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Goettingen March 6, 2017

RE: Response to reviewer comments

First, we would like to thank the reviewers for two very thorough reviews; both had excellent suggestions that will improve our final manuscript. The reviews had several commonalities but altogether presented different visions as to the focus of our paper. Although Reviewer 1 would have liked to have seen more emphasis on specific microbial groups/processes, we have chosen to maintain our original vision for this study, which, as Reviewer 2 summarized, "tests and further develops our theoretical understanding by employing these natural gradients to tease-out the synergistic effects of rainfall, nutrients, and other environmental drivers on modulating soil-atmosphere trace gas exchange." However, we have incorporated many of the specific suggestions from both reviewers as outlined below.

Reviewer 2

It would be useful if the authors could develop or speculate on the wider implications of NO uptake for local and regional atmospheric chemistry, as I do not believe that our wider community has fully engaged with the notion of soils as NO sinks, given the past emphasis on soils as NO emission sources.

We agree that our observation of regular NO uptake by soils is worth exploring, given that until now the majority of studies have reported soils acting as net NO sources. However, we do not want to overemphasize this point, as our soils experienced unique ambient conditions (i.e. high atmospheric NO and O3) that may not occur in many other sites worldwide. However, in response to this comment, we have revised this paragraph in the discussion to emphasize the importance of these unusual results. The final sentence of Section 4.4 (Line 545-549) now reads: In summary, although the soils in our study sites can be a net source of NO, particularly during the dry season (Fig. 2d) and in sites where ambient air NO concentrations are low (Fig. 5), most of the time the soils acted as net sinks of NO, signifying the importance of soil and vegetation as NO sinks (Jacob and Bakwin, 1991; Sparks et al., 2001) in areas affected by anthropogenic NO sources.

First, it would be useful if the authors could make more use of multiple regression or the mixed effects models to determine the hierarchy (i.e. relative importance) of environmental drivers for different trace gases (i.e. which are the dominant and which are the lesser environmental controls?). While the authors have outlined the dominant role of soil moisture, it would be interesting to see a clearer description of the relative importance of the other drivers. Does the hierarchy of drivers vary among sites? Do the hierarchy of drivers vary among seasons?

In response to this comment and comments from Reviewer 1 (see below), we added Table S1, which shows the hierarchy of importance of the soil factors controlling soil GHG fluxes for each season (across sites) and within each site (across seasons).

Table S1 Ranking^a of soil factors that control the soil-atmosphere trace gas exchange along orthogonal precipitation and fertility gradients in the Panama Canal watershed, central Panama (F- and P-value of the model ANOVA shown in brackets).

| | CO_2 | CH ₄ | N_2O | NO |
|------------------|--|---|-------------------------------------|----------------------|
| | | 1. Moisture (F=59.1, | | |
| | 1. NH ₄ ⁺ (F=24.5, | P<0.01) | | |
| Wet season | P<0.01) | 2. Temperature | 1. NO ₃ (F=6.1, | nc |
| (all sites) | 2. Temperature (F=9.4, | (F=10.0, P<0.01) | P=0.01) | ns |
| | P<0.01) | 3. NO_3 (F=5.6, | | |
| | | P=0.02) | | |
| | | 1. Moisture (F=10.5, | | |
| | 1. Moisture (F=52.4, | P<0.01) | | |
| Dry season | P<0.01) | 2. NO_3 (F=14.6, | ns | ns |
| (all sites) | 2. Temperature | P<0.01) | II.S | 113 |
| | (F=5.01, P=0.03) | 3. NH ₄ ⁺ (F=7.8, | | |
| | | P<0.01) | | |
| | 1. Moisture (F=38.0, | | | |
| Met | P<0.01) | ns | ns | ns |
| (wet/dry) | 2. NH ₄ ⁺ (F=13.3, | | 113 | 110 |
| | P<0.01) | | | |
| | 1. Temperature | 1. Moisture (F=33.1, | | 1. Temperature |
| P27 | (F=25.9, P<0.01) | P<0.01) | ns | (F=10.1, P<0.01) |
| (wet/dry) | 2. Moisture (F=22.7, | 2. Temperature (F=5.2, | | 2. Moisture (F=7.4, |
| | P<0.01) | P=0.03) | | P<0.01) |
| | 1. Moisture (F=25.8, | | | |
| P08 | P<0.01) | 1. Moisture (F=30.8, | 1. Moisture (F=12.8, | 1. Moisture (F=16.6, |
| (wet/dry) | 2. Temperature | P<0.01) | P<0.01) | P<0.01) |
| P19 (wet/dry) | (F=20.6, P<0.01) | | | |
| | 1. Moisture (F=44.2, | | 1. Moisture (F=27.7, | |
| | P<0.01) | 1. Moisture (F=32.5, | P<0.01) 2. NO ₃ (F=14.2, | ns |
| | 2. NH ₄ ⁺ (F=4.2, | P<0.01) | | |
| | P=0.04) | | P<0.01) | |
| | 1. Moisture (F=18.8, | 1. Moisture (F=62.5, | | |
| P32 (wet/dry) | P<0.01) | P<0.01) | 1. Moisture (F=7.2, P<0.01) | ns |
| | 2. Temperature | 2. NH ₄ ⁺ (F=7.8, | | |
| | (F=16.0, P<0.01) | P<0.01) | | |

3. NO₃ (F=4.2, P=0.04)

Second, in the section on soil CO_2 flux, I think it would be useful if the authors could revise the text to incorporate a slightly expanded discussion of how root respiration could be influencing variations in soil CO_2 fluxes (see point 10 below). For example, could the differences in respiration between this study site and others be attributed to differences in belowground biomass or root/shoot allocation? Do data exist on belowground biomass in these sites? If so, do those data help explain patterns in soil respiration?

We do not know of any data existing on root biomass in any of our present sites. From our previous work and that of others, we know root respiration can contribute 30% - 35% of the soil CO₂ efflux (van Straaten et al. 2011, Silver et al. 2005). However, we do not have any root data to base any possible contribution of roots to the soil CO₂ fluxes at our present sites. Interestingly, regardless of the contribution of autotrophic respiration to the soil CO₂ fluxes, we did not detect any significant differences in soil CO₂ fluxes among sites, but only found that across our 5 sites the temporal pattern of soil CO₂ fluxes was strongly related to soil moisture contents (Fig. 3). The range of soil moisture contents in these 5 sites (Fig. 4a) also clearly showed that the low-rainfall sites varied from the lower end up to the mid-moisture range, the high-rainfall site varied from the mid to high-moisture ranges and there was a wide overlap among sites within the mid-moisture ranges (Fig. 4a). Thus, if both autotrophic and heterotrophic responded similarly to these ranges of soil moisture contents, then their relative contributions should be less important than their overall response, or the response of soil CO₂ fluxes as a whole, to soil moisture contents.

Thus, in order to avoid any unnecessary speculative discussion, we prefer to focus our discussion on the possible causes of the generally low soil CO₂ fluxes from our present sites as compared to the other lowland forests in Panama (lines 370-379) – possibly due to low root respiration as well as considerable variation in litterfall (as a substrate for heterotrophic respiration). Here, we speculate that autotrophic respiration could be low at two of our sites since they have lower tree densities (particularly at Met and P27; see 2.1) than the old growth, lowland forests on BCI and Gigante.

To summarize, we focused our discussion on the temporal pattern, which our data have clearly shown, as well as reporting on similarities and differences with other studies from CSA lowland forests (see lines 380-389).

Third, in section 4.2 of the Discussion, the authors have identified separate sets of controls on CH4 uptake that appear to be operating on different time scales; i.e., daily fluxes of CH4 appear to be more strongly linked to soil moisture, whereas soil fertility was a stronger constraint on annual CH4 fluxes. This is an important and interesting finding, as it highlights the scale-dependency of different environmental controls, and suggests that different environmental factors may be controlling different aspects/components of trace gas cycling; e.g. in the short-term, soil moisture may be regulating transport and supply of CH4 to methanotrophs (hence, regulating instantaneous fluxes), whereas in the long-term, site fertility

^a This ranking (denoted by numbers) signifies its hierarchy of importance based on the minimal adequate LME model, using a stepwise model simplification; ns – no soil factor showed significant relationship with the soil trace gas fluxes.

may be influencing the total amount of methanotrophic biomass or the overall methanotrophic potential of these soils. It would be useful if the authors could consider a way of revising the current text to better highlight this important finding, as it has wider implications for upscaling these results or incorporating these findings into process-based models.

We have made major revisions to Section 4.2 in order to highlight better the scale-dependency of different environmental drivers of soil CH₄ fluxes.

Additional comments

1. Lines 158-165: It would be useful know the precision of the analysis; i.e. what was the coefficient of variation for the standards?

2. Lines 173-174: Ibid.

We added the detection limits of our instruments, which were calculated as 3 x standard error of the standard, which was used to check the instrument precision during the analysis. The average detection limit during the periods of our measurements was 50 ppm CO₂, 43 ppb N₂O, 45 ppb CH₄, and 0.04 ppb NO/mV (mV is the electrical signal from the produced chemiluminescence of the oxidized NO).

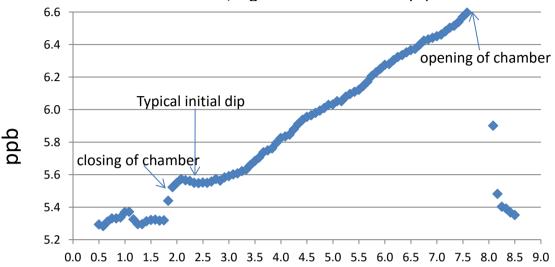
3. Lines 175-177: Were any fluxes non-linear? How were these data treated? Under more saturated soil moisture conditions, was there any evidence of ebullition? If so, how were these data treated?

Soil NO fluxes were always linear. We show below the typical soil NO fluxes where we observed net emission and net consumption. We considered the first 3-min. of linear change in NO concentrations with chamber closure time.

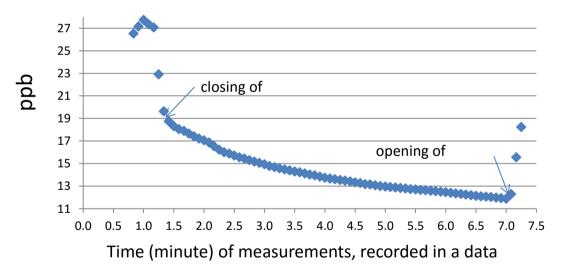
For soil CO₂, N₂O and CH₄fluxes, all 3 gases were analyzed in our gas chromatograph sequentially from the same gas sample. Since these 3 gases come from the same sample, we based our best fit of gas concentration vs. time on the CO₂ concentration increase, as it is the gas with the highest concentration among these 3 gases. The CO₂ concentration always increased linearly with time of chamber closure. Hence, we used a linear fit for all the 3 gases, and zero fluxes and negative fluxes (i.e. for N₂O and CH₄) were all included in our data analysis. This linear increase was not surprising, considering that the large volume of our chambers (11 L) decreases the likelihood of feedbacks on the diffusion gradient with increasing concentration; additionally, there was generally low soil CO₂ and N₂O fluxes at our sites (as we noted in the Discussion, lines 365-366 for CO₂, lines 462-469 for N₂O).

We also did not observe any evidence of ebullition (e.g. sudden increase of gas concentration during our 30-min chamber closure). Such a phenomenon is more likely under flooded conditions or in the transition periods to and from flooded conditions. We measured the volumetric moisture content continuously in our wettest site (plot 32) during the study period, using permanently installed water content probes (Campbell Scientific CS616, Logan, Utah), the same instrument we describe in our earlier studies in another lowland forest in Gigante, Panama (Koehler et al., 2010, Veldkamp et al. 2013, and Corre et al. 2014). The water-filled pore space in the top 10 cm, recorded in the data logger every 4 hours, did not reach saturation at any time during our measurement period.

(a) Plot 8 on June 22, 2010 (showing net NO emission, Fig. 2d of the manuscript)



(b) Metropolitano on June 16, 2010 (showing net NO consumption, Fig. 2d of the manuscript)



4. Lines 205-207: Are there any potential limitations associated with using this 15N natural abundance technique?

The potential limitations of using 15N natural abundance of the soil is its inherent high spatial variability brought about by 1) vegetation species differences, and 2) surface topography, which may drive differences in soil 15N natural abundance due to slope influences on water and solute distribution and ultimately on microbial N-cycling processes. These are the reasons why we used not only the 15N natural abundance of the surface depth but also of the 4 depth increments, and determined the overall 15N nat. abund. enrichment factor (ε) (lines 209-215), which considers the change in 15N natural abundance signature and total N concentration with

depth in relation to the surface depth, as an integrative indicator of soil N availability (shown in our previous studies, e.g. Baldos et al. 2015).

5. Lines 254-256: Have the authors considered using Box-Cox transformations to normalise the data? If successful, this would enable the authors to use parametric statistics (e.g. linear regression, multiple regression) rather than Spearman's Rank correlation. Moreover, even if the data do not fully meet the assumptions for parametric analyses, it may be useful/instructive to analyse the data using multiple regression techniques to evaluate the relative hierarchy of environmental drivers.

We have added an additional table (Table S1 above) to show the relative hierarchy of environmental drivers within sites (across seasons) and within seasons (across sites). Non-parametric statistics were only used to compare non-repeated measures with annual and seasonal averages.

6. Lines 271-286: It is worthwhile reporting the seasonal trends (or, lack of trends in NH4+) here as well. Does NH4+ show wet or dry season differences? I had assumed not given that this wasn't stated explicitly.

Line 280 states that of the four repeated measures (temperature, moisture, extractable NO_3^- and extractable NH_4^+), only moisture and extractable NO_3^- exhibited strong seasonal differences. Additionally, we have added a statement (line 294) specifically clarifying that temperature and extractable NH_4^+ exhibited between-season differences at only one site each (temperature - P8, extractable NH_4^+ - P27).

7. Lines274-275: Do you have complementary measurements of net or gross N cycling processes to help interpret these field patterns? It's possible that the reduction in NO3 during the wet season may be linked to reduced nitrification (with a growth of anoxic microsites), or an increase in NO3- reduction (e.g. DNRA or denitrification).

These lines that the reviewer is referring to are presenting the total soil N, which is commonly 4 orders of magnitude (Table 2) higher than the mineral N (Table 3), the latter reflecting the actively cycling fraction of the total N. Thus, the rate of soil N cycling, being small compared to the total soil N, cannot make a big change to the amount of total N. Total soil N reflects the long-term accumulation of N in these sites.

We indeed have measured gross rates of soil-N cycling in the same sites and replicate plots in the wet season 2010 (Nov.) and the dry season 2011 (May). We do not report them here, as they are included in a separate paper focusing on patterns of soil-N cycling and soil N availability along these orthogonal gradients of soil fertility and precipitation.

However, our interpretations in the present paper were considered in light of the rates of soil-N cycling that we measured. Across sites, gross N mineralization rates correlated with soil microbial biomass N, total soil N, 15N nat. abund. enrichment factor (ε), and 15N nat. abundance (Spearman rank correlation coefficients of 0.48-0.80, n=20, P<0.05). The patterns of microbial N and total N followed that of increasing annual precipitation, while 15N nat. abund. enrichment factor and 15N nat. abundance were low at the low- and high-rainfall sites and peaked at the mid-rainfall sites (Table 2). These patterns were opposite to those of soil pH, ECEC and exchangeable bases across sites (higher values at the low-rainfall sites with less-

weathered soils than at the mid- and high-rainfall sites with highly weathered soils; all $P \le 0.05$; Tables 2). Thus, our interpretation of this pattern of total soil N with increasing precipitation was that the higher the total N (with increasing precip.), the higher the amount of microbial N and the higher the soil N availability, as indicated by the rate of actively cycling N (i.e. gross rates of N mineralization) and mineral N (i.e. soil NH4+ levels, Table 3).

Across sites and seasons, gross N mineralization was not correlated with gross nitrification but instead with NH4+ immobilization (suggesting that heterotrophic nitrification was possibly important rather than autotrophic nitrification). We cannot merely attribute the reduction of NO3- in the wet season to reduced nitrification because gross nitrification was only measured once in the wet and once in the dry season, and we did not see significant differences between wet and dry seasons across sites nor at each site. Additionally, gross nitrification was correlated with NO3- immobilization, but not with DNRA, suggesting that when there was high NO3- availability, this was preferably assimilated by the microbial biomass. On the other hand, the soil NO3- levels we show in Table 3 were measured repeatedly, parallel to soil trace gas flux measurement, over our 21-month study period (as opposed to the gross rate of soil-N cycling which, due to the intensive labor and cost required, was only measured twice). The soil NO3- levels (Table 3) reflected the concurrently occurring NO3- production and consumption processes, and our discussion on the role of soil NO3- levels on the soil trace gas fluxes always considered the soil NO3- patterns between seasons (lines 281-282), among sites (lines 287-289), the inverse correlations of NO3- and soil moisture (lines 290-292), and the correlations of NO3- with soil CO2, CH4, N2O and NO fluxes at a particular site.

8. Lines 293-297: Were these data from bivariate regressions or from a multiple regression model? If the second, it would be useful to indicate, based on the sum of squares, which variables accounted for a larger proportion of the variance and which variables accounted for less, in order to clearly establish the hierarchy of drivers.

As shown above, we have included a table (Table S1) showing the relative hierarchy of environmental drivers. As suggested by Reviewer 2 above, we used the minimum adequate LME models in analyzing the hierarchy of drivers. This statistical analysis is also described in the revised manuscript (line 260-267).

9. Lines 317-319: Increased evidence for nutrient limitation of methanotrophy? What are the implications of this for process models (could be discussed in the Discussion)?

This question of the reviewer is related to his 3rd general comment above (please see our answer above as well). The sentences following these lines 317-319 presented the possible reasons (through correlations with controlling factors) for this pattern of differences among sites (see lines 318-331) and are discussed in lines 438-459 of the original manuscript.

The most important controlling factor on the long-term pattern of soil CH₄ fluxes across sites was soil fertility. Specifically, as shown by the strong inverse correlation between soil 15N natural abundance signatures and exchangeable cations (Table 5), the positive correlation between soil CH4 flux and fertility (Fig. 4b) likely reflected the long-term effects of soil development (Tables 1 and 2) - more CH4 uptake occurred in highly weathered soils with less rock-derived nutrients but high soil N availability (i.e. high 15N natural abundance signatures) (Tables 4 and 5). When separated by season, the correlation between average soil CH4 fluxes and soil 15N natural abundance was stronger in the dry season than the wet season (Table S2),

supporting our claim that soil N availability enhanced CH4 uptake in soils when gas diffusion was favorable (dry season).

10. Lines 339-341: Evidence for very active nitrifiers? Perhaps this could be explored further in the discussion.

This question is related to comment #7 above (please refer to our extended answer there). Our measured gross nitrification rates (measured once in the wet and once in the dry season at all sites) did not show significant differences among sites nor between seasons at each site. Thus, we cannot simply attribute the results presented in these lines (339-341) that the reviewer is asking (i.e. positive correlations between soil N2O emissions and moisture and negative correlations between soil N2O emissions and NO3- concentrations at the mid-rainfall sites (P8 andP19) to be due to active nitrifiers. This pattern is discussed in lines 474-489 of the original manuscript.

11. Lines 362-363: To what extent is inter-annual variability modified/affected by differences in belowground allocation and variations in root-rhizosphere respiration? Do data exist on the belowground biomass across your gradient or differences in root/shoot allocation? If so, this may help tease out the extent to which differences in total soil respiration are affected by differences in the fluxes from individual respiration components.

This question is related to the question about CO_2 above (please refer to our extended answer there). In brief, we do not know of any available datasets that could answer this question. However, we do think our results highlight another interesting facet of CO_2 emissions in these sites, namely, that despite the differences in soil factors between sites, we did not see differences in CO_2 fluxes. However, we did see strong temporal patterns, and therefore focused our discussion on short-term changes over time, as well as reporting on similarities and differences with other studies from CSA lowland forests (see lines 380-389).

Reviewer 1

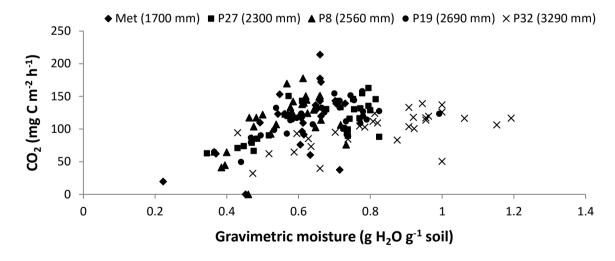
(1) Since in tropical ecosystems soil moisture is highly variable, while temperature is fairly constant (can be seen in your dataset: while gravimetric soil moisture changed from 1.2 to 0.4, soil temperature changed from 27 to 23°C. In other words 66% change of moisture, while temperature changed 15%.). Based on that it can be expected that changing soil moisture is the major driver of trace gas emissions. However, in your study a co-correlation of soil moisture and soil temperature is discussed. This is highly interesting, but not yet well presented. You should be able to demonstrate that air temperature at your sites was fairly constant, therefore the most of the change in soil temperature should be attributed to co-correlation to soil moisture changes. Based on theoretical considerations (e.g. Q10 value) you should be able to give an estimate about how much of the change in CO2, CH4, N2O emission could be caused by temperature only and by the combined soil moisture/temperature effect.

Our objective for this study was to use these natural gradients to explore the interactive effects of soil and climatic factors on soil-atmosphere trace gas exchange. However, in response to this comment and the first comment of Reviewer 2 (see above), we have used minimal adequate linear mixed effects models to identify a hierarchy of importance of the environmental drivers within and between sites/seasons, which also addresses this concern of

Reviewer 1 by giving more details about the relative importance of moisture, temperature and extractable mineral N over our 21-month measurement period (Table S1).

(2) The general "parabolic relationship" of CO2 and soil moisture might be influenced by combining all data point from all sites. It seems actually that the emission follow more actual soil moisture than rainfall gradient. For a more comprehensive analysis, it might be helpful to include correlation coefficients for rainfall, soil moisture, soil temperature, NO3- and NH4+. Since in the whole paper all figures show data points with individual symbols for each site, it seems reasonable to use different symbols for each site (Fig3).

In response to this comment, we include below a revised version of Fig. 3 to show individual symbols for each site, which we also use to replace the previous version of Fig. 3 in the manuscript. As we discussed in lines 388-389 (original manuscript), such parabolic relationships have also been observed by other studies in tropical forests of Costa Rica, Panama and Brazil. Indeed, we conducted correlation tests between annual soil CO2 emissions and annual rainfall but this was not statistically significant because their relationship was parabolic and not linearly correlated. We stated this in lines 305-308 (original manuscript). For this parabolic relationship with annual rainfall, we decided it was better to present Fig. 3 as essentially the same pattern is depicted, but Figure 3 is better, as it depicts the actual measured daily values. All the other soil factors were indeed tested for correlation with soil CO2 fluxes, and we have reported their relationships in lines 299-304 (original manuscript). They are also summarized in Tables S1 (see above.



Furthermore, is there a reason why N2O is not shown in relationship to soil moisture? It might be helpful for a more process based discussion and the role of aerobic CH4 oxidation coupled to denitrification in this soils? Predominantly the soils are a net-sink for CH4, and you measured N2O and NO3 but did not discuss the coupling of processes yet (see e.g. Zhu et al. 2016 aerobic methane oxidation coupled to denitrification).

As we mentioned in the text (lines 366-341 of the original manuscript), soil moisture was only significantly correlated with soil N_2O fluxes at two sites (P8 and P19), so including a figure showing the relationship with moisture across sites would not add anything important to the results we were presenting. We have, indeed, analyzed our data to explore whether there was a link between soil N2O and CH4 fluxes – which would support what Reviewer 1 is asking here

However, neither Table 5 nor Table S2 support such a link (i.e. no significant correlation). Thus, we would have no basis to include this aspect in our discussion without being overly speculative. We do note, though, that the roles of NO3- on soil N2O fluxes as well as on CH4 uptake were discussed extensively (lines 426-437 and 478-489 of the original manuscript).

It would be more appropriate to convert gravimetric soil moisture into either who or WFPS to normalize somehow for the soils from different site.

We are unable to convert the gravimetric to WFPS mainly because the Metropolitan and P27 sites, whose parent materials are agglomerates, had fine stones making measurement of soil bulk density erroneous. We tried to do a more accurate estimate of soil bulk density but we were not confident that we were able to get out all of the gravel in these heavy clay soils (average soil texture within the top 50 cm was 60-62%) from the soil cores we used to measure soil bulk density. On the other hand, we made very careful measurements of gravimetric measure contents every time we took subsamples from the soils that were concurrently sampled during each soil gas flux measurements. Thus, our gravimetric moisture measurements were more reliable than converting to WFPS.

(3) If soil temperature, soil moisture, and soil properties would dominate the CO2, CH4, N2O, and NO fluxes, the data points (Fig.3) should result separate functions over time. The fact, that they are overlaying each other suggests, that other parameters, which are not yet discussed might affect CO2, CH4, N2O, and NO fluxes. As such it should be discussed how abundance (and activity?) of functional microbial groups will change within the rainfall and fertility transect?

As mentioned above, we have now provided more information as to the relative hierarchy of the environmental drivers that we monitored. We agree that the abundance/activity of functional microbial groups would play a role and that such a dataset would definitely provide additional insight into trace gas fluxes along these gradients. However, as we did not take those measurements as part of this study, discussing how they may have affected our results would be purely speculative. We have added a sentence into the discussion to specifically mention that point (i.e. that in future studies, measurement of functional groups could add additional insight).

(4) Without any additional literature reference the transfer from Tamai et al., 2003 for methanotrophs to methanogens is hard to buy. In Tamai et al., 2003 a negative correlation between CH4 uptake rate and Al was found. Table 2 shows that your inhibition might be possible for P8, P19, P32, but not for the others. However, these 3 sites show actually the lowest CH4 fluxes in the rain season 2011 (Fig. 2). Shouldn't a correlation of net flux and Al result in a positive correlation if inhibition of methanotrophs based on Tamai et al., 2003 is assumed? If your assumption would be valid, how can you explain a simultaneous inhibition of methanotrophs which could cancel out your inhibition of methanogens? Since methanotrophs and methanogens are different functional groups of microbes, I think this is speculative.

We agree that this was speculative, and so this comment was removed in the revised version of section 4.2.

(5) For me it seems more plausible that a combination of pH, BS and ECEC which show strong correlations as well, might result a stronger impact for CH4 flux. And a correlation of 15N

might point towards coupled methane oxidation and denitrification (e.g. Zhu et al., 2016)?

As mentioned above, we have substantially altered Section 4.2, which is the section of the discussion related to CH_4 fluxes. However, as we outlined in the comment above, our results do not show any correlation between CH_4 and N_2O fluxes, in the annual or seasonal averages, so we chose not to incorporate that into the discussion.

Based on the microbial processes it can be assumed that CH4 oxidation should contribute to CO2 formation. However, this is indicated by a correlation of only -0.24 (CH4 and CO2) in Table 5. Consequently, a potential coupling of aerobic methane oxidation and denitrification might result only -0.07 (CH4 and N2O) in table 5.

The correlation coefficients referred by Reviewer 1 here are not statistically significant and therefore we chose not to incorporate them into the discussion. Additionally, even granting that this assumption of CH4 oxidation contributing to CO2 formation is valid, by looking at the magnitude of soil CO2 fluxes in comparison to soil CH4 uptake (Figs. 2a-b), such a contribution would be minute compared to the more conventional contributions of heterotrophic (oxidation of organic C with O2) and autotrophic (plant roots) respiration. As to possible coupling of CH4 oxidation with denitrification, please see our answer to the same comment above.

Finally the introduction and discussion would highly benefit to be focused more on microbial processes.

The purpose of the introduction was to introduce the objective of our study (i.e. testing both temporal and spatial patterns of soil trace gas fluxes). Therefore, the introduction starts with a general comment about trace gases from Central and South American forests, and then moves on to introduce known temporal and spatial controls, followed by an explanation of how using these natural gradients helps to address our objectives.

Minor comments:

Introduction

It might be better for the reader to follow the different microbial processes which cause the production and consumption of each trace gas rather than jump from effects of temperature to moisture to soil properties on CO2, CH4, N2O and NO? Overall the introduction is missing a clear structure.

See comment above.

You are writing about methanotrophs and methanogens, but for the other trace gases you don't include any information about the processes and functional microbial groups.

We use the introduction to introduce topics that we bring back later into the discussion. As we did not measure functional groups, we also didn't include them in the introduction.

Line 40: Studies (without references) either include references or refer to a comprehensive list in supplement.

This sentence has been revised so that the vague reference to "studies" is gone. (However, annual soil trace gas fluxes in Central and South American (CSA) tropical lowland forests can vary significantly; in one study...)

Line 65/66: take care of terminology, maybe define once? Net CH4 flux consists of production (positive) and consumption (negative). Furthermore, it should be mentioned that production occurs even under negative net CH4 flux, but consumption is predominant.

This sentence has been revised as follows: Soil CH₄ fluxes (predominant flux indicated by positive values (net emissions) or negative values (net consumption)) in CSA tropical lowland forests...

Material and Methods

Line 149 "soil trace gas flux measurement": you can only measure mixing ratios. Fluxes are the result of a second order calculation.

Line 150 "fluxes were measured"?

We have changed the title of section 2.2 to "soil trace gas flux calculation" and altered the wording in that section to indicate that we determined fluxes rather than directly measuring them.

Line 168 Please specify what gas did you flow through the chambers? Ambient air, synthetic air?

Line 170 specifies "air from the chamber", which is indicating ambient air.

I recommend including the formulas to calculate CO2, CH4, N2O (static) and NO (dynamic), plus the trapezoid rule to calculate the annual fluxes that the reader does not have to look up several other papers to follow the calculations.

Fluxes are calculated using the linear change in concentration over time. This is a standard calculation for trace gas flux studies. Similarly, the trapezoid rule is an established method of filling in gaps between sample dates by assuming a linear relationship in gas fluxes between those two dates. Neither of these calculations uses a specific formula.

Results

The results are majorly focusing on the descriptive correlations. Why the major results of CO2, CH4, N2O, NO fluxes is not presented here? For me these are the major results obtained from the field by hard work (Fig1 and Fig2).

The raw data can be made available for teams developing models and/or needing more specific information, but as the data was presented in Figure 1 and Figure 2, we chose to focus the results and discussions on patterns that we found in the data.

Line 291 Due to different soil properties for each site, it seems not very helpful to present Fig. 3 and talk about a "parabolic relationship".

Please see our related comment above. As shown in the figure above, even once the different sites are identified with unique symbols, the data do not separate out, but instead, together, exhibit this parabolic relationship. It is also shown in Table S2 that moisture was a major controlling factor during the dry season and within each individual site.

Discussion

Statement about what might cause the NO3- differences? Wet deposition, if yes, are there values from literature?

We have measured the gross rates of soil-N cycling at these sites. The rates of gross N mineralization (2-5 mg N kg⁻¹ d⁻¹, or about 68-170 mg N/m2/day in the top 5-cm depth, using our measured soil bulk density, averaged across sites, of 0.68 g/cm³) and gross nitrification (1.2-2.4 mg N kg⁻¹ d⁻¹, or about 41-82 mg N/m2/day in the top 5-cm depth) were much higher than our measured wet N deposition (9 kg N/ha/yr or only 2.4 mg N/m2/day) at the Gigante site (see map in Fig. S1; Gigante is across the Panama canal from our present sites). We would actually not assume that the mineral N in the soil is directly influenced by the external N input via wet N deposition. The soil N cycling rates are much larger than the wet deposition, based on our previous sites in Gigante (e.g. Corre et al. 2010, 2014), Ecuador (Baldos et al. 2015), and Indonesia (Allen et al. 2016).

We discussed the pattern of the soil NO3- levels among sites, or the mineral N pool for that matter, in perspective of the soil-N cycling, which influences this mineral N levels, and ultimately reflected in our overall index of soil N availability status (low or high N availability), 15N natural abundance enrichment factor (which has been shown to correlate with soil N availability; see lines 213-215 of the original manuscript).

Thus, we decided not to include in our discussion about wet deposition, which obviously will not directly influence the soil NO₃⁻ levels, but discussed the patterns of NO₃⁻ among sites with regards to soil N availability status of the sites (see lines 464-473 of the original manuscript).

The connection of the trace gas fluxes to microbial processes is missing. E.g. the correlation of CH4 fluxes (net uptake) is negatively correlated to 15N natural abundance. Does this point towards a CH4 production coupled to denitrification? And could this coupling be less relevant in the dry season versus the wet season and thereby result amplified correlations in the dry season?

The negative correlation of CH4 fluxes (net uptake) with 15N natural abundance was indeed discussed (lines 449-457 of the original manuscript). However, please refer to our explanation above as to why we don't think that this relationships points towards CH4 production being coupled to denitrification.

Figures:

Error bars are missing for Fig 3, 4, and 5

Fig. 4 a, b, c should include a 0 line for easier understanding. Fig. 4a might be better to bin data into moisture classes of 10%. Less data points will make the figure easier to understand and better show trends. Error bars can be included. Would it make more sense to average the

single points and report error bars to highlight the grouping in different fertilizer regimes Fig 4b? That might be helpful for discussion?

We chose not to put error bars on the scatterplots, as their purpose was to highlight trends, which may have been masked by including so much additional information. However, in response to this comment, we have included a zero line for 4a (zero occurred at the top of b and c). We are reluctant to average the data, however, as the current figures allow readers to see the exact spread found within each site rather than simply the standard deviation shown on error bars.

Fig. 5: Where was the NO ambient mixing ratio measured? Close to the ground (chamber height) or 2m height? Are there references available for such high NO ambient mixing ratios and possible sources? Based on Remde et al (1989) it might be helpful to plot NO release rate versus ambient NO mixing ratio at same moisture and temperature for each site. Furthermore, only data points for a range of soil moisture and soil temperature should be selected.

The NO ambient mixing ratio was measured at a height of 2 m above the ground (prior to each chamber measurement) near to each of the 4 chamber locations at each of the 4 replicate plots per site on each sampling day.

As to the last comments (to plot NO release rate versus ambient NO mixing ratio at same moisture and temperature for each site; only data points for a range of soil moisture and soil temperature should be selected), this would not be meaningful for our data sets, because such a way of analyzing data is driven by an inherent assumption that the ambient NO mixing ratio is influenced by biological processes in the soil. This is not the case at our study sites where anthropogenic ambient NO levels are prevalent, especially the site near to the Panama city and even the other sites along the Panamal canal, brought about by large shipping traffic. Such high NOx emission was also reported by Hietz et al. 2011.

References

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