Referee' comments

Referee #1's comments 1: "Most perplexing, since quite unusual in context of the large amount of publication on methods used to quantify soil greenhouse gas (GHG) exchange in forest ecosystems, in the presented manuscript one site is synonym with one soil flux chamber. In general at least 5-10 chambers per site are necessary to capture spatial variation in soil CO2, CH4 and to a lesser degree N2O exchange. ..."

Referee #1's comments 2: "Another problem is the type of statistical analysis used..."

Authors' response

Authors' response - The referee highlighted the difference between "site" and "plot" which we did not treat carefully when preparing the MS. What we want to demonstrate is: we conducted this study at the burned site with four replicate plots. We considered the stand conditions (eucalypt forest with moderate density), location (middle slope) to ensure the similarity of the four selected plots. To avoid any misunderstanding, we changed the term of "site" into "plot" throughout the article.

Authors' response - we appreciate the referee's comments based on his/her professional knowledge. In the submitted MS, we did not give enough details about our statistical methods. What's more, we should not mention it as "repeated measures ANOVA" while we actually used the "one way ANOVA" method (this is described in the caption of Table 1). Specifically, one-way ANOVA was applied to compare gas fluxes in the burned plots before and after the burning, and also applied to compare fluxes between the burned and unburned plots (the 4 additional selected plots) in Aug and Nov 2014, respectively. Therefore, we did not treat the 4 unburned plots located in green islands as pre-burning conditions, and these plots were only taken as additional reference plots to discuss the impacts of burning.

To clarify this point, we modified the section "2.5 Statistical analysis" as detailed below:

"All statistical analyses were performed using IBM SPSS STATISTICS (version 20) software. One-way ANOVA was introduced to examine

statistically significant differences between soil gas fluxes measured before and after the burning in the burned plots. This analysis was also applied to compare the fluxes between burned and unburned plots in Aug 2014 and Nov 2014, respectively."

Referee #1's comments 3: "no data were collected in the first 2-3 months directly after the burn, ..., there are no data presented to put the measuring campaigns into a climatic context..."

Authors' response - we understand the referee's concerns of higher sampling frequency during the first several months after burning. Our first sampling after burning was carried out on 27 Aug 2014, which was actually within 3 months after the burning (27 Aug 2014). We did not conduct earlier samplings after burning due to accessibility of the plots. Meanwhile, the sampling dates in Aug 2013 and Aug 2014 were intended to minimize the impacts of changes in climate conditions based on a primary assumption that temperature, radiation and precipitation would not change much for the same periods of a year in this study.

Referee #1's comments 4: "manuscript would also need to be edited by a native English speaker and will need to be completely rewritten in plain English..."

Authors' response - Prof. Zhihong Xu has carefully gone through the paper and revised the English expressions substantially. Prof. Xu has been working and studying in Australia for about 30 years, and he is also an Editor-in-Chief or Editor of major international journals. Please turn to the attached revised MS below for details.

Responses to Anonymous referee #2's comments

Referee #2's comments 1: The study employed a before-after/control-treatment experiment design to evaluate prescribed burning effects. The authors firstly collected gas flux data before the burning (Aug. 2013) and then on two post-burning dates (Aug. 2014 vs. Nov. 2014). This dataset was compared with a one-way ANOVA to assess burning effects

Author's response: we appreciate that the referee kindly pointed out the problems in clearly describing the statistical analysis methods and presenting the results. About the one way ANOVA analysis, we first applied it to the gas fluxes measured before and after the burning at burned plots to test whether there were any temporal variations. Then we use one way ANOVA to compare the fluxes measured in burned and adjacent unburned

on CH4, CO2, and N2O exchange rates. To account for the confounding effects from the inherent temporal dynamics of those fluxes, the authors collected a second suite of dataset on four unburned replicates and conducted a second-round ANOVA. However, after carefully examining Table 1, one could draw a conclusion that the inherent temporal dynamics of those fluxes exerted much greater influences than prescribed burning. The more accurate depiction of burning effect size and magnitude can only be derived from the second-round ANOVA. For example, although CO2 emission rate in the burning site was reduced on Aug. 2014. However, this reduction cannot be attributed to the prescribed burning because Aug. 2014 CO2 rates measured in the burned plots were not significantly different from that in the unburned plots. The authors did make such distinctions in their abstract and conclusion, but they did a poor job in the results section (especially section 3.1)

I am not very concerned about the pseudo-replicate issue as long as the authors can state clearly in the manuscript that the results only reflect the effects of this particular prescribed burning. However, the presentation of their ANOVA interpretations should be carefully revised to avoid inflicting any unwanted confusions

English presentation is problematic. Some paragraphs read smoothly, but a number of paragraphs are still rough. Please see below for an incomplete list of language suggestions.

plots in Aug and Nov 2014, respectively. We have rewritten this section to clarify this point (as detailed in response to referee #1's comments).

Authors' response: we appreciate that the referee pointed out the problems in English presentation and gave a detailed list of examples. To improve the MS's English presentation, Prof. Zhihong Xu has gone through the MS very

P4L7, the reference of Zhao et al. 2013 is mainly about greenhouse gas emissions from Three Gorges Reservoir of China. It doesn't seem to be a global climate change study. Suggest to delete this citation from the list

P4L18, change altering to alter

P4L18, change "decomposition of organic matters" to "organic matter decomposition"

P4L22, to reduce the repetitive usage of the same words over and over, suggest to

change "soil CO2, CH4 and N2O fluxes" into "those soil greenhouse gas fluxes"

P4L27, change "wildfires of" to "wildfires in"

P5L9-L10, What do you mean "As temperature and moisture reflect the seasonal variations in CO2 emissions"? It has been documented that wildfire can change soil temperature and moisture over a relatively long time period. Please clarify

P5L24-25, change to "but there are only few published studies and their results are

inconsistent"

P10L15, change to "significantly"

P10L17, change to "CH4 uptake rate became similar to that before the burning"

P10L18, change to "relatively stable"

P10L19, delete "in uptake rate"

P13L7, change "moderate" to "affect"

P13L9, "at an insignificant level"

carefully and revised the MS substantially. Prof. Xu has been working and studying in Australia for about 30 years, and he is also an Editor-in-Chief or Editor of major international journals. Here below are the modifications to the suggestions pointed out by referee # 2:

P4L7: the reference reviewed some studies on global climate change and GHG emissions but there are not the authors' original research, we accept the referee's comments and deleted the reference.

P4L18: we changed "altering" to "alter";

P4L18: we changed "decomposition of organic matters" to "organic matter decomposition"

P4L22: we changed to use "these greenhouse gas" to avoid repetitive usage of "Soil CO2, CH4 and N2O"

P4L27: we changed "wildfires of" to "wildfires in"

P5L9-10: we reorganized the sentence to clarify the point –"As the altered temperature and moisture could change the amplitude of seasonal variations in CO_2 emissions, reduced fine root activities after fires are more responsible for the decreased CO_2 "

P5L24-25: we changed this sentence according to referee's comments.

P10L15: we changed "significant" to "significantly";

P10L17: we changed "CH4 uptake had similar CH4 uptake rate as that before the burning" to "CH4 uptake rate became similar to that before burning.";

P10L18: we changed "relative stable" to "relatively stable";

P10L19: we deleted "in uptake rate";

P13L7: we changed "moderate" to "affect";

P13L9: we changed "at insignificant level" to "at an insignificant level";

P19L14, delete "to manipulate fires"

P19L22-L23, change "it was a combination of burning introduced variation and natural

seasonal variations" to "it was largely caused by natural annual variations"

P19L14: we deleted "to manipulate the fires";

P19L22-23: we changed "it was a combination of burning introduced variation and natural ..." to "it was largely caused by natural annual variation".

We also attached the revised copy of the MS below which listed other modifications highlighted.

Response to referee #1's interactive response

In response to the reply to comment 1: Changing the nomenclature form '"site" to "plot" does not change the low replication number or the pseudo-replication issue. All statistics are based on only 4 chamber locations this is simply to lower a number to make any of the conclusions proposed by the authors. As outlined in our initial comments to capture the spatial variation in soil greenhouse gas (GHG) fluxes at one plot at least 5 chambers would be necessary. The authors try to describe a whole forest system with 4 chambers.

The response given by the authors also highlights that this is a completely nonreplicated experiment there is now 1 site with 4 plots and 1 chamber per plot. If we ignore the chamber replication and spatial variation issue for a moment this type of experimental design might work in a treatment vs. control type of experiment. However, in the case of the experiment outlined in the manuscript control plots were not measured or established before the burning event. Another point that the authors have not clarified is if they measured GHG before and

Authors' response: we agree the referee's suggestion that more chambers at one plot would result in more robust conclusions.

In this study we did not mean to describe the whole forest system (Toohey forest) with the 4 deployed chambers, we wanted to explore the potential impacts of burning conducted on 27 May 2014 within a relative small regions (less than 300m*300m). Yes, there were other deployed chamber rings but were designed for nutrition transformation studies leaded by Yuzhe Wang (co-author). We treated our 4 plots as replicates because of the relative homogenous stand conditions of the study area. We understood the spatial variations in both soil properties and greenhouse gas production, therefore we fully considered the stand condition, tree density, understory conditions and apparent soil conditions when chose these plots. This could be partly assessed with the parameters which measured for the 4 consecutive days during each sampling events, which already presented in Figure 2. Especially for CH4 fluxes and soil temperature (new added to Figure 2), the relative standard errors (std/mean) were less than 10% for most of the occasions.

It is impossible to conduct the sampling at exactly the same location before

after the burning event at the exact same location what appears unlikely since chambers were probably removed before the burning event. Again, since no consideration has been given to generate plot means of multiple chambers per plot for the measured GHG fluxes the differences between measurements taken before and after the burn might largely be confounded by spatial variability.

and after burning because we have to collect the PVC rings and chambers back before the burning, and this could also destroyed the surface soil structure. The plots after burning were located within 2-3 meters of original locations. While the selected unburned plots were also within 5 meters to before the burning plots.

The response given by the authors highlights that an inappropriate statistical test was used to analyse the data. This dataset is not suitable to be analysed with a on way ANOVA since the measurements according to the authors were repeated measurements of the same subject over time. Potentially a linear mixed model might be appropriate to analyse these type of data, however; this might not be possible given the low replication number and the limited number of measurement events. The experimental design is simply not strong enough for any of these analysis and as highlighted in the first response all that was archived is to determine that soil GHG fluxes a different at different times of the year. Furthermore the correlation analysis (no information what test was used) are based on only 3 timedata points. In addition as outlined in our first set of comments some very unusual type of measurements have been correlated with the GHG fluxes in question especially gravimetric moisture content has very little use in this type of analysis Therefore it is in our eyes not possible to talk about "recovery" in the way the authors do. It is also

Authors' response: we have accepted referee #1's comments and modified the section to clarify the statistical methods we used. As concluded by the referee in the "additional comments", a one way ANOVA in this study could only tell there was a temporal variation in the dataset, this was what we wanted to present in applying one way ANOVA to the gas fluxes in burned plots before and after burning. We then conducted another ANOVA analysis to compare the fluxes between the burned and unburned plots to further explore whether the temporal variations were the natural dynamics or burning induced impacts.

Pearson correlation analysis was applied to detect any potential driving factors on soil greenhouse gas emissions. All measured soil properties and gas fluxes at the 4 replicate plots during the three sampling events were pooled together for this analysis. We clarified this point in the statistical analysis section, as described below:

"Collected soil properties and gas fluxes at the four replicate plots during the three sampling events were also pooled together for Pearson correlation analysis to detect possible effects of soil environmental variables on soil unclear in the result section when the authors compare the before and after burn measurements and when they compare the after burn burned vs. after burn unburnt measurements.

CO2, CH4 and N2O fluxes."

Please outline the reasons why you would not have had access to the plots in the first 2-3 month after the burn. The investigated forest is opposite the University campus and prescribed burn areas are generally accessible to the public in within 48 hours after a burn or even directly after a burn if research permits are requested. Furthermore, assuming that soil GHG fluxes are the same in the same month of each year is quite incorrect since their seasonal and inter-annual dynamic depends on the weather (especially soil temperature and moisture), which may differ quite largely between years.

Author's responses: We started planning to do this research in Aug 2013 because we were informed by the government that there would be a burning in that month, we chose the sampling plots before the burning and made the first measurement, our origin plan was to make another measurement right after the burning which also suggested by the referee. However, the planned burning did not happen in that August due to the impropriate weather condition. While in 2014, we were informed about the burning only several days before the burning. Therefore we turned to the assumption that soil condition and gas production were similar in the same period of each year and treated the results measured in Aug 2013 as background values and selected the unburned plots in green island as reference to explore the burning impacts.

We agree that inter-annual dynamics in weather conditions could affect soil gas fluxes, however, we believe it was reasonable that soil gas fluxes of the same period of each year should be comparable without significant disturbance and under similar weather conditions. To support this assumption, we added the soil temperature measurements for the 4 consecutive sampling days at the 4 sampling plots of 3 sampling events in Figure 2 to show the similarity of soil conditions. We also added a Table 1 to show detailed weather conditions during the sampling events: generally, the sampling events were conducted in clear weather conditions and there was very little precipitation either 30 days or 90 days before the sampling events (antecedent precipitation).

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Response to Referee #1's additional comments	
At no stage is discussed if there might have been runoff	Author's response: we have included the weather data in Table 1 and the
mediated lateral flow of nutrients or ash from burnt to unburnt	antecedent precipitation should able to partly address this point. There were
areas in the 1 year timeframe after the burn	only limited small precipitation events (always less than 3 mm) either 30
	days or 90 days before the sampling events, especially for the one on Aug
	2014. This weather condition made it less likely that nutrient or ashes in
	burned areas had transferred to unburned areas by runoff.
An coefficient of variance analyses that would indicate how	Authors' response: we plotted the mean fluxes with standard deviations as
spatially variable soil GHG fluxes and other variables for each	error bars in figure 1 to show the variance among the 4 plots of the 4
measuring date are is missing	sampling days during each sampling period. Mean ±std also listed in table 1
	and 2 to address the variance. To specify this point, we accepted the
	referee's comments and calculated the CVs in the "Result" section, the
	sentences are also listed below:
	"Coefficient of variance (CV, ratio of the standard deviation to the mean)
	among the plots during the 4 sampling days ranged from 14%-68% (mean
	32%), 9%-15% (mean 10%) and 10%-28% (mean 16%), for Aug 2013, Aug
	2014 and Nov 2014, respectively"
	"Soil CO_2 flux showed relative higher variance as indicated by the higher
	standard deviations (Figure 2) and CVs (ranged from 43% to 50% during
	the three sampling periods)"
The data have not been put into any climatic context – please	Authors' response: we accept the referee's comments and added the
provide weather data (precipitation, air temp, RH) for the study	weather data in table 1.
period and put this in context with your soil moisture and	
temperature measurements	
Please provide soil bulk density and particle size analysis data	Authors' response: we did not measure bulk density in the plots however

for the observe alots	the sails in the masion were relatively home can ave with some anovals and
for the study plots	the soils in the region were relatively homogeneous with some gravels and
	small stones.
For anyone to make an informed decision if your spatial	Authors' response: we have presented our GHG measurements of every
replication was sufficient please provide the raw data of your	single day in Figure 2. The mean and std values calculated should partially
GHG measurements	reflect the spatial replications.
Please provide information what type of regression analysis you	Authors' response: Pearson correlation analysis was applied to detect any
have used and provide adjusted R2 values instead of R values.	potential driving factors on soil greenhouse gas emission. All measured soil
Also please reconsider if 3 data points are enough to do this	properties and gas fluxes at the 4 replicate plots during the three sampling
type of analysis and if you meet the underlying assumptions of	events were pooled together for this analysis. We clarified this point in the
the test used	statistical analysis section, as described below:
	"Collected soil properties and gas fluxes at the four replicate plots during
	the three sampling events were also pooled together for Pearson correlation
	analysis to detect possible effects of soil environmental variables on soil
	CO2, CH4 and N2O fluxes."
Some of the references are not up to date and some of the newer	Author' response: we updated the reference list with a recent published
literature on wildfire or prescribed burning effects on soil GHG	paper about planned burning on temperate eucalypt forest system.
fluxes in eucalypt fluxes is missing	
	"Fest, B. J., Livesley, S. J., Fischer, J. C., and Arnadt, S. K. Repeated fuel
	reduction burns have little long-term impact on soil greenhouse gas
	exchange in a dry sclerophyll eucalypt forest. Agr Forest Meteorol, 201,
	17-25, 2015."
	1 '

1 Impacts of prescribed burning on soil greenhouse gas

2 fluxes in a suburban native forest of south-eastern

3 Queensland, Australia

4

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Abstract

- 18 Prescribed burning is a forest management practice that is widely used in Australia to reduce
- 19 the risk of damaging wildfires. Ht-Prescribed burning can affect both carbon (C) and nitrogen
- 20 (N) cycling in the forest and thereby influence the soil-atmosphere exchange of major
- 21 greenhouse gases, i.e. carbon dioxide (CO₂), methane (CH₄) and nitrous oxide (N₂O). To
- 22 quantify the impact of a prescribed burning (conducted on 27 May 2014) on greenhouse gas
- 23 exchange and the potential controlling mechanisms, we carried out a series of field
- measurements before (August 2013) and after (August 2014 and November 2014) the fire.
- 25 Gas exchange rates were determined at-in 4 replicate sites-plots which were burned during the
- 26 combustion and <u>in</u> another 4 adjacent unburned <u>sites plots</u> located in green islands, using a set
- 27 of static chambers. Surface soil properties including temperature, pH, moisture, soil C and N
- 28 pools were also determined either by in situ measurement or by analysing surface 10 cm soil
- 29 samples. All of the chamber measurements indicated a net sink of atmospheric CH₄, with

mean CH₄ uptake ranging from 1.15 to 1.99 mg m⁻² d⁻¹. The Prescribed burning significantly enhanced CH₄ uptake as indicated by the significant higher CH₄ uptake rates at in the burned sites plots measured in August 2014. While within In the next following 3 months, the CH₄ uptake rate was recovered to the pre-burning levels. Mean CO₂ emission from the forest soils ranged from 2721.76 to 7113.49 mg m⁻² d⁻¹. The effect of prescribed burning on CO₂ emission was limited within the first 3 months, as no significant difference was observed between the burned and the adjacent unburned sites plots in both August and November 2014. The temporal dynamics of the CO₂ emissions presented showed more seasonal variations, rather than the burning effects of prescribed burning. The N₂O emission at in the studied sitesplots was quite low, and no significant impact of prescribed burning was observed. The changes in understory plants and litter layers, surface soil temperature, C and N substrate availability and microbial activities, resulting from following the prescribed burning, were the factors that controlled the greenhouse gas exchanges. Our results suggested that the low intensity prescribed burning would decrease soil CO2 emission and increase CH4 uptake, however, but this effect would be present within a relative short period. Only slight changes in the surface soil properties during the combustion and very limited damages impacts of prescribed burning in on the mineral soils supported the quick rapid recovery of the greenhouse gas exchange rates.

1 Introduction

As the a result of continuously increasing greenhouse gas emissions, global climate change studies have predicted a drier future with high probability of increasing temperatures, lower average rainfall and increase in the frequency and severity of droughts and extreme weather events (Zhao et al., 2013; Sherwood and Fu, 2014; Fu et al., 2015). As for In Australia, climate changes were also identified as key drivers of the increases in days with high fire risk weather and probability of severe wildfires (Murphy and Timbal, 2008; Fest, 2013). In response to these predictions, the use of prescribed burning is increased in Australia forest management to protect both native and plantation forests from the risk of damaging wildfires (Wang et al., 2014). The Prescribed burns are generally targeted at the understorey vegetation and surface litters, while aiming for minimum damage to overstorey trees. Despite the controlled burning conditions, prescribed burning can still have significant effects on altering environmental factors including soil water content and soil temperature. The combustion

event would also result in amounts of charcoal and dying tree roots (Kim et al., 2011; 1 2 Sullivan et al., 2011) and therefore altering alter root activities, decomposition of organic 3 matters decomposition, availability of substrate and soil N dynamics (Weber, 1990; Certini, 2005; Livesley et al., 2011; Wang et al., 2014). All these parameters are closely related to 4 three major greenhouse gas exchanges at soil-atmosphere interface, namely carbon dioxide 5 6 (CO_2) , methane (CH_4) and nitrous oxide (N_2O) . Studies have paid special attentions to soil 7 CO₂, CH₄ and N₂O these greenhouse gas fluxes, not only because of the warming effect caused by CO₂, CH₄ and N₂O in the atmosphere globally (Zhao et al., 2013; Sherwood and 8 Fu, 2014), but also because of their use as very effective indicators for evaluating soil C and 9 N pools and soil microbial activities (Weber, 1990). Many studies have been conducted to 10 quantify CO₂, CH₄ and N₂O exchanges at forest soil-atmosphere interface and the impact of 11 intensive wildfires of in different climate regions, but very few works have reported the 12 effects of prescribed burning on soil greenhouse gas emissions, especially in Australia. 13 14 The CO₂ is the primary greenhouse gas and account for a major part of anticipated global warming (Sommerfeld et al., 1993). Within the forest ecosystem, soil is the major C reservoir 15 while soil respiration is an important mechanism that releases the fixed C into atmosphere 16 (Seidl et al., 2014). Forest fires are generally reported to decrease soil CO₂ efflux (Weber, 17 18 1990; Burke et al., 1997; Kim et al., 2011; Livesley et al., 2011). The reported key driving 19 factors of fires on CO2 efflux are the changes in soil temperature, moisture and fine root activities. As the altered temperature and moisture reflect could change the amplitude of the 20 seasonal variations in CO2 emissions, reduced fine root activities after fires are more 21 responsible for the decreased CO₂ (Kim et al., 2011; Sullivan et al., 2011). Sullivan et al. 22 (2011) also concluded that reduced microbial respiration indicated by microbial biomass after 23 24 prescribed burning could further contribute to the decreased CO₂ efflux. Unlike with this "decrease" effect, Fest et al. (2015) also reported that low intensity burning slightly increased 25 soil CO₂ flux in temperature eucalypt forest systems. This is attributed to the higher inputs of 26 easily decomposable compounds, higher surface temperature and soil nutrient depletion after 27 burning treatments (Fest et al., 2015). 28 29 The CH₄ is the second most important greenhouse gas forcing climate change, with a global warming potential of 25 times greater than CO2. Aerated forest soils are one of the most 30 important biological sink of CH₄ as the results of oxidation of atmospheric CH₄ by 31 methanotrophic bacteria. Studies have reported both positive (Livesley et al., 2011; Sullivan 32 et al., 2011; Fest, 2013) and no significant impacts of fires on forest soil CH₄ uptake (Kim et 33

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forest soil system, the prescribed burning altered soil moisture condition, and removal of 2 3 litter layer and soil O horizon would weaken or eliminate the barrier effect of the surface soil and thereby increase diffusion of CH₄ into soil profile (Sullivan et al., 2011; Fest, 2013). 4 The N₂O emission is less reported in forest soil studies, despite the greater climate warming 5 potential of N₂O than CO₂ and CH₄ and the large contribution of forest soil N₂O emission to 6 the global atmospheric N₂O budget (Kiese and Butterbach-Bahl, 2002; Kiese et al., 2003). 7 Fires would affect soil N₂O emission by altering the rates of nitrification and denitrification 8 9 processes but few published studies have reported inconsistent results; there are only few published studies and their results are inconsistent. Fires could change forest soil N storages 10 but the mechanism of driving N₂O emission changes is unclear (Certini, 2005; Nave et al., 11 2011). Available N substrate (Kiese et al., 2003), soil moisture and temperature (Fest et al., 12 2009), water filled pore space (Kiese and Butterbach-Bahl, 2002) and stand conditions 13 (Butterbach-Bahl et al., 1997) are potential factors which could affect N₂O emission rates. 14 15 The eucalypt dominated natural forest ecosystem studied in this experiment is quite typical 16 across most of Australia's forest areas, with a total of 92 million hectares or 74% of of Australia's forest area (Department Agriculture, 17 http://www.agriculture.gov.au/abares/forestsaustralia/profiles/eucalypt-forest). These forests 18 19 hold important C storage over the country and also provide important ecosystem services 20 such as biodiversity, recreation, water resource and wood products (Fest et al., 2009). Prescribed burning is one of the most important management tools in Australia to protect 21 these forests from firestorm and maintain their functions, including forest regeneration, site 22 preparation, fuel reduction and habitat management (Guinto et al., 2000; Bai et al., 2012; 23 Wang et al., 2014). Some studies have reported the greenhouse gas emissions from Australian 24 25 forest soils (Kiese and Butterbach-Bahl, 2002; Dalal et al., 2003), but the impact of prescribed burning on Australian eucalyptus forests are rarely studied. Therefore, we have 26 limited understanding about the magnitude and direction of the effect of prescribed burning 27 28 on the greenhouse gas exchange which is critical to understand the interaction between burned ecosystem and the atmosphere. In this study, we setup four sampling sites plots which 29 had similar stand conditions to address the following questions: (1) would prescribed fire 30 burning affect greenhouse gas emissions at the soil atmosphere interface? (2) And if so, 31 hHow long would these effects last? and (3) wWhat would be the controlling factors? To 32 address these questions, we conducted a series of field measurements of CH₄, CO₂ and N₂O 33

al., 2011). Since soil gas diffusivity is one of the key regulators of soil CH₄ uptake in the

- 1 exchange at surface soil before and after a-prescribed burning. To examine the potential
- 2 driving factors and mechanisms we also collected surface soil samples for analysing
- 3 biological, chemical and physical variables which might be altered by the prescribed burning.

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2 Methods

2.1 Site description

- 7 The study was carried out in Toohey Forest (27°30'S, 135°02'E), located 10 km south of
- 8 Brisbane in south-eastern Queensland, Australia (Figure 1Figure 1). This forest accounts for
- 9 about 600 hectares dominated by different species of eucalypt and contains some 460 species
- 10 of vascular plants. About half of this forest is a local government conservation reserve and
- surrounded by suburban areas (Catterall and Wallace, 1987; Catterall et al., 2001; Farmer et
- 12 al., 2004). The climate for the region and around is characterized as subtropical with a
- dominant weather pattern of hot, wet summers and cool, dry winters. The mean annual
- 14 rainfall is about 1000 mm, with the majority received during winter summer months.
- 15 Temperatures generally ranged between approximately 30 and 35 °C in summer and 10 and
- 16 15 $^{\circ}$ C in winter.
- 17 Patterns of burning prior to the 1950s are unknown, but from the 1950s to the early 1970s,
- individual fires probably burned across a large proportion of the area. From the early 1970s
- 19 individual fires were confined to more localized areas and created a spatially heterogeneous
- 20 | fire regime. Since 1993, 27 blocks within Toohey Forest haves been conducted with regular
- 21 <u>prescribed</u> burnings (Wang et al., 2015). Prescribed burnings are generally low intensity cool
- burns, and usually occur at the end of the dry season in winter. Generally, every burning
- event would last for several hours (usually overnight) from ignition to extinguishing of any
- small fires. The fire was monitored and controlled during the burning.
- 25 The prescribed burning related withof this study was conducted on 27 May 2014. Last
- 26 recorded burning in this block was in 1 June 1999. Before the prescribed burning, we selected
- 27 4 sites plots (30 m between each other, figure 1) with similar stand conditions for sampling.
- 28 The understory of these sites plots was burned out during the recent prescribed burning, left a
- 29 layer of wood charcoal on the ground. After the <u>prescribed</u> burning, these 4 <u>sites plots</u> were
- measured repeatedly at three months (August 2014) and six months (November 2014) later.
- 31 Detailed sampling dates and weather conditions for the selected sampling events were listed

- in Table 1. Basically, the sampling events were conducted under clear weather condition and there were no major precipitation events either 30 days or 90 days before the sampling events. Additionally Furthermore, another 4 unburned sites—plots adjacent to the burned plots as paired plots were selected to further examine the impacts of prescribed burning. These sites unburned plots were located in some—the adjacent green islands of those not touched by the recent prescribed burning—and were near the existing four burned sites.
- 7 (Figure 1)

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2.2 Sampling method

As previously described, 4 plots was selected for sampling. We treated these 4 plots as replicates and a series of experiments, including impacts of burning on soil greenhouse gas emission, N transformation and litter decomposition, were carried out in each plot, a total of 11 chambers (rings) were established for specific purposes through introducing different treatments (e.g. water or solution application, N isotope enrichment), only the chamber for soil gas sampling was left untreated and the results were presented here.

We adopted a static chamber method to measure CO2, CH4 and N2O emissions simultaneously. A PVC chamber (cylinder with a diameter of 245 mm, wall thickness approximately 4 mm and length 300 mm) was installed in each of the 4 sitesplots. The chambers were sharpened at the bottom edge and were pushed 10 cm into the soil using a hammer. Weed control was performed within and around the chamber to prevent the impacts of grasses. Sampling was conducted a week later after the chambers were set. Gas fluxes across the soil-atmosphere interface were determined daily by sampling air in the headspace of PVC chambers during the 4-day field measurement. The internal volume of a chamber was 9.4 L when placed 10 cm deep into forest floor. There were 4 holes evenly distributed on the chamber (10 cm above ground) to help the inner atmosphere fully mixed with outside, on all sampling occasions prior to each gas sampling, these holes were sealed with a set of rubber plugs. The top of the chamber was also covered with a cap fastened with black rubber band to prevent any gas exchange between the inner headspace and the outside. After covering the cap, 15 ml gas samples were taken from the sampling port at the centre of the chamber top at 0 and 60 min after chamber deployment. A 25 ml syringe was attached to the sampling port and the plunger of the syringe was pumped up and down several times to mix the gases in the chamber before taking a sample. Gas samples were immediately injected into pre-evacuated 15-ml tubes capped with butyl rubber stoppers and prepare for analysis of CO₂, CH₄ and N₂O. Gas sampling was conducted for 4 consecutive days to capture consistent emission patterns from the 4 plots and the data was pooled for statistical analysis (Lennon, 2011).

All gas samplings were conducted during daylight hours from approximately 8:00 to 12:00. However, considering of the great spatial heterogeneity of soil gas fluxes reported by existing studies (Kim et al., 2011; Sullivan et al., 2011), the acquired results in the present study might be tangled with burning induced impacts and spatial related variabilities due to the relative large distance among the plots (Prosser, 2010). We considered the similarity of soil and stand conditions (eucalypt forest with moderate density) and plot location (middle slope) when selecting the plots, which would partly offset this potential impact.

2.3 Soil properties property analysis

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The top 10 cm soil in the chamber was collected with a shovel. Collected soil samples were thoroughly mixed and passed through a 2 mm sieve. Soil moisture was measured gravimetrically after drying at 105 °C for 48 hours. pH was measured with a 1:5 aqueous solutions after shaking for 30 min. Soil inorganic N concentrations were extracted with 2 M KCL and measured using a modified micro-diffusion method (Wang et al., 2015) and a Discrete Chemistry Analyser (Westco Smartchem SC 200, Discrete Wet Chemistry Analyser). To determine water soluble organic C (WSOC) and total N (WSTN), 7 g fresh soil was added to 35 ml distilled water in a 50 ml plastic centrifuge vials, the suspension was then shaken by an end-over-end shaker for 5 min followed by centrifuging at 10000 rpm for 10 min. The suspension was then filtered through a Whatman 42 filter paper and a 33 mm Millex syringedriven 0.45 µm filter successively before analysed by with a Shimadzu TOC-VCSH/CSN 21 TOC/N analyser. Similarly, hot water extractable organic C (HWEOC) and total N (HWETN) were also measured, while the only difference was, 1:5 soil water solution, was incubated in a capped and sealed tube at 70 $\,^{\circ}$ C for 18 hours.

Soil microbial biomass C (MBC) and N (MBN) were determined using the fumigation-25 26 extraction method described by Vance et al. (1987) and Brookes et al. (1985). Briefly, fumigated and non-fumigated soils (5 g dry weight equivalent) were extracted with 25 ml of 27 0.5 M K₂SO₄ (soil/extractant ratio 1:5). The fumigation lasted for 16 hours. Samples were 28 shaken for 30 minutes and then filtered through a Whatman 42 filter paper. Soluble organic C 29 and total soluble N (TSN) in the fumigated and non-fumigated samples were determined 30 31 using a Shimadzu TOC-VCSH/CSN TOC/N analyser. MBC and MBN were calculated using

- 1 conversion factors of 2.64 and 2.22 for C (Vance et al., 1987) and N (Brookes et al., 1985),
- 2 respectively.

3 2.4 Gas analysis

- 4 Collected gas samples were sent to laboratory for gas chromatography (GC) analysis for CO₂,
- 5 CH₄ and N₂O concentrations right shortly after the field campaign sampling. The
- 6 concentrations of CO₂ and CH₄ was were measured using a GC system (GC-2010 PLUS
- 7 Shimadzu) with Flame Ionization Detector and N₂O was measured using the same GC system
- 8 with Electron Capture Detector. The standards (0.5 ppm for CH₄, 400.5 ppm for CO₂ and 0.3
- 9 ppm for N₂O) were run before and after each set of samples to ensure the reproducibility of
- 10 measurements. Gas fluxes for CO₂, CH₄ and N₂O were determined from a regression analysis
- with gas concentrations change within the chamber versus time (Zhao et al., 2013).

2.5 Statistical analysis

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- 13 All statistical analyses were performed using IBM SPSS STATISTICS (version 20) software.
- 14 One-way ANOVA was introduced to examine statistically significant differences between
- 15 soil gas fluxes measured before and after the burning in the burned plots. This analysis was
- also applied to compare the fluxes between burned and unburned plots in Aug 2014 and Nov
- 17 <u>2014, respectively.Repeated measures ANOVA was used to examine statistically significant</u>
- 18 differences and changing patterns of soil gas fluxes and soil variables following the
- 19 prescribed burning with the measurement date as the repeated factor. Collected soil properties
- and gas fluxes at the four replicate plots during the three sampling events were also pooled
- 21 <u>together for Pearson correlation analysis Correlation analysis was tested for to detect possible</u>
- 22 effects of soil environmental variables on soil CO₂, CH₄ and N₂O fluxes.

3 Results

3.1 Greenhouse gas exchange rates before and after prescribed burning

- 26 Average CH₄, CO₂ and N₂O emissions rates of the 4 replicate sites plots for each sampling
- event were listed in Table 1 Table 2. While temporal patterns of gas exchange for the 4-day
- sampling of the 3 sampling periods were shown in Figure 2. Coefficient of variance
- 29 (CV, ratio of the standard deviation to the mean) among the plots during the 4 sampling days
- 30 ranged from 14%-68% (mean 32%), 9%-15% (mean 10%) and 10%-28% (mean 16%), for

Aug 2013, Aug 2014 and Nov 2014, respectively. All the sampling plotssites showed 1 negative CH₄ emissions rates during the three sampling events, or uptake atmospheric CH₄. 2 At In the burned plotssites, mean CH₄ uptake was significant significantly increased by 64% 3 three months after the prescribed burning (p < 0.001), while during the third sampling period, 4 5 CH_4 uptake had similar CH₄ uptake rate as that became similar to that before the burning (p = 6 0.843). At-In the unburned plotssites, CH₄ uptake was relative-relatively stable during the dates of each the sampling periods and also showed less variation in uptake rate between 7 8 August 2014 and November 2014. The significant difference in mean \underline{CH}_4 uptake rate in August 2014 (p < 0.001) but similar rates in November 2014 (p = 0.921) also confirmed that 9 10 the CH₄ uptake increased at the first three months but was recovered to the pre-burning level about six months after the prescribed burning. 11 Soil CO₂ flux showed relative higher variance as indicated by the higher standard deviations 12 (Figure 2) and CVs (ranged from 43% to 50% during the three sampling periods). Mean CO₂ 13 emission from all sampling burned sites plots was significantly decreased by 41% in August 14 2014 (p < 0.001). In November 2014, CO_2 efflux rates had exceeded that before the burning 15 by 28% but the difference was not significant (p = 0.392). Similar CO₂ emission rates 16 between the burned and adjacent unburned plotseites during the sampling dates in-of August 17 18 2014 (p = 0.549) and in November 2014 (p = 0.218) were also observed. This might indicated 19 that the temporal dynamics detected at the burned plots reflected more natural variations 20 rather than burning induced impacts. As for The lower N2O, lower emission rates, compared to that in August 2013, were found 21 both in August 2014 (p = 0.003) and November 2014 (p < 0.001). During the three sampling 22 periods, the study sites plots were not solely performed as a source of atmospheric N₂O, on 23 27 August 2013, 6 August 2014 and most days enof November 2014, but the plotssites also 24 25 took up N₂O from the atmosphere. No observed significant difference in N₂O emission was observed between the burned and unburned sites plots in both August and November 2014. 26 (Table **12**) 27

(Figure 2) 28

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Soil basic properties and their relationships with gas exchange rates

After the prescribed burning, mean-soil moisture-of the surface soil showed no significant difference between burned and unburned plotssites (p = 0.804), although most of the

sampling plotssites (5 out of 8 for the two sampling events in 2014) had relative higher values. Soil temperature was slightly higher during for most sampling dates at in the burned plotssites, but no significant difference was found in August 2014 (p = 0.644) and November 2014 (p = 0.751). The pH in the surface soil was higher in 2014 than in 2013, and the values at in all burned plotssites were slightly higher than those of unburned sites (p = 0.293). NO₃-N was quite low both before and after the prescribed burning, but NH₄-N was significantly increased after the prescribed burning. When relating these soil parameters to greenhouse gas emissions from the soil surface, soil temperature showed a positive correlation with the CH_4 uptake (R = 0.232, p = 0.044) and CO_2 efflux (R = 0.47, p < 0.000) and a negative correlation with N_2O emission (R = -0.284, p = 0.011). pH was negatively correlated with the CH_4 uptake (R = -0.595, P = 0.006) and CO_2 (R = -0.591, p = 0.006) emission. NH₄-N was negatively correlated with N₂O emission (R = -0.591, p = 0.006)0.533, p = 0.015). (Table 23)

3.3 Soil C and N dynamics before and after burning

MBN had the same dynamics as MBC.

There was no significant difference in WSOC at-in the burned plotssites between August 2013 and August 2014, and only slightly decreased WSOC was observed in August 2014. However, WSOC was significant higher in November 2014 (p = 0.034).-Comparing to the unburned sites, WSOC in most burned sites (3 out of 4) was lower in August 2014 (p = 0.387) while higher in November 2014 (p = 0.237). No significant difference was found between any sampling periods for WSTN, despite higher WSTN at-in some burned plotssites than those before the prescribed burning and in the unburned plotssites. HWEOC was significantly increased in August 2014 than that in August 2013 (p < 0.001) and in November 2014 it was recovered to the level before the prescribed burning (p = 0.929). The difference in HWEOC between the burned and adjacent unburned plotssites were also significant in August 2014 (p = 0.0361) but insignificant in November 2014. The situation was similar for HWETN.

Mean-MBC at in the burned plotssites in August 2014 was 378.94 mg kg⁻¹, which was lower than that in August 2013 (522.45 mg kg⁻¹, p = 0.069), and this value did not change much in November 2014 (380.37 mg kg⁻¹). Burned sites also showed lower MBC values when compared to the unburned sites both in August (p = 0.121) and November (p = 0.516) 2014.

- 1 The correlation analysis between soil C or N pools and gas emissions showed that CH₄
- 2 uptake was negatively correlated with WSOC (R = 0.523, p = 0.018). CO_2 efflux has had
- 3 negative correlation with HWEOC (R = -0.690, p = 0.001) and HWETN (R = -0.730, p <
- 4 0.001). N₂O emission was positively correlated with MBN (R = 0.565, p = 0.009).

4 Discussions

4.1 Burning ilmpacts of prescribed burning on soil properties

The pPrescribed burning has resulted in a slightly increase in the surface soil temperature of this study, which is in the agreement with most existing of the literature results (Burke et al., 1997; Certini, 2005). The burning of the understory vegetative vegetation cover, together with the resulted consumption of fuel loadss, removal of litter layer and increased charred materials on the soil surface would all moderate affect soil temperatures by intercepting direct sunlight and moderating the loss of soil heat by radiation. However, the controlled burning condition or low fire intensity limited this difference at thean insignificant level. Meanwhile, the 4 selected plotssites in the study did not show consistent fluctuations in the surface soil moisture before and after the prescribed burning, which would generally be expected to decrease after a fire (Burke et al., 1997; Kim et al., 2011; Sullivan et al., 2011). Generally, increased soil temperature, combined with the reduced shade, would result in higher evaporation rates and therefore restricts the movement of water into soil profile (Burke et al., 1997; Certini, 2005). This be might attributed to the representativeness of the measurements and one measurement for each sampling period might not suffice be adequate to represent the physical state of water in the soil (Weber, 1990).

An increase in <u>soil</u> pH values was found <u>at-in</u> the burned a<u>plotsreas</u> in August 2014 and it was returned to a <u>comparative similar</u> level in November 2014. Although no significant difference was found between <u>the</u> burned and <u>the referenceadjacent</u> unburned <u>plotssites</u> in 2014, <u>pH</u> values for the burned <u>plotssites</u> were still higher than those at the unburned <u>plotssites</u>. The increased pH after the <u>prescribed</u> burning would be probably due to the release of extractable basic cations from the deposited ashes during the burning. Several studies also <u>find-reported</u> increased pH after <u>the</u> fire (Guinto et al., 1999; Certini, 2005; Kim et al., 2011; Xue et al., 2014) and the increased pH would either <u>be</u> recovered to <u>the pre-burning unburned-level</u> within a year (Rhoades et al., 2004; Xue et al., 2014) or last for longer periods (Arocena and Opio, 2003; Ponder Jr et al., 2009; Granged et al., 2011), depending on the site condition and burning intensities.

NH₄-N was significantly increased after the <u>prescribed</u> burning, but no significant changes were observed for NO₃-N₄- <u>s</u>Since NH₄-N <u>wais</u> a direct product of combustion and NO₃- <u>wais</u> formed from NH₄⁺ some weeks or months later as a result of nitrification (Covington and Sackett, 1992; Diaz-Raviña et al., 1992; Wang et al., 2015). Hence, the increase in NH₄-N was probably due to the transformation of organic N during the combustion. Also the deposition of organic N in ash and enhanced ammonification would also contribute to the increased NH₄⁺ (Knoepp and Swank, 1993; Wan et al., 2001). This was also supported by the similar <u>phenomenon found by findings of</u> Nardoto and Bustamante (2003) in savannas of Central Brazil and Covington and Sackett (1992) in a ponderosa pine forest in USA.

The <u>prescribed</u> burning <u>has</u>-significantly reduced MBC in the surface soil and it showed no apparent sign of recovery six months <u>later</u>-after the <u>prescribed</u> burning. Decreased MBC after prescribed burning or wildfires have been reported and it would normally last for several years (Prieto-Fernández et al., 1998). As for the other two soil liable organic C pool indicators, WSOC showed no significant change before and after burning while HWEOC <u>was</u> significantly increased in August 2014 and returned to <u>the pre-burning level in Novemebr 2014</u>. While the low intensity of <u>the prescribed burning may only cause volatilization of organic C to a limited extent, soil microbes might be decreased due to their sensitiveness to temperature (Hernández et al., 1997; Neary et al., 1999). This microbial lysis, as well as the heat-induced alterations of soil organic matter, contributed to the release of carbohydrates which were reflected by the initial increase in HWEOC.</u>

4.2 Variations in greenhouse gas exchanges and their driving factors

4.2.1 CH₄ uptake

The CH₄ uptake rates before <u>prescribed</u> burning and six months after <u>the</u> burning from <u>the</u> burned <u>plotssites</u> and all fluxes from <u>the</u> unburned <u>plotssites</u> fall in the range of CH₄ fluxes <u>obtained reported</u> by Kiese et al. (2003) (<u>varies from 0.84-1.63 mg m⁻² d⁻¹</u>) and a recent study by Rowlings et al. (2012) which were conducted in a similar forest ecosystem in Australia. While the high uptake rate of CH₄ three months after the burning was also comparable to the results obtained in Australia forests under extreme dry conditions (Fest et al., 2009; Rowlings et al., 2012). The prescribed burning increased CH₄ uptake in this study. The same effect has also been reported by Burke et al. (1997) and Sullivan et al. (2011). However, unlike most studies reporting that the enhanced CH₄ uptake may last for several years, our results

indicated that CH₄ uptake rate was returned to the pre-burning level within six months after the prescribed burning. We obtained this conclusion from the similar CH₄ uptake rates in November 2014 when compared to the CH₄ uptake at in the unburned plotssites and the rates before the burning atin the burned plotssites. The low fire intensity of the prescribed burning in this study may might cause less impact on the system and therefore shorten the required time to recover to the pre-burning conditions for the studied forest. Studies have found that fire intensity has significant effect on forest soil CH₄ consumption and CO₂ emissions while severe wildfires always impact gas exchange rates for the subsequent several years (Burke et al., 1997; Neary et al., 1999; Sullivan et al., 2011). Kim et al. (2011) also found a quick recovery of CH₄ uptake that after 2 years of low intensity burnings in a Japanese forest.

Soil moisture has been shown to be a key parameter controlling CH₄ consumption by the soils through limiting the transport of atmospheric CH₄ to microbial communities living at depth in the soil (Burke et al., 1997). However, we observed no significant relationships between soil moisture and CH₄ uptake as reported by other studies (Sommerfeld et al., 1993; Kiese et al., 2003; Livesley et al., 2011). This was 7 probably due to the low intensity burning, and hence the prescribed burning did not affect the soil water conditions in the soil horizons relevant to the CH₄ oxidation, or the soil moisture was partly recovered to the pre-burning level and was also lower than the range of strong moisture control. Castro et al. (1994) found that moisture control was strongest when between 60% and 100% of available soil pore space was water filled. Meanwhile, surface soil temperature appeared to show more significant influence on CH₄ uptake in this study. We also found weak but significant relationship between CH₄ uptake and soil pH. The mechanism of how increased pH would affect soil CH₄ uptake after fire is not clear, and Jaatinen et al. (2004) estimated that the increased pH after fire caused any change in the methanotroph community and would not be directly responsible for the increased uptake rates. Therefore, the increased pH in our study would probably indirectly affect CH₄ uptake together with other fire introduced changes.

The relative high and significant correlation between CH₄ uptake and WSOC indicates that the decreased soil C may might have increased CH₄ diffusion into the soil profile. Removal of the C rich O horizon caused by the prescribed burning eliminated a barrier of for CH₄ diffusion. This is also supported by the decreasing CH₄ uptake and recovered WSOC in November 2014, combined with recovered litter deposit and ground plants regrowth. This effect of prescribed burning reduced thickness of organic layer to CH₄ uptake was also found in similar forest ecosystems (Saari et al., 1998; Steinkamp et al., 2001). Another possible

reason for the observed characteristics of CH₄ uptake is the physical changes in the surface soil. Although not measured in the current study, literature has shown that low to moderate fires would increase soil structure stability due to the formation of the dydrophobic film on the external surface of aggregates (Mataix-Solera and Doerr, 2004). With lack of the protection of ground plants and litter layers, surface soil was more likely to loose more fine fractions and lead to soil coarsening by the increased erosion (Certini, 2005). These physical changes in the surface soil would all create aform channels ideal for diffusion of atmosphere CH₄ into the soil profile and thereby increase CH₄ oxidation rates.

4.2.2 CO₂ effluxes

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The studied plotssites acted as a persistent source of atmospheric CO₂ before and after the prescribed burning, while the CO₂ emission rates, either before or after the burning, were similar to the results obtained by Carlyle and Than (1988) in a native forest with low soil moisture (about 5520 mg m⁻² d⁻¹) and by Rowlings et al. (2012) in an Australian subtropical rainforest (around 3600 mg m⁻² d⁻¹). However, the CO₂ emission values were much lower than the reported high soil respirations (over 20000 mg m⁻² d⁻¹) in various Australian forest ecosystems with high soil moisture and temperature (Carlyle and Than, 1988; Fest et al., 2009; Rowlings et al., 2012). A number of existing studies have reported reduced CO₂ emissions after the fire due to diminished root activity and lower root respirations (Czimczik et al., 2006; Kim et al., 2011; Sullivan et al., 2011), and the effects would last for various length from less than 2 years (Weber, 1990; Irvine et al., 2007) to longer periods (Burke et al., 1997). However, in this study, it seems that CO₂ emission started recovering three months after the burning. This is supported by the similar CO_2 emission rates in August 2014 (p = 0.218) and November 2014 (p = 0.549) between the burned and the adjacent unburned plotssites. The decreased CO₂ flux three months after the burning, which is only 41% of that before burning, might be attributed to the reduced root respiration and decomposition activities related to soil microbial communities. The observed high HWEOC value in August 2014 probably indicated increased dead fine roots after the combustion of ground biomass, which was in agreement with the estimation that after removing the aboveground biomass, most fine roots would die within 2 months by Fahey and Arthur (1994). Meanwhile, studies have reported that root respiration could contribute up to 50% of the total soil respiration (Irvine and Law, 2002; Zerva and Mencuccini, 2005). Decomposition of the dead fine roots could lead to the flush of C substrate but this was limited by the decreased microbial

activities in this study, suggested by the significant lower MBC and MBN values in August and November 2014. Meanwhile, lower MBC inat the burned plotssites compared to the unburned plotssites probably also suggested a reduced heterotrophic contribution to total soil respiration. Similar finding was also reported by Sullivan et al. (2011) who concluded that MBC explained a large proportion of the variation in soil CO2 flux at the burned site than at the unburned sites in a ponderosa forest in south-western USA. Considering the positive relationship between CO₂ efflux and WSOC, which was an important part of soil labile C, the microbial biomass was likely to be limited by the amount of labile C available for assimilation into microbial biomass. Maheswaran and Attiwill (1989) and Zerva and Mencuccini (2005) also reported reduced CO₂ emission which was related to reduced microbial populations limited by an available source of C after the fire. Such explanation can be further supported by the subsequent recovery ofing CO₂ flux six months after the burning, combined with recovered MBC to the near pre-burning level and higher WSOC levels than before the burning. It was also reasonable that higher temperature in November 2014 had stimulated the surface soil respiration and therefore could contribute to the higher CO2 emission rate.

4.2.3 N₂O emissions

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The soil-atmosphere fluxes of N₂O measured in the current-study were very small (-0.21 to 18 0.54 mg m⁻² d⁻¹ before the burning and -0.18-0.11 mg m⁻² d⁻¹ after the burning). These low 19 fluxes were similar to the small N₂O emission reported by Fest et al. (2009) and Livesley et al. 20 (2011), but was much lower than the range of 0.75-8.19 mg m⁻² d⁻¹ recorded by Kiese et al. 21 (2003) in a tropical rainforest and the range of 0.62-1.57 mg m⁻² d⁻¹ by Rowlings et al. (2012) 22 in a subtropical rainforest. No significant effect of prescribed burning was observed on the 23 N₂O emission. Since forest soils were generally accepted as a source of atmospheric N₂O 24 (Butterbach-Bahl et al., 1997), the negative values we measured might be attributed to the 25 changes in N₂O concentration during the chamber employment were quite low during the 26 chamber employment, and these changes were below the detection limit of the GC system. 27 Even though the dry and well aerated soil of the sampled sites makesplots would make it 28 prone to nitrification rather than denitrification, the observed small inorganic N pool (NH₄⁺ < 29 than 10 mg N kg⁻¹ while $NO_3^- < 0.1$ mg N kg⁻¹) dominated by NH_4^+ also limited the 30 nitrification processes. Although there was a significant increase in NH₄⁺ three months (p = 31 0.009) and six months (p = 0.009) after the burning, nitrification was still negligible. This 32

could be attributed to enhanced situation of low soil water availability and dry conditions after prescribed burning, due to since the removal of understory plants and litter layers and increased evapotranspiration rates would, limited the activities of soil nitrifiers (Livesley et al., 2011). Also the prescribed burning induced charcoal at the soil surface which would also supress N₂O exchange rates which wereas reported in a recent controlled experiment in Japan (Kim et al., 2011). However, the accumulated N substrate, either NH₄⁺ or NO₃⁻, might cause further high N₂O emissions with under appropriate conditions, for example, wet after precipitations.

4.3 Contribution of the gas emissions to the burning introduced greenhouse gas effect

Although consistently consuming atmospheric CH₄, the forest soil in Toohey Forest still acteds as a net C source to the atmosphere, due to the greater CO₂ emission rates during the studied period. However, the burning induced lower CO₂ emission and higher CH₄ uptake rates could significantly reduce the amount of C released into atmosphere, especially when extending these effects to the first several months after the burning. This reduced C emission could partly compensate the greenhouse gas effect during the operation of the burning prescribed burning could cause eruption of CO₂ into the atmosphere by combusting photosynthetic fixed C embedded in understory plants, litter layers, surface soil organic C and also the consumption of fossil fuels to manipulate the fires. Data on the C burnedloss due to prescribed burning, fuel consumed and continuous measurement of soil gas exchanges are required to quantify the burning—caused greenhouse effect in future studies.

5 Conclusion

 The low intensity prescribed burning in Toohey Forest caused changes in both soil properties and greenhouse gas exchange rates. Soil CH₄ uptake was significantly enhanced due to the increased CH₄ diffusivity into the soil profiles. The removal of litter layer and surface soil organic materials and the altered soil physical structural caused by the prescribed burning were the major factors contributing to the increased CH₄ diffusion. The CO₂ emission was largely decreased but it was a combination of burning introduced variation and natural seasonal variations largely caused by natural annual variations. Changes in root respiration and soil microbial community were the two controlling factors related to burning the effect of prescribed burning on CO₂ emission. Due to the controlled condition of the prescribed

- 1 burning, both CH₄ uptake and CO₂ emission started to recover about three months after the
- 2 burning and it would appears that the gas exchange rates were recovered to the pre-burning
- 3 level about six months after the burning. This quick-rapid recovery was closely related to the
- 4 limited effect of prescribed burning on the soil and no dramatic damages in the mineral soils.
- 5 However, the decreased CO₂ emission and increased CH₄ uptake during this period could still
- 6 partly compensate the greenhouse gas effect caused by the combustion of C during the
- 7 | burning. The N₂O emission was quite low at in the studied sites plots and showed no obvious
- 8 impacts from the of prescribed burning. Finally, a continuous monitoring of soil properties
- 9 and soil greenhouse gas exchanges and even ecosystem gas exchange rates before and after
- 10 burning is important to reveal the key mechanisms and quantify the complex impacts of
- 11 burning on forest ecosystem and regional climate.

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References

- 19 Arocena, J. M., and Opio, C.: Prescribed fire-induced changes in properties of sub-boreal
- 20 forest soils, Geoderma, 113, 1-16, 2003.
- 21 Bai, H. S., Sun, F., Xu, Z., Blumfield, T., Chen, C., and Wild, C.: Appraisal of ¹⁵N
- 22 enrichment and ¹⁵N natural abundance methods for estimating N₂ fixation by understorey
- 23 Acacia leiocalyx and A. disparimma in a native forest of subtropical Australia, J Soils
- 24 Sediments, 12, 653-662, 2012.
- 25 Brookes, P. C., Landman, A., Pruden, G., and Jenkinson, D. S.: Chloroform fumigation and
- 26 the release of soil nitrogen: a rapid direct extraction method to measure microbial biomass
- 27 nitrogen in soil., Soil Biol Biochem, 17, 837-842, 1985.
- Burke, R. A., Zepp, R. G., Tarr, M. A., Miller, W. L., and Stocks, B. J.: Effect of fire on soil-
- 29 atmosphere exchange of methane and carbon dioxide in Canadian boreal forest sites, J
- 30 Geophys Res-Atmos, 102, 29289-29300, 1997.

- 1 Butterbach-Bahl, K., Gasche, R., Breuer, L., and Papen, H.: Fluxes of NO and N₂O from
- 2 temperate forest soils: Impact of forest type, N deposition and of liming on the NO and N₂O
- 3 emissions, Nutr Cycl Agroecosys, 48, 79-90, 1997.
- 4 Carlyle, J. C., and Than, U. B.: Abiotic controls of soil respiration beneath an eighteen-year-
- old pinus radiata stand in south-eastern Australia, J Ecol, 76, 654-662, 1988.
- 6 Castro, M. S., Melillo, J. M., Steudler, P. A., and Chapman, J. W.: Soil moisture as a
- 7 predictor of methane uptake by temperate forest soils, Can J Forest Res, 24, 1805-1810, 1994.
- 8 Catterall, C. P., and Wallace, C. J.: An Island in Suburbia: the Natural and Social History of
- 9 Toohey Forest., Institute of Applied Environmental Research, Griffith University., Brisbane,
- 10 1987.
- 11 Catterall, C. P., Piper, S. D., Bunn, S. E., and Arthur, J. M.: Flora and fauna assemblages vary
- with local topography in a subtropical eucalypt forest, Austral Ecol, 26, 56-69, 2001.
- 13 Certini, G.: Effects of fire on properties of forest soils: a review, Oecologia, 143, 1-10, 2005.
- 14 Covington, W. W., and Sackett, S. S.: Soil mineral nitrogen changes following prescribed
- burning in ponderosa pine, Forest Ecol Manag, 54, 175-191, 1992.
- 16 Czimczik, C. I., Trumbore, S. E., Carbone, M. S., and Winston, G. C.: Changing sources of
- soil respiration with time since fire in a boreal forest, Glob Change Biol, 12, 957-971, 2006.
- Dalal, R. C., Wang, W., Robertson, G. P., and Parton, W. J.: Nitrous oxide emission from
- 19 Australian agricultural lands and mitigation options: a review, Aust J Soil Res, 41, 165-195,
- 20 2003.
- 21 Diaz-Raviña, M., Prieto, A., Acea, M. J., and Carballas, T.: Fumigation-extraction method to
- estimate microbial biomass in heated soils, Soil Biol Biochem, 24, 259-264, 1992.
- 23 Fahey, T. J., and Arthur, M. A.: Further-studies of root decomposition following harvest of a
- 24 northern hardwoods forest, For Sci, 40, 618-629, 1994.
- 25 Farmer, D., Catterall, C. P., and Piper, S. D.: Abundance patterns across months and locations,
- and their differences between migrant and resident landbirds in lowland subtropical eucalypt
- 27 forest, Emu, 104, 283-296, 2004.
- 28 Fest, B.: The impact of fire disturbance and simulated climate change conditions on soil
- 29 methane exchange in eucalypt forests of south-eastern Australia, Department of Forest and

- 1 Ecosystem Science, Melbourne School of Land and Environment, University of Melbourne,
- 2 2013.
- 3 Fest, B. J., Livesley, S. J., Drösler, M., van Gorsel, E., and Arndt, S. K.: Soil-atmosphere
- 4 greenhouse gas exchange in a cool, temperate Eucalyptus delegatensis forest in south-eastern
- 5 Australia, Agr Forest Meteorol, 149, 393-406, 2009.
- 6 Fest, B. J., Livesley, S. J., Fischer, J. C., and Arnadt, S. K. Repeated fuel reduction burns
- 7 have little long-term impact on soil greenhouse gas exchange in a dry sclerophyll eucalypt
- 8 <u>forest. Agr Forest Meteorol, 201, 17-25, 2015.</u>
- 9 Fu, L., Zhao, Y., Xu, Z. H., and Wu, B. F.: spatial and temporal dynamics of forest
- 10 aboveground carbon stocks in rsponse to climate and environmental changes, J Soils
- 11 Sediments, 15, 249-259, 2015.
- 12 Granged, A. J. P., Jordán, A., Zavala, L. M., Muñoz-Rojas, M., and Mataix-Solera, J.: Short-
- 13 term effects of experimental fire for a soil under eucalyptus forest (SE Australia), Geoderma,
- 14 167–168, 125-134, 2011.
- Guinto, D. F., Xu, Z. H., Saffigna, P. G., House, A. P. N., and Perera, M. C. S.: Soil nitrogen
- mineralisation and organic matter composition revealed by ¹³C NMR spectroscopy under
- 17 repeated prescribed burning in eucalypt forests of south-east Queensland, Aust J Soil Ress,
- 18 37, 123-136, 1999.
- 19 Guinto, D. F., Xu, Z., House, A. P. N., and Saffigna, P. G.: Assessment of N₂ fixation by
- 20 understorey acacias in recurrently burnt eucalypt forests of subtropical Australia using ¹⁵N
- 21 isotope dilution techniques, Can J Forest Res, 30, 112-121, 2000.
- Hern ández, T., Garc á, C., and Reinhardt, I.: Short-term effect of wildfire on the chemical,
- 23 biochemical and microbiological properties of Mediterranean pine forest soils, Biol Fert Soils,
- 24 25, 109-116, 1997.
- 25 Irvine, J., and Law, B. E.: Contrasting soil respiration in young and old-growth ponderosa
- 26 pine forests, Glob Change Biol, 8, 1183-1194, 2002.
- 27 Irvine, J., Law, B. E., and Hibbard, K. A.: Postfire carbon pools and fluxes in semiarid
- ponderosa pine in Central Oregon, Glob Change Biol, 13, 1748-1760, 2007.
- Jaatinen, K., Knief, C., Dunfield, P. F., Yrj ål å, K., and Fritze, H.: Methanotrophic bacteria in
- 30 boreal forest soil after fire, FEMS Microbiol Ecol, 50, 195-202, 2004.

- 1 Kiese, R., and Butterbach-Bahl, K.: N₂O and CO₂ emissions from three different tropical
- 2 forest sites in the wet tropics of Queensland, Australia, Soil Biol Biochem, 34, 975-987, 2002.
- 3 Kiese, R., Hewett, B., Graham, A., and Butterbach-Bahl, K.: Seasonal variability of N₂O
- 4 emissions and CH₄ uptake by tropical rainforest soils of Queensland, Australia, Glob
- 5 Biogeochem Cy, 17, n/a-n/a, 2003.
- 6 Kim, Y. S., Makoto, K., Takakai, F., Shibata, H., Satomura, T., Takagi, K., Hatano, R., and
- 7 Koike, T.: Greenhouse gas emissions after a prescribed fire in white birch-dwarf bamboo
- 8 stands in northern Japan, focusing on the role of charcoal, Eur J Forest Res, 130, 1031-1044,
- 9 2011.
- 10 Knoepp, J. D., and Swank, W. T.: Site preparation burning to improve southern Appalachian
- 11 pine-hardwood stands: nitrogen responses in soil, soil water, and streams, Can J Forest Res,
- 12 23, 2263-2270, 1993.
- 13 Lennon, J. T.: Replication, lies and lesser-known truths regarding experimental design in
- 14 environmental microbiology, Environ. Microbiol, 13, 1383-1386, 2011.
- Livesley, S. J., Grover, S., Hutley, L. B., Jamali, H., Butterbach-Bahl, K., Fest, B., Beringer,
- 16 J., and Arndt, S. K.: Seasonal variation and fire effects on CH₄, N₂O and CO₂ exchange in
- savanna soils of northern Australia, Agr Forest Meteorol, 151, 1440-1452, 2011.
- 18 Maheswaran, J., and Attiwill, P. M.: Soil respiration in eucalypt forests of southeastern
- 19 Australia, Biol Fert Soils 8, 154-159, 1989.
- 20 Mataix-Solera, J., and Doerr, S. H.: Hydrophobicity and aggregate stability in calcareous
- 21 topsoils from fire-affected pine forests in southeastern Spain, Geoderma, 118, 77-88, 2004.
- 22 Murphy, B. F., and Timbal, B.: A review of recent climate variability and climate change in
- southeastern Australia, Int J Climatol, 28, 859-879, 2008.
- 24 Nardoto, G. B., and Bustamante, M. M. C.: Effects of fire on soil nitrogen dynamics and
- 25 microbial biomass in savannas of Central Brazil, Pesqui Agropecu Bras, 38, 955-962, 2003.
- Nave, L. E., Vance, E. D., Swanston, C. W., and Curtis, P. S.: Fire effects on temperate forest
- 27 soil C and N storage, Ecol Appl, 21, 1189-1201, 2011.
- 28 Neary, D. G., Klopatek, C. C., DeBano, L. F., and Ffolliott, P. F.: Fire effects on
- belowground sustainability: a review and synthesis, Forest Ecol Manag, 122, 51-71, 1999.

- 1 Ponder Jr, F., Tadros, M., and Loewenstein, E. F.: Microbial properties and litter and soil
- 2 nutrients after two prescribed fires in developing savannas in an upland Missouri Ozark
- 3 Forest, Forest Ecol Manag, 257, 755-763, 2009.
- 4 Prieto-Fern ández, A., Acea, M. J., and Carballas, T.: Soil microbial and extractable C and N
- 5 after wildfire, Biol Fert Soils, 27, 132-142, 1998.
- 6 Prosser, J. I.: Replicate or lie, Environ. Microbiol, 12, 1806-1810, 2010.
- 7 Rhoades, C. C., Meier, A. J., and Rebertus, A. J.: Soil properties in fire-consumed log
- 8 burnout openings in a Missouri oak savanna, Forest Ecol Manag, 192, 277-284, 2004.
- 9 Rowlings, D. W., Grace, P. R., Kiese, R., and Weier, K. L.: Environmental factors controlling
- temporal and spatial variability in the soil-atmosphere exchange of CO₂, CH₄ and N₂O from
- an Australian subtropical rainforest, Glob Change Biol, 18, 726-738, 2012.
- 12 Saari, A., Heiskanen, J., and Martikainen, P. J.: Effect of the organic horizon on methane
- oxidation and uptake in soil of a boreal Scots pine forest, FEMS Microbiol Ecol, 26, 245-255,
- 14 1998.
- 15 Seidl, R., Schelhaas, M.-J., Rammer, W., and Verkerk, P. J.: Increasing forest disturbances in
- Europe and their impact on carbon storage, Nature Climate Change, 4, 806-810, 2014.
- 17 Sherwood, S., and Fu, Q.: A Drier Future?, Science, 343, 737-739, 2014.
- 18 Sommerfeld, R. A., Mosier, A. R., and Musselman, R. C.: CO₂, CH₄ and N₂O flux through a
- 19 Wyoming snowpack and implications for global budgets, Nature, 361, 140-142, 1993.
- 20 Steinkamp, R., Butterbach-Bahl, K., and Papen, H.: Methane oxidation by soils of an N
- 21 limited and N fertilized spruce forest in the Black Forest, Germany, Soil Biol Biochem, 33,
- 22 145-153, 2001.
- Sullivan, B. W., Kolb, T. E., Hart, S. C., Kaye, J. P., Hungate, B. A., Dore, S., and Montes-
- 24 Helu, M.: Wildfire reduces carbon dioxide efflux and increases methane uptake in ponderosa
- pine forest soils of the southwestern USA, Biogeochemistry, 104, 251-265, 2011.
- Vance, E. D., Brookes, P. C., and Jenkinson, D. S.: An extraction method for measuring soil
- 27 microbial biomass C, Soil Bio. Biochem, 19, 703-707, 1987.
- 28 Wan, S., Hui, D., and Luo, Y.: Fire effects on nitrogen pools and dynamics in terrestrial
- 29 ecosystems: a meta-analysis, Ecol Appl, 11, 1349-1365, 2001.

- 1 Wang, Y., Xu, Z., and Zhou, Q.: Impact of fire on soil gross nitrogen transformations in
- 2 forest ecosystems, J. Soils Sediments, 14, 1030-1040, 2014.
- Wang, Y., Xu, Z., Zheng, J., Abdullah, K., and Zhou, Q.: δ^{15} N of soil nitrogen pools and their
- 4 dynamics under decomposing leaf litters in a suburban native forest subject to repeated
- 5 prescribed burning in southeast Queensland, Australia, J Soils Sediments, 15, 1063-1074,
- 6 2015.
- 7 Weber, M. G.: Forest soil respiration after cutting and burning in immature aspen ecosystems,
- 8 Forest Ecol Manag, 31, 1-14, 1990.
- 9 Xue, L., Li, Q., and Chen, H.: Effects of a wildfire on selected physical, chemical and
- 10 biochemical soil properties in a pinus massoniana forest in south China, Forests, 5, 2947-
- 11 2966, 2014.
- 12 Zerva, A., and Mencuccini, M.: Short-term effects of clearfelling on soil CO₂, CH₄, and N₂O
- fluxes in a Sitka spruce plantation, Soil Biol Biochem, 37, 2025-2036, 2005.
- 14 Zhao, Y., Wu, B. F., and Zeng, Y.: Spatial and temporal patterns of greenhouse gas emissions
- 15 from Three Gorges Reservoir of China, Biogeosciences, 10, 1219–1230, 2013.

Table 1. Weather conditions for the three sampling events, precipitation was recorded by the Mt Gravatt Alert weather station (27.55° S, 153.07° E, ~2 km from the sampling plots) and the data were collected at the website of Bureau of Meteorology (http://www.bom.gov.au).

Sampling events	Sampling dates	Antecedent precipitation* (30 days, mm)	Antecedent precipitation (3 months, mm)	Rainfall during sampling period	Temperature measured on- site
Aug 2013	<u>27-30</u>	<u>4</u>	<u>71</u>	<u>0</u>	24.76
Aug 2014	<u>5-8</u>	10^{1}	<u>22</u>	<u>0</u>	<u>23</u>
Nov 2014	<u>10-13</u>	<u>11</u>	<u>85²</u>	<u>0</u>	<u>29.88</u>

* Total rainfall for the indicated periods

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- 5 <u>1 highest daily rainfall was 7 mm recorded on 27 Jul 2014</u>
 - 2 most rainfall for this period was recorded on 17 August 2014 (69 mm)

Sites Dates		CH ₄ mg m ⁻² d ⁻¹	CO ₂ mg m ⁻² d ⁻¹	N ₂ O mg m ⁻² d ⁻¹		
	Aug 2013	-1.21 (0.42)a	5009.17 (2657.67)a	0.21 (0.24)a		
Burned	Aug 2014	-1.99 (0.51)bA	2974.24 (895.78)bA	0.00 (0.10)aA		
	Nov 2014	-1.17 (0.78)aC	5835.69 (2639.99)aB	-0.04 (0.07)bB		
T.I	Aug 2014	-1.28 (0.26)B	2721.76 (1360.24)A	0.02 (0.11)A		
Unburned	Nov 2014	-1.15 (0.16)C	7113.49 (3086.07)B	-0.01 (0.09)B		

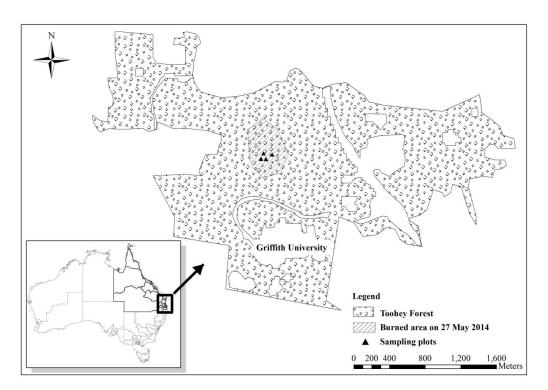
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Table 23. Surface soil properties in Toohey Forest before and after the prescribed burning. Values in parentheses indicate standard errors for the 4 replicates replicate plots of each sampling period. Soil moisture is presented in %. Other parameters (except pH) are presented in mg kg⁻¹

Sites	Date	Moisture	pН	NH ₄ -N	NO ₃ -N	WSOC	WSTN	HWEOC	HWETN	MBC	MBN
	Aug 2013	12.3	4.33	1.78	0.02	88.83	7.10	875.44	67.80	522.45	56.37
	Aug 2013	(4.4)	(0.10)	(0.55)	(0.03)	(13.54)	(0.83)	(180.32)	(10.38)	(76.18)	(14.2)
Durnad	Aug 2014	10.3	5.76	6.76	0.09	80.00	6.81	2809.99	183.75	378.94	35.77
Burned		(2.7)	(0.17)	(2.30)	(0.05)	(20.20)	(2.16)	(479.18)	(39.10)	(103.58)	(10.00)
	NOV 2014	10.3	4.88	10.63	0.10	148.09	7.38	893.47	54.08	444.68	48.02
		(3.0)	(0.24)	(4.18)	(0.12)	(38.25)	(2.97)	(310.29)	(19.32)	(45.27)	(7.15)
	Aug 2014	10.1	5.44	3.79	0.03	97.27	7.22	3638.29	203.91	493.62	45.26
Unburned		(2.0)	(0.24)	(1.67)	(0.04)	(28.36)	(1.19)	(571.01)	(17.70)	(73.81)	(8.19)
Onburned	NOV 2014	9.8	4.67	6.44	0.04	114.35	5.14	942.56	51.94	406.07	42.76
		(2.3)	(0.16)	(1.83)	(0.07)	(28.30)	(1.29)	(254.30)	(13.02)	(249.90)	(14.56)



2 Figure 1. Map of the study site in Toohey Forest located in south-eastern Queensland, Australia.

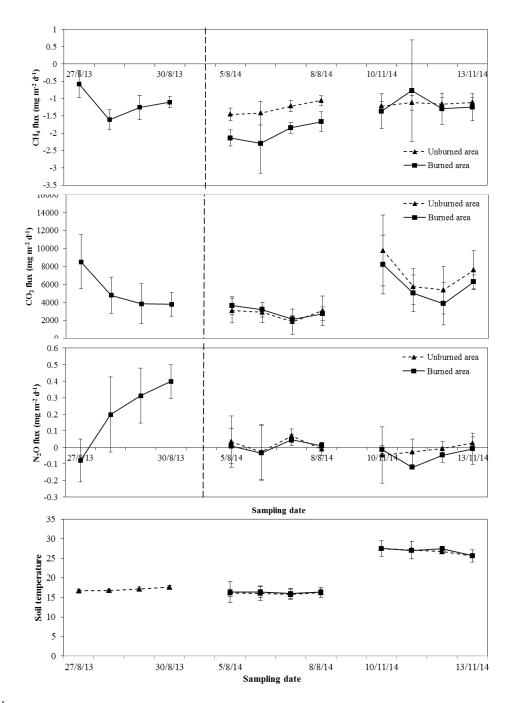


Figure 2. CH₄, CO₂ and N₂O exchange rates <u>and on-site measured soil temperature</u> before and after the prescribed burning. The dash line indicated the date of burning conducted on 27

- 1 May 2014. Each sampling period lasted for 4 days and the values were averaged from the 4
- 2 selected sites plots each day. The vertical bars indicated the standard error of the mean.