection of system malfunction or non-linearity of data. The comparisons made showed that TRASORs respiration rates were comparable to those measured with commercial systems. © 1997 Elsevier Science B.V.

*Keywords:* Infrared gas analysis; Soil respiration; Field measurements

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## 1. Introduction

The soil is a significant part of the terrestrial surface involved in the exchange of CO<sub>2</sub> and it holds an important role in the planetary carbon cycle. The measurement of the respiration rate of the root-soil-organisms system is in this regard of importance in order to quantify the Carbon budget in natural and agricultural ecosystems (Norman et al., 1992).

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The respiration data in relation to environmental parameters also provides useful indications about the biological state of the soil. The metabolic ratio, or specific respiration (Pirt, 1975; Anderson and Domsch, 1993), can highlight a stress factor acting on the soil biomass or reveal the evolutionary degree of an ecosystem (Insam and Haselwandter, 1989).  $\text{CO}_2/\text{organic-C}$  ratio is another parameter that may quantify a stressful condition on the soil biomass, leading to a low efficiency in the use of the soil organic matter.

TRASOR is a portable dedicated system developed for the measurement of the rate of soil  $\text{CO}_2$  emission in situ, featuring the following characteristics: total automation of the measurement and data recording; configurability of the length and number and graphic monitoring of the transients; ease of replication—using any portable gas analyzer with 1 ppm accuracy and 1 s response time, and a datalogger—at low cost.

## 2. Closed versus open systems for soil respiration measurement

In closed systems, the biological system under study is enclosed in a chamber (cuvette) of known volume and a sub sample of the air contained in the enclosure is fed to the infrared gas analyzer (IRGA). Assimilation or respiration rates may be established by calculating the slope of carbon dioxide concentration ( $[\text{CO}_2]$ ) over time, and extrapolating to the initial time (Reicosky et al., 1990). The measurements are performed in short time spans, to avoid excessive system disturbance. They are sensitive to air leaks, which must be carefully minimized. The main objection leveled at closed systems is that because  $[\text{CO}_2]$  builds up in the enclosure, steady-state conditions are never reached during the measurement. This is of importance for monitoring leaf activity but not for soil respiration where the magnitude of the gradient driving the diffusion process is little influenced, since soil  $[\text{CO}_2]$  in the soil surface layers is typically several thousands parts per million.

Steady state open systems perform measurements under equilibrium conditions. A rather smaller enclosure is subjected to a measured continuous air flow, and  $[\text{CO}_2]$  in the air entering and leaving the cuvette is monitored for mass balance calculations. Leaks outward do not pose problems, since part of the mass may escape the system without causing errors, provided that the flow rate entering the cuvette, which is slightly pressurized to avoid inward leaks, is measured. Open systems perform measurements at steady-state conditions, i.e. when no variation in time of state variables occurs. Since some kind of enclosure must be used to isolate a portion of the system, the original environment is somewhat disturbed while steady state conditions reestablish, and the new equilibrium may differ substantially from the original state. In the case of soil, temperature and moisture content are likely to be affected.

Perhaps the key factor to be controlled for correct soil respiration estimates is cuvette internal barometric pressure. Differences of the order of microbars between the inside and the outside of the chamber can cause significant errors in flux measurements (Kimball and Lemon, 1971; Kanemasu and Sij, 1974).  $\text{CO}_2$  diffusion

rate was found to be positively correlated to the air flow provided by the chamber air stirring fan, if this was blowing upward, negatively if it was placed to blow downward (Rick Garcia, personal communication). A positive pressure of 30–40  $\mu$ bar, caused by wind blowing toward the vent port of the cuvette, was sufficient to inhibit any measured  $\text{CO}_2$  evolution in multiple open soil cuvettes, sequentially scanned in an automated configuration (Werner Kutsch, personal communication).

A vent port, with a capillary tube attached or a pressure relief valve, should be present in closed system cuvettes, to allow for pressure equilibration. While this implies a small leak, in most cases it is not important with respect to the fact that the soil does not make a tight seal at the bottom.

Open systems seem to be more prone to these causes of errors, beside requiring longer to reach equilibrium conditions. For such reason, they are better suited to prolonged observations at a given site rather than being moved around for fast, repeated measurements, aimed to average out soil spatial variability. In this regard, they also lend themselves to be scanned in sequence in unmanned automated operations.

Another problem is posed by the evaporation from the soil surface, during a measurement, when in very wet conditions. Interference of water vapor with the absorption of infrared radiation is almost entirely eliminated by optical filtering in front of the IRGA detector. Dilution and even band-broadening effects may be corrected for via software (Burch et al., 1962; Wolfe and Zissis, 1985) but the cuvette may soon become pressurized. This factor is more of concern in closed systems - the cuvette air is continuously refreshed in steady state apparatus and excess water vapor removed - and constitutes another reason for venting the cuvette to allow equilibration with outside pressure. Cuvette vapor pressure should therefore be controlled or at least monitored, and measurements should not last longer than 60–90 s.

From the above discussion, it can be concluded that, for most practical applications, closed systems should be preferred to assess soil respiration rates. They can be made portable and moved around the site under study for repeated and short time samplings. If properly designed and managed, closed system soil respirometers are at least as accurate and dependable as open systems.

### 3. A comparison with commercially available closed system soil respirometers

At the present time, most popular commercial soil respirometers are the LI-COR 6200 (LI-COR, Lincoln, NE) and the PP Systems SRC-1 (PP Systems, Hitchin, Hertfordshire, UK) which are both closed systems.

The LI-COR 6200 is made of a stainless steel soil chamber (model 6000–09), housing the temperature and humidity sensors and connected to the IRGAs console. After installing the chamber on the soil, the internal scrubber is manually turned on and  $[\text{CO}_2]$  is drawn below ambient concentration. Upon turning off the scrubber, the soil respiration causes  $[\text{CO}_2]$  to rise. The rates are logged at regular intervals and the flux is calculated by regressing rates measured in a range spanning

the ambient  $[\text{CO}_2]$  (Reicosky et al., 1990). The procedure must then be repeated several times to ensure that the measurements are repeatable.

A drawback of this instrument, which was not originally designed for operation with a soil cuvette, is that  $[\text{CO}_2]$  drawdown cannot be monitored on the LCD display, so that a guess has to be made about the proper time to switch. A useful feature is on the other hand, the possibility to control the water vapor partial pressure by diverting part of the flow through a humidity scrubber column. At high soil moisture content, evaporation may in fact cause overpressure in the cuvette and interfere with  $[\text{CO}_2]$  measurement, as previously discussed.

The PPSystems instrument, based on the work of Parkinson (1981), performs transients in series:  $[\text{CO}_2]$  is scanned every 8 s and statistical analysis is performed following the third reading (about 20 s). A quadratic equation is fitted to data to determine the rate of increase at time zero, and the user is warned in the case of excessive non linearity. The width of the scans is fixed and monitoring the process is limited to the numerical display of chamber  $[\text{CO}_2]$  and calculated rates, every 8 s.

If repeated readings are needed,  $[\text{CO}_2]$  may increase up to 60 ppm above ambient. Since  $\text{CO}_2$  scrubbing is not possible, repeating the measurement implies extracting the cuvette and venting it until ambient  $[\text{CO}_2]$  is reached. Reinserting in the soil may cause a disturbance that affects the successive measurements.

The transit-time principle of operation constitutes the main difference between TRASOR and the above described instruments and overcomes some of their limitations.

## 4. Description of TRASOR

### 4.1. Principle of operation

Transit time leaf porometers have been popular instruments among plant physiologists, in the last decade. They are aimed at measuring plant stomatal conductance by monitoring humidity changes in a cuvette clamped to a leaf. The LI-60 diffusive resistance meter was a simple device developed by LI-COR (Lincoln, NE), following the papers by van Bavel et al. (1965); Kanemasu et al. (1969); Kanemasu and Sij (1974). To make a measurement, the sensor cup was placed over a leaf and the time of needle movement between two points of an arbitrary scale analog resistance meter was recorded with a stop watch. The cup was then dried by manually pumping air forced through a drying tube containing silica-gel, and the cycle repeated several times. Leaf stomatal conductance was later calculated by referring to a calibration developed in the laboratory on a resistance plate having pores to simulate leaf resistance.

An evolution of this instrument is the MK-3 automatic porometer by Delta-T Devices (Cambridge, UK). Again, the water vapor emitted by the transpiring leaf surface causes the relative humidity (RH) within the cup to rise, but in this case the instrument automatically times the RH rise over a fixed interval. Cycles are

achieved by means of a pump blowing dry air into the cup after each timing measurement and taking the RH down below the timing interval starting point. Calibration procedures are similar to those used for the LI-COR instrument.

The scheme of operation for TRASOR is identical in principle to the Delta-T porometer. The CO<sub>2</sub> transients (lasting typically 2 s for the Delta-T porometer) are slightly longer since air samples must be sent to an external sensor, the cell of the IRGA, and the volume of the cuvette acts as a buffer.

#### *4.2. Parts and connections*

TRASOR is composed of a 10 cm diameter, 14 cm height plexiglas soil cuvette and a case (Fig. 1). The case houses an IRGA, a data logger, a 2.5 l/min air pump, a 2-way valve, a flowmeter, flow regulators, and a CO<sub>2</sub> scrubber column. The valve (ETO-3-12, Clippard Instruments Laboratory, Inc, Cincinnati OHIO, USA) is switched via a solid-state relay when a digital port of the CR10 data logger (Campbell Scientific Ltd, Shepshed, Leics, UK) is set high under program control. In the same way, the air pump (NMP 30 KNDC, KNF Italy, Milano, Italy) is switched off via software, after the end of the measurement. A further digital channel of the logger is set high when the measurement is completed, to short circuit two selected channels of the 15 pins D-type IRGA output connector, allowing for the auto-zero routine to be performed, as described later. Four of the data logger analog input channels are connected to the soil temperature probe type thermocouple (Omega Engineering Inc., Stamford, CT), IRGA output signal, cuvette thermistor air temperature and capacitive relative humidity probe (model Hi-3602 A, HYCAL, El Monte, CA). Other sensors may be connected to the remaining available channels.

The IRGA is a single board device (SBA-2, PPSystems, Hitchin, Hertfordshire, UK) with built-in infrared sensor cell featuring a 0–1000 ppm measuring range and an accuracy of 1% of full scale. Such specifications are sufficient for TRASOR operation, since absolute determination of CO<sub>2</sub> partial pressure is of little concern and measurements must be performed at ambient concentrations.

The electric power is provided by one or two 2-A-h rechargeable batteries which may be fitted in the bottom of the case for 1.5 or 3 h of continuous use respectively, while a 9 V alkaline battery provides backup power for data logger program and data.

The dataloggers external keyboard/display or a notebook PC are used to control the measurements, although the first option does not allow for graphical monitoring of the transients.

#### *4.3. Software description and system functioning*

The data logger program was developed using 6.2 EDLOG data logger programming software (Campbell Scientific Ltd.). When the program is downloaded into the logger via the PC serial port, default values of variables are loaded but the program is not executed until a valid plot number is entered via keyboard.

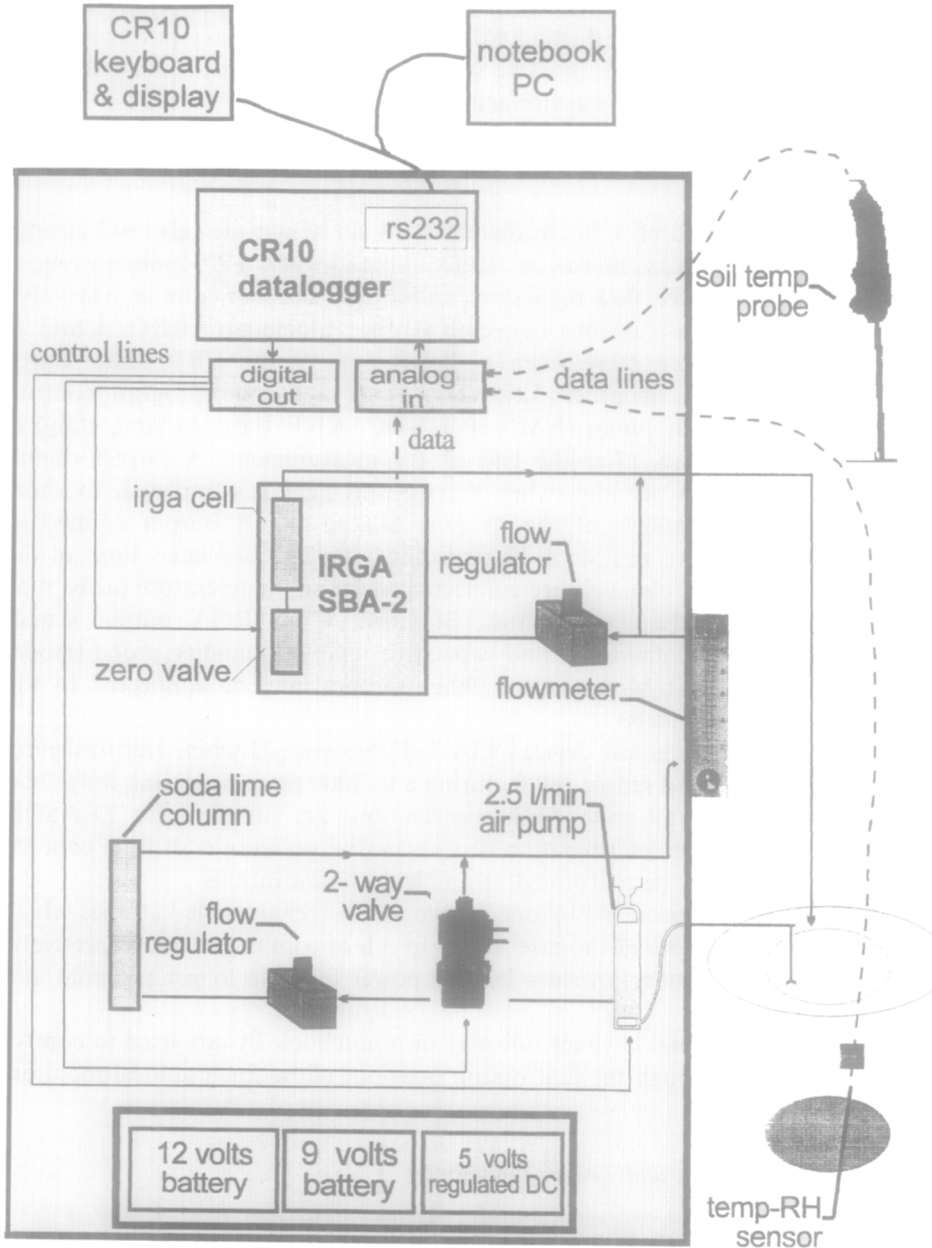


Fig. 1. Block diagram showing parts and connections of TRASOR. Air tubings are thick lines; digital signals thin lines; analog sensor outputs dotted lines. 12 V DC power (connections not shown) goes from battery to valve, air pump, IRGA and datalogger; 5 V DC power to temperature-RH probe.

A subroutine may be initiated to measure  $[\text{CO}_2]$  at the soil surface. The knowledge of the environmental  $[\text{CO}_2]$  is useful to set the upper and the lower setpoints in order to bracket the measured concentration, as suggested by Norman et al. (1992), to minimize  $\text{CO}_2$  leaks by diffusion along the concentration gradient.

When a measurement is started, a preset number of transient cycles are performed, before mean data are output to final storage. A typical sequence of transients is shown in the bottom chart of Fig. 3, a screen image captured by the Campbell Scientific Gpaphterm PC-210 software communication package.

When the cuvette is gently driven in the soil, the  $\text{CO}_2$  produced by macro and microorganisms respiration activity diffuses into the enclosure. The enriched air is circulated in the system by the pump and a 0,5 l/min subsample is fed to the IRGA. When the cuvette  $[\text{CO}_2]$  exceeds the preset upper threshold, the valve is switched and the flow is diverted to pass through a soda-lime scrubber column, with the effect of a fall in  $[\text{CO}_2]$ . The speed of drawdown may be controlled by the flow regulator to avoid depletion taking place too fast when respiration rates are low, since, for the required accuracy of timing, each transient should not last less than 10–15 s.

When the  $[\text{CO}_2]$  reaches the lower threshold, the valve is switched back to the 'enrichment position' so that concentration in the system builds up again. The elapsed time to travel between the lower and the upper setpoint is measured and recorded and the cycle is repeated until the completion of the desired number of replicates.

The transit time and the average values of soil temperature, air temperature, and relative humidity are sent to final storage at the end of each transient. The mean transit time and its standard deviation across the replicates are output at the end of the measurement.

At that time the analyzer is allowed to auto-zero, a built-in routine automatically executed whenever a null voltage is supplied to pin 6 of the IRGA output connector. The analyzer's built-in valve is switched to route the incoming air through a scrubber column (not shown in Fig. 1), then into the cell. After a further 30 s, the pump is switched off to save on battery power. The program will no longer be executed until a new plot number is specified.

The transients may be graphically monitored with the Gpaphterm software, if a notebook PC is used instead of the CR10 external keyboard. This has the advantage that non linearity in the transients and other visual anomalies may alert the user to the presence of leaks or of system malfunction. If the slope of  $[\text{CO}_2]$  increase differs among the transients, such non-steady state conditions during the measurement are likely to be due to disturbances induced by the cuvette on the natural process of  $\text{CO}_2$  diffusion.

#### 4.4. Calibration

Respiration rates can be calculated in real time by applying the  $\text{CO}_2$  mass balance, via the ideal gas law, knowing air temperature and the total volume of cuvette, IRGA cell and tubing.

However, the authors believe that a more appropriate approach, for collecting reliable data, is to a field calibration as follows. The cuvette should be placed on a layer of sterilized sand, under field operating conditions of  $[\text{CO}_2]$ , wind, temperature etc. A precision syringe pump (or mass flow regulator), filled with pure  $\text{CO}_2$  should then be connected to a short tube whose other end is placed underneath the cuvette, on the top of the sand. Different  $\text{CO}_2$  delivery rates may then be applied, spanning the range of expected soil respiration rates. Under these conditions, the  $\text{CO}_2$  artificially added by the syringe, effectively simulates the actual soil activity, so that rates of cuvette  $[\text{CO}_2]$  increase, as measured by TRASOR, can be regressed on  $\text{CO}_2$  injection rate, to yield a calibration function. This can be incorporated in the datalogger program, or simply used to back-calculate fluxes on the basis of raw data measured in the field.

The calibration procedure described offers several advantages. The uncertainties connected with determining total system volume for the various configurations the user may employ (different cuvettes, IRGA, dead volumes), are no longer a problem. Every possible cause of systematic error—especially leaks, including that at the vent port—is accounted for. The procedure may be repeated for different environmental conditions, to yield a family of calibration curves.

## 5. Field trials

A prototype version of TRASOR, featuring a Campbell Scientific 21X logger and an ADC LCA-2 portable IRGA (Analytical Development Company, Hoddesdon, UK), logged field data on a potato crop during the 1995 growth season on a sandy soil at about 25% volumetric soil moisture content. Measured rates were lower in carbon dioxide enriched (FACE) plots than in controls (8.64 and 12.01  $\mu\text{mol}/\text{m}^2/\text{s}$ , respectively;  $\text{LSD}_{0.05} = 2.00$ ).

A non replicated comparison trial was made on the occasion of the International Workshop on Field Techniques for Ecophysiology held in Rapolano (Siena, Italy), during September 1995, between this TRASOR prototype and a Li-6200 system operated by LI-COR Instruments staff.

Two patches of grass were cleared of the sod, and the upper soil layers loosened. A Motorola Venturi pressure differential device (Motorola Ltd., Milton Keynes, UK), with a range of 0–1000  $\mu\text{bar}$  and a resolution of 1  $\mu\text{bar}$  was connected to a plastic tube protruding from the soil in the center of the plot. Measurements were taken on the plot # 1 with both instruments. The equipment was then moved to plot # 2 and the comparison repeated immediately, then again after 60 min when the burst of  $\text{CO}_2$ , due to mixing the soil, was declining. A fan was placed blowing on the cuvette at the distance of 2 m, to investigate the influence of wind on the cuvette internal barometric pressure. Measured rates were similar for the two instruments (Fig. 2).

Pressure fluctuations (Fig. 3, middle chart) were negligible in the TRASOR cuvette, which is fitted with a 0.5 mm internal diameter capillary tube attached to the vent port. A mean depression of about 90  $\mu\text{bar}$  (Fig. 3, top chart) however,

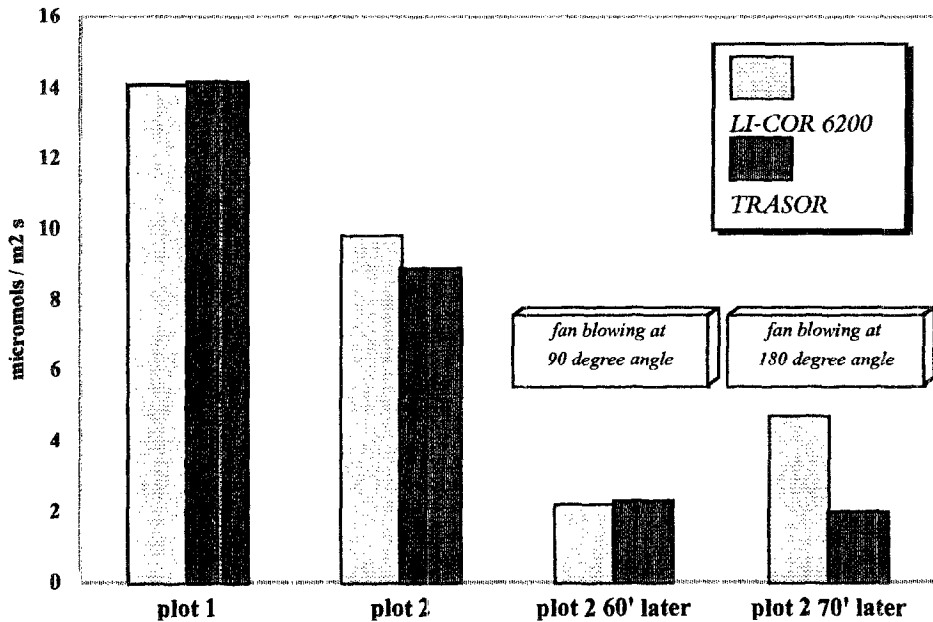


Fig. 2. Results of a comparison trial made at the 'International Workshop on Field Instrumentation for Ecophysiological Measurements' between TRASOR and a LI-COR 6200 system.

developed in the LI-COR cuvette when the built-in pressure relief fitting was oriented opposite to the fan, so that measured rates were doubled.

A further comparison was made, under laboratory conditions, of present day TRASOR with a PP Systems SRC-1. Six pots were prepared containing a mix of sand and organic topsoil in various proportions, to yield a range of different respiration rates. Such a range was further extended, after performing a set of measurements, increasing soil temperature by putting the pots under a 1000 W metal halide lamp. Alternated measurements with the two instruments were taken on each pot. Results are shown in Fig. 4.

Rates were correlated ( $r = 0.74$ ) but data are scattered around the 1:1 line. This is due to the different diameter of the cuvette between the two instruments, which caused soil disturbance following each reading on a pot and biased the rates measured by the successive unit. This can not be eliminated other than using the same cuvette for both instruments, which would make the comparison less meaningful, or by randomly using each unit for the first measurement on a pot. The latter approach was adopted, but data dispersal was a consequence.

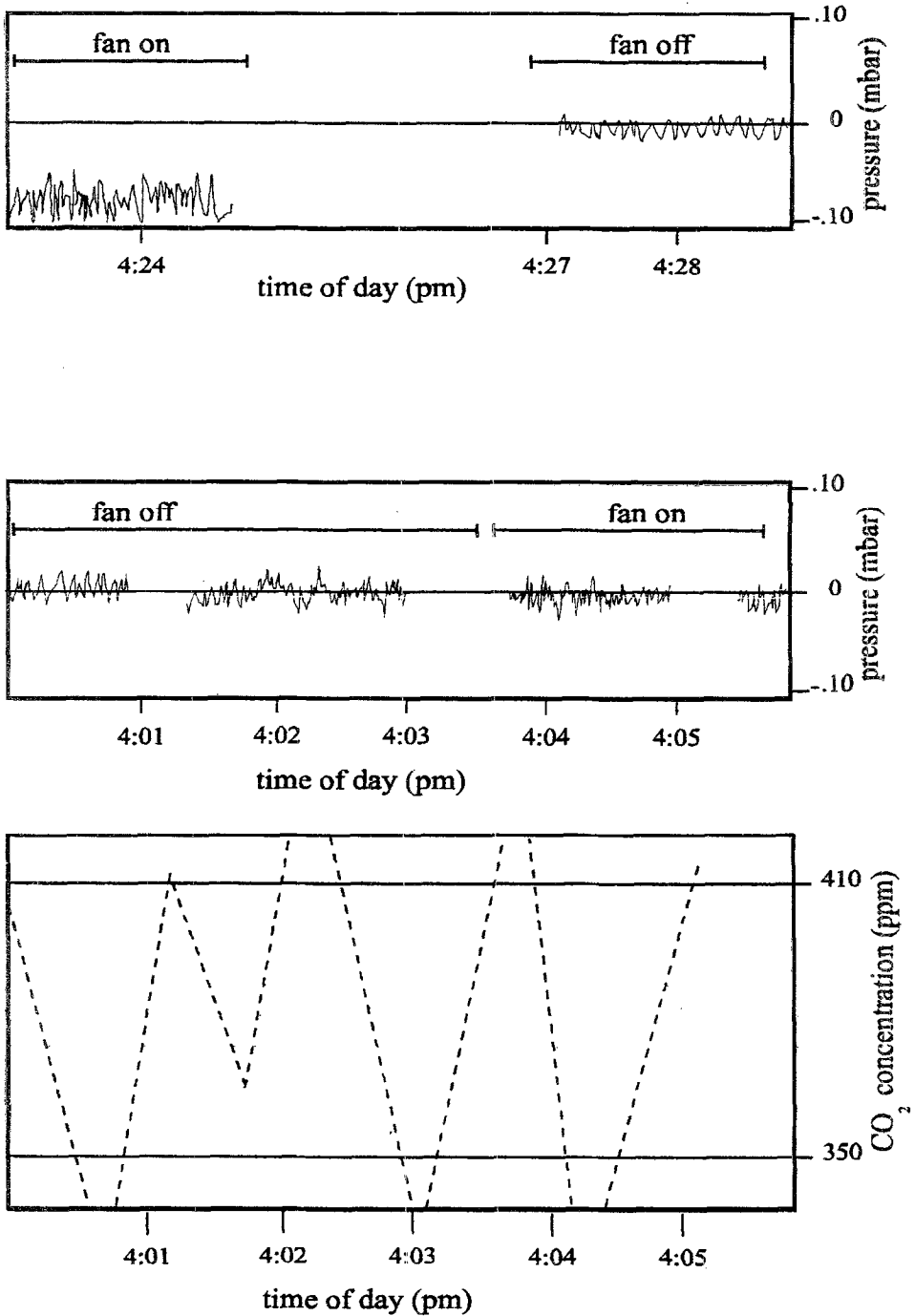


Fig. 3. Chart of LI-COR 6200 (top) and TRASOR (middle) barometric pressure difference between the inside and outside of the soil cuvette, during the last two measurements of Fig. 2. The left side of the upper chart refers to a measurement made with a fan blowing at right angles with respect to the pressure relief valve; the right side to the measurement repeated without moving the cuvette, but with the fan angled at 180°. The bottom part of the figure shows the trend of the CO<sub>2</sub> transients of TRASOR.

## 6. Future developments

A more compact and simplified version of TRASOR is being developed, which is entirely PC-based. The four analog channels built in the SBA-2 IRGA board, which are accessed at the output connector (Section 4.2) are used to accept external sensors signals. The board software has been factory modified to output, every second, a string with the IRGA reading and such voltages on the serial communication output pin, to a notebook PC RS-232 port. A Labview 4.0 (National Instruments Italia, Milan, Italy) executable file is run to pilot the system and provide the graphical interface and data acquisition. Two of the pins of the PC serial port are used to switch valves and pumps. Notebook requirements are 486 or Power Macintosh CPU, with at least 8 Mb RAM.

Soil moisture content is an important extra variable to monitor. The  $\text{CO}_2$  evolution rate is in fact strongly dependent on the water content, since it affects the microbial respiration and the release of the  $\text{CO}_2$  from the soil solution. A probe such as Delta-T NC1 (Delta-T Devices, Cambridge, UK) can be used with TRASOR to provide real time estimates of the average soil water content in the top 0.1–0.15 m. Such probe is based on the ‘time domain reflectometry’ principle, a technology originally developed for cable testing and later adopted to soil moisture content determination (Topp et al., 1980).

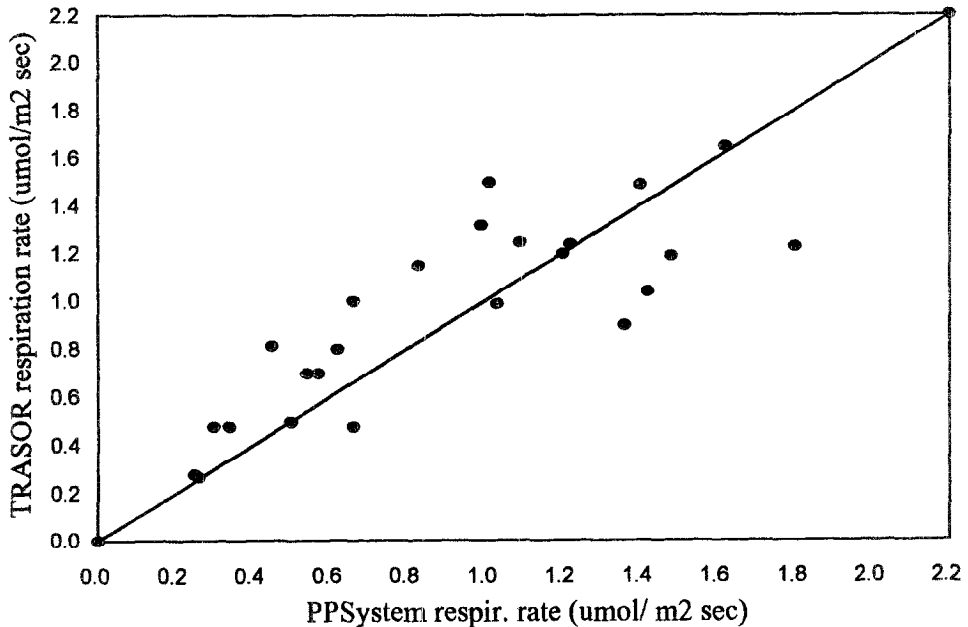


Fig. 4. Results of a laboratory comparison trial between TRASOR and a PP Systems SRC-1.

## 7. Conclusions

TRASOR offers advantages over existing systems. While made of parts frequently available in ecophysiology laboratories, it can be made self-contained and lightweight, less than 4 kg, allowing for a single person operation. The transit-time principle of operation, allows for repeated short-time transients to be performed in sequence and graphically monitored. Set-points may be modified in real time to adapt to different respiration rates and chambers with different area/volume ratios may be used for the same optimization purpose.

Comparisons of this relatively low cost instrument made against a commercial system, showed that their performances were similar.

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