1   Promotion of full-scale constructed wetlands in the wine sector:

2   comparison of greenhouse gas emissions with activated sludge systems

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Abstract

The aim of this study was to quantify and compare greenhouse gas (GHG) (i.e. carbon dioxide (CO_2), nitrous oxide (N_2O) and methane (CH_4)) emissions from two full-scale winery wastewater and sludge treatment systems (i.e. constructed wetlands (CWs) and activated sludge system) located in Galicia (Spain). GHG fluxes were measured using the static chamber method in combination with an on-site Fourier transform infrared spectroscopy (FTIR) gas analyser in the CWs system. These on-site innovative techniques proved to be very accurate and reliable. In the activated sludge treatment systems, the floating chamber method in combination with the FTIR gas analyser was used. Measurements were carried out during the vintage season, when winery wastewater has the highest flow and loads, and the rest of the year. Emission rates of CO_2, N_2O and CH_4 in the CWs units (i.e. vertical flow, horizontal subsurface flow and sludge treatment wetlands) ranged from 1.35E+02 to 7.54E+04, 1.70E-01 to 3.09E+01 and -3.05E+01 to 1.79E+03 mg m^{-2} day^{-1}, respectively. In the case of the activated sludge units (i.e. reactor, secondary settler and sludge storage tank) emission rates of CO_2, N_2O and CH_4 ranged from 1.56E+04 to 1.43E+05, 1.13E+01 to 4.75E+01 and 2.52E+01 to 1.01E+03 mg m^{-2} day^{-1}, respectively. Seasonally, daily and instantaneous variability in emissions as well as spatial variability was found. Comparing CWs with the activated sludge system, surface emission rates were lower in the CWs system in both seasons considered. Results highlighted that CWs are suitable technologies that can help to reduce GHG emissions associated with winery wastewater treatment.

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1. Introduction

Constructed wetlands (CWs) are a state of the art solution for wastewater and sludge (biosolids) treatment. Moreover, the application of these systems is becoming wider in the treatment of different wastewater including domestic, municipal, urban and agricultural drainage, landfill leachate, farming and fishing industry and many other industrial sectors (Vymazal, 2018). There is evidence from previous researches that CWs are a suitable solution for winery wastewater and sludge treatment (Flores et al., 2019; Serrano et al., 2011; Vymazal, 2014). Winery effluents have a huge variability of flows and organic loads throughout the year due to the seasonality of wine production, which is concentrated during the vintage season, about 20-30 days per year (Agustina et al., 2008; Flores et al., 2019; Masi et al., 2015). These strong changes in flows and loads make CWs a very suitable technology from the technical point of view due to their configuration in the form of fixed bed bioreactors.

However, sustainability of these systems is also an important factor beyond technical aspects to choose the most appropriate treatment technology for each specific case (Flores et al., 2020). Thus, it is important to quantify their environmental impacts and their greenhouse gases (GHG) emission rates, including carbon dioxide (CO_2), nitrous oxide (N_2O) and methane (CH_4). To our knowledge, GHG emissions from winery wastewater and sludge treatment have not been quantified yet and the amount of data on CWs in this specific sector is low in comparison to other sectors (e.g. municipal wastewater) (Mander et al., 2014).

72   The aim of this work was to quantify greenhouse gas emissions (CO_2, CH_4 and

73 N_2O) from a full-scale winery CWs system already in operation. Results were compared

74 with a conventional treatment system (activated sludge system) implemented in another

75 winery in which emissions were simultaneously measured. The methodology used in this

76 paper for gas emissions quantification is novel in the field of the CWs. Furthermore,

77 spatial as well as temporal (seasonally, daily and instantaneously) emissions were studied.

78 The emissions from the CWs system in this study were also compared to other emissions

79 from CWs found in the literature.

80   This research has been done in two wineries located in Galicia (Spain) and has

81 been carried out in the frame of the WETWINE project (http://wetwine.eu/en/), which

82 aims to promote environmentally friendly solutions to treat winery effluents in the South-

83 West of Europe (SUDOE Programme).

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85 **2. Materials and methods**

86 **2.1 Wastewater treatment plants description**

87 **2.1.1 Constructed wetlands system**

88   The CWs system is located in a winery in Galicia (Spain) and started operating in

89 July 2017. The winery produces around 368,000 L year^{-1} of white wine and has a

90 wastewater production of 1,400 m^3 per year. The wastewater treatment system (Figure 1)

91 consists of a hydrolytic upflow sludge blanket (HUSB) reactor of 1.5 m^3, followed by

92 two parallel vertical subsurface flow (VF) CWs (15 m^2 each), and a horizontal subsurface

93 flow (HSSF) CW (30 m^2). The excess sludge from the HUSB reactor is pumped to four

94 sludge treatment wetlands (STWs) of 5 m^2 each. Treated wastewater is discharged into

95 the municipal sewer system, while stabilized sludge is reused as a fertilizer or soil

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96   conditioner in the vineyards (Flores et al., 2019). CWs were planted with *Phragmites*

97   *australis* (common reed) in addition to some *Iris pseudacorus* in the STWs. The average

98   inflow to the CWs system was 1 m^3 day^{-1} during the vintage season and 2 m^3 day^{-1} during

99   the rest of the year. During the non-vintage season, wastewater going to the CWs system

100   was mostly coming from bottling and washing processes and had a low organic load.

101   When this wastewater was not enough for the treatment system, it was mixed with treated

102   wastewater recirculated from the outflow so as to ensure that there was a minimum flow

103   and cover the evapotranspiration process. The surface organic loading rate measured for

104   the VF CWs was in average 138 g COD m^{-2} day^{-1} during the vintage season and 27 g

105   COD m^{-2} day^{-1} during the rest of the year. In the case of the HSSF CW the average loading

106   rate measured was 51 and 15 g COD m^{-2} day^{-1} for the vintage season and the rest of the

107   year, respectively. The average sludge loading rate of the STWs was 3.15 kg DS m^{-2} year^{-1}

108   ^{1}. The average total organic rate entering the system was 5 kg COD day^{-1} during the

109   vintage season and 0.5 kg COD day^{-1} during the rest of the year. The average porosity of

110   the filter media was 40% in the VF CWs and 47% in the HSSF CW.

111   The operation of the system changed depending on the season. During the vintage

112   season, one VF CW was fed with pulses during 3 days, and then there was a resting period

113   of another 3 days while the other VF CW was being fed. So, as usual, the functioning of

114   the VF CWs was alternative. The STWs were fed once a week approximately and they

115   had also a resting period of at least one week. During the rest of the year, the feeding and

116   resting periods for the VF CWs were extended up to 7 days each, and the STWs were

117   only fed once due to the low sludge content in the wastewater.

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119   **2.1.2 Activated sludge system**

120 The activated sludge system is implemented in a winery also located in Galicia

121 (Spain) with a production of 4,832 m^3 of wastewater per year and a production of

122 3,850,000 L $year^{-1}$ of white and red wine. The system consists of a conventional pre-

123 treatment and a homogenization tank followed by an activated sludge reactor with

124 extended aeration (200 m^3) and a secondary settler (26 m^3). Treated wastewater is

125 discharged into the municipal sewage system and the sludge from the secondary settler is

126 stored in a tank (18 m^3), and then centrifuged and treated outside of the plant (Flores et

127 al., 2019). The aerated reactor, the secondary settler and the sludge tank were all open

128 tanks. The measured average loading rate of the plant was 430 g COD m^{-3} day^{-1} (86 kg

129 COD day^{-1}) during the vintage season and 100 g COD m^{-3} day^{-1} (20 kg COD day^{-1}) during

130 the rest of the year. Some chemicals such as sodium hydroxide, urea, phosphoric acid and

131 flocculant were used during the treatment for regulating pH, providing nutrients and

132 increase the sedimentation efficiency.

133

134 **2.2 Greenhouse gas emissions measurements**

135 The measurements of CO_2, CH_4 and N_2O fluxes were done using the static

136 chamber method for the CWs system (Chen et al., 1997; De la Varga et al., 2015; Rapson

137 and Dacres, 2014; Rolston et al., 1993; Uggetti et al., 2012) and the floating chamber

138 method for the activated sludge treatment plant (Chandran, 2010; Czepiel et al., 1995;

139 Hwang et al., 2016; Ribera-Guardia et al., 2019).

140 In the static chamber method, a closed PVC chamber of approximately 68 L (diameter:

141 39 – 48.5 cm, height: 45 cm) and a Fourier transform infrared spectroscopy (FTIR) gas

142 analyser (Gasmet DX4015) were used to collect and analyse the gas fluxes. In the

143   activated sludge system, a floating stainless steel gas collection hood (AC'SCENT® Flux

144   Hood, 40L) connected to the FTIR gas analyser was used.

145   The measuring range for the FTIR gas analyser was $0 – 2,000$ ppm for CO_2, $0 –$

146   100 ppm for CH_4 and $0 – 5$ ppm for N_2O. Moreover, as the FTIR gas analyser also

147   measured carbon monoxide (CO) and ammonia (NH_3) gas concentrations, they were also

148   considered in this study. Although CO and NH_3 are not GHG, they can be a potential

149   hazard in high concentrations.

150   Two sampling campaigns were conducted in 2018 considering the most important

151   seasons (activities) of the year in the wineries: vintage season (26 days during

152   August/September) and the rest of the year (33 days during February/March). The two

153   periods (i.e. vintage season and the rest of the year) selected for the campaigns were

154   considered representative in terms of wastewater characteristics and plants cycle. In fact,

155   the vintage season corresponded to the warmer months in which the plants are in a

156   growing phase. On the other hand, the rest of the year corresponded to the colder months

157   in which plants are in a translocating and dormant phase. Plant coverage was not fully

158   developed and was around 50% during the vintage season. During the rest of the year,

159   plant coverage was around 90% in the VFCWs and the STWs and 100% in the HSSF

160   CW. The sludge layer depth in the STW during these sampling campaigns was in average

161   5 cm.

162   In the CWs system, GHG emissions were measured in the following treatment

163   units: one of the two VF CWs, the HSSF CW and one of the STWs. To consider spatial

164   variability in the CWs, 2 or 3 points have been sampled in each wetland for a period of

165   time. The sampled points changed depending on the type of CW (Figure 2): for the VF

166   CW, 2 points next to the feeding zones and 1 far from these zones were selected; for the

167   HSSF CW, 3 points distributed along the wetland following the water path; and for the

168   STW, 1 point beside to the feeding zone and 1 point far from this zone. Measurements

169   were done during the whole day (daylight and night) to study the daily variability of the

170   emissions. Furthermore, in the case of the VF CW and the STW, feeding and resting

171   periods and in between feeding pulses periods were considered for measurements. In each

172   campaign, between 13 and 21 measurements were done in every unit depending on the

173   operation regime of each CW (e.g. in the VF CW and the STW more measurements were

174   done to consider feeding, resting and between feeding pulses periods). The chamber was

175   placed ensuring that air was confined inside it and isolated from the outside. The chamber

176   was buried 4.5 cm in the VF CW and 2.5 cm in the STW. In the case of the HSSF CW,

177   the chamber was buried 2.5 cm in order to reach the water surface. The chamber was also

178   covered with an isolating material (a thermal blanket made of polyethylene terephthalate

179   and aluminium) during the sunny days to protect it from the solar radiation and prevent

180   heating. The Teflon tube of the FTIR gas analyser was introduced through a septum into

181   the chamber for measurements (Figure 3). There was a second tube which returned the

182   sampled air into the chamber. In this way, the gas was accumulated inside the chamber

183   without any other mass exchange. At the same time, there were two fans working inside

184   the chamber so as to guarantee complete mixing and a thin tube (inner diameter of 0.3

185   cm) placed in the septum to prevent development of underpressure in the chamber. A

186   temperature probe (model 109 from Campbell Scientific) was installed inside the

187   chamber connected to a datalogger to record the temperature. Gas pressure inside the

188   chamber was also measured with the FTIR gas analyser. Gas measurements were taken

189   every minute and measurements in each sampling point ranged from 3 to 6 hours

190   depending on the intensity of the gas accumulation rate.

191   The volume of air contained inside the chamber (V_g) was obtained by geometric

192   calculations as follows:

193    $$V_g = \frac{\pi}{3} \cdot (H - H_b) \cdot (R'^2 + r^2 + R' \cdot r)$$ (1)

194   where H and H_b were respectively the total height and the buried height of the chamber,

195   R' was the inferior radius on the surface of the wetland and r the superior radius of the

196   chamber (Figure 3). The volume of the plants was not taken into account in calculations

197   as they were cut previously so as to install the chamber properly. De la Varga et al. (2015)

198   measured GHG emissions with and without plants and no significant variations were

199   found in the results.

200   In the activated sludge treatment plant, GHG emissions were measured in

201   different points in the aerated reactor, the secondary settler and the sludge storage tank.

202   The Teflon tube of the FTIR gas analyser was also introduced inside the chamber through

203   a septum for gas measurements. In this system the gas was not accumulated inside the

204   chamber, so there was no need of a returning tube. Temperature inside the chamber and

205   air flow were also measured. Gas measurements were taken every minute and the period

206   for measurements in each point depended on the working hours of the winery. Gas

207   measurements in this system were done during three consecutive days in each campaign.

208   In the case of the CWs system, emission rates were calculated from the slope

209   obtained from the linear increase of the gas concentration inside the chamber during each

210   measurement. Measurements recorded from the FTIR gas analyser were in ppm (mL m^{-}

211   ^{3}). For this reason, to calculate the surface emission rate (SER) of each gas in mg m^{-2} day^{-}

212   ^{1} the Ideal Gas Law was adapted to convert volume units (mL) into mass units (mg):

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214    $$SER \ (\mathrm{mg \ m^{-2} \ day^{-1}}) = \frac{\text{slope}}{S_{chamber}} \cdot \frac{V_g \cdot P \cdot m_m}{R \cdot (T_a + 273.15)} \cdot 1.44$$ (2)

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where "slope" is the coefficient of the equation obtained from the lineal regression

analysis of the corresponding gas against time (ppm min^{-1}), V_g is the volume of gas inside

the chamber (m^3), P is the pressure inside the chamber (bar), R is the ideal gases constant

($8.314 \cdot 10^{-5}$ bar m^3 mol^{-1} K^{-1}), $S_{chamber}$ is the collection surface (m^2), T_a is the average

temperature inside the chamber (ºC), m_m is the molar mass (CH$_4$: 16 g mol^{-1}, CO$_2$: 44 g

mol^{-1}, N$_2$O: 44 g mol^{-1}) and 1.44 is a unit conversion factor.

To calculate the SER from the activated sludge system, the following equation

was applied (Chandran, 2010), where also volume units (mL) can be converted into mass

units (mg):

$$SER \ (\text{mg m}^{-2} \ \text{day}^{-1}) = \frac{Q_{emission} \cdot C}{S_{chamber}} \cdot \frac{P \cdot m_m}{R \cdot (T_a + 273.15)} \cdot 10^{-6} \tag{3}$$

where $Q_{emission}$ is the gas flux (L day^{-1}), C is the gas concentration inside the chamber

(ppm$_v$), P is the atmospheric pressure (bar), R is the ideal gases constant ($8.314 \cdot 10^{-5}$ bar

m^3 mol^{-1} K^{-1}), $S_{chamber}$ is the collection surface (m^2), T_a is the average temperature (ºC),

m_m is the molar mass (CH$_4$: 16 g mol^{-1}, CO$_2$: 44 g mol^{-1}, N$_2$O: 44 g mol^{-1}) and 10^{-6} is a

unit conversion factor.

The SER was calculated from average values from the emissions measured in

different sampling points and in different temporal scales. Results with a low coefficient

of determination ($R^2 < 0.8$) were not considered. All the results are expressed with three

significant numbers.

Moreover, water flow from the two treatment systems was also recorded in order

to study the relationship between hydraulics and GHG emissions. The volumetric method

239   was used to estimate the flow after a feeding pulse in the outlet of the VF CW and the

240   HSSF CW. In the case of the activated sludge system, there was an automatic flowmeter

241   at the outlet of the plant.

242

243   **3. Results and Discussion**

244   The static chamber method in combination with the FTIR gas analyser proved to

245   be a suitable tool to study emissions (fluxes) from the system. There was a remarkable

246   linearity between the gas concentration inside the chamber and time (Figure 4). This study

247   is the first in which the FTIR on-site methodology is used for the measurement of GHG

248   emissions in CWs. With this technique errors due to sample transportation to the

249   laboratory and sample manipulation are highly minimised. Also, instantaneously

250   information about multiple different gases can be directly obtained on-site. However, less

251   measurements can be done at the same time, but they are of a greater quality.

252   Average surface emission rates in the vintage season and in the rest of the year are

253   shown in Tables 1 and 2, respectively. Note that emission rates are expressed in mass per

254   surface area ($mg\ m^{-2}\ day^{-1}$) as well as in mass per flow treated ($g\ m^{-3}$). Spatial as well as

255   temporal variability in emissions among the same wetland unit were detected. Global

256   emissions of CO_2 in the VF CW ranged from 5.83E+02 to 7.54E+04 $mg\ CO_2\ m^{-2}\ day^{-1}$,

257   with higher average values during the vintage season. Similar average values have been

258   reported in the VF CWs from the Kõo system treating municipal wastewater in Estonia

259   (Søvik et al., 2006). The average CO_2 recorded in the vintage season was approximately

260   two times the values obtained from other VF CWs treating municipal wastewater found

261   in literature (Mander et al., 2014). As complete operation cycles were taken into account,

262   it is worth mentioning that there were differences greater than 2 times in CO_2 emissions

263 during feeding and resting periods, with average values of 4.24E+04 and 2.06E+04 mg

264 CO_2 m^{-2} day $^{-1}$, respectively during the vintage season, and 5.89E+03 and 2.00E+03,

265 respectively during the rest of the year.

266   The range of N_2O emissions from the VF CW was from 1.70E-01 to 3.09E+01

267 mg N_2O m^{-2} day $^{-1}$, with also higher average values during the vintage season. These

268 results were within the range of a review study on VF CWs considering urban wastewater

269 (Mander et al., 2014) and in a lower range than the values calculated in other studies on

270 VF CWs treating urban wastewater (Filali et al., 2017; Søvik et al., 2006). N_2O emissions

271 during feeding and resting periods in the VF CW were 5.10E+00 and 1.28E+01 mg N_2O

272 m^{-2} day $^{-1}$ during the vintage season, respectively and 8.82E-01 and 2.54E-01 during the

273 rest of the year, respectively. During high organic loading rates (i.e. vintage season), N_2O

274 emissions were higher during resting periods such as observed in other studies of CWs

275 treating municipal wastewater and sludge (Filali et al., 2017; Uggetti et al., 2012).

276   CH_4 emissions were also detected and ranged from 6.95E-01 to 5.25E+02 mg CH_4

277 m^{-2} day $^{-1}$, with average values very similar around the year. Higher values were obtained

278 in this study in comparison with previous ones, although the average values from the

279 present study were very similar (Mander et al., 2014; Søvik et al., 2006). During feeding

280 periods, CH_4 emissions were higher (average values of 1.37E+02 and 1.60E+02 mg CH_4

281 m^{-2} day $^{-1}$ during the vintage season and the rest of the year, respectively) in comparison

282 with resting periods, where CH_4 emissions were almost negligible (average values of

283 1.31E+00 and 2.29E+00 mg CH_4 m^{-2} day $^{-1}$ during the vintage season and the rest of the

284 year, respectively). Previous studies on VF CWs treating domestic wastewaters found

285 that CH_4 emissions were negligible (Pan et al., 2011; Wang et al., 2013; Yan et al., 2012).

286 We have not a direct evidence on the reasons behind detected CH_4 emissions from the

VF CW, however, they could be the result of the synergistic effect of concomitant factors. They could be due to the fact that (i) wastewater coming from the HUSB reactor had anaerobic conditions and when reached the VF CW, CH_4 was released to the atmosphere and/or, (ii) there were anaerobic microsites in the VF CW. CH_4 emissions were quite constant, but when there was a feeding pulse, emissions increased up to 40 times the next 20 minutes after the pulse (Figure 5). This trend suggests that release after wastewater load could be a very important factor on CH_4 emissions. There was a clear daily variability in the VF CW: during the morning and early afternoon emissions increased and during the evening and night emissions decreased up to 2 times (average 1.6 times, Figure 6). This tendency was observed during the whole year and not depended on feeding-resting periods. No significant spatial variations were found in the VF CW, which means that wastewater was homogeneously distributed along the wetland surface (Filali et al., 2017).

In the case of the HSSF CW, the range of CO_2 and CH_4 emissions varied from 1.35E+02 to 8.90E+03 and from 4.41E+00 to 1.79E+03 mg m^{-2} day $^{-1}$, respectively, during the vintage season. During the rest of the year CO_2 and CH_4 emissions varied from 2.46E+02 to 2.25E+03 and -3.05E+01 to 3.74E+01 mg m^{-2} day $^{-1}$, respectively. Negative emission values reflected that there is absorption instead of emission. There were no emissions of N_2O in the HSSF CW as they were not accumulated inside the chamber (R^2<0.2, Figure 7). Average CO_2 and CH_4 results were in accordance with previous studies on HSSF CWs for urban wastewater treatment. On the other hand, the lack of N_2O emissions was not in agreement with previous studies (Corbella and Puigagut, 2014; De la Varga et al., 2015; Mander et al., 2014; Søvik et al., 2006). The main reason why there were not emissions of N_2O in the HSSF CW is because winery wastewaters have a very

311 low content of nitrogen and phosphorous in comparison with domestic wastewater and

312 was mostly eliminated in the VF CW (Arienzo et al., 2009; Flores et al., 2019). CO_2 and

313 CH_4 emissions also had a daily variability in the HSSF CW. Emissions increased from

314 the morning until the early afternoon, when a peak was found and then emissions

315 decreased from the afternoon and during the night (Figure 6). However, a previous study

316 reported that there was no significant daily variation in CH_4 emissions (De la Varga et

317 al., 2015). CH_4 emissions were found higher near the inlet of the HSSF CW and decreased

318 along the wetland, as has also been reported previously (De la Varga et al., 2015; Søvik

319 et al., 2006; Teiter and Mander, 2005). During the vintage season, in average, CH_4

320 emissions were 8 times higher at the inlet than in the outlet zone. The higher CH_4

321 emissions near the inlet is likely related to higher substrate concentrations and organic

322 load (Corbella and Puigagut, 2014). CO_2 emissions were maintained with similar values

323 across the entire surface of the HSSF CW which was not in accordance with other studies

324 considering HSSF CWs treating urban wastewater (Søvik et al., 2006; Teiter and Mander,

325 2005).

326 In the STW during the vintage season the range of CO_2 emissions was from

327 2.05E+03 to 7.39E+04 mg CO_2 m^{-2} day $^{-1}$, N_2O emissions varied from 1.90E-01 to

328 2.56E+01 mg N_2O m^{-2} day $^{-1}$ and CH_4 emissions ranged from 7.07E-01 to >5.00E+02 mg

329 CH_4 m^{-2} day $^{-1}$. During feeding events CO_2, N_2O and CH_4 emissions increased with

330 average values of 3.32E+04, 1.16E+01 and 3.32E+02 mg m^{-2} day $^{-1}$, respectively. After

331 feeding, CO_2, N_2O and CH_4 emissions decreased progressively with average values

332 during resting periods of 9.31E+03, 6.55E+00 and 8.73E+00 mg m^{-2} day $^{-1}$. This

333 behaviour was also observed in another study which considered STW treating sludge

334 from urban wastewater (Uggetti et al., 2012); however, GHG emissions (i.e. N_2O and

335   CH_4) were in a higher range than those obtained in the present study. During the rest of

336   the year, CO_2, N_2O and CH_4 emissions were negligible, due to the fact that sludge

337   produced and, hence, the sludge fed to the STW was minimal. Note that during the rest

338   of the year wineries mainly produce very lightly loaded wastewater from the bottling and

339   washing processes. Unlike the other CWs, there was no evidence of daily variability in

340   emissions in the STW.

341   During the two campaigns, CO and NH_3 emissions were also measured in the

342   CWs (VF, HSSF and STW) but they were negligible or inexistent. Small traces of CO

343   and NH_3 were detected, but they were not accumulated inside the chamber (low R^2).

344   Overall, the highest SER of CO_2 and N_2O were found in the VF CW, followed by

345   the STW and then the HSSF. However, the HSSF CW had the highest SER of CH_4 (only

346   during the vintage season).

347   In the aerated reactor of the activated sludge system, CO_2 emissions were in the

348   range of 5.47E+04 to 1.43E+05 mg CO_2 m^{-2} day $^{-1}$. The large amount of CO_2 emissions

349   produced was due to the respiration of organic matter in the reactor for the biodegradation

350   processes (Daelman et al., 2012). Emissions of N_2O and CH_4 in the reactor ranged from

351   1.73E+01 to 4.75E+01 and from 3.17E+01 to 1.28E+02 mg m^{-2} day^{-1} respectively. The

352   presence of CH_4 emissions was probably due to the existence of anaerobic microsites

353   inside the activated sludge flocs. There were also emissions of CO and NH_3 in the reactor.

354   CO ranged from 0.00E+00 to 6.59E+01 mg CO m^{-2} day $^{-1}$, with average values of 2.57E-

355   01 and 6.13E+01 mg CO m^{-2} day $^{-1}$ during the vintage season and the rest of the year,

356   respectively. NH_3 ranged from 0.00E+00 to 5.18E+01 mg NH_3 m^{-2} day $^{-1}$ with average

357   values of 2.55E+00 and 4.87E+01 mg NH_3 m^{-2} day $^{-1}$ during the vintage season and the

358   rest of the year, respectively. As mentioned above, winery wastewater has a low content

359   of nitrogen and phosphorous, so urea and phosphoric acid are used along with other

360   chemicals for maintaining organic degradation by bacteria. For this reason, N_2O and NH_3

361   were emitted.

362   CO_2 emissions from the secondary settler ranged from 1.96E+04 to 3.91E+04 mg

363   CO_2 m^{-2} day $^{-1}$. N_2O emissions in the secondary settler ranged from 1.40E+01 to

364   2.60E+01 mg N_2O m^{-2} day $^{-1}$. CH_4 was also present in the secondary settler, with values

365   ranging from 2.52E+01 to 5.76E+02 mg CH_4 m^{-2} day $^{-1}$. Low emissions of NH_3 were

366   detected (ranging from 0.00E+00 to 8.00E+00 mg NH_3 m^{-2} day $^{-1}$, average value of

367   2.27E+00 and 1.80E+00 mg NH_3 m^{-2} day $^{-1}$ during the vintage season and the rest of the

368   year, respectively). CO emissions were not detected during measurements in the

369   secondary settler.

370   Emissions from the sludge storage tank in the activated sludge system were also

371   measured. The range of CO_2 was 1.56E+04 – 5.06E+04 mg CO_2 m^{-2} day $^{-1}$. N_2O

372   emissions ranged from 1.13E+01 to 2.32E+01 mg N_2O m^{-2} day $^{-1}$. There was a high

373   concentration of CH_4 emissions in the sludge storage tank (ranging between 1.32E+02

374   and 1.01E+03 mg CH_4 m^{-2} day $^{-1}$) due to the fermentation of the accumulated sludge

375   stored during several days without any aeration. NH_3 fluxes ranged from 0.00E+00 to

376   1.02E+01 mg NH_3 m^{-2} day $^{-1}$, with an average value of 3.38E+00 mg NH_3 m^{-2} day $^{-1}$

377   during the vintage season and 4.79E-01 mg NH_3 m^{-2} day $^{-1}$ during the rest of the year.

378   There was also no presence of CO emissions in the sludge storage tank.

379   Overall, the highest emission rates of CO_2 and N_2O were found in the aerated

380   reactor, followed by the secondary settler and the sludge tank with similar values.

381   However, the sludge tank had the highest emissions rates of CH_4, followed by the

382   secondary settler during the vintage season.

383   Total emission rates per m^3 of treated water of CO_2, N_2O and CH_4 in the CWs

384   system were 17, 58 and 6 times higher during the vintage season than the rest of the year,

385   respectively. In the case of the activated sludge system, total emission rates per m^3 of

386   treated water of CO_2, N_2O and CH_4 were 0.8, 1 and 2 times higher during the vintage

387   season than the rest of the year, respectively (Tables 1 and 2).

388   To sum up, SER were lower in the CWs system than in the activated sludge system

389   in both seasons considered (Figure 8). During the vintage season, total SER of CO_2, N_2O

390   and CH_4 were 1.3, 1.8 and 2 times lower in the CWs system than in the activated sludge

391   system, respectively. During the rest of the year, SER of CO_2, N_2O and CH_4 were 12, 34

392   and 1.6 times lower in the CWs system than in the activated sludge system, respectively.

393   Emission rates per m^3 of treated water were higher in the CWs system than in the activated

394   sludge system. However, Flores et al., 2020 found that the activated sludge system

395   contributed the most to global GHG emissions due to indirect emissions from energy and

396   chemical consumption during the operation of the plants and transportation, which are

397   out of the scope of the present study.

398   Furthermore, GHG emission rates associated with sludge treatment were between

399   1 and 16,300 times higher in the sludge tank of the activated sludge system than in the

400   STWs. To reduce GHG emissions from sludge treatment, a suitable solution could be to

401   implement STWs in those wineries already operating with activated sludge systems

402   (Flores et al., 2019).

403   Additionally, further studies should be carried out to improve wastewater quality

404   entering the CWs in order to reduce GHG emissions in the VF CWs. For instance, a

405   different pre-treatment might be studied so as to avoid anaerobic conditions in the primary

406   treatment reactor.

Finally, the methodology used for the measurement of GHG emissions from CWs using the static chamber in combination with the on-site FTIR gas analyser resulted to be very accurate and reliable in comparison with other techniques (e.g. gas sampling through syringes and off-site laboratory analysis). This method allowed to obtain good quality data as well as to register instantaneous changes on GHG emissions (i.e. spontaneous high or low emission peaks) that other methods (e.g. punctual sampling through syringes) do not achieve.

4. Conclusions

This study quantified and compared CO_2, N_2O and CH_4 emissions from two full-scale winery wastewater treatment systems (e.g. CWs and activated sludge systems). The novel methodology used with the static chamber in combination with the FTIR gas analyser allowed to study spatial and temporal (seasonally, daily and instantaneously) variability in GHG emissions in the CWs system. Emission rates resulted to be higher in the activated sludge system than in CWs. Emission rates of CO_2, N_2O and CH_4 in the CWs units (i.e. VF, HSSF and STW) ranged from 1.35E+02 to 7.54E+04, 1.70E-01 to 3.09E+01 and -3.05E+01 to 1.79E+03 mg m^{-2} day^{-1}, respectively. In the case of the activated sludge units (i.e. reactor, secondary settler and sludge storage tank) emission rates of CO_2, N_2O and CH_4 ranged from 1.56E+04 to 1.43E+05, 1.13E+01 to 4.75E+01 and 2.52E+01 to 1.01E+03 mg m^{-2} day^{-1}, respectively. These results demonstrated that the implementation of CWs can be as competitive as conventional technologies (i.e. activated sludge) for winery wastewater and sludge treatment, providing a sustainable solution for waste management in the wine sector.

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534   **Table 1.** Emission rates results of carbon dioxide (CO_2), nitrous oxide (N_2O) and methane (CH_4) in the constructed wetlands (CWs) and

535   the activated sludge system during the vintage season. Results are presented as surface emission rates (SER) and emission rate per m^3 of

536 treated water. Except for the total values, the rest of the values correspond to one treatment unit. VF – vertical flow CW, HSSF – horizontal

537   subsurface flow CW, STW – sludge treatment wetland.

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System	CO_2		N_2O		CH_4	
	SER (mean $\pm$ S.D.)	Emission rate per m^3 of treated water	SER (mean $\pm$ S.D.)	Emission rate per m^3 of treated water	SER (mean $\pm$ S.D.)	Emission rate per m^3 of treated water
	(mg CO_2 m^{-2} day^{-1})	(g m^{-3})	(mg N_2O m^{-2} day^{-1})	(g m^{-3})	(mg CH_4 m^{-2} day^{-1})	(g m^{-3})
VF	3.36E+04 $\pm$ 1.79E+04	7.87E+02	7.83E+00 $\pm$ 8.20E+00	2.24E-01	9.74E+01 $\pm$ 1.37E+02	1.73E+00
HSSF	3.65E+03 $\pm$ 2.68E+03	7.65E+01	0.00E+00 $\pm$ 0.00E+00	0.00E+00	3.52E+02 $\pm$ 4.85E+02	7.38E+00
STW	1.45E+04 $\pm$ 1.90E+04	1.19E+02	7.48E+00 $\pm$ 7.50E+00	5.80E-02	1.86E+01 $\pm$ 3.72E+01	1.44E-01
Total CWs	**1.29E+05**	**2.13E+03**	**4.56E+01**	**6.78E-01**	**6.21E+02**	**1.14E+01**
Reactor	8.68E+04 $\pm$ 2.87E+04	2.17E+02	2.84E+01 $\pm$ 8.38E+00	7.11E-02	5.48E+01 $\pm$ 1.17E+01	1.37E-01
Secondary Settler	3.70E+04 $\pm$ 3.38E+03	1.20E+01	2.38E+01 $\pm$ 2.22E+00	7.72E-03	3.73E+02 $\pm$ 1.77E+02	1.21E-01
Sludge Storage tank	4.54E+04 $\pm$ 1.01E+04	1.03E+01	2.88E+01 $\pm$ 3.97E+00	6.55E-03	8.32E+02 $\pm$ 4.17E+02	1.89E-01
Total Activated Sludge	**1.69E+05**	**2.39E+02**	**8.10E+01**	**8.53E-02**	**1.26E+03**	**4.48E-01**

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550 **Table 2.** Emission rates results of carbon dioxide (CO_2), nitrous oxide (N_2O) and methane (CH_4) in the constructed wetlands (CWs) and
551 the activated sludge system during the rest of the year. Results are presented as surface emission rates (SER) and emission rate per m^3 of
552 treated water. Except for the total values, the rest of the values correspond to one treatment unit. VF – vertical flow CW, HSSF – horizontal
553 subsurface flow CW, STW – sludge treatment wetland.

554

| System	CO_2		N_2O		CH_4	
	SER (mean ± S.D.)	Emission rate per m^3 of treated water	SER (mean ± S.D.)	Emission rate per m^3 of treated water	SER (mean ± S.D.)	Emission rate per m^3 of treated water
	(mg CO_2 m^{-2} day^{-1})	(g m^{-3})	(mg N_2O m^{-2} day^{-1})	(g m^{-3})	(mg CH_4 m^{-2} day^{-1})	(g m^{-3})
VF	4.41E+03 ± 4.78E+03	3.68E+01	7.140E-01 ± 6.72E-01	5.29E-03	1.07E+02 ± 1.30E+02	7.54E-01
HSSF	1.08E+03 ± 7.80E+02	5.41E+01	0.00E+00 ± 0.00E+00	0.00E+00	9.21E+00 ± 2.19E+01	4.60E-01
STW	0.00E+00 ± 0.00E+00	0.00E+00	0.00E+00 ± 0.00E+00	0.00E+00	0.00E+00 ± 0.00E+00	0.00E+00
Total CWs	**9.90E+03**	**1.28E+02**	**1.43E+00**	**1.16E-02**	**2.24E+02**	**1.97E+00**
Reactor	8.48E+04 ± 4.29E+03	2.70E+02	2.11E+01 ± 2.16E+00	6.72E-02	3.60E+01 ± 1.22E+00	1.15E-01
Secondary Settler	2.00E+04 ± 5.52E+02	7.80E+00	1.49E+01 ± 1.12E+00	5.82E-03	2.76E+01 ± 2.27E+00	1.08E-02
Sludge Storage tank	1.63E+04 ± 9.33E+02	4.45E+00	1.19E+01 ± 6.80E-01	3.26E-03	3.02E+02 ± 2.56E+02	8.25E-02
Total Activated Sludge	**1.21E+05**	**2.82E+02**	**4.80E+01**	**7.63E-02**	**3.66E+02**	**2.08E-01**

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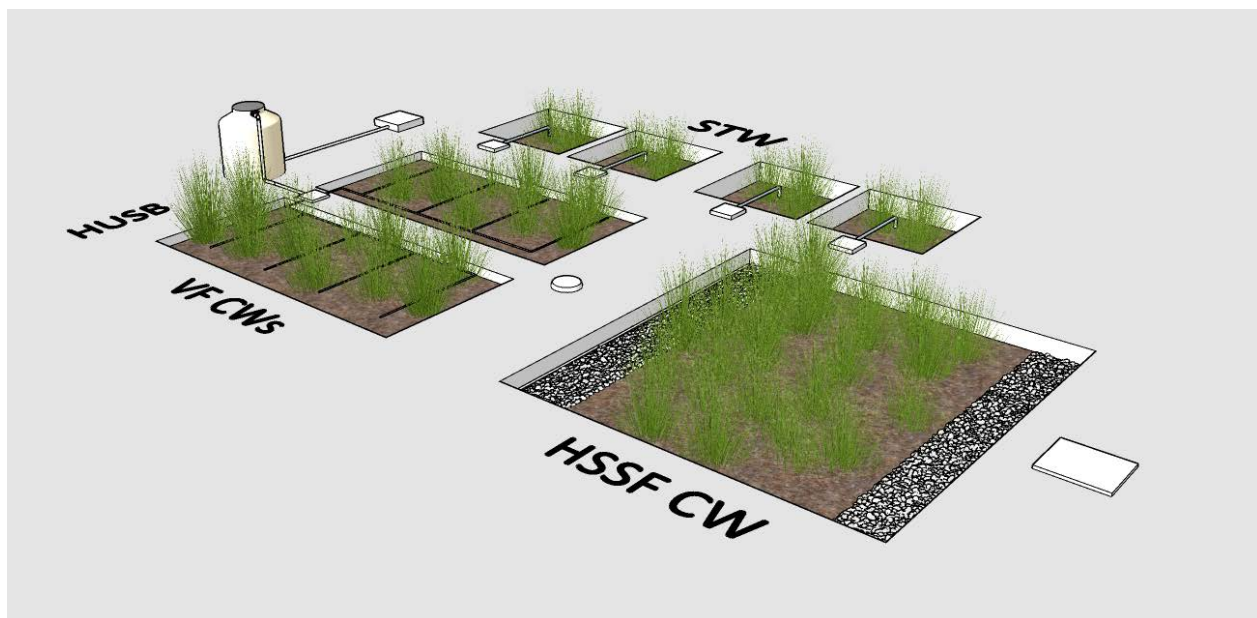
Figure 1. 3D representation of the constructed wetlands (CWs) system in the winery located in Galicia (Spain). HUSB – hydrolytic upflow sludge blanket reactor, VF – vertical flow CWs, HSSF – horizontal subsurface flow CW, STW – sludge treatment wetlands.
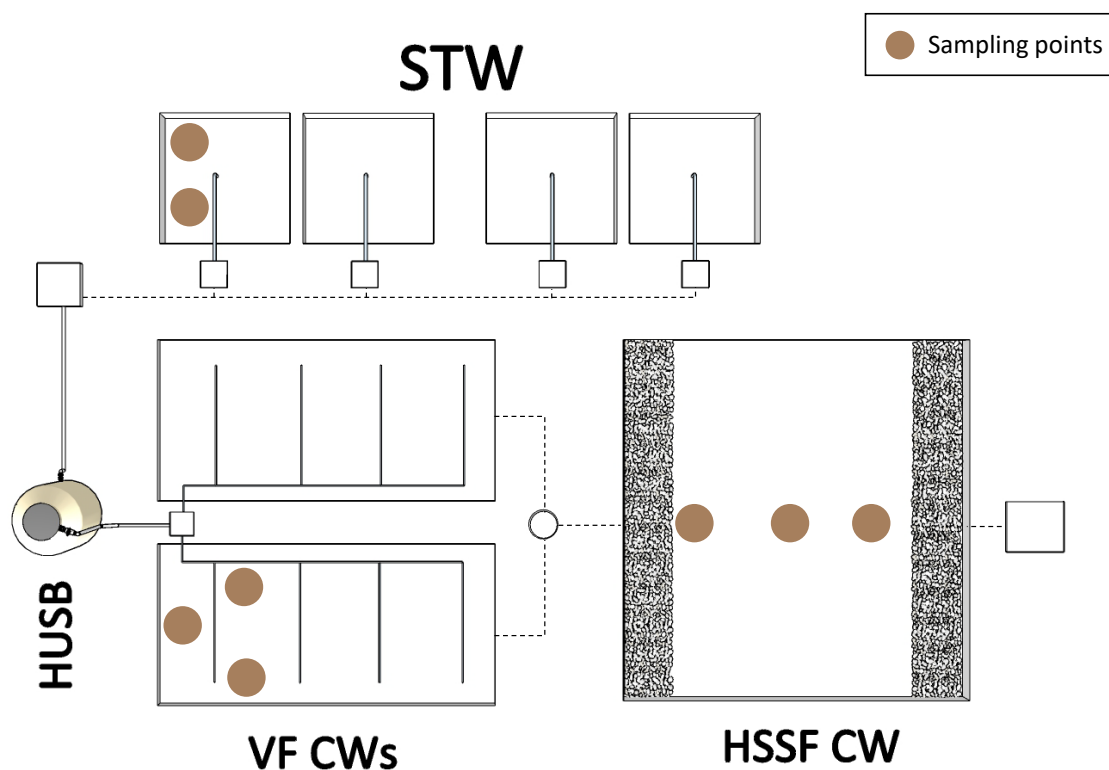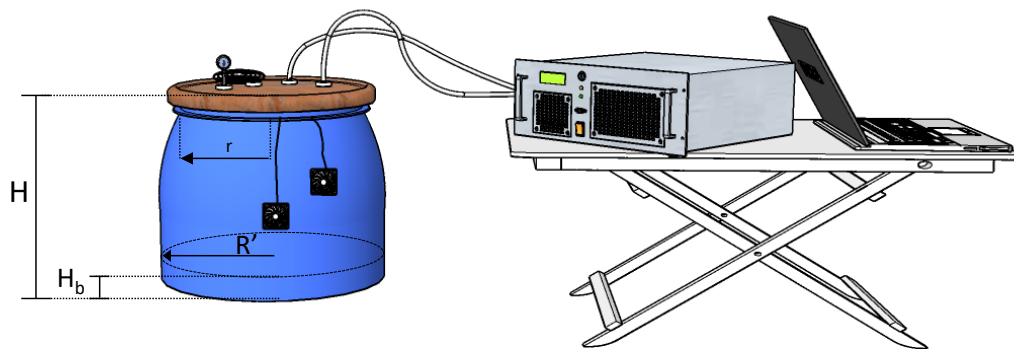
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Figure 2. Plan view of the constructed wetlands (CWs) system pointing at sampling

570 points of the greenhouse gas emissions measurements. HUSB – hydrolytic upflow sludge

571 blanket reactor, VF – vertical flow CWs, HSSF – horizontal subsurface flow CW, STW

572 – sludge treatment wetlands.

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576 **Figure 3**. Set up for greenhouse gas measurements using the static chamber method with

577 an on-site Fourier transform infrared spectroscopy gas analyser in the constructed

578 wetlands system.

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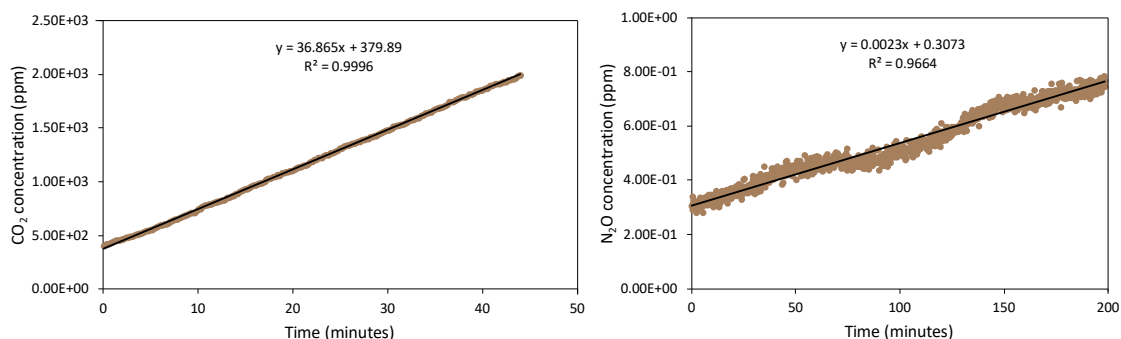
581 **Figure 4**. Examples of the linear regression of the gas concentration (CO_2 in the left and

582 N_2O in the right) inside the chamber versus time. R^2 is the coefficient of determination.

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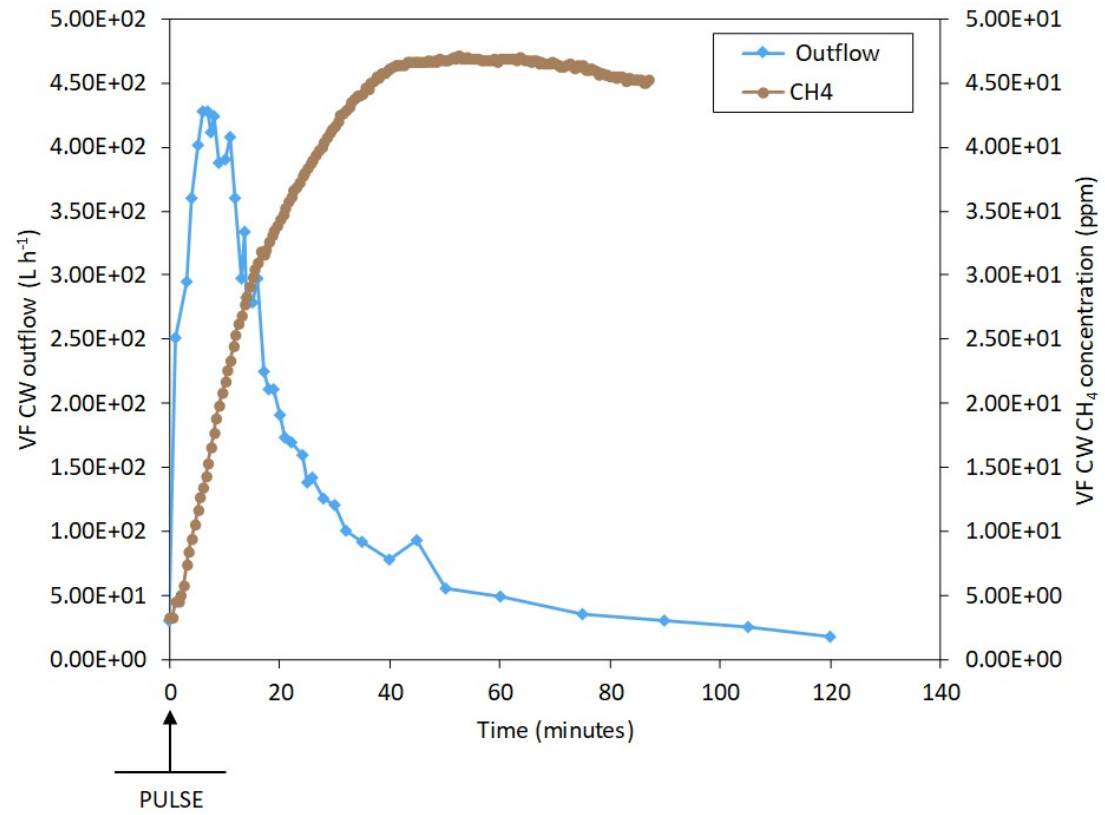
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586 **Figure 5.** Relation between the outflow and CH4 concentration after a feeding pulse in
587 the vertical flow constructed wetland (VF CW).
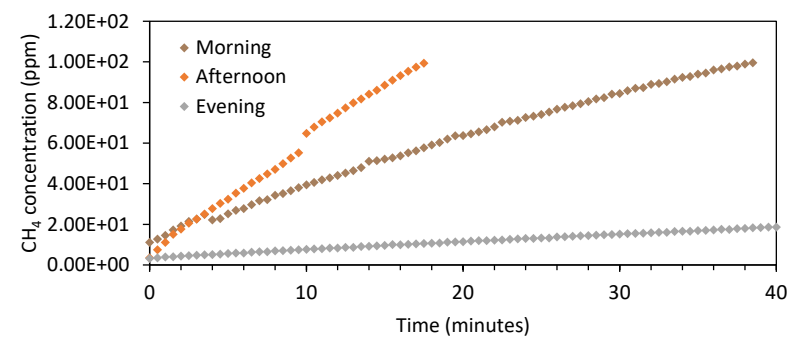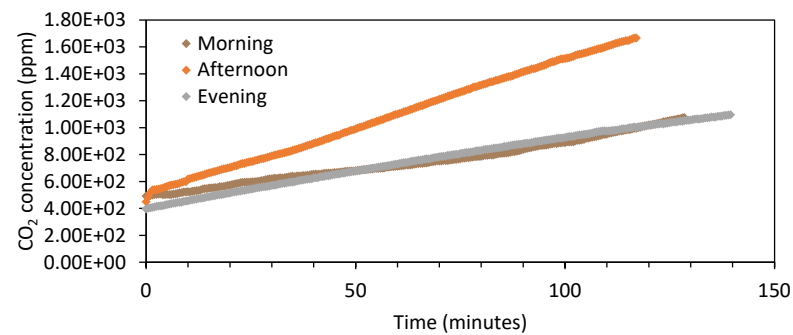
a) VF
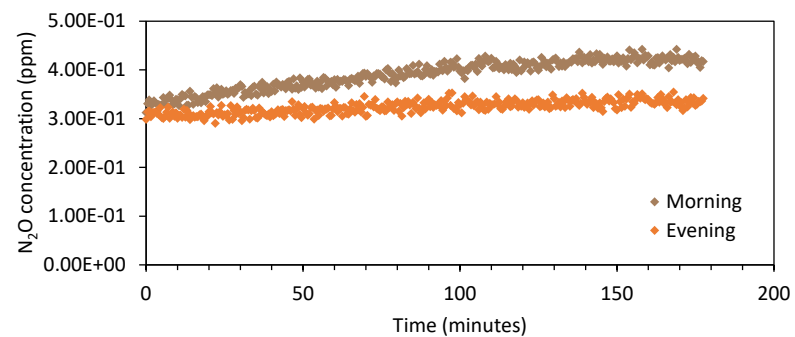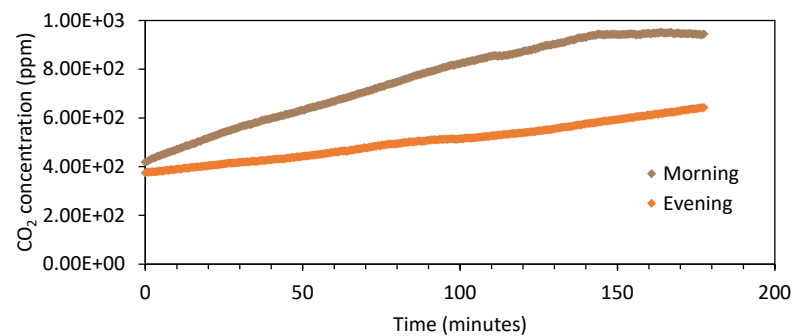
b) HSSF

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590 **Figure 6.** Daily variability of greenhouse gas emissions in the vertical flow constructed wetland (VF, left column) and in the horizontal
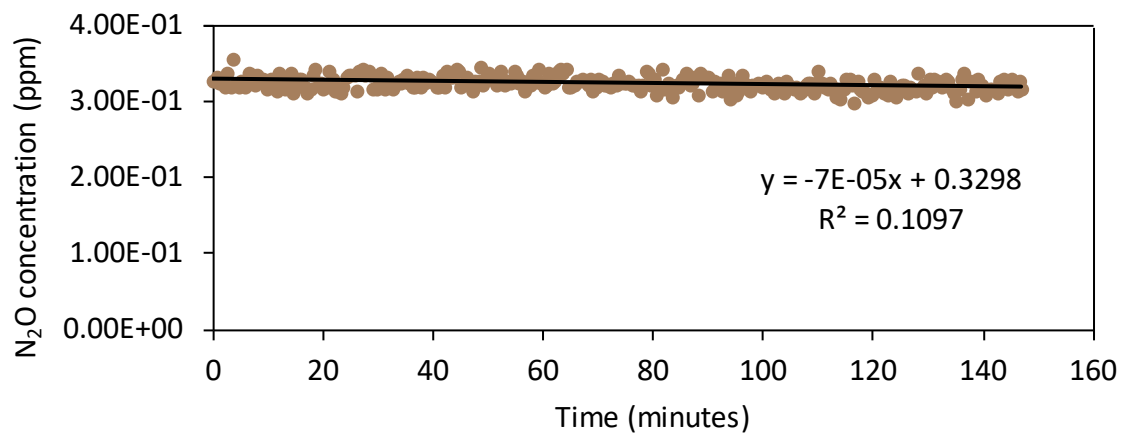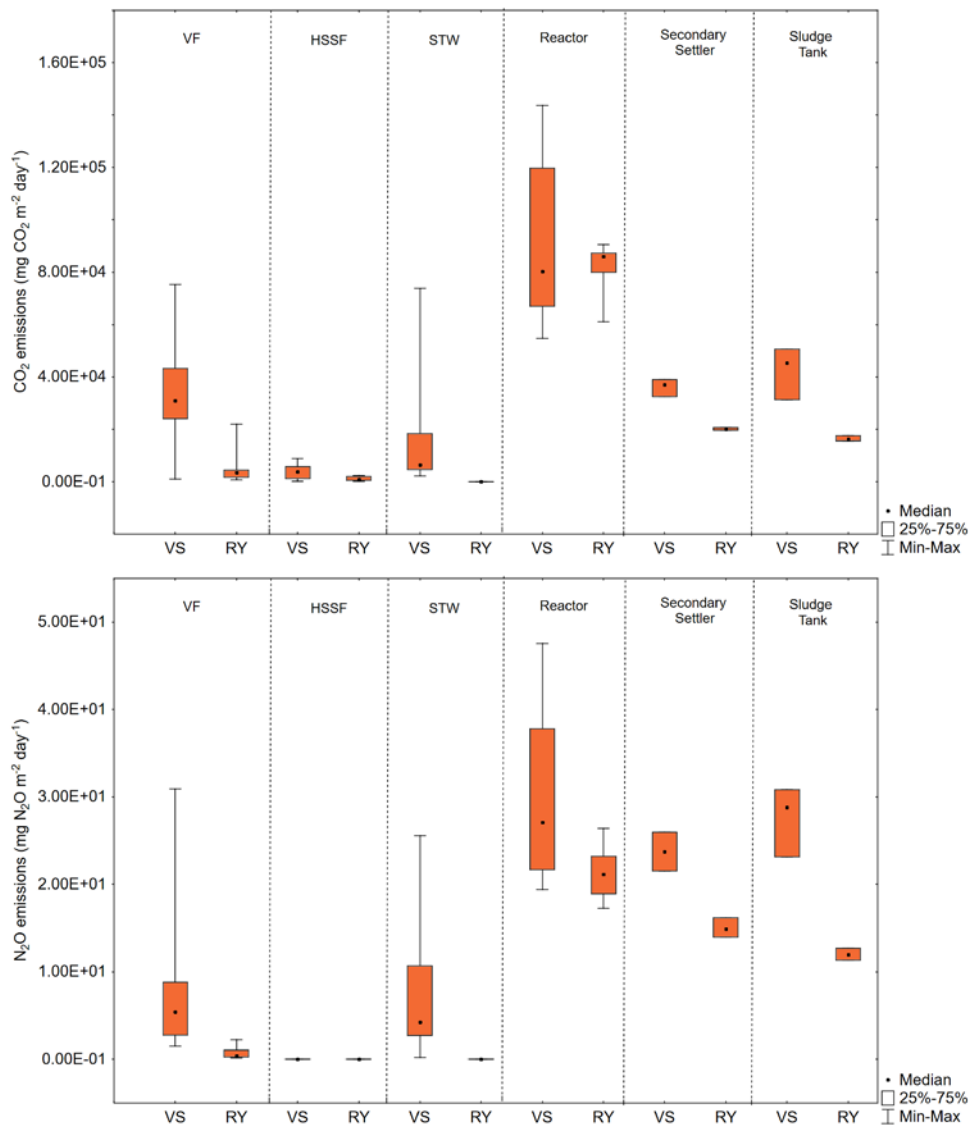591 subsurface flow constructed wetland (HSSF, right column).

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Figure 7. N_2O concentration inside the chamber in the horizontal subsurface flow constructed wetland versus time. R^2 is the coefficient of determination.
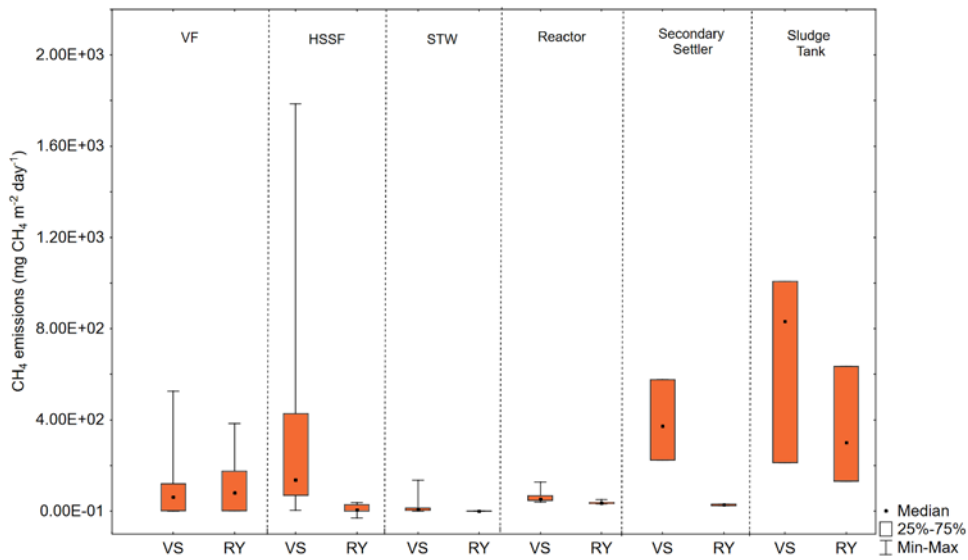
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602 **Figure 8**. Median, 25% and 75% quartile and min/max values of measured emissions of
603 carbon dioxide (CO_2, upper plot), nitrous oxide (N_2O, middle plot) and methane (CH_4,
604 lower plot) in the constructed wetlands and activated sludge systems during the vintage
605 season (VS) and the rest of the year (RY). The values shown in the graphs correspond to
606 one unit of the treatment system. VF – vertical flow CW, HSSF – horizontal subsurface
607 flow CW, STW – sludge treatment wetland.