

## ***Interactive comment on “The role of termite CH<sub>4</sub> emissions on ecosystem scale: a case study in the Amazon rain forest” by Hella van Asperen et al.***

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### **Response to Anonymous Referee 2**

*Interactive comment on “The role of termite CH<sub>4</sub> emissions on ecosystem scale: a case study in the Amazon rain forest” by Hella van Asperen et al, Received and published: 22 December 2020*

**This manuscript presents a well thought out study to quantify methane emissions by termites in the Amazon rain forest. The authors reviewed the literature extensively and compared/discussed with their findings. I have some comments**

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**that I think will make the study more valuable.**

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Thank you for your kind words and the time you spent on reviewing our manuscript. We are grateful for your suggestions, which we have used to improve the manuscript. Below you will find a point to point response to each of your raised concerns and, if applicable, the corrected and improved manuscript text.

In addition, we would like to point out that the given termite emission estimates have changed due to an improved termite weight determination.

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**1. Please provide the estimate of CH<sub>4</sub> emissions by termite and put in context with the overall CH<sub>4</sub> budget globally or in the Amazon. This manuscript presents CH<sub>4</sub> emission factors only. Without knowing how many termite mounds in Amazon, it's difficult to imagine the scale of the global CH<sub>4</sub> budget. I think this is one of a key messages for readers.**

Thank you for this interesting point. Below we will:

- elaborate on our considerations regarding the termite mound-upscaling;
- provide a 'back of the envelope' estimate on the role of termite emissions in the Amazon CH<sub>4</sub> budget, and in the global CH<sub>4</sub> budget;
- show the revised manuscript text.

**Termite mound upscaling:** Based on mound density numbers, it is difficult to state

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a termite CH<sub>4</sub> emission estimate for the whole Amazon. As stated in the discussion (§4.3), mound density numbers vary largely between ecosystems. There is only little data available on mound density numbers, and most Amazon studies were performed in close proximity to our fieldsite (due to the research activities of local institute INPA). While this relatively large amount of local studies is unique and useful for the upscaling for our *local* ecosystem, it is unwise to assume that these mound density numbers apply to the whole Amazon. For this reason, we choose to only state a mound CH<sub>4</sub> emission estimate for our specific ecosystem, and to inform the readers about the limitations of this estimate.

**Back-of-the-envelope estimate for the global CH<sub>4</sub> budget:** By use of the data presented in the comprehensive modeling study of Kirsche et al. (2013), the following back-of-the-envelope estimate can be made:

Kirsche et al (2013) (Table 1) stated an annual global termite emission of 11 Tg CH<sub>4</sub> year<sup>-1</sup>. They state that 36% of termite emissions originate from the region 'tropical South America' (p 818, first sentence), which calculates to 3.96 Tg CH<sub>4</sub> year. Substituting the used termite emission factor of 2.8 μg CH<sub>4</sub> g<sub>termite</sub><sup>-1</sup> h<sup>-1</sup> by the value found in our study of 5.6 μg CH<sub>4</sub> g<sub>termite</sub><sup>-1</sup> h<sup>-1</sup>, would lead to a doubling of the regions estimated termite emission, namely 7.92 Tg instead of 3.96. The global estimate would increase from 11 Tg to 14.96 Tg.

The termite emission factor is a practical estimate of the average termite emission, which can be used for CH<sub>4</sub> budget studies. Since our study only measured one termite species, and there is likely a variation between species and ecosystems, we do not suggest that the currently used termite emission factor of 2.8 μg CH<sub>4</sub> g<sub>termite</sub><sup>-1</sup> h<sup>-1</sup> should be replaced by our value. We do however want to show and point out that the termite emission factor is still an uncertain part in the tropical CH<sub>4</sub> budget.

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To include the reader in this train-of-thought, we have revised this part of the manuscript:

**Revised text in §4.3:** As a 'back-of-the-envelope' calculation, based on Kirsche et al. (2013): 36% of global termite emission (11 Tg) is expected to come from the region of 'tropical South America' (0.36\*11=3.96 Tg). Substituting the emission factor of 2.8 with the newly found 5.6 μg CH<sub>4</sub> g<sub>termite</sub><sup>-1</sup> h<sup>-1</sup> would increase this regions estimate to 7.92 Tg, and the global estimate to 14.96 Tg.

Our study points out that termite emissions are still an uncertain source in the CH<sub>4</sub> budget, and are especially poorly quantified for the Amazon rain forest. Measurement of CH<sub>4</sub> emissions from different termite species, preferably covering species of different feeding or nesting habits, in combination with more precise termite distribution and abundance data, would allow more precise estimates and a better understanding of the role of termites in the CH<sub>4</sub> budget.

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**2. The first sentence in the Introduction section, it says "Methane (CH<sub>4</sub>) is the second most important long-lived anthropogenic greenhouse gas." I think CH<sub>4</sub> has been recognized to be "short-lived" climate pollutant.**

Thank you for pointing this out. We have changed the first sentence to:

**Revised text:** Methane (CH<sub>4</sub>) is the second most important anthropogenic greenhouse gas, but its natural sources are still not well understood.

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**3. In the Introduction section, Line 35, it says "Recently, it was shown that termites have a mitigating effect during droughts in tropical rain forests". Please elaborate what mitigating effect.**

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Ashton et al. (2019) performed a termite suppression experiment and found that termite activity increased during drought, resulting in accelerated litter decomposition, elevated soil moisture, greater soil nutrient heterogeneity, and higher seedling survival rates. The authors suggested different underlying mechanisms for this response such as more favorable conditions for tunneling (e.g., drier, less-waterlogged ground), increased foraging ability above ground in the absence of heavy rain, and/or reduced predation pressure from ants.

We have changed the text in the Introduction to:

**Previous text:** Recently, it was shown that termites have a mitigating effect during droughts in tropical rain forests.

**Revised text:** Recently, it was shown that termites increase their activity during droughts, resulting, among others, in enhanced litter decomposition, elevated soil moisture and higher seedling survival rates, thereby demonstrating a mitigating effect during droughts in tropical rain forests.

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**4. In the Introduction and in Appendix, the authors touched on N<sub>2</sub>O emissions from termite but didn't give conclusive results.**

We agree that this point is not sufficiently discussed. An elaboration on this subject can be found at point 7.

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**5. Section 2.3, Line 129, LGR GHG analyzer was mentioned to be the instrument deployed to quantify CH<sub>4</sub> emissions in flux chambers. I think authors should add brief instrument performance specifications and details of what calibration and drift evaluation have been done in Amazon. While the absolute CH<sub>4</sub> concentrations in flux chamber measurements are not very critical, since it's to measure the CH<sub>4</sub> concentration increase, but the manuscript does not provide the measured concentrations and jumped directly to the emission factor estimates. For example, LGR UGGA precision is about 2 ppb. Does it perform the same in Amazon? Also, what CH<sub>4</sub> concentration increments measured in the flux chambers? If it was only 2 ppb, then that data would not be useful. I think it should be many times more than the instrument precision and drift.**

Thank you for raising this point. During the campaigns, we have set the Los Gatos instrument to the 10-second averaging modus. Calibration gases were measured every second day for 5 minutes, resulting in a precision ( $1\sigma$ ) of  $\sim 0.7$  ppm and  $\sim 3.0$  ppb for respectively CO<sub>2</sub> and CH<sub>4</sub>.

The concentration increases during the 20 min chamber closure were large. Concentrations were climbing from forest concentrations to concentrations of up to 5750 ppb CH<sub>4</sub>, and up to 1950 ppm CO<sub>2</sub>, thereby far exceeding the measurement precision of the Los Gatos instrument.

We have added the following lines to the revised manuscript (beginning of Results):

**Revised text in §3.1:** Headspace concentrations increased strongly during chamber closure, and chamber concentrations reached up to 5750 nmol CH<sub>4</sub> mol and 1950  $\mu$ mol CO<sub>2</sub> mol<sup>-1</sup>.

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**6. Well-designed flux chambers should have a small mixing fan or internal distribution tubing to quantify fluxes. §2.5 describes how LGR sampling tubes were connected on top of a 220 L chamber, if the air inside is not well mixed, the two fittings on top of the chamber may not detect CH<sub>4</sub> at the bottom of the chamber.**

Thank you for raising this point. Below we will:

- clarify the locations of the inlet fittings;
- elaborate on why we did not install a fan, and how we ensured mixed chamber air;
- give the revised manuscript text.

The 220 L chamber had two fittings on each side of the bucket while the smaller soil chamber had the two fittings on top of the chamber. Re-reading §2.5, we agree with the reviewer that the text is confusing, and we have revised this part.

As a small side note, termite mounds emit CH<sub>4</sub> from its entire surface, thereby presenting a sphere-shaped source of 45-65 cm height *inside* the chamber head space. Therefore, we do not expect a large difference between CH<sub>4</sub> concentrations at the top and the bottom of the chamber headspace.

We were hesitant about installing a small mixing fan. On the one hand, the absence of a mixing fan might lead to an underestimation of the flux (Christiansen et al. 2011). On the other hand, a mixing fan might lead to turbulence in the head space (Janssens

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et al. (2000), Pumpanen (2004)), which possibly induces unrepresentatively high CH<sub>4</sub> emissions from the mound.

Since we wanted to avoid overestimation of termite mound CH<sub>4</sub> fluxes, we decided to not install a mixing fan. Instead we installed a 4 inlet vertical sampling tube inside the chamber head space, a technique to minimize the effects of gas concentration gradients in the head space (Clough et al, 2020). Inside the chamber at fitting height (~30 cm), a T-piece with two 20 cm-long Teflon tubing was positioned vertically, and two small incisions were made, so that head space air was sampled from 4 different heights (approx. at 10, 25, 35 and 50 cm height from the soil). The sampling tube was tested in the lab to verify whether air was sampled from all 4 inlets.

We have added the following lines and references to the revised manuscript:

**Revised text in §2.3:** Two one-touch fittings (1/4 inch, SMC Pneumatics) were installed on each side of the bucket. To minimize the possible effects of gas concentration gradients in the headspace, we installed a 4 inlet vertical sampling tube inside the chamber, so that air was sampled from different heights (~10, ~25, ~35 and ~50 cm) in the headspace (Clough et al, 2020).

**Revised text in §2.5:** To be able to connect the Los Gatos instrument, the soil chamber had two one-touch fittings on top.

#### References

- Christiansen, Jesper Riis, et al. "Assessing the effects of chamber placement, manual sampling and headspace mixing on CH<sub>4</sub> fluxes in a laboratory experiment." *Plant and soil* 343.1-2 (2011): 171-185.
- Clough, Timothy J., et al. "Global Research Alliance N<sub>2</sub>O chamber methodology guidelines: Design considerations." *Journal of Environmental Quality* 49.5 (2020): 1081-1091.
- Janssens, Ivan A., et al. "Assessing forest soil CO<sub>2</sub> efflux: an in situ comparison of four techniques." *Tree physiology* 20.1 (2000): 23-32.
- Pumpanen, Jukka, et al. "Comparison of different chamber techniques for measuring soil CO<sub>2</sub> efflux." *Agricultural and Forest Meteorology* 123.3-4 (2004): 159-176

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7. Appendix A1 and A2 talk about N<sub>2</sub>O calibrations and measured concentrations. The measured N<sub>2</sub>O concentrations are outside of the calibration range. While the lower range (333.7 ppb) is similar to NOAA's measurements in Brazil, the manuscript does not provide the FTIR instrument precision and therefore, it's difficult to determine whether the detected range (333.7-342.4 ppb) is within instrument drift or it's actually an increment of N<sub>2</sub>O. I don't think the authors can conclude there isn't N<sub>2</sub>O emissions.

Thank you for pointing this out. Below we will:

- explain why FTIR N<sub>2</sub>O concentration measurements outside the calibration range can be used, by stating the precision and linearity of this instrument;
- explain why we can conclude that there are low N<sub>2</sub>O emissions, by calculating the methods detection limit;
- support our statement (very low N<sub>2</sub>O emissions) with additional data.

First of all, to clarify, the mentioned range of 333.7-342.4 ppb was measured over *all* chambers during the whole week. Actual increments during individual chamber closures were a lot smaller, as discussed here below. We have clarified this in the revised manuscript text.

The FTIR-instrument has the following reported precision ( $1\sigma$ ) for 10 minute-averaged spectral analyses: 0.02  $\mu\text{mol mol}^{-1}$ , 0.2 nmol mol<sup>-1</sup>, 0.2 nmol mol<sup>-1</sup>, 0.06 nmol mol<sup>-1</sup>, and 0.04 permil, for respectively CO<sub>2</sub>, CH<sub>4</sub>, N<sub>2</sub>O, CO, and  $\sigma^{13}\text{C}$  of CO<sub>2</sub>.

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Measurements performed during the campaign week were set to 5 minutes, so that a precision of  $1/\sqrt{N}$  is achieved, which is 0.09 nmol mol<sup>-1</sup> for N<sub>2</sub>O.

The FTIR instrument has been shown to be linear for all gases in the ambient concentration range, and linearity was tested for N<sub>2</sub>O in the range 300-350 ppb. So while the choice of calibration gases was not optimal, we are confident that the FTIR-instrument still performs well in this concentration range.

**Detection limit of measurements:**

As also requested by reviewer 1, we calculated the minimum N<sub>2</sub>O flux detectable by this instrument and method:

- Assuming bag samples taken at 2, 5 and 8 minutes during chamber closure.
- Given: collar area 0.25 m<sup>2</sup>, chamber volume 220 L, mound volume 50 L, headspace volume 220-50 = 170 L.
- Assuming: molar volume of 24.5 L mol<sup>-1</sup> (1 atm, 25 °C).
- Minimum detectable concentration difference is ( $2\sigma$ ) 0.18 nmol mol<sup>-1</sup>.
- A concentration difference between t=2 min and t=5 min of 0.18 nmol mol<sup>-1</sup> is caused by a flux of 0.027 nmol collar/mound<sup>-1</sup> s<sup>-1</sup>.

So, given the parameters above, the chamber set up has a detection limit of 0.027 nmol mound<sup>-1</sup> s<sup>-1</sup>.

The FTIR-instrument has a cross sensitivity with CO<sub>2</sub>, which is well determined for CO<sub>2</sub> <800  $\mu\text{mol mol}^{-1}$ , but is less certain for unnaturally high CO<sub>2</sub> concentrations.

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For this reason, we preferred to only use the N<sub>2</sub>O headspace concentration measurements with CO<sub>2</sub> <800 μmol mol<sup>-1</sup>. Only 5 mound chamber closures had two consecutive N<sub>2</sub>O concentration points (t=2min and t=5min) with CO<sub>2</sub> <800 μmol mol<sup>-1</sup>, and only 3 sets of two-consecutive concentration points passed the minimum concentration difference of 0.18 nmol mol<sup>-1</sup>. These differences were ~0.2, ~0.3 and ~0.7 nmol mol<sup>-1</sup>, leading to a calculated N<sub>2</sub>O flux of ~0.03 - ~0.11 nmol mound<sup>-1</sup> s<sup>-1</sup>.

**Additional measurements to support statement ‘very low N<sub>2</sub>O emissions’:** In October 2020, additional valley soil N<sub>2</sub>O flux measurement were performed with the same chamber system and collars (5 collars, 3 repetitions), but with a longer closing time (35 min), without termite mounds (so lower CO<sub>2</sub>), and with 4 measurements per chamber closure. Also during these measurements, concentration increases were very low. Out of 15 measurements, 8 measurements had an R<sup>2</sup>>0.90, and calculated fluxes ranged between 0.008-0.106 nmol m<sup>-2</sup> s<sup>-1</sup> (average=0.032 nmol m<sup>-2</sup> s<sup>-1</sup>, sd=0.33). Since the valleys are known to be low on nitrogen (Quesada et al., 2010), such low fluxes are expected, and similar N<sub>2</sub>O valley soil fluxes were found by Matson et al (1987) in a fieldsite closeby.

Since the 3 calculated mound N<sub>2</sub>O flux measurements are based on only 2 consecutive headspace concentration points, no uncertainty can be given, wherefore we preferred not to state the fluxes in the previous manuscript. For the revised manuscript, we have stated the detection limit, explain why not all mound fluxes could be calculated, and support our observation of low N<sub>2</sub>O mound fluxes by the additional soil N<sub>2</sub>O flux measurements:

**Appendix A2:** Gas samples (3 samples per chamber closure) revealed stable N<sub>2</sub>O concentrations, and headspace concentrations ranged between 333.7 and 342.4 nmol mol<sup>-1</sup> over the different chamber closures. Since headspace CO<sub>2</sub> concentrations sometimes exceeded 800 μmol mol<sup>-1</sup>, and N<sub>2</sub>O-CO<sub>2</sub>

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cross-sensitivity becomes uncertain at higher CO<sub>2</sub> concentrations, not all 3 headspace samples per chamber closure could be used, wherefore qualitative N<sub>2</sub>O flux estimates cannot be reported. As a back-of-the-envelope calculation, N<sub>2</sub>O fluxes were calculated if 2 consecutive headspace samples were with CO<sub>2</sub> <800 μmol mol<sup>-1</sup>, and if a minimum N<sub>2</sub>O concentration difference of 0.18 nmol mol<sup>-1</sup> was found (FTIR precision (σ) for 5 min spectra is 0.09 nmol mol<sup>-1</sup>), which gave us 3 mound flux estimates ranging between ~0.03 and ~0.11 nmol N<sub>2</sub>O mound<sup>-1</sup> s<sup>-1</sup>. Similarly low fluxes were found during additionally performed flux measurements, performed as part of a substudy, which showed valley soil fluxes ranging between 0.008-0.106 nmol N<sub>2</sub>O m<sup>-2</sup> s<sup>-1</sup>. The low mound fluxes would be in agreement with a previous study which suggested that termite mound N<sub>2</sub>O emissions are dependent on the N-content of the termites diet (Brauman et al., 2015), which is expected to be low in the valleys of this ecosystem (Quesada et al., 2010).

**References:**

-Matson, Pamela A., and Peter M. Vitousek. "Cross-system comparisons of soil nitrogen transformations and nitrous oxide flux in tropical forest ecosystems." *Global Biogeochemical Cycles* 1.2 (1987): 163-170.

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