**Methane emissions from created and restored freshwater and brackish marshes
in southwest Florida, USA**

Xiaoyu Li^{a,b}, William J. Mitsch^{b*}

^a Key Laboratory of Wetland Ecology and Environment, Northeast Institute of
Geography and Agroecology, Chinese Academy of Science, Changchun, China,
130102

^b Everglades Wetland Research Park, Florida Gulf Coast University, 4940 Bayshore
Drive, Naples, FL, USA, 34112

*Corresponding author: phone: +1 239 325 1365; fax: +1 239 732 7043;

email: wmitsch@fgcu.edu

Abstract

It is important to estimate greenhouse gas emissions from newly created and restored wetlands so that we learn how to design them to minimize these emissions. Spatial and temporal patterns of methane emissions were measured in three wetland marsh complexes in southwest Florida: a created freshwater marsh on a university campus, a restored brackish/salt marsh on the fringe of a mangrove (*Rhizophora mangle*) coastal swamp; and a natural freshwater marsh adjacent to a cypress (*Taxodium distichum*) strand. Non-steady-state rigid chambers were used twice a day (morning and afternoon) on a monthly schedule for 13 months. The mean $\pm$ standard error (median) of methane emissions from permanently and intermittently flooded areas were 124 ± 47 (0) and 0.8 ± 0.7 (0) g-CH₄-C m⁻² y⁻¹ in the created freshwater marsh, and 58 ± 14 (9.4) and -0.1 ± 0.3 (0) g-CH₄-C m⁻² y⁻¹ in restored brackish marsh. Methane missions

1 were 0.8 ± 0.5 (0) g-CH₄-C m⁻² y⁻¹ in the intermittently flooded natural freshwater
2 marsh that served as our control. In general, average methane emissions were higher
3 from permanently than intermittently flooding areas ($P < 0.05$); our data suggested
4 that ebullition may have occurred in permanently flooding areas and caused up to 6.3
5 g-CH₄-C m⁻² d⁻¹ in pulses in the created marsh and 1.7 g-CH₄-C m⁻² d⁻¹ in restored
6 marsh. Removing ebullition outliers, methane emission estimates decreased by one-
7 fourth to one-half the annual rates given above to 30 g-CH₄-C m⁻² y⁻¹ in permanent
8 created freshwater marshes and 27 g-CH₄-C m⁻² y⁻¹ in the restored brackish marshes.
9 There were no significant differences in methane emissions from created, restored or
10 natural marshes in intermittently flooding areas ($P > 0.05$). CH₄ emissions exhibited
11 strong quadratic and linear relationships with water depth when all sites are compared
12 ($P < 0.05$), but no significant relationships with soil temperature or salinity. These
13 results give useful information on ways to minimize methane emissions when creating
14 and restoring wetlands. But these results should be only one of the considerations for
15 wetland design as other ecosystem services should also be considered.

16 **Key words:** wetland creation and restoration, wetland hydrology; tropical/subtropical
17 methane emissions; methane ebullition; *Taxodium distichum* swamp; *Spartina bakeri*
18 marsh

19

20 1. Introduction

21 Methane (CH₄) is the third most abundant greenhouse gas (GHG), following water
22 vapor and carbon dioxide (CO₂), in the atmosphere (Nahlik and Mitsch, 2011; Mitsch
23 and Gosselink, 2015). CH₄ concentrations have increased 150% (from 722 ppb to
24 1803 ppb) since the beginning of the industrial era in the late 18th century and the
25 global warming potential (GWP) of CH₄ is now estimated to be 28 times greater than

1 CO₂ over a 100-year time scale without climate-carbon feedbacks (IPCC, 2014).
2 Therefore, even relatively low rates of CH₄ emissions could offset the benefit of CO₂
3 sequestration in terms of the net GHG effect (Knox et al., 2015), although Mitsch et
4 al. (2013) illustrated that is not a simple matter of using the ratio of the GWPs to
5 determine the net impact of wetlands on climate.

6 A majority of the natural CH₄ emissions are from tropical/subtropical wetlands
7 (Bergamaschi et al., 2007; Bloom et al., 2010; Mitsch et al., 2010, 2013). Therefore, it
8 is important to do further research on methane emissions from tropical and
9 subtropical wetlands. Also, although wetlands constructed or restored for water
10 quality enhancement or habitat replacement could act as carbon sinks (Mitsch et al.,
11 2013; de Klein and van der Werf, 2014), most studies on methane emission from
12 wetlands have been done at natural wetlands such as peatlands, floodplains, coastal
13 marshes, and freshwater marshes (Baldocchi et al., 2012; Livesley and Andrusiak,
14 2012; Trudeau et al., 2013; Sun et al., 2013; Tong et al., 2013; Chen et al., 2013) and
15 some methane emission studies were carried out at created marshes in temperate zone
16 central Ohio (Altor and Mitsch, 2006, 2008; Nahlik and Mitsch, 2010; Sha et al.,
17 2011; Waletzko and Mitsch, 2014; Stefanik and Mitsch, 2014; Morin et al., 2014;
18 Brooker et al., 2014; Chowdhury et al., 2014).. But few if any measurements of CH₄
19 emissions have been made from restored or created wetlands in tropical or subtropical
20 regions.

21 CH₄ emissions from wetlands to the atmosphere are affected by several
22 environmental variables (Wang and Han, 2005; Bergamaschi et al., 2007). The water
23 table affects the spatial variation of CH₄ flux by controlling the spatial distribution of
24 vascular plant species (Sun et al., 2013) and the overall extent of wetlands. Water
25 level also affects GHG emissions directly through its influence on redox potential

1 (Ding et al., 2002, 2003, Nahlik and Mitsch, 2011, Yang et al., 2013). Low
2 temperature reduces CH₄ emission directly and indirectly by decreased the
3 decomposition of organic carbon and plant photosynthesis (Ding and Cai, 2007). CH₄
4 flux is negatively affected by salinity (Bartlett et al., 1988, Bartlett and Harriss, 1993,
5 Poffenbarger et al., 2011. Livesley and Andrusiak, 2012, Marton et al., 2012) so
6 coastal salt marshes and mangrove swamps routinely have lower CH₄ emissions than
7 do freshwater marshes (Ding and Cai, 2007; Tong et al., 2013; Sun et al., 2013). This
8 is due to substrate competition between sulfate reducing bacteria and methanogens,
9 with less CH₄ produced in high salinity (Ding and Cai, 2007).

10 Our objective is to investigate the spatial and temporal variations of methane
11 emissions in created/restored freshwater and brackish marshes in subtropical south
12 Florida USA, and compared them to nearby natural freshwater marshes.
13 Environmental parameters such as water depth, soil temperature and salinity were
14 also measured to determine their effects on CH₄ emissions. Our hypotheses are as
15 follows:

- 16 1. CH₄ emissions will be lower from brackish marshes than from freshwater
17 marshes, and both will be lower than from natural marsh;
- 18 2. CH₄ emissions will be higher in permanently flooded zones than in
19 intermittently flooded zones; and
- 20 3. Water depth and salinity are the main controlling factors influencing CH₄
21 emissions in these subtropical wetlands.

22

23 **2. Methods**

24 **2.1. Study sites**

25 The three marshes in this study are located in Southwest Florida USA (Figure 1)
26 and described in Table 1. The created freshwater marshes (26°27'N, 81°46'W) are

1 located north of the main entrance road to the Florida Gulf Coast University campus
2 in Fort Myers, Florida, adjacent to the Ben Hill Griffin Parkway. Before the campus
3 was constructed, an extensive ecological inventory was carried out in 1995 Campus
4 Master Plan Inventory report (FGCU, 2010). Because campus construction and its
5 associated infrastructure impacted a variety of upland and wetland systems on this
6 site, a comprehensive mitigation plan was developed and was implemented. To offset
7 impacts, 35.5 ha of wetlands were created, restored and preserved. This freshwater
8 marsh was created as part of that mitigation in 1995 (FGCU, 2010). Rainfall in wet
9 season and runoff from adjacent campus areas are the main water sources recharging
10 the wetlands. With increasing water depth, the dominated vegetation transitions from
11 *Cyperus* spp. to *Utricularia vulgaris*, *Nymphaea tetragona*, and *Eleocharis palustris*.
12 Populations of *Nymphaea* spp. and *Utricularia vulgaris* were positively correlated
13 with the areas of permanently flooding, which expand in the rainy season.

14 The 23-ha restored brackish marsh is part of the 65-ha Naples Botanical Garden in
15 Naples, Florida, USA (26°06'N, 81°46'W). Restoration of the site took place in 2007
16 and it has been used for environmental education for garden visitors and for wetland
17 research by university students since then. The restored brackish marsh is on the
18 fringe of an extensive mangrove (*Rhizophora mangle*) coastal swamp on Naples Bay.
19 The salinity of the brackish marsh depends on the mixture of salty water from tidal
20 change of mangrove creeks and freshwater from rainfall, surface runoff from the
21 botanical garden. The brackish marsh is dominated by sand cordgrass *Spartina bakeri*,
22 and accompanied by other accompanying species, such as *Spartina patens*, *Cladium*
23 *jamaicense*, *Acrostichum danaeifolium* and *Eleocharis cellulosa*. Some mangrove
24 trees, e.g., *Rhizophora mangle* and *Avicennia germinans*, are beginning to colonize in
25 the marsh.

1 Our natural freshwater marsh that was used as a reference site was at the Audubon
2 Society's Corkscrew Swamp Sanctuary (CSS) in southwest Florida (26°23'N,
3 81°35'W). CSS is a 5260-ha nature preserve and home to the largest remaining stand
4 of old growth bald cypress (*Taxodium distichum*) in North America (Fig.1, Table 1)
5 and was designated as a wetland of international importance by the Ramsar
6 Convention in 2010. CSS consisted of pine flatwoods, wet prairie (marshes), and an
7 old-growth bald cypress forest. The dominant macrophyte in the marshes used in this
8 study site was *Eleocharis palustris*.

9 2.2. Experimental design

10 Four plots were chosen at the created freshwater marshes, two in intermittently
11 flooded sites (dominated by *Cyperus* spp.) and two in permanently flooded sites
12 (dominated by *Utricularia vulgaris*, *Nymphaea tetragona*, and *Eleocharis palustris*).
13 Four plots were established at the restored brackish marsh, two at intermittently
14 flooded sites dominated by *Spartina patens*, and two at permanently flooded sites
15 (open water). Two plots were chosen as intermittently flooded reference marsh
16 dominated by *Eleocharis palustris*. There were no permanent flooding areas in
17 reference natural marshes.

18 CH₄ samples were taken monthly from February 2014 to June 2014 and again from
19 October 2014 to February 2015. Rectangular plastic storage containers (L×W×H =
20 0.4 m × 0.56 m × 0.3 m) with bottoms removed (20-cm left) were used as non-steady-
21 state chamber frames. Two non-steady-state chamber frames were inserted 10 cm into
22 the ground at each intermittently flooded plot when the water level was below 0 cm in
23 January 2014, to serve as permanent bases. The distance was no more than 30 cm
24 between two chambers. A portable floating chamber deployed in permanently flooded
25 zones was encircled by foam around the top edges for buoyancy and wrapped with

1 duct tape for waterproofing.

2 *2.3. Methane flux measurements and analysis*

3 Gas was collected in the morning and afternoon during each sampling time for each
4 chamber. Samples were taken from each chamber every 5 min over 25 min after the
5 chamber was closed, through a septa with a 20-ml syringe and injected into pre-
6 evacuated, 10-ml glass vials sealed with grey butyl septa (T0-T5, T0 was background
7 CH₄ concentrations). Thermometers were installed within the chambers and chamber
8 temperature was recorded when each sample was extracted.

9 CH₄ concentrations were analyzed on a Shimadzu GC-14A gas chromatograph.
10 Matheson methane standards were used to perform six-point calibration curves. Two
11 check standards were injected at a rate of approximately one check standard per 30
12 field samples as described by Sha et al. (2011). CH₄ volume/volume concentrations
13 (C_v , $\mu\text{L CH}_4 \text{ L}^{-1}$) were converted into mass/volume concentration (C_m , $\text{mg CH}_4\text{-C m}^{-3}$),
14 methane emissions rates got from a linear regression was used to calculate hourly
15 CH₄ flux ($\text{mg CH}_4\text{-C m}^{-2} \text{ h}^{-1}$), according to the methods described by Villa and Mitsch
16 (2014). Average hourly methane emission rates were calculated by two replicates
17 from averaging morning and afternoon fluxes.

18 *2.4. Environmental and climate factors*

19 Soil temperature at 5-cm depth was recorded by thermometers during each
20 sampling time. Water depth (Ott Thalimedes) and salinity (YSI multi-parameter
21 sondes) were determined in open water in the created freshwater marsh and restored
22 brackish marsh, which data recorded every ten minutes. At the freshwater natural
23 marsh, water depth was estimated by a meter stick at each sampling. Precipitation and
24 air temperature data were from Immokalee County, Southwest International Airport

(Fort Myers, Florida) and Naples Florida (downloaded from WU-weather underground website), the closest meteorological stations to the study sites.

2.5. Statistical analysis

SPSS 22.0 and SigmaPlot 12.0 were used to analyze and plot the data. One-way ANOVA was used to test differences of climate and environmental factors. Non-parametric tests, Kruskal-Wallis and Mann-Whitney tests were used to compare methane emissions of different sites, different flooded zones and seasons. Box plots were used to describe the spatial and seasonal variation of methane emissions, with boxes edges indicating 25 and 75 percentiles. Whiskers extended to the maximum and minimum of methane emissions. Outliers were showed outside the 10th and 90th percentiles using dots. Spearman's correlations were used to determine relationship between mean methane emissions and sites properties. All significance tests and correlations were conducted at a level of $\alpha=0.05$.

3. Results

3.1. Physical factors

Although the annual precipitation of restored brackish marsh (1233 mm yr⁻¹) and created freshwater marsh (1100 yr⁻¹) sites were higher than the natural freshwater marsh (770 mm yr⁻¹), there were no significant differences on average monthly precipitation during sampling period ($F=0.27$, $P > 0.05$, Table 1, Fig. 2). Average air temperatures were also similar among the three sites ($F=0.21$, $P > 0.05$), but the range of air temperatures from natural freshwater marsh was higher than at the other sites (Table 1).

There were no significant differences in water depth among the sites in permanently

1 (Fig. 2, $F=0.05$, $P > 0.05$) and intermittently flooded zones ($F=2.55$, $P > 0.05$).
2 Salinity was 3.93 ± 0.02 ppt (mean $\pm$ standard error) in the brackish marsh, 28 times
3 higher than that in the created freshwater marsh (0.14 ± 0.0004 ppt) ($P < 0.05$).
4 Average soil temperature was similar at the created freshwater (23.2°C) and restored
5 brackish (22.2°C) marshes, but both were higher than soil temperature at the
6 freshwater natural marsh (19.1°C) ($F=10.68$, $P < 0.05$).

7 *3.2. Spatial and temporal patterns of CH_4 emissions*

8 Hourly CH_4 emissions over the entire measurement period are shown in Fig. 2 and
9 summarized in Fig. 3. Some ebullition occurred in permanently flooding areas.
10 Therefore methane emissions in Fig. 2 and correlation with environmental factors in
11 Fig. 4 were done with outliers and without outliers. Methane emission ranges were
12 $0.07 - 262.48$, $-2.06 - 70.86$, $0 - 2.15$ $\text{mg CH}_4\text{-C m}^{-2}\text{h}^{-1}$ with outliers and $-0.07 - 21.92$,
13 $-2.06 - 16.80$, $0 - 2.15$ $\text{mg CH}_4\text{-C m}^{-2}\text{h}^{-1}$ without outliers at the created freshwater,
14 restored brackish and natural freshwater marshes, respectively (Figs. 2 and 3). With
15 outliers, average hourly CH_4 fluxes (mean $\pm$ standard error, $\text{mg CH}_4\text{-C m}^{-2}\text{h}^{-1}$) were
16 14.14 ± 5.35 (permanently flooded) and 0.09 ± 0.08 (intermittently flooded) from the
17 created freshwater marsh, 6.59 ± 1.57 (permanently flooded) and -0.01 ± 0.03
18 (intermittently flooded) from the restored brackish marsh, and 0.10 ± 0.05
19 (intermittently flooded) from natural freshwater marsh. Without outliers, average
20 hourly CH_4 fluxes ($\text{mg CH}_4\text{-C m}^{-2}\text{h}^{-1}$) were 3.49 ± 0.76 and 0.09 ± 0.08 from the
21 created freshwater marsh, 3.16 ± 0.08 and -0.01 ± 0.03 from the restored brackish
22 marsh, and 0.10 ± 0.05 from natural freshwater marsh. CH_4 fluxes were higher from
23 permanently flooded zones than from the intermittently flooded zones (Fig.3,
24 $X^2=28.92$, $P < 0.05$; NBG, $X^2=45.77$, $P < 0.05$). Significant seasonal variability was
25 found in permanently flooded zones in both created ($X^2=21.46$, $P < 0.05$) and restored

1 ($X^2=31.08$, $P < 0.05$) marshes (Fig. 2). Therefore, methane emissions from created
2 and restored marshes with permanently flooding areas were significantly higher than
3 those from the natural freshwater marsh intermittently flooding areas ($X^2=9.76$, $P <$
4 0.05). But there were no significant differences among intermittently flooding areas
5 (Fig. 3).

6 *3.3. Factors of effecting CH₄ emissions*

7 CH₄ fluxes at the created freshwater marsh had positive correlations with water depth,
8 and negative correlations with soil temperature ($P < 0.05$), no correlation with salinity
9 ($p = 0.18$). CH₄ fluxes at the restored brackish marsh had positively correlations with
10 water depth and soil temperature ($P < 0.05$). Interestingly, no significant correlation
11 was found between methane emissions and water depth or soil temperature at the
12 natural marsh ($P > 0.05$). Linear and nonlinear regressions were used to compare the
13 relationship between methane emission and environmental factors when five sites
14 from three location were compared. There were no linear or nonlinear relationships of
15 methane emission with soil temperature (Fig. 4, $P > 0.05$). Methane emissions
16 exhibited strong quadratic relationships and linear relationships ($P < 0.01$) with water
17 depths when outliers were removed, but only significant linear relationships with
18 water depths when outliers were included (Table 2).

19

20 **4. Discussion**

21 *4.1. Seasonal and spatial variability of methane emission*

22 Methane emissions showed significant seasonal and spatial variability in our study,
23 not unlike patterns seen in many other methane emission studies where climate is
24 seasonal (Sha et al., 2011; Ford et al., 2012). Our study showed that methane

1 emissions from our created or restored marshes were significant higher than those
2 from a natural marsh in southwest Florida. Created wetlands had been shown to have
3 lower CH₄ emissions than natural wetlands in temperate zone Ohio (Nahlik and
4 Mitsch, 2010; Mitsch et al., 2014), attributed to the fact that soil organic carbon was
5 still accumulating in the created wetlands. Annual carbon production by emergent and
6 submerged vegetation in created wetlands was shown to be less than half that of
7 reference wetlands in a brackish marsh in the northwestern Gulf of Mexico (Madrid et
8 al., 2012). However, unlike the natural marsh, the created and restored marshes in our
9 study had permanently flooded areas. Seasonally aerobic soil can prevent CH₄
10 produced in deeper layers from being emitted into the atmosphere (Herbst et al.,
11 2011). Husin et al. (1995) found that more methane is produced in permanently
12 flooded rice paddies than in intermittently flooded paddies.

13 The CH₄ emissions were low and similar in the intermittently flooded area of all
14 three sites. Methane emissions from the natural marsh were only 0.10 ± 0.05 mg CH₄-
15 C m⁻² h⁻¹. Water depth and soil temperatures did not change much during our
16 sampling time. The salinity of the brackish marsh was lower than adjacent coastal
17 mangroves, but high enough to inhibit methanogenesis. Methane production is
18 inhibited by salinity (Marton et al., 2012; Mitsch and Gosselink, 2015), primarily
19 because sulfates in salt water lead to sulfate reducing bacteria that compete with
20 methanogens for access to organic substrate (Dowrick et al., 2006). However the
21 mean or median of methane emissions from the restored brackish marsh was not
22 lower than from the created freshwater marsh,.

23 *4.2. Methane emission comparison*

24 To better understand the significance of our methane emissions rates, data were
25 compared with and without outliers. Outliers were judged to be outside of 10% and

1 90% percentiles of all data. After removing the outliers, the mean and range of
2 methane emissions were more similar between the created and restored marshes in
3 permanently flooded area as well as in intermittently flooded areas and both were
4 higher than emissions from the natural marsh. The mean annual emissions of CH₄
5 observed were 123.9 (30.6) and 57.7 (27.7) g CH₄-C m⁻² y⁻¹ in permanently flooded
6 areas of created and restored wetlands (with outliers removed in parentheses), and 0.8,
7 -0.1, 0.8 g CH₄-C m⁻² y⁻¹ in intermittently flooded areas of created, restored and
8 natural marshes respectively.

9 Methane emissions from our created and restored marshes were within ranges seen
10 in other natural wetlands in tropical or subtropical zones (Table 3). The fluxes of
11 permanently flooding marshes in our study were lower than those from permanent
12 freshwater marsh in western Costa Rica (Nahlik and Mitsch, 2011) and pulsing inland
13 delta in Botswana (Mitsch et al., 2013; Gondwe and Masamba, 2014). Yet they were
14 higher than rates estimated for the nearby Florida Everglades several decades earlier
15 (Bartlett et al., 1989). All methane fluxes in intermittently flooding marshes in our
16 study were lower than those measured in an earlier study in Corkscrew Swamp (Villa
17 and Mitsch, 2014), and Lago Calado, Amazon Basin, Brazil (Bartlett et al., 1988).
18 Both our created freshwater and restored brackish marshes had lower methane
19 emissions than did the freshwater tidal marsh and brackish tidal marsh in Louisiana
20 (DeLaune et al., 1983), but they had higher rates than a tidal salt marsh there.
21 Therefore, created or restored wetlands can be large or small sources of methane
22 depending on the environmental and management conditions involved and methane
23 fluxes from created or restored marshes cannot be ignored.

24 *4.3. Methane emissions and wetland hydrology*

25 In both our freshwater and brackish marshes, methane emissions were positively

1 correlated with water depth. Water depth has been routinely shown to be the primary
2 controlling factor of CH₄ emissions from wetlands, affected the spatial variation of
3 CH₄ flux as well as affecting the spatial distribution of vascular plant species (Sun et
4 al., 2013; Malone et al., 2013). Water depth decreased in the permanently flooded
5 areas from February to June 2014 and from October 2014 to February 2015, and
6 essentially dried up in the intermittently flooded areas. Couwenberg et al. (2011)
7 reviewed several publications on CH₄ emissions and found a significant relationship
8 between mean annual water depth and annual CH₄ emissions. The methane emissions
9 from our natural marsh did not correlate with water depth, mainly because the water
10 depth did not change markedly in our sampling period. Our results that methane
11 fluxes exhibited strong quadratic and linear relationships with water depth agreed
12 with results found by Bartlett et al. (1983) in the nearby Florida Everglades and by
13 Villa and Mitsch (2014) in the wet prairie at south Florida's Corkscrew Swamp.

14 *4.4. Methane emissions and soil temperature*

15 Karki et al. (2014) indicated the importance of soil temperature in controlling
16 microbial activities when conditions are favorable for CH₄ production and most
17 methane production occurs in the top 10 cm of the soil (Crozier et al., 1995). A clear
18 relation between CH₄ and soil temperature can only be expected for situations where
19 CH₄ oxidation in the top soil is negligible (Herbst et al., 2011). Interestingly, negative,
20 positive and no correlations were found separately in our created freshwater marsh,
21 restored brackish marsh and natural marsh with soil temperature. Measured methane
22 emissions have showed clear relationships with soil temperature,(Ding and Cai, 2007;
23 de Klein and van der Werf, 2014) as we found in the brackish marsh.

24 Methanogenic bacteria are mesophilic with an optimal range for growth between
25 30-40°C (Zinder, 1993). Seasonal temperature variation in tropical and subtropical

1 wetlands is far less pronounced than in boreal wetlands. The prolonged periods at
2 optimal temperature for methanogen growth at our sites may have resulted in lower
3 correlations between CH₄ fluxes and soil temperature (Jauhiainen et al., 2005; Bloom
4 et al., 2010).

5 When all sites were compared, no linear relationship between methane emissions
6 and soil temperature were found in our study. In the upper soils of wetlands, methane
7 emission rates may be impacted by differences in soil organic carbon (Nahlik and
8 Mitsch, 2011). Organic soils were found to be less responsive to diurnal temperature
9 changes than were sandy soils (Wang and Han, 2005) and a negative correlation
10 between methane emissions and anaerobiosis has been reported (Huang et al., 2005).
11 Further research on the relationships among the soil carbon pool, vegetation
12 productivity, and methane emissions is needed.

13 *4.5. Wetlands creation and restoration and climate change*

14 Although re-flooding drained wetlands may increase methane emissions, reflooding at
15 restored wetlands has been reported to inhibit ecosystem respiration, making the
16 wetlands net CO₂ sinks (Knox et al., 2015). The controlling mechanisms of methane
17 emission from restored wetlands are more complex than those reported for natural
18 wetlands, since they included both management activities and slow adaptive processes
19 related to changes in vegetation and hydrology (Herbst et al., 2011). Both the
20 management regime and the time scale of research can affect methane emissions (De
21 Klein and van der Werf, 2014). Intermittent inundation of hydric soils was shown to
22 produce significantly lower methane fluxes than continuous inundation in created
23 marshes in Ohio (Altor and Mitsch, 2008) and more recently from forested wetland
24 soils in Florida (Pereyra and Mitsch, in review). Similar results were found in the
25 created freshwater marsh and the restored brackish marsh, with lower CH₄ fluxes

1 emitted in shallow water than deep water. Today, more attention has been focused on
2 the “design of sustainable ecosystems” which is the definition of ecological
3 engineering (Mitsch, 2012). Climate change need to be considered when large scale
4 restoration and creation of wetlands are considered as in the Florida Everglades
5 restoration (NRC, 2014). Therefore, optimizing the design, control options, and
6 management of wetland hydrology is key to restoring and creating wetlands while
7 minimized greenhouse gas emissions. Our study suggests that, from a methane
8 emission perspective, created and restored marshes should have fewer deep water
9 areas in their design. But other ecosystem services such as habitat diversity and flood
10 mitigation may suggest the opposite strategy.

11

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23

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12

1 Table 1 Description of three marsh study sites in southwest Florida

Wetland site name; wetland type	Co-ordinates	Wetland type	Area (ha)	Creation/Restoration time	Total Precipitation (mm)	Air Temperature Range (°C)	Average Water Temperature (°C)
Florida Gulf Coast University (FGCU); freshwater created marsh	26°27'N, 81°46'W	Created freshwater marsh	35.5	1995	1100.33	18.28-28.94	23.95
Naples Botanical Garden (NBG); brackish restored marsh	26°06'N, 81°46'W	Restored brackish marsh	64.8	2007	1252.98	18.86-28.34	26.57
Corkscrew Swamp Sanctuary (CSS); freshwater natural marsh	26°23'N, 81°35'W	Natural freshwater marsh/prairie	5260	---	811.28	16.20-30.90	----

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2 Table 2 Regression analysis of methane emission and water depth

Regression	Model Summary					Equation
	R ²	F	df1	df2	Sig.	
<i>Without outliers</i>						
Linear	0.97	95.46	1	3	0.002**	Y = 0.07X - 0.58
Logarithmic	0.76	9.42	1	3	0.06	
Quadratic	0.99	1632	2	2	0.001**	Y = 0.002X ² - 0.03X + 0.17
<i>With outliers</i>						
Linear	0.82	13.77	1	3	0.03*	Y = 0.23X - 2.1
Logarithmic	0.64	5.24	1	3	0.11	
Quadratic	0.87	6.84	2	2	0.13	

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Table 3 Comparison of methane emissions (mean $\pm$ standard error) from subtropical/tropical natural and restored/created marshes

Wetland type	Methane flux (g CH ₄ -C m ⁻² yr ⁻¹)	Study location	Reference
Natural Wetlands			
Intermittent freshwater marsh	0.8 $\pm$ 0.4	Corkscrew Swamp, SW Florida, USA	This study
	19.5 $\pm$ 9.7	Corkscrew Swamp, SW Florida, USA	Villa and Mitsch, 2014
Intermittent freshwater lake	7.3 $\pm$ 1.5	Lago Calado, Amazon Basin, Brazil	Bartlett et al., 1988
	11.0 $\pm$ 3.3	Amazon Basin (near Manaus), Brazil	Devol et al., 1990
	6.4	Orinoco River Floodplain, Venezuela	Smith et al., 2000
Seasonally flooded inland delta	152.6 $\pm$ 14.6	Okavango Delta, Botswana	Gondwe and Masamba, 2014
	96 $\pm$ 11	Okavango Delta, Botswana	Mitsch et al., 2013
Isolated wetland in tropical rain forest	293 $\pm$ 86	La Selva, Costa Rica	Mitsch et al., 2013
Disturbed floodplain wetland	350 $\pm$ 86	Palo Verde, Costa Rica	Mitsch et al., 2013
Dwarf cypress/sawgrass swamp	2.7 $\pm$ 0.4	Everglades, Florida, USA	Bartlett et al., 1989
Spikerush community	10.7 $\pm$ 1.2	Everglades, Florida, USA	Bartlett et al., 1989
Sawgrass (< 1m) community	14.2 $\pm$ 1.1	Everglades, Florida, USA	Bartlett et al., 1989
Sawgrass (> 1m) community	26.2 $\pm$ 3.6	Everglades, Florida, USA	Bartlett et al., 1989
Sawgrass/spikerush/periphyton	16.5 $\pm$ 1.6	Everglades, Florida, USA	Bartlett et al., 1989
Swamp forest	25.1 $\pm$ 8.2	Everglades, Florida, USA	Bartlett et al., 1989
Tidal freshwater marsh (<i>Panicum hemitomon</i>)	213.3	Barataria Basin, Louisiana, USA	DeLaune et al., 1983
Tidal brackish marsh (<i>Spartina patens</i>)	97.3	Barataria Basin, Louisiana, USA	DeLaune et al., 1983
Tidal salt marsh (<i>Spartina alterniflora</i>)	5.7	Barataria Basin, Louisiana, USA	DeLaune et al., 1983
Created/restored Wetlands			
Permanent flooded freshwater marsh	123.9 $\pm$ 46.9	FGCU campus, SW Florida, USA	This study
Intermittent flooded freshwater marsh	0.8 $\pm$ 0.7	FGCU campus, SW Florida, USA	This study
Permanently flooded brackish marsh	57.7 $\pm$ 13.8	Naples Botanical Garden, SW Florida, USA	This study
Intermittently flooded brackish marsh	-0.1 $\pm$ 0.3	Naples Botanical Garden, SW Florida, USA	This study

Figure legends

Fig. 1 Location of the three research sites in southwest Florida: freshwater created marsh at Florida Gulf Coast University (FGCU); brackish restored marsh at Naples Botanical Garden (NBG); freshwater natural marsh (reference marsh) at Corkscrew Swamp Sanctuary (CSS).

Fig. 2 Temporal patterns of methane emissions (rows D and E) and supporting environmental variables including total monthly precipitation (row A), mean air temperature (row B) and mean water level (row C). Study marshes are freshwater created marsh in column 1; brackish restored marsh in column 2; and freshwater natural marsh (reference marsh) in column 3. Methane emissions are shown as means (triangles) and medians (circles) with outliers (row D) and without outliers (row E) in permanently flooded (P) and intermittently (I) flooded areas.

Fig. 3 Mean (dashes) and median (solid line) values of CH_4 emissions ($\text{mg CH}_4\text{-C m}^{-2} \text{ h}^{-1}$) from permanently flooded (P) and intermittently flooded (I) zones separately in three location. Edges in the boxes represent 25% and 75% percentiles; whiskers extend to the minimum and maximum, dots indicate outliers outside the 10th and 90th percentiles. Different letters indicate significant differences of CH_4 emissions among sites.

Fig. 4 Relationships between CH_4 emissions and water level (A, C), and soil temperature (B, D) for all sites. A and B showed data with outliers; C and D showed data without outliers. FGCU: freshwater created marsh at Florida Gulf Coast University; NBG: brackish restored marsh at Naples Botanical Garden; CSS: freshwater natural marsh (reference marsh) at Corkscrew Swamp Sanctuary. I: intermittently flooded; P: permanently flooded.

Figure 1

Figure 2

Figure 3

Figure 4