

Referee 1

The authors themselves say that this is a work providing preliminary results and that is true; the work is quite descriptive and superficial. It is unclear how this present study makes a substantial contribution in Pantanal GHG studies.

There is little work on GHG emissions in the Pantanal. But above all, we do not wish to consider the Pantanal as a whole, because, as mentioned in the text, it is made up of much contrasted sub-regions, mainly from a chemical point of view. We focused on Nhecolândia, the only sub-region of the Pantanal that evolves clearly in an alkaline pathway, and in which, the factor of concentration of surface waters is a major axis of environmental diversity (previous studies have shown this). Some recent studies in Nhecolândia have focused on the variability of gas emissions in space within the same lakes. These works are mentioned in our manuscript, and the annual emission budgets (in this case methane) are compared to our results. In our case, we study the variability over time, during the seasons, and based on daily cycles (when cycles have not been interrupted by weather conditions). We also emphasized the opposition between green and black-water lakes. Particularly for the most reactive green water lakes, we mention dissolved methane values of the order of 60 $\mu\text{mol} / \text{L}$. These extreme values are not common, outside the classical framework of lake studies and are worth reporting. Moreover, for similar dissolved methane values, we show that methane emissions can be multiplied by 10 because of the appearance of a new phenomenon, the exceeding of the O₂ bubble point, a very frequent phenomenon in some lakes of Nhecolândia. Again, it is a process known to physicists, but its environmental consequences have never been studied, and it deserves to be mentioned. In terms of contribution to GHG studies in Pantanal, the development of a regional emissions assessment will necessarily involve understanding the variability of emissions in space and time. This study contributes to this, focusing on daily and seasonal variability.

The main problem is in the study design, especially the gas emission studies. After reading the manuscript several times, I still don't know how many times the gas measurements were carried out. In methods section there is no indication about dates, time etc. of sampling.

We agree with this remark, and this aspect has been modified in the new version of the manuscript. Time of gas measurement, dates, depth of the water column (etc.) were mentioned. In addition, picture of data collection are supplied as supplement material. Below is the new text for "study design" section

"Gas fluxes from the lake to the atmosphere were measured using 32-L polyethylene floating chambers, having a base area of 0.195 m². The main conditions during the field campaigns are summarized in table 1. Two procedures were used for these measurements with fixed or slowly moving chambers. The procedure using slowly moving chambers (Photo 2 Supplement S1) was favored when the water level was sufficient and the lake diameter not too large to allow to cross from one bank to another. In this case, depending on the lake diameter, a train of 3 to 6 floating chambers was attached, leaving a gap of 10 meters between two successive floating chambers. Floating chambers were placed in the water every minute at a distance of about 30 m from the lake shore, and then slowly pulled toward the opposite bank at a maximum rate of 5 m min⁻¹. This experimental design allows for scanning the various water column heights, with the least turbulence disruption to the lake surface. To minimize artificial turbulence effects, foam elements were adjusted so that a maximum of 2 cm of the chamber penetrated below the water surface. The collects were carried out once each chamber reached a distance of about 30 m from the opposite bank. The collection times were variable since the first chamber reached the other margin in approximately 20 to 25 minutes, whereas the last chamber took about 35 to 40 minutes. When the water level was too low, or the lake too wide, we opted for a procedure with fixed floating chambers (Photo 3 Supplement S1). In order not to disturb the sediment just below the chamber, they were anchored with a 10-m line to avoid drifting. The line was equipped with a float to the vertical of the anchor. The chambers were located from the center to the border of the lake, and the collects were carried out after 20 min from an inflatable boat with shallow draft. Due to the low

water column, it was not possible to place a bubble shield to prevent bubbles from reaching the chamber. Therefore, the results represent the sum of both fluxes by diffusion and ebullition. For each chamber, gas samples were collected in duplicate (about 2 minutes apart) through a 60-mL syringe. Then they were transferred into 30-mL glass bottles, previously capped with gas-tight, 10-mm thick butyl rubber septa and aluminum caps, and evacuated with a hand vacuum pump at 0.75 kPa. Air samples were also collected at the departure of the chamber train for the ambient gas levels. Gas fluxes were calculated by the linear change in the amount of gas in the chambers as a function of sampled time. Thus, for example for a 6-chambers protocol, the mean and standard deviation on 12 measurements are presented as single gas emission value and error bars, respectively, for a given hour that corresponds to the launching of the first chambers. This operation was repeated every two or three hours or in order to present a complete 24-hour cycle.

Gas concentrations in the liquid phase were estimated indirectly using a headspace displacement method (Hope et al., 1995) with a 120-mL syringe and an air:water volume proportion of 1:3 (30:90 mL). For this, water samples were collected 5 cm below the surface, about 30 m from the edge of the lake. To equilibrate the headspace with the liquid phase, the syringe was shaken for 2 min by hand before injecting the headspace gas into the 30-mL glass bottle. For CH₄, the coefficient of gas transfer velocity (K_{600} , m d⁻¹) was calculated from the flux, the dissolved CH₄ concentration in water and the CH₄ partial pressure in the floating chamber as described by Bastviken et al. (2004)."

Below is Table 1: with the requested information

Table 1: Date, location, lake characteristics and general conditions during greenhouse gas emission monitoring.

Date	Type of lake (name) Surface km ²	Weather conditions	Phyt. Bloom conditions	EC range µS.cm ⁻¹	pH range	DOC mg.L ⁻¹	Procedure Numb of chambers	Water column range meter	Time of gas coll. Minute
Sept. 13, 2012	Black (P) 0.087	Sunny	-	1400-1599	8.81-8.99	51	Fixed 3	0.3 – 0.8	20
Sept. 14, 2012	Green (V) 0.109	Sunny	Moderate	2420-2888	9.48-9.73	236	Fixed 3	0.1 – 0.4	20
Aug. 30, 2013	Black (P) 0.091	Sunny	-	1715-1855	9.21-9.33	37	Fixed 3	0.3 – 1.1	20
Sept. 1, 2013	Green (V) 0.109	Partially cloudy	Strong	2302-2410	9.67-9.78	265	Fixed 3	0.1 – 0.5	20
Dec. 2, 2014	Green (M) 0.053	sunny	No	2014-2204	9.37-9.51	102	Sl. moving 6	0.1 – 0.4	23 to 43
Jul. 7, 2015	Green (M) 0.055	sunny	No	1940-2030	9.28-9.37	82	Sl. moving 3	0.1 – 0.4	21 to 37
Sept. 10, 2015	Green (G) 0.285	Sunny (evening storm)	Strong	34000- 35100	10.3- 10.44	326	Fixed 3	0.1 – 0.2	20
Sept. 12, 2015	Black (P) 0.093	Strongly rainy	-	1382-1450	9.3-9.4	36	Fixed 3	0.4 – 0.7	20

And below a picture added as supplement material:



Photo 2: Gas collection from a train of 6 slowly moving chambers on green water lake M in the absence of cyanobacteria bloom (December 2014). The first floating chamber has just reached the point of collection. Two samples will be collected in each chamber. The average of these 12 samples will provide 1 flux data for each gas (CH_4 , CO_2 and N_2O).

The studied systems appeared very shallow and thus they most probably are hot spots for ebullition, but ebullition was not studied at all (although it is discussed quite a lot). I find it very surprising that ebullition was ignored. From chambers the samples were drawn only at the beginning and in the end of the measuring period – which as such is strange – so the ebullition is included in the results, but in a proper study you should still measure it separately.

We also agree that emissions by diffusion and ebullition should be separated. Generally, this is done by installing a bubble shield, installed at a depth of about 50 cm, and which prevents the bubbles from reaching some chambers, the separation between diffusive and ebullitive flows being made by subtraction between the results from the equipped and non-equipped chambers. We added a photo to illustrate the collection, and this photo emphasizes that the studied lakes are often very shallow, particularly (but not only) during low water period. The installation of such a structure below some chambers was simply impossible, as it would have disrupted the sediment, releasing gases artificially. Therefore, our results include both diffusive and ebullitive fluxes, as already mentioned in the manuscript. “*Due to the low water column, it was not possible to place a bubble shield to prevent bubbles from reaching the chamber. Therefore, the results represent the sum of both fluxes by diffusion and ebullition.*”

In addition, installing a bubble shield prevents only bubbles that come from below the shield to reach the chamber. This system is functional for bubbles emanating from the sediment. In our study, we discuss the role of ebullition from the water column (in relation to exceeding of the O_2 bubble point) and not from the sediment. Indeed, the microbubbles (when they begin to be visible they have a diameter of about 0.1 mm) do not emanate from the sediment. A shield would therefore give very different values depending on its depth in the water column below the chamber. It is not the information sought.

The authors are well aware of the importance of hydrology and thus weather for their study system, but there is nothing about these basic measurements indicating that they were not monitored at all during the study period.

We have a meteorological station near the sites that allowed to acquire the basic climatic data. These regional data have been complemented by local data (wind speed and air temperature) since

the temperature contrast between the lake and the surrounding forest is known to locally generate a breeze regime (Quenol et al., 2009).

When discussing the results, the importance of evaporation for gas fluxes is clearly stated, but despite this, heat fluxes were not measured during the study.

We did not mention heat flux because it is clear that the temperature increases gradually, while the increase in methane emissions is abrupt for green water lakes, and that this increase matches with (abrupt) exceeding of the O₂ bubble point. It is this phenomenon that has been suspected as responsible for the increase in methane emissions. We argue with a new figure where CH₄ K₆₀₀ are presented, comparing lake V (strong bloom without O₂ supersaturation at only 450 %, i.e. without reaching the bubble point, temp. max. about 37°C) and lake G (Strong bloom with dissolved O₂ > 500 % and ebullition, temp. max. about 39°C). We mention in the discussion that “The consistent change in the calculated K₆₀₀ values (Fig. 6), which coincided with the occurrence of the abrupt generalized ebullition of lake G, emphasize that CH₄ behave quite differently in these 2 lakes.”

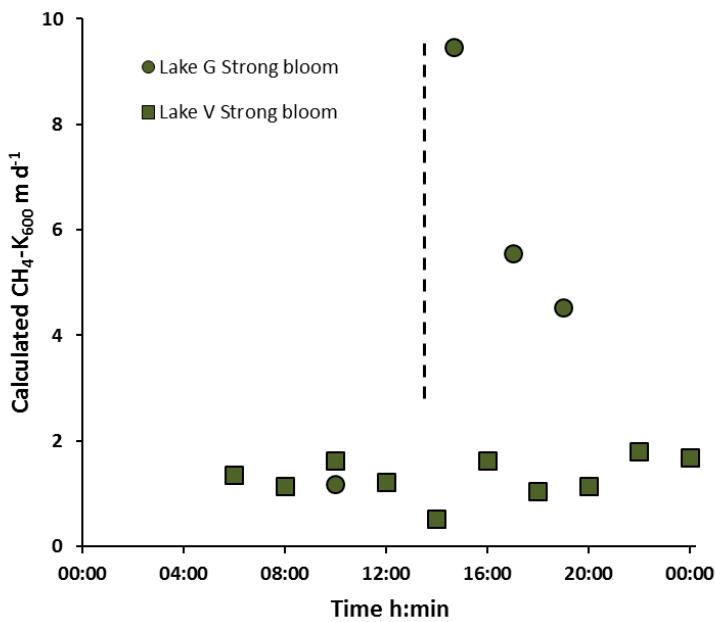


Figure 6: Calculated exchange gas coefficient for Methane in lakes V and G in strong bloom condition. The dashed line represents the beginning of the ebullition in lake G (13:20).

There is no explanation for the selection of studies lakes, i.e. why only one freshwater lake was chosen. There is very limited background data on the lakes. It is said that the lakes are shallow, but no bathymetric maps are available. The surface area of the lakes is not presented and cannot be estimated from figure 1, since in the aerial photographs there is no scale. In general, no information about the morphometry of the lakes is available.

First, we can present a practical aspect to answer this comment. The entire region is made up of private properties. Research can only be done with the agreement of the owners and managers on site. This is the main constraint on the choice of lakes, and this constraint can change from year to year following the sale of a farm to a new owner that could be opposed to our work on its land. Second, before each trip, it is impossible to predict which lake will be able to bring additional and complementary data to the already existing dataset. A lake collected during a field campaign may be in the same situation during the next mission, whereas a neighboring lake may have evolved towards a stronger bloom, for example. In this case, the team will decide to collect the neighboring lake. We have focused on the acquisition of a set of data that can cover the most diverse situations.

In any case, with the restructuring of the manuscript, we have retained only 4 lakes, removing F and G for which no emission measurements had been made. The dimensions and depths are mentioned

in the text and in the table. We also mention the range of collecting depths, in the case of fixed or moving floating chambers. A scale, which was not visible in the first version, was added to Figure 1. See below the bottom part of Fig. 1.

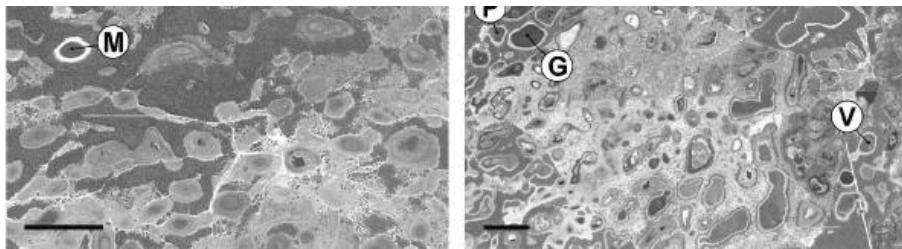


Figure 1: Location of the Pantanal wetland, Nhecolândia region, Nhumirim and Centenario farms and studied lakes. Satellite images are from Google Earth™ (bar = 1 km).

The lakes are divided into two classes, green and black lakes, but it left unclear where the name especially of the black lakes comes from. Are they dark coloured due to DOC loading?

Actually both, green or black water lakes have high DOC values. These values are in the range 30 - 320 mg C / L., now mentioned in the table. Blackwater lakes have the lowest values of DOC, 30-50 mg C / L, but also show clay particles dispersed in water due to high pH and high sodium levels. For this reason, the turbidity is generally greater than 100 NTU, opaque to light and their color is distinctly dark brown. Green-water lakes have higher DOC values but no clay particles in suspension. In the absence of bloom, they are brown in color but have a lower turbidity (<20 NTU). From the beginning of the development of the bloom, the green color dominates. See also below photo 1 in supplement material S1.



Photo 1: Aerial picture illustrating the contrast between a lake with black waters and a lake with green waters. Here the bloom is moderate to strong. The two lakes are about a hundred meters apart (source, matuete.com).

No explanation is given for the fact that only three lakes were chosen for the sediment studies. Why these three?

With the restructuring of the manuscript suggested by different reviewers, we have refocused the data on 4 lakes. We provide the sedimentation data for these 4 lakes. We already had data on Lake M, we did analysis from the three other lakes. The results obtained are presented, they only allow to note, unlike expected, the absence of significant difference between the two types of lakes (Green or black water).

The lakes were sampled for gas concentrations in the water, but nothing is said about the location of these sampling points and sampling depths?

We agree with this comment, location and depth of sampling have been mentioned in the new ms: "For this, water samples were collected 5 cm below the surface, about 30 m from the edge of the lake."

Temperatures were measured inside and outside of the chambers but it is not explained how these data were used. The calculations of fluxes were not explained.

We agree, the calculation is now explained in the text. The temperature was used for the calculation of the CH₄-K600 from the head space data and the CH₄ concentration in the chambers.

It is said that oxidation-reduction potential was measured also in the water (why?), not only in the sediment. These results are not shown.

We agree. As geochemists, we are used to measure redox potential in water, which is giving a much wider range (and therefore wider information) than dissolved O₂. Anyways, these data have not been used in the study and are now removed from the "material and methods" section.

Nothing is said about the calibration of the fluorometer.

Again we agree with the comment. Anyway, the results given by the fluorometer are not relevant as for most of the case, value is over-range. It has been removed from the manuscript.

The size of the gas bubbles is given, but it is totally unclear how the bubble studies were made. The gas emission part of results is not well structured, and needs to be rewritten to clarify the findings.

The objective was not to study the bubbles, but to show that the appearance of ebullition affects the gas emissions during a few hours in the afternoon. Anyway, we agree that this part was confused and it has been re-written, with separate paragraph for CH₄, CO₂ and N₂O, and figures have been modified accordingly grouping all information for a given gas on the same figure. See for example fig of CO₂ emission below.

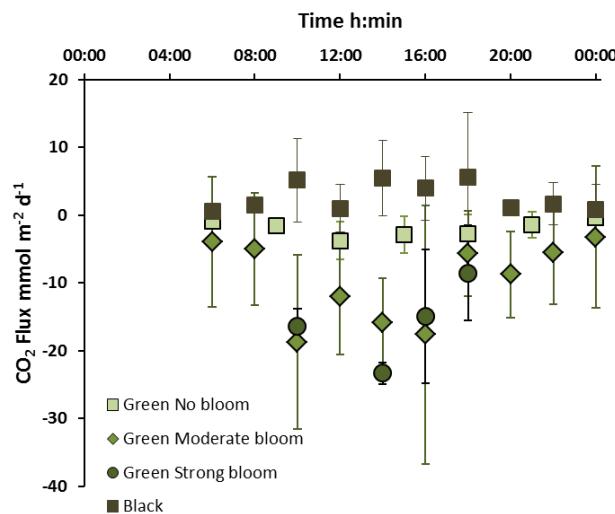


Figure 7: Daily cycle of carbon dioxide fluxes showing emission from black water lake (P), and increasing consumption with increasing magnitude of the cyanobacterial bloom in green water lakes for no- (lake M), moderate- (lake V) and strong (lake G) bloom conditions.

In discussion gas bubbles and especially microbubbles are emphasized. However, bubbles were not studied at all and thus there is no evidence on these phenomena in the Pantanal small lakes. The advice is to be very cautious when discussing bubbles.

A picture of the lake surface taken when the ebullition starts is added (Supplement material S1). Initially we were reluctant to introduce this illustration because the size of the bubbles on the

surface of the lake does not correspond to the size of the bubbles that form in the water column. What appears here on the picture is the grouping of hundreds of microbubbles (<0.1 mm) into larger bubbles (~1cm). These bubbles appear abruptly at the lake surface when dissolved O₂ level exceeds 500% saturation, i.e. when the bubble point is reached.



Photo 4: Detail of ebullition in strong bloom condition after exceeding the O₂ bubble point (> 500 % saturation). Microbubbles (~ 0.1 mm) do not arise from the sediment but form in the water column, and gather into larger bubbles on the surface of the lake. The abrupt emergence of ebullition, which will continue for about 3 hours during the afternoon, matches to a significant tenfold increase in methane emissions.

There is also a section for the influence of rainfall. Similarly to bubbles, no information on rainfall or weather in general, so there is no proper ground for this kind of discussion.

This part has been shifted to the end of the discussion and in the “future direction” section. The death of the phytoplankton probably impacts gas emission, and it will be checked in the future, with other complementary data and different situations:

“Immediately after the first rain of the season, the bloom disappeared in a few hours, and substrates to methanogens, in the form of very labile organic carbon, deposited massively at the bottom of the lake. It resulted in a drastic decrease in pH in the upper part of the sediments probably due to the release of organic acids that accompany the onset of mineralization. For a few days, the drop in the sediment pH (about 2 units) created conditions more favorable to the synthesis of CH₄, although specific methanogens inherent to alkaline grow and produce methane at pH above 9. The proximity between the sediments and the water-atmosphere exchange surface due to a thin water column (generally <0.5 m) is likely to favor methane diffusion and emissions. Unfortunately, due to the beginning of the flooding of the Pantanal, it was not possible to continue the measures, and no value of GHG flux is associated with this period, which will have to be monitored in the future.”

Besides gas emission part of the results section, at least the section ‘studied area’ should be restructured and divided at least to two paragraphs.

The section has been re-written and divided into 4 parts: General information on Pantanal and Nhecolândia, abiotic chemical characteristics of lakes, aspects of lake biogeochemistry, and choice of study sites.