Comments to the Manuscript egusphere-2023-605 "Driving and limiting factors of CH_4 and CO_2 emissions from coastal brackish-water wetlands in temperate regions"

General Comments

- This interesting study addresses the relevant scientific question of the quantifications of possible emission of GHGs (CO_2 and CH_4) from coastal wetlands.

- The attention is focused on a study area at the Italian Adriatic Seaside, even comprehensive of four different study sites. This study area can be considered representative of possible conditions referable to the relevant Mediterranean Area.

- It should be positively pointed out the overall methodological approach to combine the fundamental method of the accumulation chamber to monitor the CO_2 and CH_4 flux emissions with a large number of complementary and needed measures in terms of soil properties and water levels, surface, and groundwater physical-chemical parameters (as temperature, pH, electrical conductivity, and sulphate concentrations of water).

- The statistical elaboration and the evaluation and discussion of all these monitoring data sets are appropriate.

- The English style and grammar are appropriate.

We thank the reviewer for the positive comments.

- Some Figures, that contain a deep visualization of interesting data and elaborations, could be eventually reorganized to favour their increased readability.

Thanks for the comment. We believe we have now improved the readability of the figures, reorganizing some of them. In specific Fig. 1 reporting the study area has been separated in two images, and more details have been added to increase readability. In Fig. 3-4-5, some elements have been removed, and moved as separate images into the Supplementary Material.

- In the following, some specific suggestions to improve the expected final revised version of the manuscript.

Specific Comments

Dear Reviewer,

Thank you for taking the time to review our work. We appreciate your valuable feedback and constructive comments. In this response, we address each of your concerns and provide explanations and clarifications regarding the raised points.

"Abstract":

- Page 1, line 11: it should be "…. greenhouse gas (GHG) emissions…"

- Page 1, line 13: no need to repeat again the complete wording "… methane (CH_4)…" as it was already extensively introduced at page 1, line 9

The sentences have been corrected as suggested.

"Introduction":

- Page 2, line 27: "gases" instead of "gasses"

- Page 2, line 34: please, be consistent with a selected format only for the units throughout the manuscript and related Tables and Figures. Therefore, for instance 0.109 g/m2/day or 0.109 g m^{-2} day^{-1}, make a choice only.

- Page 2, line 35: it should be "… CH_4…"

- Page 2, line 53: it should be "… on GHG emissions…" instead of "… on GHGs emissions…"

- Page 2, line 59: it should be better specified as "… CH_4 and CO_2 emission fluxes…"

- Page 3, line 64: "modelling" and "models" in "… modelling the C cycle accounting in temperate coastal wetlands models…", please try to avoid wording repetitions in a sentence

The sentences have been corrected, and formatting has been apported as suggested.

Section 2.1:

- Page 3, line 73: to be consistent with the Section title, it should be here "study area" instead of "study site"

- Page 3, line 76: The citations "EEC 1979; 1992" should clearly appear with the same acronym (EEC) in the "References" Section

- Page 3, lines 77-78: Please verify that, according to the Journal Editing Style, pertaining web site links should be reported along the manuscript text or, differently, properly listed in the "References" Section

- Page 3, lines 79: please, provide a pertaining citation for the reported mean annual temperature of 13.3°C in the study area

Corrections, and additional bibliography have been apported along the suggested improvements.

Section 2.1.1:

- Page 5, lines 112 and 114: the acronym citation "RER" should be properly reported as "RER (Regione Emilia Romagna)" in the "References" Section

Corrections have been apported to the section. Conformity of reference list has been checked.

Section 2.2.1:

- Page 6, lines 161-162: the meaning of the following sentence should be explained better: "… a value of 0.05 mol/m2/day was assigned to all fluxes larger than zero and lower than this value to avoid over estimation."

- Page 6, lines 162-163: concerning the negative or zero measurements, is it eventually possible to have effective absence of CH_4 or CO_2 emissions from some points of a wetland?

- Page 6, line 166: is it suggested to write "… loss-on-ignition analysis: …"

The sentence at line 161-162 had been improved to enhance clarity as follows: "*All measurements have been recorded after a minimum of 90 seconds analysis. Based on the lowest sensitivity limit of the instrument indicated by the manufacture, a value of 0.05 mol/m2/day was assigned to all fluxes larger than zero and lower than the sensitivity limit to avoid errors." To answer the reviewers comment we specify that it is possible to have effective absence of emissions, or even report negative value of CO2. This can be further looked by substituting the dark chamber with an opaque device and measure the net flux. Since our instrument is not sensitive enough to discriminate between very low fluxes and zero, to avoid error we decided to apply the instrument detection limit to all fluxes, accordingly to the manufacturer's manual cited in the bibliography. "*

Section 2.2.2:

- Page 7, line 176: please report extensively the meaning of the acronym "ARPAE"

The issue has been addressed

Section 2.5:

- Page 7, lines 182-183: is it appropriate the comma in "… and, for homoscedasticity…"

- Page 7, line 188: "plain" or, better, "plane"?

The sentences have been corrected as suggested.

Section 3.1:

- Page 7, line 200: it is suggested "GHG fluxes" instead of "GHGs fluxes" (see also the title of Section 3.1.2)

- Page 8, lines 231-233: please, specify that the mentioned mean values refer to CE site

Sentences have been improved as suggested: *"CH_4 and CO_2 fluxes in PA are always lower than those recorded in CE, while being both sites characterized by the presence of freshwater. During SS in particular, PA has the lowest mean flux of CH_4 of the whole study area (6.04 g/m^2/day), while CE the highest (254.09 g/m^2/day) (Tab.2). "*

Fig.1:

- Eventually, to increase in particular the visibility and readability of the four EC diagrams at the selected piezometers, the authors could consider the alternative option to split the original figure into two separated figures (one with the overall map even specifying the identifying numbers of these piezometers, the other with the mentioned four diagrams)

Map of the study area has been changed based on the suggestions. The two figure have been separated. Also, references to the piezometers have been added to the map, and the readability of the diagrams has been improved in terms of font size and colors.

Table 2

- **Not "DV. ST." while "ST. DV." or even better only "SD"**
- **Moreover, please add a legend by specifying that: ST. DV. (or SD) = standard deviation; CV = coefficient of variation**

Tables have been changed.

Section 3.2.1:

- Page 10, line 249: please specify the meaning of "In the first case" (it should be referring to CH_4 ?)

The issue has been addressed and improved in the text . The sentence now is more clearly referred to CH_4 emissions *"For CH_4 in particular the two components explain 63.9% of the total variance, and specifically PC1 explains 38.5% and PC2 25.4% of variance (Fig. S3). "*

Figg. 3, 4, and 5:

- Eventually, to increase the overall readability of these interesting figures, the authors could consider the alternative option to insert and combine in all the respective histograms and scree plot as an independent figure.

Figures have been improved along the suggestions made. The scree plot has been removed and added as independent figures in the Supplementary Material.

1. ***Discussion:***

- Page 18, line 362: please delete one of the "..".

Done

1. ***and 5. Sections:***

- Page 19, lines 422-431: indeed, this paragraph should be included and harmonized within the final "5. Conclusions" Section

The text has been harmonized with the Conclusion section as suggested by the reviewer.

RC2: **'Comment on egusphere-2023-605', Anonymous Referee #2, 23 Sep 2023**

The #ms (2023-605) is aimed to identify the driving and limiting environmental factors for CH4 and CO2 production in temperate coastal wetlands with varying water salinity. The results are interesting and interpretations are mapped primarily through PCA test. Strategically the field study was well conducted as seen by sample size is robust, seasonal captures, point data. However, there are areas in the ms that requires more clarity and hopefully authors could improve the revised version by addressing the comments appended below:

The authors thank the reviewer for taking the time to review our work and highly appreciate the valuable feedback given. Hereafter, we will answer point by point to all the risen issues.

Abstract – It needs to be largely improved. Please provide some quantitative results in the abstract. Suggest adding flux of CO2 & CH4 as per zonation. Also Avoid making shallow sentences like – soil properties are determined by collecting soil samples- what is new here in the sentence? Everyone knows soil collection is essential for their lab-based property determination.

Quantitative results have been added to the manuscript, also syntax has been revised along the suggestion as follows (lines 8-22):

Coastal wetlands play a fundamental role in mitigating climate change thanks to their ability to store large amounts of organic carbon in the soil. However, degraded freshwater wetlands are also known to be the first natural emitter of methane (CH_4). Salinity is known to inhibit CH_4 production, but its effect in brackish ecosystems is still poorly understood. This study provides a contribution to understanding how environmental variables may affect greenhouse gas emissions (GHG) in coastal temperate wetlands. We present the results of over one year of measurements performed in four wetlands located along a salinity gradient on the northeast Adriatic coast near Ravenna, Italy. Soil properties were determined by coring soil samples, while carbon dioxide (CO_2) and CH_4 fluxes from soils and standing waters were monthly monitored by a portable gas flux-meter. Additionally, water levels and surface and groundwater physical-chemical parameters (temperature, pH, electrical conductivity, and sulphate concentrations of water) were monthly monitored by multiparametric probes. Specifically, we observed a substantial reduction in CH_4 emissions when water depths exceeded the critical threshold of 50 cm, regardless of the water's salinity level. In deeply flooded freshwater ecosystems, the mean CH_4 flux measured at 5.04 g/m^2/day, while in brackish environments, this value increased to 12.27 g/m^2/day. In contrast, in shallower freshwater areas with depths less than 50 cm, CH_4 fluxes exhibited a remarkable spike, reaching an average of 196.98 g/m^2/day. Similar trends were evident for CO_2 emissions, with lower fluxes observed in freshwater environments when water depths exceeded the critical thresholds, averaging at 5.67 g/m^2/day. Conversely, in waters shallower than 50 cm, CO_2 fluxes could reach an average as high as 71.42 g/m^2/day. Furthermore, CO_2 fluxes decreased with increasing salinity, with an average of 15.47 g/m^2/day within brackish environments. Temperature and irradiance strongly influenced CH_4 emissions from water and soil, resulting in higher rates during summer and spring.

Results and Discussion:

I did not see the implication of bulk density and SOC in the entire manuscript despite there being scope for determining their importance as regulatory customers of GHGs in soil (SOC in particular). Authors should perform additional statistical analysis either as ANOVA or multiple regression incorporating all the observed

physical and biogeochemical parameters in soil to examine their role as determinants of CO2 and CH4 emission and explain in more detail the underlying reasons for such trends.

In the original design of the field sampling, the measurements performed to investigate the soil organic matter content (BD and SOC) were solely intended to allow a general characterization of the study sites. Therefore, since soil samples were only collected seasonally (and not monthly), these data were not included in the statistical analysis. If we were to use these data in statistical analyses now, we would be compelled to repeatedly replicate the same values with negative effects on the PCA analysis, which would bring to possible misleading interpretations.

However, during the lab analyses performed for the present manuscript, we decided to perform more laboratory analyses focused on the crucial involvement of carbon and organic matter in the methanogenesis process and, for this reason, we have started a more detailed study specifically focused on carbon, sulfur compounds and microbial community composition (retrieved using metabarcoding techniques). This work is now concluded and will be submitted shortly for publication.
This topic is outside the purview and goals of this article, calling for a more in-depth and narrowly focused debate. Therefore, even though we cannot show these new results in the current manuscript, we will clarify the regulatory importance of bulk density and SOC in soil in the Discussion section, anticipating that new results will be soon part of another paper (lines 448-453):

"Research that focuses on microbial community structure, interaction between microbial communities and carbon-functional composition, and ecological factors influencing both microbial communities and carbon-functional composition are essential if we aim to fully understand the complex process of methanogenesis in coastal environments. Future research will be conducted to accomplish these goals, primarily concentrating on biogeochemistry and the organization of microbial communities. Through this project, want to promote a comprehensive knowledge of the complex processes that underlie methanogenesis in coastal environments. "

Previous research shows role of Chl-a and its positive correlation with wetland CH4 emission boosted by organic matter availability and primary productivity. Since no Chl-a data was shown, it is prudent also to highlight additional determinants of CH4 emission other than the observed parameters (refers doi.org/10.3389/fevo.2019.00032)

Thank you for this insight. Following this suggestion, we will further emphasize in the Discussion that the methanogenesis is a complex process involving multiple environmental variables, more than those listed in our study. Unfortunately, our lab equipment does not provide the necessary instrumentation to measure Chl-a. For this purpose, the text has been improved as follows, with the suggested inputs (lines 431-434):

"However, methanogenesis is a complex interplay of environmental factors and site-specific conditions (Kotsyurbenko et al., 2019) and other variables not considered in this study could potentially prove to be significant. Vegetation composition, primary productivity and Chlorophyll-a in waters, can influence or be the indicator for the organic matter supplied to sediments, influencing methanogenesis rates in wetlands sediments both in freshwater and saltwater ecosystems (Grasset et al., 2018; Huertas et al., 2019). "

Where are the conc data of GHGs? Flux is a computed result from conc and piston velocity (for water), temp, volume, change with incubation time (for soil). Unless the conc data of CH4 and CO2 are shown, this is always difficult to explain flux pattern. For instance, up to 50cm water depth, CH4 flux was maximum then dropped, but was the conc also followed the same trend? This is good to show GHGs conc data in the results and displays. Also if 50cm depth threshold is of a first of a kind findings (line 405-06) then this has to be reflected in the abstract and conclusion. However, authors should also consult with the similar piece of work done in lake by Grossart et al., 2011 (Microbial methane production in oxygenated water column of an oligotrophic lake)

As explained in lines 155-173, we used a portable CH4-CO2 flux-meter (West Systems srl, Pontedera, Italy) to directly measure gas fluxes. The method is reported in the manufacturer manual (Giovenali et al., 2013, https://www.westgroupnews.com/wp-content/uploads/2016/02/570p_The_FLUXMETER_IMPLEMENTATION_Full-Research.pdf) and has been widely used for other studies (e.g. Capaccioni et al., 2020, https://doi.org/10.1016/j.wasman.2010.10.004, Hu et al., 2022 https://doi.org/10.1016/j.ecoleng.2022.106793).

Fluxes are computed from conentration expressed in ppm/sec, as stated in lines 158-159, so fluxes measurements are a direct representation of the concentration.

In the methodology reported the equation to convert the concentration into a flux is presented. According to Chiodini et al. (1998)(https://doi.org/10.1016/S0883-2927(97)00076-0), the flux (expressed as moles/m^2/day) is proportional to the gradient concentration C (measured as ppm/sec) while the proportionality constant depends on the height of the accumulation chamber (H=V/A) if the chamber has a cylinder or parallelogram shape), air pressure, and air temperature. The flow of the gas is determined by the gradient dC/dt since the accumulation chamber's form is fixed and air pressure and air temperature are measurably large.

Such flux measurements were performed both on soils and standing waters, and the results were computed with the same method. Dataset will be accessible on Zenodo platform, after publishing.

The study of Grossart et al., 2011, while being of high interest and offering new perspectives, does not propose comparable environmental conditions to our study. In fact, it describes an oligotrophic lake, with a maxim depth of 69.5 m, while our study is set up in a much shallower water body (max 1.2 m of depth) with high organic matter. Since methanogenesis is highly ecosystem dependent, factors that control methane emissions from oligotrophic lakes may be different from those that control methane emissions from eutrophic system like ours (Regione Emilia Romagna, 2018). As a possible comparison to our results, we have considered Calabrese et al., 2021 (10.1088/1748-9326/abedea) because the authors examine environments similar to those that we selected for our study, including freshwater and saltwater ecosystems in midlatitude climate conditions.

A section highlighting fluxes measured from standing waters as been added in 3.3. and included in the abstract following the reviewer's comment. Thank you.

Additional comment

In the graphical, the water column image is unclear – why in the seaward water column is lower than upland! or was it groundwater table? Suggest adding texts/legends for clarity.

We thank the reviewer for the comment. In the graphical abstract, when representing salinity effect, water was represented graphically with a color scale to represent the transition from freshwater (blue) to saltwater (red). Since such color scale may be confusing, water is now represented with the same color in the three pictures. We have added text to clarify.

Suggest improving Fig 3-7. Hard time for eyes to understand texts. Show clear visibility of units (avoid grey text, use black). Bubble Heatmap could be shown in better way for more visibility.

Figures 3-7 have been improved in readability along the suggestions in terms of colors and font size. Color scale has been corrected using black instead of grey in bubble heatmaps. All fonts in the images have been increased in resolution.

Suggest adding SD in Tab. 1. With number of discrete samples analyzed (soil part, e.g.,)

Tab. 1 has been modified adding to the mean value ± its standard deviation and sample size. Thanks for the suggestion.

Suggest consulting with language correction agencies or similar as the present version is full of grammatical mistakes with misplaced wordings, formatting in flux units etc.

The text has been improved by checking for typos with a dedicated software, and reviewing syntax and form with external supervision of a native language speaker.

The authors thank the reviewer for the time and efforts put into this review. We believe that the suggestions greatly improved the quality of the manuscript.