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Determining Gas Sampling Timelines for Estimating Emissions in Small Chamber Incubation Experiments

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Abstract: - A laboratory experiment was set up in small chambers for monitoring greenhouse gas emissions and determining the most suitable time for sampling. A six-treatment experiment was conducted, including a one week pre-incubation and a week for incubation. Timelines for sampling were 1, 2, 3, 6 and 24 hours after closing the lid of the incubation chambers. Variation in greenhouse gas fluxes was high due to the time of sampling. The rates of gas emissions increased in first three hours and decreased afterward. The rates of greenhouse gas emissions at 3 hours after closing lids was close to the mean for the 24-h period.

Keywords: *- biochar, chamber, Ferrosol soil, incubation, sampling*

I. INTRODUCTION

Research on greenhouse gas (GHG) emissions in agricultural science has been motivated in recent decades by mitigating climate change and global warming. This is because GHG emissions have an impact on the broader environment (through global warming) as well as its contribution to carbon and nutrient losses. To evaluate GHG emissions (carbon dioxide, methane and nitrous oxide), both laboratory and field experiments are required. Laboratory experiments play an important role in researching GHG emissions as they provide information on processes controlling emissions under controlled temperature and moisture conditions. This type of research can be conducted in incubators and glasshouses.

In laboratory incubations, 0.5 L glass chambers [1, 2], 1.0 L [3] or 5.0 L glass jars [4] (hereafter "chambers") have been used for soil incubation which are sealed and gas samples taken from the chambers. The volume of the chambers varies due to the weight of soil that is used in the experiments and the purpose of the research. For example, a large amount of soil provides less variability, as well as the ability to take soil sub-sample for analysis during the incubation.

For the timing of gas sampling, different timelines have been used for their measurement of GHG emissions. If the incubation time is too long, the concentration of emissions will be saturated reducing oxygen in the headspace which affects the microbial activity which produces carbon dioxide and nitrous oxide. Gas samples have been taken after closing for a half hour [5-7], one hour [3-6, 8], two hours [2, 9] and four hours [10]. The time for sampling chosen by different authors has not following any previous studies. It has led to confusion for other researchers setting up new experiments. The aim of this study is to determine the suitable timeline for sampling in small chamber laboratory incubations.

II. MATERIALS AND METHODS

Soil was collected from Wollongbar (28^{0}49' S, 153^0 25'E), NSW, Australia. This soil is classified as red Ferrosol [11]. Top soil was collected from the cultivated surface (0-20 cm), air-dried and sieved through a 2-mm stainless steel mesh. It after that was stored in fridge at 4^{0}C until used. Soil bulk density was 1.02 g cm^{-3}

Biochar used in this experiment was produced from rice husks after pyrolysis and supplied by Barmac Industries Pty Ltd. Chemical properties of rice husk biochar (hereafter biochar) are listed as total C (%) 46.5, N (%) 0.62, pH (1:5 H_2O) 9.1 and CEC 17.9 (cmol/kg) [9]

A laboratory experiment was set up with six treatments with and without biochar applied at the equivalent of 0, 20 and 50 tonnes biochar ha^{-1} and two water contents (60 and 90% water-filled pore space). Water-filled pore space (WFPS) is calculated as:

$$WFPS\ (\%) = \frac{GWC \times BD}{1 - \frac{BD}{2.65}}$$

where:
- GWC: gravimetric water content (%)
- BD: soil bulk density (g/cm^3)
- 2.65 assumes the soil particle density (g/cm^3)

The six treatments (with four replicates) were (i) 0 biochar 60% WFPS, (ii) 0 biochar 90% WFPS, (iii) 20t biochar 60% WFPS, (iv) 20t biochar 90% WFPS, (v) 50t biochar 60% WFPS, and (vi) 50t biochar 90% WFPS.

Approximately 225 g air-dried soil (200 cm^3 equivalent) was well mixed with the given amount of biochar and re-packed into a PVC core (25 cm high, 5.05 cm internal diameter). The cores were placed in a 1-L glass chamber (jar). The soil was treated with deionized water to adjust water content to 60% WFPS. All the chambers were pre-incubated at 25^{0}C for seven days to stabilize the activities of micro-organisms. After pre-incubation, the water content was adjusted to 60 or 90% WFPS (i.e. near saturation), respectively by adding deionized water and checked regularly every 3 days by weighing. The rate of biochar was applied based on the surface area (1 ha = 10,000 m^2) with a core surface area is 20 cm^2.

The headspace air was observed after 1, 3 and 7 days incubation. Preparing for gas sampling, the jars were capped by a septum lids. Samples were taken at 1, 2, 3, 6, and 24 hours after the lid closure for three days by using a gas-tight syringe inserted through the rubber septum. After sampling, the jars were opened for air exchange. Gas samples taken in the syringe were immediately transferred to an evacuated exetainer (Labco Ltd, Buckinghamshire, UK).

Concentrations of N_2O and CO_2 was measured by a gas chromatograph (Shimadzu, Japan) equipped with two detectors. N_2O was detected by electron capture detector (ECD) and CO_2 was detected by hydrogen flame ionization detector (FID). Flux rates of N_2O and CO_2 were calculated using equation 1 and 2 which were described in van Zwieten *et al.* [4].

III. RESULTS AND DISCUSSION

The fluxes of CO_2 and N_2O varied during the time of sampling (Figure 1). In Day 1 and Day 3, the fluxes were reducing for every hour. The fluxes of greenhouse gases could be distinguished into two groups. The emissions were low in the low water content (60% WFPS) meanwhile the higher water content (90% WFPS) emitted the higher GHG emissions.

At Day 7, the concentration of CO_2 was similar trend to other days before but the N_2O flux increased in the first 3 hours and then reduced in the sixth hour and continuously reduced until the 24th hour.

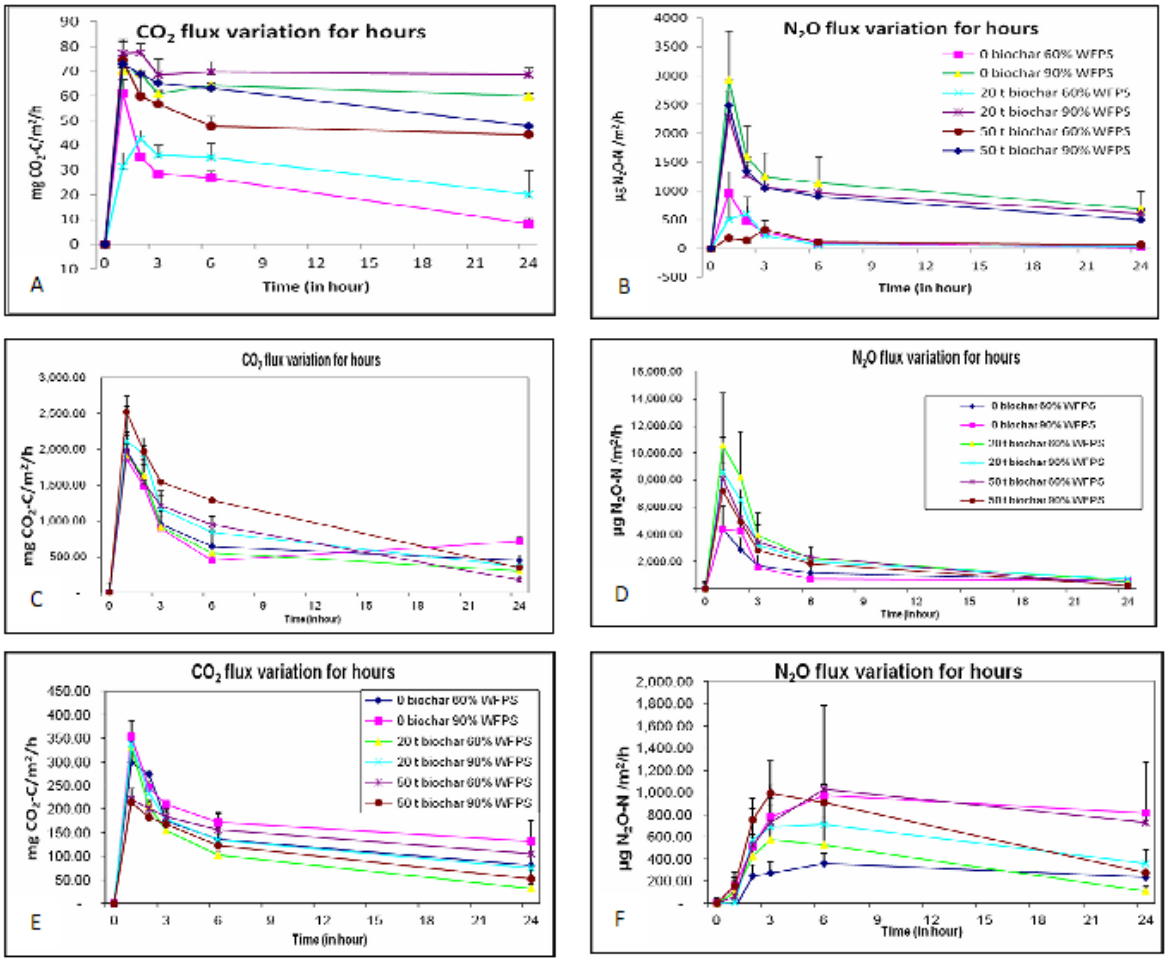

Figure 1. Comparison GHG fluxes for one, three and seven days after incubating at 25^{0}C. Error bars present one standard error. Note: A&B present Day 1, C&D express Day 3 and E&F show Day 7.

For CO_2 emission, it changed from 31-74 mg CO_2-C/m^2/h at one hour to 8-68 mg CO_2-C /m^2/h at 24 hours in Day 1 (referred in Figure 1). The N_2O flux rate dropped from 179-2,929 µg N_2O-N/m^2/h in the first

hour to 27-691 µg N_2O-N/m^2/h at 24 hours in Day 1. The flux rates at three hours were similar to the mean of the 24-h period incubation.

In general, the rates of CO_2 and N_2O emissions were higher in the 90% WFPS treatments than those of 60% WFPS. This is similar to the result of Dobbie and Smith [10]. Moreover, the N_2O emissions in Day 3 (Figure 1 C&D) increased higher in Day 1 and Day 7 which supported the result from Singh *et al.* [6].

The relationships between sampling time and greenhouse gas emissions were negatively correlated (Table 1). The R^2 were more than 0.5 to 0.8 (except 0.31 in the 0 biochar 90% WFPS). When the time of closure was too long, the gas emissions did not increase. This may be due to saturation of gas in the headspace and low oxygen concentrations affecting microbial activities.

Sampling chamber headspace at three hours after closure of the lids gave the best results for estimating gas emissions. The timelines for sampling would be implied in the studies with small chamber from 0.5 L to 5 L in the incubation condition.

Table 1. Relationship between time of sampling and GHG flux rates. * The regressions (R^2) are significant at $P<0.05$.

	CO_2 emissions			N_2O emissions		
Treatments	Slope	Intercept	R^{2*}	Slope	Intercept	R^{2*}
0 t biochar 60% WFPS	-19.32	588.52	0.54	-45.01	1,229.0	0.53
0 t biochar 90% WFPS	-12.76	548.27	0.31	-57.42	1,902.2	0.50
20 t biochar 60% WFPS	-21.28	574.50	0.58	-117.22	2,751.9	0.59
20 t biochar 90% WFPS	-23.63	686.06	0.66	-98.49	2,683.1	0.59
50 t biochar 60% WFPS	-23.20	634.05	0.83	-79.94	2,123.0	0.71
50 t biochar 90% WFPS	-27.86	782.85	0.82	-95.84	2,451.9	0.68

IV. CONCLUSIONS

This is the first research to examine the effect of sampling time on GHG emissions in laboratory incubations. Using small (1.0 L) chambers in laboratory incubations, flux rates were estimated after headspace sampling at 1, 2, 3, 6 and 24 hours. After lids were closed for three hours was determined the best choice for gas sampling.

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