

## Comments R2:

### General comments:

In a world dealing with climate change, there is a need to better understand all ecosystems. Studies like this one, investigating GHG exchange in understudied ecosystems like non-tidal salt marches are relevant and important. The combination of in-situ CO<sub>2</sub> – CH<sub>4</sub> soil fluxes with CO<sub>2</sub> vegetation fluxes in the different habitats results in interesting insights and a valuable addition to the laboratory studies with controlled conditions previously carried out. The study highlights the seasonal variability and the differences between species well.

Thank you for your positive feedback on the value of the study.

The Materials and Methods sections could be more detailed. Many elements are not mentioned here such as soil information, salinity, more specific climate data, the amount of data points taken and details about calculations are also left out. Possible additions and suggestions are mentioned in the attached file.

We have included information about salinity and climatic data in section 2.1. We have also added more specific climate data (Figure S2), and a table showing mean soil SOC, TN and bulk density parameters obtained for the three studied habitats (Table S3). Detailed information about the samplings performed are shown in the Supplementary Material tables, while further explanation about the mineralization quotient calculations has also been added. Lines where this information has been included are specified in the responses to the specific comments (see below).

The Carbon mineralization quotient is not entirely clearly explained for me in the method section and not much explanation is given in the result and conclusion section. I think more explanation is needed around the mineralization quotient calculation and some discussion is needed around the carbon sequestration potential of the habitats as this result is interesting but not well supported.

The mineralization quotient has been more thoroughly explained in the Material and Methods section and commented in detail in the Discussion section. The lines where this information has been included are specified in the responses to the corresponding specific comments (see below).

The authors mention large discrepancies between measurement methods (GC, soda lime) and between in situ and laboratory experiments, therefore this information should be added when comparing to literature. Especially for the soil fluxes it should be mentioned which method and closure time is used in the literature you compare to. In this study, the closure time of the chamber for the GC method is very long, with only 2 data points (before and at the end of the closure time), so this will seriously influence the fluxes.

We have always compared our results with previous studies of soil carbon fluxes conducted under field conditions. This has been clarified in the caption of Table 1. We have also added information about the methodology used in these previous studies (Table 1), emphasizing the limitations of comparing data obtained by means of different methodologies (lines 420-423 and 427-428). Moreover, we have specified which previous studies used the same chamber closure time (lines 453-455) and we have highlighted the reliability of the soda lime method to estimate integrated soil CO<sub>2</sub> fluxes over long time periods, such as the one used in the present study (lines 128-129 and 151-154).

Data presentation can be improved by the inclusion of a table with soil parameters, a table with the main results, a map of the study region and graph of climate data (either in supplementary or in the main text).

We have included all this information as Supplementary Material.

More comments and also some technical corrections and suggestions for readability are included in the attached document.

### **Specific comments:**

#### ***Introduction***

Line 68: How extensive are the salt marshes worldwide/in this region and what is the proportion of tidal vs non-tidal in this region?

This information has been added to the text in lines 72-74 (*"Hence, considering the extensive coverage of non-tidal salt marshes in the Mediterranean Basin, which has been estimated in approximately 19 million hectares (around 2.5% of the total area of the 27 Mediterranean countries and 1 to 2% of wetlands in the world; Geijzendorffer et al., 2018)"*).

#### ***Materials and Methods***

Line 79: Authors could add a map of the region

Following your suggestion, we have added a map of the study zone in the Supplementary Material (Fig. S1).

Line 92: Authors could add some climate data of the region both in numbers and a graph. Annually average rainfall, mean temperatures, ... also add some soil data if this is available.

We have included the lowest and the highest mean temperatures in the text (line 98) and we have added a Figure (supplementary material, Figure S2) showing monthly mean daily temperatures and total rainfall for the previous 10 years and for the study year (2017). Besides, we have included the groundwater salinity in section 2.1 (lines 103-105). Soil salinity differences between habitats and seasons (represented as variations in soil electrical conductivity) are shown in section 3.2.1 (Figure 3).

How many months is the soil flooded on average? Is this different between the different habitats?

We have included a sentence providing information on the flooding duration of each habitat in lines 101-103 (*"The duration of flooding varies among habitats, with the shortest duration in the salt meadow (a few days at most), an intermediate duration in the halophilous scrub (several weeks) and the longest duration in the glasswort sward (ranging from several weeks to several months) (Pascual and Martinoy, 2017)"*).

Be sure to add the bulk density, SOC, C and N from the three habitats as these are important in the discussion

We have added a table to the Supplementary Material (Table S3) showing mean SOC, TN and bulk density values for the three studied habitats.

Section 2.2: Is the NER measured in situ with the leaves and tissues attached to the plants?

All measures of net CO<sub>2</sub> fluxes from vegetation were conducted in the field using intact and attached plant tissues. We have clarified this in the text in lines 108-109 (*“Measurements were performed in the field, using living attached green and woody plant tissues”*).

How many leaves and tissues were measured per plant and how many plants were measured per species per session and in total?

The number of plants measured per species and the frequency of samplings, considering the time of the day, are detailed in Table S1. A sentence has been added to inform readers about this Table (lines 116-117).

Section 2.3: I’m don’t know the soda lime method very well, but was the amount of soda lime needed for the chambers tested to be sure of complete absorption of the efflux or was this based this on previous literature?

The amount of soda lime required for this study was previously tested by one of the coauthors in earlier works (unpublished data).

I also think the statement starting in line 137 should be more nuanced or better supported, as in the same reference (Lou and Zhou, 2006), there is also indicated “The method (soda lime) tends to overestimate soil CO<sub>2</sub> efflux in its low range and underestimate it in its high range compared-with dynamic methods (Yim *et al.* 2002). The technique can potentially underestimate soil surface CO<sub>2</sub> effluxes by 10 to 100% (Norman *et al.* 1992, rochette *et al.* 1992, Haynes and Gower 1995, Nay *et al.* 1994).”

The sentence used in this article is based on the sentence from (Lou and Zhou, 2006) “The GC method can potentially underestimate the rate of soil CO<sub>2</sub> fluxes in comparison with other methods by up to 45% (Knoepp and Vose 2002).” But if you continue to Knoepp and Vose 2002, it seems to me that the SODA method used in this study is not that good either compared to the other methods and underestimates the CO<sub>2</sub> emissions even more than the GC method.

We agree that the statement about gas chromatography has not enough support, and thus we have decided to remove the sentence “it has been observed that gas chromatography can underestimate CO<sub>2</sub> emission rates by up to 45% in comparison with the soda-lime method (Lou and Zhou, 2006)”. Instead, we have explained why we chose to use the soda-lime method, rather than gas chromatography, to measure soil CO<sub>2</sub> fluxes when the soil was not flooded, see lines 151-154 (*“Gas chromatography analyses were not used to estimate soil respiration when the soil was not flooded because temperature and humidity variations throughout the day and night could affect the concentration of gas components in the sample, not being this a problem by using the soda-lime method, which can integrate soil CO<sub>2</sub> fluxes over long periods, such as 24h (Keith and Wong, 2006)”*).

In this study serious underestimation is however a possible issue as the closure time is 24h. This is a long time in which saturation in the headspace can occur. The accumulation of the gas inside the chamber can limit the further emission. However this underestimation is not linked to the GC method but rather to the closure time of the chamber.

Previous studies (see for instance Keith and Wong, 2006) support the reliability of the soda-lime method for measuring soil CO<sub>2</sub> emission after long periods of chamber closure (such as 24h). One study cited in the text also kept the chamber closed for 24h (Hirota *et al.*, 2007), although measurements were not performed using the soda lime methodology.

Mention how many measurements were taken in flooded state and how many in non-flooded state and how this is different for different habitats.

The number of measurements performed in flooded and non-flooded soils for each habitat is detailed in Table S2. We have added a sentence to let readers know (lines 154-155).

$W_f$  and  $W_i$  were estimated from volumetric concentration (%) considering the air volume inside the chamber in each sampling date. Is it meant here inclusion of temperature and pressure measurement from the chamber on the sampling date to transform the ppm/ppb measurements from the GC to g CO<sub>2</sub>/CH<sub>4</sub>? If yes, which temperature and pressure is used?

No, we did not include temperature and pressure in the calculations. We converted the volumetric concentration (ml CO<sub>2</sub>/100 ml air) obtained from the GC results to mg CO<sub>2</sub> using the chamber volume and CO<sub>2</sub> density. We estimated the amount of CO<sub>2</sub> (in millilitres) within the chamber and then we converted these ml to g CO<sub>2</sub> by multiplying by the CO<sub>2</sub> density.

Line 146: very small comment but the unit of SMF is g CH<sub>4</sub> m<sup>-2</sup> d<sup>-1</sup> here but later on the unit mg CH<sub>4</sub> m<sup>-2</sup> d<sup>-1</sup> is consistently used.

Thank you. We changed the units in the CH<sub>4</sub> flux equation to mg CH<sub>4</sub> m<sup>-2</sup> d<sup>-1</sup>.

Line 164: The bulk density of soil is never mentioned before, so authors could add the values. Also mention the SOC values used (see also previous comment on line 92).

As previously mentioned, we have added Table S3 in Supplementary Material, which provides information on SOC, TN and bulk density for the same plots where we performed the carbon flux measurements.

I'm not familiar with the carbon mineralization quotient. How were the C\_CO<sub>2</sub> and C\_CH<sub>4</sub> values calculated?

The carbon content of CO<sub>2</sub> (C\_CO<sub>2</sub>) and CH<sub>4</sub> (C\_CH<sub>4</sub>) was calculated using the atomic and molecular weights of these molecules. For example, to calculate C\_CO<sub>2</sub>: from X grams of CO<sub>2</sub>, we estimated the amount of carbon in these X grams by considering that 44 g of CO<sub>2</sub> have 12 g of C. Thus, we multiplied the grams of CO<sub>2</sub> emitted by 12 and we divided by 44. C\_CH<sub>4</sub> values were calculated similarly.

How was the transformation from the unit of "g CH<sub>4</sub> m<sup>-2</sup> d<sup>-1</sup>" and "g CO<sub>2</sub> m<sup>-2</sup> d<sup>-1</sup>" to "mg C g soil<sup>-1</sup> d<sup>-1</sup>"?

The calculation of mg C is explained in the previous response. To convert emissions per unit area to emissions per grams of soil, the volume of soil under the chamber was first estimated by multiplying the area of the chamber by 20 cm, which was the soil depth considered. Then, using the estimated soil volume and bulk density (g soil/cm<sup>3</sup>), we calculated the grams of soil under the chamber (i.e. the soil from which the carbon emission measured comes from).

We added information about these calculations in lines 179-185 ("*C\_CO<sub>2</sub> and C\_CH<sub>4</sub> were calculated multiplying the amount of CO<sub>2</sub> and CH<sub>4</sub> emitted by 12/44 and 12/16, respectively, being 12 the molecular weight of carbon, 44 the molecular weight of CO<sub>2</sub>, and 16 the molecular weight of CH<sub>4</sub>. SOC values were taken from previous measurements performed*").

*in July 2015 and 2016 in the same experiment (Table S3), after observing that these values exhibited stability and remained constant over the studied years (Carrasco-Barea et al., 2023). To convert emissions per unit area to emissions per grams of soil, we estimated the volume of soil beneath the chamber by multiplying the chamber area by the considered soil depth (20 cm), and then multiplying this volume by soil bulk density (g soil cm<sup>-3</sup>.)”*

## **Results**

Concerning the correlation between SR and SOC and TN found in July: there is one value of SOC and TN for each habitat, so three in total? Or is there a value for each plot?

To avoid pseudoreplication, we used the mean of SOC and TOC for each habitat (n=5 per habitat), with each habitat having a single representative value.

## **Discussion**

The authors could add a table (maybe in supplementary) with mean/min/max values of water use efficiency, photosynthetic rates, ...

Following your suggestion, we have added a table to the Supplementary Material with mean/min/max values of instantaneous net CO<sub>2</sub> exchange rates (NER) of vegetation (Table S4), another with stomatal conductances and intrinsic water use efficiencies (Table S5) and a third one with carbon fluxes and mineralization quotients of non-flooded soils (Table S6).

Section 5.2.2.

The soil C and N content is put forward as possible explanation in line 372 and 377 for the higher SR in HS and SM than in GS, so it think the values of SOC and TN should be added in the paper (maybe in supplementary).

As commented before, we have added a table with the mean values of SOC, TN and bulk density to the Supplementary Material (Table S3).

Line 375-378: Accordingly, a positive correlation was found between July SR and SOC or TN content at the halophilous scrub and the salt meadow of La Pletera salt marsh, since these two habitats had higher content of SOC and TN than the glasswort sward (Carrasco-Barea et al., 2023).

Was there only a positive correlation found between SR and SOC and TN for HS and SW because in the results this is not specified (see my one remark in the section Results above). Also if you have one value of SOC or TN per habitat, then how is a correlation per habitat found, which brings me back to my previous question in Result section?

Maybe this needs to be rephrased, framing that the positive correlation found between July SR and SOC or TN content across the habitats together with the fact that HS and SM have higher SOC and TN underpin the statement that the differences in SR might be related to the soil C and/or N content.

Thank you for your comment, since there was an error in the redaction of these sentence. As you noticed, it seems that we did a correlation per each habitat, when, in fact, we did a correlation considering one value (the mean) per habitat. We rephrased the sentence in order to clarify this as follows (lines 393-396): *“In our study, a positive correlation was found between July SR and SOC or TN content reinforcing the idea that differences in SR among*

*habitats would be related with the higher SOC and TN content found in the halophilous scrub and the salt meadow compared to the glasswort sward (Table S3)*”.

Line 394: can the “occasional tide” be more specific. Are we talking about flooding during several weeks in specific months or also small occasional floods once every week?

We have clarified the meaning of “occasional tide”, including the frequency with which this salt marsh is typically flooded and the shortest and longest flood durations across the different habitats in lines 417-418 (*“In tidal salt marshes, flooding occurs once or twice every day, while it is occasional at La Pletera (1-2 times per year, remaining the soil flooded from some days in the salt meadow to several weeks or even months in the glasswort sward) (Pascual and Martinoy, 2017).”*). This information has also been added to the Materials and Methods section (lines 101-103).

Line 406-413: The way this section is written makes it seem like there is mainly absorption of methane and then some sudden high emission peaks, while it is actually mainly emissions that are measured. Maybe a percentage of negative fluxes to total fluxes can be given.

This part of the discussion has been rewritten. We have now focused the discussion on the seasonal changes and the comparison between habitats and with previous studies performed in other marshes (lines 429-455).

Line 411-413: “In the glasswort sward, peaks in CH<sub>4</sub> emissions were observed both when the soil was not flooded (110± 59 mg CH<sub>4</sub> m<sup>-2</sup> d<sup>-1</sup>) and when it was flooded (131 ± 45 mg CH<sub>4</sub> m<sup>-2</sup> d<sup>-1</sup>), highlighting that methane oxidation in the overlying water column would not be happening.”

I’m not sure that emissions during both non flooded and flooded states prove that there is no oxidation in the overlying water column. It states that the methane oxidation is rather limited or actually that the methane oxidation is not a big factor as the net emission is still large.

This part of the discussion was removed following a suggestion from reviewer 1.

Line 439-442: Not much is said about this mineralization quotient. Is there an explanation for the higher sequestration potential of the HS and SM or the lower sequestration potential of GS? I assume that the SOC amount of GS is very low compared to the SOC amount of HS and SM (be sure to put these values in).

As commented above, we have added SOC values to Table S3 and discussed the results in more detail in the discussion, relating the mineralization quotient to the SOC values in lines 404-409 (*“Despite the halophilous scrub and the salt meadow had higher soil CO<sub>2</sub> emissions than the glasswort sward, they showed lower mineralization quotients due to a much greater amount of SOC (Table S3), which was in accordance with a much higher aboveground, belowground and litter biomass (Carrasco-Barea et al., 2023). Hence, our results would indicate that soils of the halophilous scrub and the salt meadow would have a higher carbon sequestration potential, despite their higher soil carbon emissions”*).

Table 1. The authors mention large discrepancies between measurement methods and between in situ or laboratory experiments, therefore this information can maybe be added in the table.

We have added to Table 1 the methodology used in each study and we have specified in the table caption that all the studies cited were conducted *in situ*.

I would suggest to move this table to supplementary material and instead incorporate some tables with the data gathered from this study. For readers who want to quickly scan the paper, a table with the mean results of the study would be handy.

We think that Table 1 helps readers to quickly compare present data with data from previous studies and easily locate values discussed in the text. For this reason, we have decided to keep it in the main manuscript. However, following your suggestion, we have included three tables (Tables S4, S5 and S6) with the main results, although they have been added to the Supplementary Material to avoid redundancy with the Figures in the manuscript.

#### **Technical corrections:**

##### **Abstract**

Line 19-21: Regarding the studied habitats, the halophilous scrub and the salt meadow showed higher soil CO<sub>2</sub> emissions than the glasswort sward, ~~being these values, in general, and the overall emissions~~ were higher than those previously reported for tidal salt marshes.

Done.

##### **Introduction**

Line 29: ~~in relation~~ compared to the atmospheric concentration

Done (line 31).

Line 30-32: In this context of continuous global warming, ecosystems play an important role in global climate regulation, ~~being thus~~ Therefore, it is essential to determine net emissions of greenhouse gases of ecosystems to estimate their effects on global warming.

Done (lines 31-33).

Line 34: (Laffoley & Grimsditch, 2009) is not present in the references.

We have included this citation in the reference's list.

Line 35-39: ~~Regarding net primary productivity, p~~Previous studies on the photosynthetic capacity of salt marsh halophytic species have mainly focused on the effect of salinity on photosynthetic rates, ~~being and most of these studies mostly were~~ performed under controlled conditions (Davy et al., 2006; Duarte et al., 2014; Kuramoto and Brest, 1979; Nieva et al., 1999; Pearcy and Ustin, 1984; Redondo-Gómez et al., 2007) and less frequently under field conditions (Drake, 1989; Maricle and Maricle, 2018; Warren and Brockelman, 1989).

Done (lines 36-40).

Line 45-47: Photosynthetic rates also depend on abiotic factors, such as light, temperature, flooding regime, salinity or nutrient availability (Drake, 1989; Huckle et al., 2000). ~~being in~~

general it is assumed that the highest plant photosynthetic activity occurs during the hours of the day with the highest solar radiation (midday).

Done (lines 45-48).

Line 55-56: In salt marshes, flooding also has a major effect on CO<sub>2</sub> and CH<sub>4</sub> emissions, since it determines which process, aerobic respiration or anaerobic metabolism, prevails.

Done (lines 56-57).

Line 60-62: Nevertheless, ~~in general~~, generally soil CH<sub>4</sub> emissions are negatively affected by salinity (Bartlett and Harriss, 1993; Livesley and Andrusiak, 2012; Poffenbarger et al., 2011), since in saline environments sulphate-reducing bacteria use to compete with methanogens for energy sources, and consequently disfavor and even inhibit methane production.

Done (lines 61-64).

Line 65-68: ~~However, despite the importance that of soil carbon fluxes can potentially have~~ in climate regulation, few studies have characterized these fluxes in Mediterranean salt marshes (Wang, 2018), and, to our knowledge, not one study has been performed in non-tidal salt marshes (tides range from 0.1 to 1 m, in contrast to 1-10 m of tidal salt marshes) of the Mediterranean Basin.

Done (69-72).

### **Materials and Methods**

Line 79-80: The study was performed at La Pletera, a coastal Mediterranean non-tidal salt marsh located in the north of the river mouth of the Ter ~~mouth~~ in the municipality of Torroella de Montgrí.

Changed to "... in the north of the mouth of the Ter river ..." (lines 84-85).

Line 86-87: being all these species C3 -> , all these species being C3

Done (lines 91-92).

Line 136: the air volume inside the chamber ~~in~~ on each sampling date.

Done (lines 149-150).

Line 151: air volume inside the chamber ~~in~~ on each sampling date.

Done (line 167).

Line 157 : stored in the soil (SOC) ~~for~~ at/above a certain depth

Done (lines 173-174).

### **Results**

Line 203: Differences among species in instantaneous ~~net CO<sub>2</sub> exchange rates (NER)~~ from green tissues

Done (line 222).

Line 205: after sunrise ~~(with no significant differences with *S. fruticosa* in April (Fig. 1a),~~



Done (lines 223-224).

Line 232: During most of the year, *E. atherica* showed the highest values of stomatal conductance (gs) at midday

Done (line 151).

Line 272: Daily soil respiration (SR) for non-flooded soils of the three

Done (line 291).

Line 273-274: ~~On the contrary,~~ CO<sub>2</sub> emissions were remarkably lower when soils were flooded.

Done (line 292).

Line 276-277: Remarkably high peaks of soil CH<sub>4</sub> emissions were recorded in the three habitats, ~~despite but also~~ negative values, (indicating net CH<sub>4</sub> consumption) were ~~also~~ observed (Fig. 4b).

Done (lines 295-296).

Line 277-279: In the halophilous scrub, soil CH<sub>4</sub> emissions were detected in April, June (with high values) and September, with ~~and~~ the highest CH<sub>4</sub> absorption ~~being was~~ observed in February, when the soil was flooded.

Done (lines 296-297).

Line 279-281: Maximum soil CH<sub>4</sub> emissions for the salt meadow were recorded in July for the salt meadow, and for the glasswort sward in March or June in flooded and non-flooded soils, respectively; ~~for the glasswort sward.~~

Done (lines 298-299).

Line 285: SR and daily soil methane fluxes (SMF) were

Done (line 303).

## **Discussion**

Line 316-319: The same ~~occurred with~~ was true for *E. atherica* and *S. patula*. ~~being~~ their maximum photosynthetic rates at La Pletera ( $29.1 \pm 2.4$  and  $20.8 \pm 2.9$   $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ , respectively) were higher than those previously reported for *E. atherica* ( $18 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ , Rozema & Diggelen 1991) and for the annual species *Salicornia ramosissima* ( $14 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ , Pérez-Romero et al. 2018) grown under controlled conditions.

Done (lines 335-338).

Line 322-324: ~~Therefore,~~ Studies reporting photosynthetic rates of dominant salt marsh plant species under field conditions are scarce, and the values obtained often diverge substantially from those recorded under controlled conditions.

Done (lines 341-343).

Line 335-338: Interestingly, photosynthetic rates of the studied species at La Pletera were much lower in autumn than in spring, despite the environmental parameters, such as temperature and soil moisture, ~~were also~~ being favorable to for photosynthesis in autumn,

(especially in October, where maximum temperature was 21°C and soil VWC was even higher than in March and April;-(Pascual, 2022). A possible explanation might be related ~~with to~~ the high accumulation of ions and soluble carbohydrates ~~that in~~ these species ~~would present~~ after a salt stress period, ~~such as the one occurring~~ which occurs in the Mediterranean salt marshes during summer.

Done (lines 354-359).

Line 346-348: especially in March and May and before sunset, with values of photosynthesis reaching 12  $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ . ~~These values are in agreement with data reported for Californian evergreen species (Saveyn et al., 2010) or for savannah shrubs and trees (Cernusak et al., 2006; Levy and Jarvis, 1998).~~

Done (lines 365-367).

Line 357-358: Regarding night respiration rates, the highest values for the four species were recorded in summer (August) and/or autumn (November), ~~being especially elevated with~~ those found for the green tissues of *S. fruticosa* and *E. atherica* during these months ~~being especially elevated.~~

Done (lines 375-377).

Line 362: references are large

We have reduced the font size.

Line 364-365: In November, respiration rates were also very high despite ~~that~~ the minimum temperature was much colder (4.6 °C) than in August (22.2 °C) and similar to February (5.9°C) (Pascual, 2022).

Done (lines 382-383).

Line 384-386: On the contrary, the sparse vegetation, (which is only alive during few months) of the glasswort sward and the poorly developed root system of its dominant species, *S. patula*, would make ~~negligible~~ the contribution of roots to soil respiration in this habitat ~~negligible.~~

Done (lines 402-404).

Line 437: This would not be the case ~~of in~~ the salt meadow, the most distant habitat from the sea,

Done (lines 450-451).