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Ratio of CO_2 and O_2 as index for categorising soil biological activity in sugarcane areas under contrasting straw management regimes

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Abstract. This study was developed in a sugarcane area under contrasting management regimes defined by mechanical green harvesting (GH) and burning harvesting (BH) to test the hypotheses that the ratio of carbon dioxide (CO_2) and oxygen (O_2), known as the apparent respiratory quotient (ARQ), can be used to categorise soil biological activity. The study aimed to (i) examine the profile and relationship between the CO_2 flux (FCO_2) and the O_2 flux (FO_2) in a sugarcane area under mechanical harvesting with straw burning (BH) and mechanical harvesting with maintenance of straw (GH), considering soil moisture; (ii) and suggest the use of ARQ as an index for categorising the biological activity of soils. Our results showed consistently lower FCO_2 for soil moisture in the range of 6.0–8.6% for both management regimes. The soil moisture increments triggered a decrease in FO_2 and an increase in FCO_2 and ARQ. The FCO_2 and FO_2 were positively correlated under BH. The BH yielded a cumulative CO_2 emission of 53.68% higher than for GH. Overall, our findings revealed that soil moisture affected the O_2 uptake and CO_2 emission profile of soil, limiting O_2 uptake and increasing CO_2 releases for water-filled porosity below 70%. The GH management system, which incorporates sugarcane residues into the superficial layer of the soil, can help protect against soil erosion. The ARQ can be used as an index to categorise biological activity in soil, where ARQ values close to 1 can be considered a reflection of aerobic activity with balance between CO_2 production and O_2 consumption.

Additional keywords: CO_2 emission, sugarcane straw management, O_2 uptake, respiratory quotient, soil biological activity.

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Introduction

Measurements of oxygen flux (FO_2) in soils are related to the metabolic status of microorganisms and carbon accumulation or loss, especially in environments where these processes are driven by aerobic microbial activity (Stern *et al*. 1999), considering FO_2 as a reflection of carbon dioxide flux (FCO_2) in the global carbon cycle (Keeling and Shertz 1992; Dilly 2003).

The FCO_2 is considered to be the result of biochemical processes in soil, and is directly related to the respiration of roots and decomposition of organic matter through microbial activity (Lal 2009). It is estimated that for a mixed hardwood forest ecosystem, root respiration accounts for 14–21% of FCO_2, with the remaining fraction due to the biological activity of soil microorganisms (Melillo *et al*. 2002). It is noteworthy that FCO_2 can also result from chemical activities in soil (calcareous and urea reaction; Angert *et al*. 2015), and processes of degassing of the soil solution and CO_2 desorption from the solid phase can produce soil CO_2 efflux (Smagin *et al*. 2016).

The relationship between FCO_2 and FO_2, known and described by Angert *et al*. (2015) as apparent respiratory quotient (ARQ), is an alternative means of describing and categorising soil activities (chemical, physical and biological), where there is a strong relationship between CO_2 production and O_2 consumption. Values of ARQ that are higher or lower than 1 can be interpreted as an imbalance between CO_2 production and O_2 consumption as a response to chemical and physical activities in soil (Linn and Doran 1984).

The soil pore network characteristics and soil texture directly influence FCO_2 and FO_2 exchange between soil and the atmosphere, through the presence of empty spaces between soil particles and aggregates (Chen *et al*. 2011). Furthermore, water and nutrient availability (Almeida *et al*. 2015), and tillage management (Bicalho *et al*. 2014), can also influence CO_2 production and transport, as well as O_2 consumption in soil.

Sugarcane can mechanically harvested with removal or burning of the straw or with maintenance of straw on soil without burning (De Figueiredo and La Scala 2011). Brazil is currently the largest sugarcane producer globally, with an average of 74.1 Mg ha^{-1} biomass yield annually (Conab 2014), and 20 Mg ha^{-1} of resulting residues are retained on the soil surface following harvest (Urquiaga *et al*. 1991; Oliveira *et al*. 1999).

The study aimed to (i) examine the profile and relationship between FCO_2 and FO_2 in a sugarcane area under contrasting management regimes (mechanical harvesting with straw burning versus mechanical harvesting with maintenance of straw), considering soil moisture; (ii) and suggest the use of ARQ as an index for categorising biological activity in soil.

Material and methods

Characterisation of the study area

The study was conducted in an area under sugarcane (*Saccharum* spp.) cultivation located in the state of Mato Grosso do Sul, near the municipality of Aparecida do Taboado (20°19′S and 51°13′W), Brazil, during 4–14 July 2014. The soil was classified as a dystrophic Red-Yellow Latossolo (Embrapa 2014) and Oxisol (Soil Survey Staff 2014), with a sandy clay loam texture across the 0–0.2 m depth layer (Table 1).

The region has a tropical humid climate classified as Aw (Peel *et al*. 2007), characterised by a rainy summer (September–June) and a dry winter (June–August), with an average annual rainfall of 1595 mm. During the field measurements, there were two precipitation events on 13 and 14 July with a daily rainfall of 6.1 and 1.5 mm, respectively (Climate Channel at UNESP Ilha Solteira, http://clima.feis.unesp.br).

The effect of residue management was evaluated in two sections of the production field under contrasting straw management regimes (Sections): Section 1, mechanical harvesting with straw burning (BH) and Section 2, mechanical harvesting with maintenance of straw (GH). Normally, GH adds an average of 20 Mg ha^{-1} of crop residue; however, the quantity depends on the variety and the harvest stage (Correia and Durigan 2004; Tofoli *et al*. 2009; Almeida *et al*. 2014). Both sections had a sugarcane productivity of 63 and 46 Mg ha^{-1} in 2013 and 2014, respectively.

The study area was 21.77 ha cultivated with the CTC variety of sugarcane, at a population density of 60 000 ha^{-1}. This area has been used for sugarcane production for 20+ years. The soil was prepared and sugarcane planted using the conventional system (soil disturbance) in 2012.

Fertilisation was performed along the furrow and involved the distribution of 250 kg ha^{-1} of mono-ammonium phosphate, equivalent to 120 kg ha^{-1} of P_2O_5 and 27 kg ha^{-1} of $N\text{-}NH_4^+$. Subsequently, topdressing was done using a liquid formula 05–00–13 + 0.3% Zn + 0.3% B, in the amount of 1000 L ha^{-1}, equivalent to 50 kg ha^{-1} N, 130 kg ha^{-1} of K_2O, 3 kg ha^{-1} of Zn and B, respectively. After the first cutting, ratoon fertilisation was performed using the best management practices of applying 90 kg ha^{-1} N, 30 kg ha^{-1} of P_2O_5 and 110 kg ha^{-1} of K_2O.

For the BH, sugarcane was harvested mechanically, with burning; for the GH, the harvest was also mechanical but without burning. At sampling time, the sugarcane plants were ~20 cm tall at 2 weeks after their second cutting (harvest). Therefore, the area did not include plants that displayed higher growth stages.

Field sampling was done by selecting 10 points that had at least 5 m spacing from within each management treatment, and within the four central lines of each plot. The results of soil testing from the two sections (0–0.2 m layer), including soil physical (sand, silt and clay) and chemical properties (pH, soil organic matter, phosphorus, sulfur, calcium, potassium, magnesium, aluminium and cation exchange capacity), as well as the soil porosity (macroporosity, microporosity and total porosity), are shown in Table 1. These measures were determined using methodology from Embrapa (1997).

Water-filled porosity (WFP) was calculated using Eqn 1 described by Linn and Doran (1984). Where, soil moisture was the volumetric water content (%) and the total soil porosity (TP, %) was calculated by TP = (1 − PB/PP) × 100, where the soil particle density (PP) is assumed to be 2.65 Mg m^{-3} and soil bulk density (PB) is in Mg m^{-3}.

$$\%\mathrm{WFP} = (\text{soil moisture}/\mathrm{TP}) \times 100 \qquad (1)$$

Table 1. Physical and chemical attributes of a Red-Yellow Latosol under contrasting sugarcane management regimes involving mechanised harvesting with the presence of straw (GH) and with straw burning (BH)

pH in 0.1 KCl, soil organic matter (SOM), phosphorus (P), sulfur (S), calcium (Ca^{2+}), potassium (K^+), magnesium (Mg^{2+}), aluminium (Al^{3+}), cation exchange capacity (CEC), macroporosity (Macro), microporosity (Micro), total porosity (TP) and water-filled porosity (WFP)

Straw management	Soil chemical attributes								
	pH	SOM	P	S	Ca^{2+}	K^+	Mg^{2+}	Al^{3+}	CEC
	–	(g dm^{-3})	(mg dm^{-3})		(mmol$_c$ dm^{-3})				
BH	5.11 ± 0.04	16.30 ± 0.58	8.0 ± 0.26	5.5 ± 0.22	19.7 ± 1.1	1.36 ± 0.09	9.1 ± 0.81	0.8 ± 0.2	55.06 ± 1.95
GH	5.18 ± 0.08	15.90 ± 0.23	8.2 ± 0.29	5.3 ± 0.26	22.0 ± 0.86	1.26 ± 0.13	8.7 ± 0.58	0.9 ± 0.31	53.46 ± 0.72

	Soil physical attributes						
	Sand	Silt	Clay	Macro	Micro	TP	WFP
	(g kg^{-1})			(%)			
BH	613 ± 1.0	101.0 ± 1.0	286.0 ± 0.0	14.6 ± 2.01	29.01 ± 0.60	43.69 ± 1.66	18.73 ± 5.6
GH	602 ± 0.35	111.5 ± 0.35	286.0 ± 0.0	11.1 ± 1.37	31.58 ± 1.05	42.75 ± 1.07	18.49 ± 6.4

Soil sample and variables analysed

PVC rings (10 cm in diameter and 8.5 cm in height) were previously installed and fixed at the sample points. After 24 h, FCO_2 and FO_2 were collected along with data describing soil moisture and temperature for six separate measurement days (4, 6, 8, 10, 12 and 14 July). We used this period because we wanted to observe the relationship between these variables and the soil without the confounding contribution from crop growth at later stages. Soil measurements were made during 07:00–08 : 00 hours.

To collect FCO_2 we used an IRGA (LI-COR Inc, Lincoln, NE, USA) with a closed circulation system, an internal volume of 854 mL and a soil contact area of 84 cm^2. The IRGA had an infrared (IR) system that measured the CO_2 concentration using optical–IR absorption spectroscopy. The soil temperature was monitored using a temperature probe that was integrated into the IRGA system.

The soil moisture was measured using a portable Time Domain Reflectometry system (Hydrosense™; Campbell Scientific, Garbutt, Australia) that determined the soil moisture according to the dielectric constant of the travel time for an electromagnetic pulse across the space separating the two end points (two rods, 12 cm high) inserted into the soil adjacent to the PVC ring (0–10 cm).

The soil FO_2 was monitored using an O_2 sensor (CM-021; CO_2 Meter Inc., Ormond Beach, FL, USA) with a full-scale span of 0–25% (v/v). This sensor was portable and utilised ultraviolet light fluorescence to assess the O_2 concentration. The sensor result was read using Gaslab software to calculate the soil O_2 uptake rate. With the CO_2 and O_2 results, we calculated the ARQ (mol mol^{-1}) according to Wolinska *et al*. (2011), where the ARQ is a ratio of the CO_2 emission and O_2 uptake.

Estimation of soil FO_2

Soil FO_2 rate (dO_2/dt) was calculated by a linear interpolation of the concentration values as a function of time, taking into account the atmospheric pressure, temperature and volume of the gas trapped in the chamber, using Eqn 2 as described by Smagin *et al*. (2016) and Smagin (2006):

$$FO_2\left(g \cdot m^{-2}\,s^{-1}\right) = \frac{dO_2 10^{-6} PM}{dt\ RT} H \tag{2}$$

where, dO_2/dt is the amount of O_2 (ppm) measured at time t (s); P is atmospheric pressure (Pa); M is O_2 molar mass (g m^{-3}); R is the universal gas constant (8.31 J mol^{-1} k^{-1}); T is absolute temperature (K) and $H = V/A$: for volume $(V) = 0.00066\ m^3$ and cross-sectional area $(A) = 0.008\ m^2$ of the camera above the ground (soil surface).

Data processing and statistical analysis

Soil moisture, O_2 uptake (FO_2), CO_2 emission (FCO_2) and the daily mean of ARQ were calculated using an N (number) of 10 replicates per day, compared using Student's t-test ($P \leq 0.05$) per each management sector. Consequently, FCO_2, FO_2 and total ARQ were calculated using all days observed, with $N = 60$ per treatment. An integration of the area under the FCO_2 and FO_2 curves was calculated. Thereafter, the treatment results were compared using Student's t-test ($P \leq 0.05$).

The relationships of FO_2 and FCO_2 with soil moisture were calculated using Pearson's correlation for both management regimes. The analysis of presuppositions was conducted using analysis of residuals, identifying the outliers and influent values using leverage statistics. The normality of residuals was verified by the Shapiro-Wilk test ($P \leq 0.05$) and homogeneity of variance was assessed using the Bartlett test ($P \leq 0.05$).

Results

Daily results of soil variables

Soil CO_2 emission on 4, 6, 8 and 10 July were similar, with means of 0.04, 0.06, 0.03 and 0.04 mg m^{-2} s^{-1} for GH and 0.07, 0.07, 0.06 and 0.07 mg m^{-2} s^{-1} for BH respectively (Fig. 1*d*). Both treatments, presented lower and relatively constant CO_2 emission when the soil moisture varied within 6.44–8.60% (BH) and 6.0–7.4% (GH), (Fig. 1*b*). However, after 10 July, CO_2 emission and soil moisture increased by means of 0.07 and 0.14 mg m^{-2} s^{-1} and 14.11 and 14.33% respectively for GH and BH (Fig. 1*d, b*), following a precipitation event of 6.1 mm (Fig. 1*a*).

The BH provided higher CO_2 emission across all days observed, and significantly differed from those of GH on the 4, 8, 10, 12 and 14 July. The highest BH difference was on 12 July with a CO_2 emission increase of 53.68% compared with GH (Fig. 1*d*).

The temporal variability in soil O_2 uptake was inverse compared with CO_2 emission the estimates of O_2 uptake revealed variation of 0.22–0.46 and 0.20–0.40 mg m^{-2} s^{-1} on the first days (4 and 10 July), and following the precipitation event the O_2 uptake decreased (without a significant difference between the days and treatments) (Fig. 1*c*).

The ARQ profile was very similar to the estimates of CO_2 emission, soil moisture and precipitation (Fig. 1). In other words, ARQ was constant, lower than 1 and mean variation range of 0.27–0.90 (GH) and 0.17–0.31 (BH) during 4–10 July. Contrastingly, following the precipitation event (12 July) ARQ was higher than 1, with the greatest values for ARQ in BH (1.38 ± 0.46 mol mol^{-1}) and lower than 1 in GH (0.77 ± 0.46 mol mol^{-1}).

Accumulated FCO_2, FO_2 and ARQ

Accumulated FO_2 revealed means of 202.52 ± 49.62 and 267.41 ± 73.6 g O_2 m^{-2} for the GH and BH regimes respectively (Fig. 2*a*). Comparison by t-test showed no differences in the FO_2 uptake between harvesting techniques ($P > 0.05$). However, BH demonstrated higher cumulative CO_2 emission (87.07 ± 19.45 g CO_2 m^{-2}), which was 50.0% higher but not significantly different compared with GH (52.31 ± 15.41 g CO_2 m^{-2}) (Fig. 2*a*).

The cumulative ratio of FCO_2 and FO_2, represented by ARQ, was below 1 in both treatments, having means of 0.23 ± 0.18 and 0.18 ± 0.07 for BH and GH respectively (Fig. 2*b*). The ARQs for both treatments did not significantly differ according to t-test.

Relationships of soil CO_2 emission and O_2 uptake with soil moisture

The CO_2 emission and soil moisture were positively and significant correlated for both treatments (Fig. 3*a, c*), although

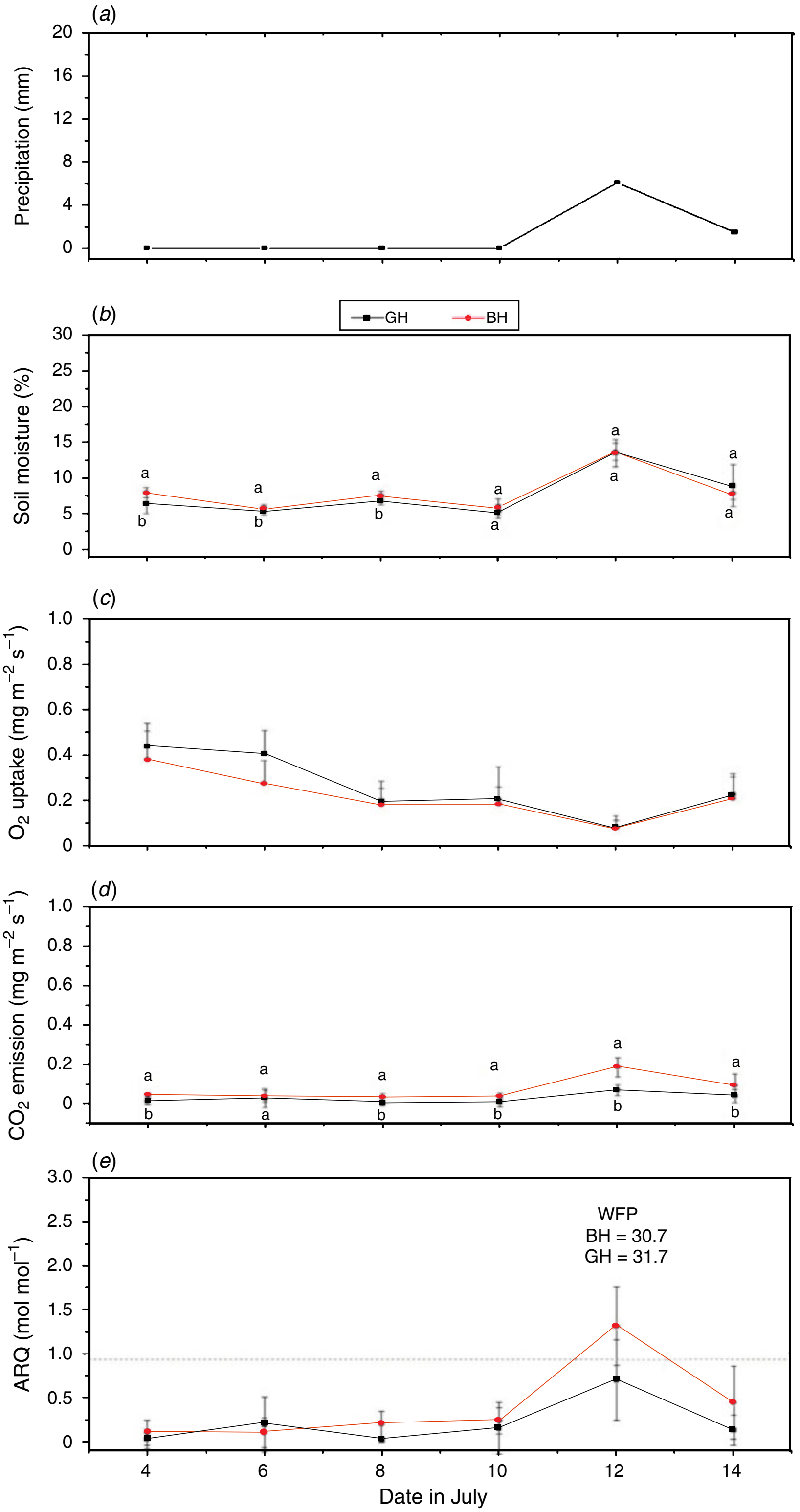

Fig. 1. (*a*) Precipitation, (*b*) soil moisture, (*c*) O_2 uptake, (*d*) CO_2 emission and (*e*) apparent respiratory quotient (ARQ) of soil under contrasting sugarcane management regimes of mechanised harvesting with straw (GH) and burned straw (BH). Note: days marked with different lower-case letters differ according to *t*-test ($P \leq 0.05$). The O_2 uptake and ARQ show no significant difference for days according to *t*-test ($P \leq 0.05$).

higher for BH ($r = 0.74$) than for GH ($r = 0.50$). The higher r for BH suggests a greater sensitivity of burned residue management. The soil moisture was negatively correlated with O_2 uptake in BH ($r = –0.43$) and GH ($r = –0.42$) (Fig. 3*b*, *d*). The CO_2 and O_2 were negatively correlated for BH ($r = –0.41$), but not correlated for GH (Fig. 4). We also noted that GH had a higher microporosity (31.6%) and lower macroporosity (11.1%) and WFP (18.73%) compared with BH (Table 1).

Discussion

CO_2 and O_2 results

The consistent magnitude of FCO_2 across 4–10 July for GH and BH suggests a corresponding stability in soil microbial activity. Typically, low and stable FCO_2 occurs after the soil carbon mineralisation of soil organic matter (Cunha *et al*. 2011; Badía *et al*. 2013; Knicker *et al*. 2013) leading to lower emissions and microbial activity (Luo *et al*. 2006).

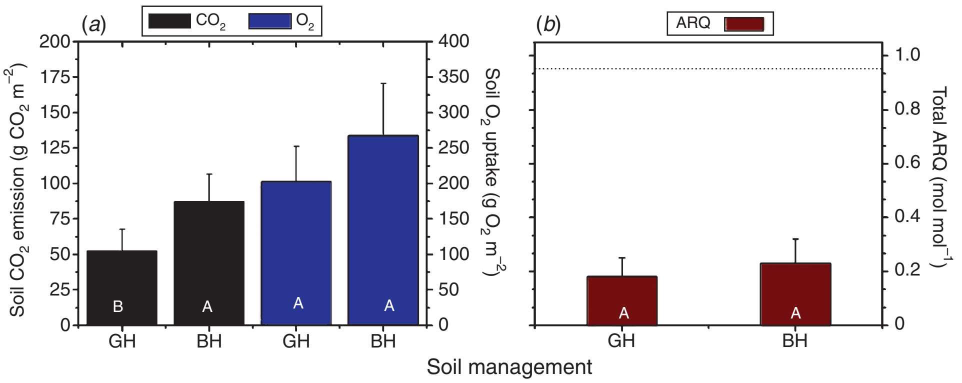

Fig. 2. (*a*) Soil cumulative CO_2 emission and soil cumulative O_2 uptake and (*b*) apparent respiratory quotient (Total ARQ) of soil under contrasting sugarcane management regimes of mechanised harvesting with straw (GH) and burned straw (BH). Note: bars identified with different upper-case letters differ according to *t*-test ($P \leq 0.05$).

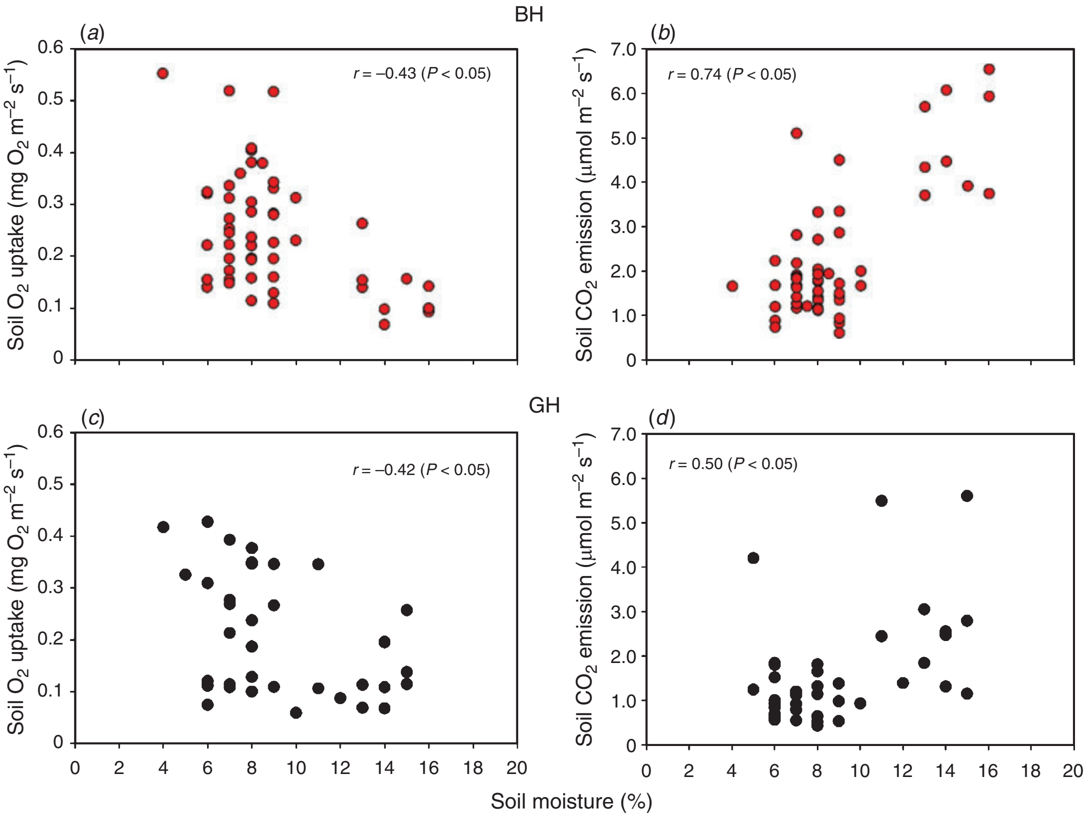

Fig. 3. Relationship of soil moisture (%) with (*a*, *c*) O_2 uptake and (*b*, *d*) CO_2 emission, within a sugarcane area under contrasting sugarcane management regimes of mechanised harvesting with straw (GH) and burned straw (BH).

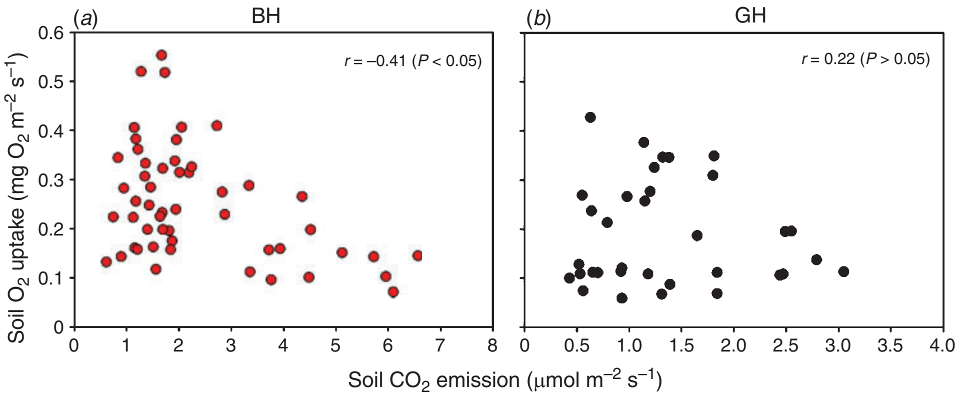

Fig. 4. Relationship between soil CO_2 emission and soil O_2 uptake within a sugarcane soil under contrasting sugarcane management regimes of mechanised harvesting (*a*) with burned straw (BH) and (*b*) straw (GH).

Higher CO_2 emission for BH compared with GH has been observed in other studies by Panosso *et al*. (2009) and Corradi *et al*. (2013), who examined a similar Oxisol and soil management regimes in São Paulo. This difference could be explained by higher nutrient availability in the BH treatment due to burning (Marques *et al*. 2009; Panosso *et al*. 2011). However, in our experiment we did not observe a significant difference in soil nutrient availability between BH and GH. This is likely to be driven by the fact the burning occurred once, and that changes to organic nutrient availability through this process depend upon the intensity and duration of burning.

Contrastingly, the BH had higher macroporosity and total porosity, and lower microporosity, compared with GH. The high porosity for BH may have resulted from burning of sugarcane residue and an increase in empty spaces in soil, causing the opening of potentially charred root channels or plugging of smaller micropores by ash deposits. It should be kept in mind that both the BH and GH treatments had the same degree of mechanical harvesting traffic and soil preparation.

The relationship between soil porosity and CO_2 has been previously reported (Xu and Qi 2001; Epron *et al*. 2006; Panosso *et al*. 2011; Bicalho *et al*. 2014). Soil porosity is responsible for soil gaseous transport (Xu and Qi 2001; Epron *et al*. 2006) and the movement of organic and inorganic solutions throughout the soil, which supports the natural habitat for microbial communities (Ranjard and Richaume 2001). Therefore, soil porosity can help to explain the degree of soil CO_2 emission (Wick *et al*. 2012).

Additionally, the lower FCO_2 estimates for GH were likely the result of sugarcane residues on the soil surface, which have been shown to present a high carbon/nitrogen ratio (Almeida *et al*. 2015), lignin (Costa *et al*. 2013) and cellulose contents (Almeida *et al*. 2009), and a lower crude protein concentration (Pereira *et al*. 2000). These characteristics are important parameters in nutrient dynamics (Lal 2004) and can consequently reduce FCO_2 because of slow residue decomposition (Almeida *et al*. 2014).

Relationships between soil CO_2 and O_2

The temporal variability in soil O_2 was inverse compared with CO_2, reflected by a decrease in O_2 and an increase in CO_2 for both treatments, following precipitation. Consequently, these results confirm that soil moisture can change the soil CO_2 and O_2 profile. According to Gardini *et al*. (1991) and Howard and Howard (1993), soil moisture is a key abiotic factor affecting the CO_2 emission and O_2 uptake processes (Gardini *et al*. 1991), as well as soil temperature (Kyaw Tha Paw *et al*. 2006).

The positive correlation between CO_2 and soil moisture was also observed by Lal (2009) and Wei *et al*. (2014), and can be described as linear response with respect to soil moisture (variations of 38–47%) at clay soil (Corradi *et al*. 2013). Soil moisture can promote increases in FCO_2 of up to 80% (Chen *et al*. 2011) as result of higher microorganism and root activity (Lal and Kimble 1997). According to Doran *et al*. (1990) and Chen *et al*. (2011) the highest soil respiration rates occur for WFP of 40–70% for the majority of soils. In our experiment, there was WFP < 70% for all treatments and days observed.

Additionally, the high correlation between FCO_2 and soil moisture for BH can be explained by higher macroporosity and lower microporosity, which has been shown to govern CO_2 transport rates and water infiltration in soil aggregations (Silva *et al*. 2005), and is typically faster in soils with higher macroporosity than microporosity (Ceddia *et al*. 1999).

Decreases in O_2 following precipitation events were also observed by Linn and Doran (1984) and Gardini *et al*. (1991), and have been explained by a reduction in the amount of O_2 in soil pores with water infiltration (Cook *et al*. 2007), which limits the O_2 exchange between soil and atmosphere (Armstrong and Drew 2002; Elberling *et al*. 2011). This suggests that water in soil pores limits O_2 uptake, but may increase CO_2 release for WFP < 70%.

Relationships between soil CO_2 emission and O_2 uptake

The FCO_2 and FO_2 were negatively correlated for the BH treatment. However, we did not observe a relationship between these factors for the GH treatment. Kyaw Tha Paw *et al*. (2006) also found a negative relationship between FCO_2 and FO_2 in soil (at 15 cm in depth) with vegetation of ~3 cm high, as consequence of respiration by roots and microorganisms and the simultaneous increase of FCO_2 concentration and O_2 depletion. We further observed that FO_2 was higher than FCO_2 for both treatments and, similarly, Angert *et al*. (2015)

found higher FO_2 compared with CO_2 in a study of temperate and alpine forest ecosystems.

To understand ARQ as an index for categorising soil activity, it must first be understood that ARQ values close to 1 are considered a reflection of aerobic activity with ARQ balance – the result of production of 1 mol CO_2 and consumption of 1 mol of O_2. However, ARQ values higher or lower than 1 indicate an imbalance between FCO_2 and FO_2. The ARQ index has similarly been used as a criterion for soil microbial activity across different WFP conditions (Stotzky 1960; Alef 1995; Dilly 2003), as a means of elucidating the relationship between FCO_2 and FO_2.

The ARQ was below 1 before the rainfall event for BH and GH, but shifted closer to 1 after precipitation (12 July) with soil moisture in the range of 6.4–14.5% and $WFP < 32.0\%$. According to Linn and Doran (1984), an increase in ARQ values of 1.3–1.7 can occur with an increase in soil water content and $WFP > 70\%$ and indicates a shift towards anaerobic metabolism. However, in our experiment the WFP value was not >70% on 12 July. Franzluebbers (1999) highlighted that the maximum respiratory activity of soil microbial biomass at WFP levels in the range of 27–68% was due to higher availability of O_2. Under this WFP condition, it is likely that there was a predominance of aerobic compared with anaerobic respiration, soil chemical reactions (presence of calcareous materials and urea fertilisation) and the degassing process. According to Smagin *et al*. (2016), processes of degassing of the soil solution and CO_2 desorption from the solid phase can produce soil CO_2 efflux. Therefore, these processes can produce CO_2 with no direct relationship to O_2 consumption, thus changing the soil CO_2 and O_2 balance.

Conclusion

Our results show that soil moisture affected the O_2 uptake and CO_2 emission profile of soil by limiting O_2 uptake and increasing the release of CO_2 for conditions of $WFP < 70\%$. The high level of soil macroporosity and low degree of soil microporosity increased CO_2 emission.

The correlation between O_2 uptake and CO_2 emission profiles depends on crop residue management and soil pore network characteristics. The BH management regime provided higher cumulative CO_2 emission with a 50.0% increase compared with GH, which added sugarcane residue to the superficial soil layers and so helped prevent soil erosion.

The ARQ can be used as an index to categorise biological activity in soil, with ARQ values close to 1 considered a reflection of aerobic activity with balance between CO_2 production and O_2 consumption.

Conflicts of interest

The authors declare no conflicts of interest.

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