

1 **Intermittent aeration to improve wastewater treatment**
2 **efficiency in pilot-scale constructed wetland**

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21

22 **Abstract**

23 Forced aeration of horizontal subsurface flow constructed wetlands (HSSF CWs) is
24 nowadays a recognized method to improve treatment efficiency, mainly in terms of
25 ammonium removal. While numerous investigations have been reported testing constant
26 aeration, scarce information can be found about the efficiency of intermittent aeration.
27 This study aims at comparing continuous and intermittent aeration, establishing if there
28 is an optimal regime that will increase treatment efficiency of HSSF CWs whilst
29 minimizing the energy requirement. Full and intermittent aeration were tested in a pilot
30 plant of three HSSF CWs (2.64 m² each) fed with primary treated wastewater. One unit
31 was fully aerated; one intermittently aerated (i.e. by setting a limit of 0.5 mg/L dissolved
32 oxygen within the bed) with the remaining unit not aerated as a control. Results indicated
33 that intermittent aeration was the most successful operating method. Indeed, the
34 coexistence of aerobic and anoxic conditions promoted by the intermittent aeration
35 resulted in the highest COD (66%), ammonium (99%) and total nitrogen (79%) removals.
36 On the other hand, continuous aeration promotes ammonium removal (99%), but resulted
37 in nitrate concentrations in the effluent of up to 27 mg/L. This study demonstrates the
38 high potential of the intermittent aeration to increase wastewater treatment efficiency of
39 CWs providing an extreme benefit in terms of the energy consumption.

40 **Keywords:** Intermittent aeration, Ammonium removal, Nitrification/Denitrification,
41 Constructed Wetlands, Horizontal sub-surface flow.
42

43 **Introduction**

44 Constructed wetlands (CWs) have been widely used in the last few decades (Vymazal,
45 2011), showing worthy efficiency in the treatment of urban wastewater, mine water,
46 landfill leachate, industrial effluents, air-strip runoff and road runoff (Kadlec and
47 Wallace, 2009). A favorable performance in terms of organic matter and ammonium
48 removal, together with the low energy requirements, a minimal maintenance requirement
49 and low operational costs are among the reasons for the wide spread implementation of
50 the technology all over the world (García et al., 2010). Moreover, the important role of
51 CWs as greenspace and wildlife habitat make them an appropriate alternative to
52 conventional wastewater treatment, mainly in wild and isolated small communities.

53 Subsurface oxygen limitation has been identified amongst the main factors compromising
54 contaminant removal in horizontal subsurface flow constructed wetlands (HSSF CWs)
55 (Brix and Schierup, 1990). Such systems promote the co-existence of different redox
56 statuses, these strongly affect the relative importance of the biochemical pathways for
57 organic matter and nutrient removal (García et al., 2004).

58 Forced or active aeration, originally developed by Wallace (2001), has received
59 increasing attention in the recent years as an efficient technique to improve removal of
60 organic matter and reduce nitrogen species in HSSF CWs (Nivala et al. 2007; Wu et al.
61 2014). This technology has been employed for industrial waste streams, including
62 contaminated groundwater (Wallace and Kadlec, 2005), coffee processing wastewater
63 (Rossmann et al., 2013), landfill leachate (Nivala et al., 2007), airstrip deicing runoff
64 (Higgins, 2003; Murphy et al. 2015), aquaculture (Webb et al., 2013) and livestock
65 wastewater (Zhu et al., 2012). Recent studies highlight the efficiency of aerated systems
66 in reducing nitrogen (Li et al., 2014), emerging contaminants (Avila et al., 2014) and
67 greenhouse gas emissions (Maltais-Landry et al., 2009). Besides this Labella et al. (2015)

68 showed that the reduction of the surface required by aerated systems counterbalances the
69 investment and power consumption of aeration, resulting in similar costs for both aerated
70 and conventional systems.

71 Most experiences with forced aeration however refer to continuous aeration, which has a
72 significant energy consumption and can hamper the development of anoxic conditions
73 (Wu et al., 2014). Anoxic conditions are needed for denitrification, which is an anaerobic
74 heterotrophic process limited by the presence of oxygen and by the organic carbon
75 availability (Fan et al., 2013).

76 In this sense, intermittent aeration controlling and adjusting the dissolved oxygen within
77 the wetland seems to offer an effective alternative to avoid excessive aeration and achieve
78 better total nitrogen removals. In fact, intermittent aeration provides environments of
79 aerobic and anoxic conditions stimulating simultaneous nitrification and denitrification
80 processes (Boog et al., 2014; Fan et al., 2013), which is considered the main N sink in
81 CWs (Tanner et al., 2002). In spite of the promising results obtained in some recent
82 studies (Fan et al., 2013; Zhang et al., 2010), currently scarce information on intermittent
83 aeration is available. Moreover, continuous and intermittent aeration have not been
84 compared yet.

85 The aim of this study was to determine the optimum forced aeration regime (i.e.
86 continuous or intermittent) of HSSF CWs in order to increase treatment efficiency and
87 reduce the energy consumption. To this end, the effect of continuous and intermittent
88 aeration on organic matter and nitrogen removal was evaluated in pilot HSSF CWs.

89 **Materials and Methods**

90 *Pilot plant*

91 The experimental plant (Figure 1) was located at the Agropolis campus of the Universitat
92 Politècnica de Catalunya·BarcelonaTech, in the municipality of Viladecans, near

93 Barcelona, Spain (41.288 N, 2.043 E UTM). The plant was built in early 2015 and set in
94 operation in May of the same year. The raw wastewater, coming from an office building
95 hosting around 50 people, was treated in a septic tank and then pumped to a continuously
96 stirred plastic tank (1.2 m³ volume) used as a reservoir for a few hours. Afterwards,
97 wastewater (here on referred to as influent) was pumped equally into three HSSF CWs in
98 parallel which provided secondary treatment. The individual CW cells were built with an
99 external steel structure supporting five composite polypropylene and glass fiber panels
100 which form the lightweight support for a butyl rubber waterproof membrane. Each CW
101 was built as a prototype for an autonomous reed bed installation as part of a larger project.
102 Each CW had a surface of 2.64 m² (2.2 m long, 1.2 m wide, 1.3 m high). A uniform gravel
103 layer (40% estimated initial porosity) was set to provide a depth of 1.10 m. The water
104 level was kept at 0.10 m below the gravel surface, giving a total water depth of 1 m. The
105 CWs were planted in April 2015 with common reed (*Phragmites australis*) at an initial
106 density of 16 plants/m². The CWs were automatically fed by means of peristaltic pumps
107 under a continuous flow regime and operated at 5.5 days of hydraulic retention time
108 (HRT), with a surface hydraulic loading rate (HLR) of about 7.2 cm/d and a cross-
109 sectional organic loading rate (OLR) around 8 gCOD/m²·d. More details about the beds
110 design and operation can be found in Table 1. During the setting-up of the system, a PVC
111 cylinder (volume of about 0.22 m³) was placed nearby the outlet zone of each bed in order
112 to provide a free gravel zone.

113

114 *Aeration system*

115 Aeration was provided in each bed by means of six aeration pipes (outer diameter of 15
116 mm) pierced with 3 mm holes at a 305 mm separation. These parameters were selected
117 based on typical values used in industrial settings. The system of pipes covering the

118 bottom of the beds was connected to a compressor injecting air at a flow rate of 12.1 m³/h
119 (Josval Serie Cierzo NK 50, Zaragoza, Spain).

120 As previously done by Labella et al. (2015), dissolved oxygen at the bottom of the three
121 wetlands was continuously monitored by means of a dissolved oxygen probe (CS512
122 Oxyguard Type III, Campbell Scientific Inc., USA) located in the gravel free area at the
123 bottom of each bed and connected to a data logger (CR1000, Campbell Scientific Inc.,
124 USA).

125 In order to assess the effect of forced aeration on the wetlands performance, the
126 experimental design as shown in Figure 1 was employed with

- 127 • one bed continuously aerated (here on referred to as fully aerated),
- 128 • one bed with intermittent aeration controlled by a minimum oxygen set point
129 concentration of 0.5 mg/l (later referred to as intermittently aerated),
- 130 • one bed not aerated (referred to as the control from this point onwards).

131 The intermittent aeration was achieved by means of a feedback option of the data logger
132 (control Deadbond version 2.5). The valve controlling air injection was opened when the
133 oxygen concentration was lower than the 0.5 mg/l set point and closed for values higher
134 than this. This configuration was established in accordance with previous results showing
135 that wastewater treatment was satisfactorily improved when oxygen concentration within
136 the wetlands was maintained at 0.5 mg/L (Labella et al., 2015).

137

138 *Physical and Chemical analysis*

139 Water quality was monitored during twelve weeks (between May and July 2015)
140 collecting 27 samples from CWs influent (effluent of the stirred plastic tank) and 27
141 samples from the CWs effluent. The surveyed water quality parameters were the total
142 chemical oxygen demand (COD), total Kjeldahl nitrogen (TKN), ammonium nitrogen

143 (NH₄⁺-N), nitrite (NO₂-N) and nitrate (NO₃⁻-N) nitrogen. Analyses were carried out
144 according to Standard Methods (APHA-AWWA-WEF, 2005) 5220 for COD and 4500 for
145 TKN. Ammonium was measured according to the Solorzano method (Solorzano, 1969),
146 while nitrites and nitrates were determined by a DIONEX ICS-1000 ion chromatograph
147 (limit of detection 0.5 ppm NO_x). COD and ammonium nitrogen were monitored two or
148 three times per week, while the others parameters were analyzed weekly.
149 For each configuration, the removal efficiencies were calculated for nitrogen species and
150 COD according to Eq. 1.

$$151 \quad \text{Removal efficiency (\%)} = \left(1 - \frac{C_e * V_e}{C_i * V_i}\right) * 100 \quad \text{Eq. 1}$$

152 Where C_e was the effluent concentration, V_e is the effluent volume, C_i was the influent
153 concentration and V_i the influent volume of the wetlands. The statistical difference of the
154 experimental results was evaluated by means 3 ways ANOVA and *post-hoc* test (Tukey's)
155 performed using SPSS statistic software 22 (IBM Corporation, Armonk, New York,
156 USA). Water temperature was continuously monitored by means of probes (Temperature
157 Probe Model 107, Campbell Scientific Inc., USA) located in the gravel free area at the
158 bottom of each bed and connected to a data logger (CR1000, Campbell Scientific Inc.,
159 USA). Meteorological data were gathered from the municipal meteorological stations of
160 Viladecans, Barcelona, Spain, located near the site.

161 In order to measure the evapotranspiration, the water flow was measured at the inlet and
162 at the outlet of each wetland by means of peristaltic pumps (at the inlet) and a flow meter
163 device located at the outlet.

164 **Results and discussion**

165 The dissolved oxygen concentration recorded in the three beds clearly displays the effect
166 of aeration (Figure 2). In the bed without air injection, oxygen concentrations were always

167 near to zero, while in the bed with intermittent air injection (0.5mg/L) oxygen
168 concentrations ranged between 0.5 and 2 mg/L (due to the excess power of the compressor
169 used for air injection). The oxygen concentration in the fully aerated bed was significantly
170 higher, fluctuating between 7 and 8 mg/L.

171 The average air temperature during the experiments was 24.7°C, ranging between
172 minimum values of 10.5°C and maximum values of 34.6 °C. In accordance with the
173 summer season in Spain, water temperatures within all the CWs varied between 21°C and
174 32°C.

175 The average COD influent concentration was 118±62 mg/L, with spot peaks of 300 mg/L
176 (Table 2). COD removal was clearly observed in the three beds, where the average
177 discharge concentrations were 68±14 mg/L and 53±12 mg/L in the intermittently and
178 fully aerated beds respectively; and 61±14 mg/L in the control bed. Such values
179 correspond to similar mass removals in all the beds (61-65%). Indeed, no significant
180 differences were recorded between the systems. The small differences observed are likely
181 due to the low loading of the beds. Similar results were found by Butterworth et al. (2013)
182 where the authors did not find differences between fully aerated beds and control beds
183 without aeration. Concerning intermittent aeration, Fan et al. (2013) and Zhang et al.
184 (2010) show scarce or a slightly positive effect on organic matter removal in synthetic
185 and domestic wastewater.

186 Total Kjeldhal Nitrogen (TKN) (data not shown) saw influent concentrations ranging
187 between 10 and 40 mg/L. Such low concentrations are attributed to the limited use of the
188 office building, only frequented during working hours. Outlet concentrations of 10-15
189 mg/L were found in the bed without aeration. On the other hand, when air was injected,
190 concentrations were reduced to about 3 mg/L. Results indicated significantly higher

191 removals in the beds with full (76%) and intermittent aeration (77%) with respect to the
192 control bed, in which only 54% of the TKN was removed. This suggested that the set
193 point of 0.5 mgO₂/L was sufficient for optimal TKN removal and even more efficient
194 than the use of full aeration.

195 Similar results were obtained for ammonium (Figure 3). In this case, the effluents of the
196 aerated beds showed significantly lower concentrations than the control bed (p<0.01). In
197 general, concentrations of 15±11 mg/L present in the influent were reduced to 7±3 mg/L
198 in the bed without aeration, while values near to zero were obtained in both the partially
199 and continuously aerated beds (Table 3). Indeed, no significant differences were found
200 between full and intermittent aeration indicating good nitrification performance in both
201 systems. In general, low removals found in the control (53%) were significantly increased
202 by full (99%) and intermittent aeration (99%). The scarce ammonium removal obtained
203 in the control bed might be attributed to the poor nitrification occurring in anoxic
204 conditions. Such results are confirmed by previous studies in which ammonium removals
205 increased from 59% in a control bed without aeration to 99% in a fully aerated bed
206 (Butterworth et al., 2013). Likewise, Fan et al. (2013) and Zang et al. (2010) showed the
207 positive effect of intermittent aeration on ammonium removal, improving removals from
208 20-24% in a control bed to 89-93% in an intermittently aerated bed.

209 The concentrations of nitrogen oxides (NO_x) provide a useful assessment of the efficacy
210 of the nitrification and denitrification processes (Figure 4). The influent presented with
211 relatively high concentrations of nitrate (8±4 mg/L). Concentrations found in the effluent
212 of the fully aerated bed (24±6 mg/L) were higher than those of the intermittently aerated
213 one (14±6 mg/L), while lower concentrations were found in the control (5±3 mg/L). Such
214 results indicated the high contribution of aeration to the nitrification process, which
215 results in high nitrate concentration. The results of this study are in accordance with the

216 pattern previously showed by Maltais-Landry et al. (2009) comparing fully aerated and
217 non-aerated beds. The authors detected net NO_x productions of about 4 mg/L in the fully
218 aerated systems, while no NO_x was produced in the control. Further investigation of this
219 effect will need to be undertaken on a system with less variation around the 0.5mg/l set
220 point to maximise the creation of zones of varying oxygen concentration.

221 Considering the total nitrogen (TN) as the sum of TKN, nitrite and nitrate (Figure 5),
222 intermittent aeration can achieve lower effluent concentrations (18±7 mg/L) than full
223 aeration (27±6 mg/L). In term of removals, the control reached 61%, while, due to the
224 high amount of nitrate, the fully aerated bed only reaches 50%. The intermittently aerated
225 bed shows better performance, obtaining an average removal of 66% over the
226 experimental period. Our results are in accordance with the literature, with a previous
227 study showing 49% higher TN removals in intermittent aerated beds than in the control
228 (Zhang et al., 2010), while others authors do not detect differences between fully aerated
229 and control beds (Maltais-Landry et al., 2009).

230 It is important to highlight that, even if TN removals were similar for the aerated and non-
231 aerated systems, in the control bed the nitrogen was mainly present in form of ammonium.
232 In this sense, aeration is important for ammonium removal, which is much more harmful
233 for the environment than the other nitrogen forms. Indeed, ammonium toxic effect on
234 zooplankton community has been widely reported (Ankeley et al., 1995; Monda et al.,
235 1995; Puigagut et al., 2005). According to the results found in this study, partial aeration
236 is the most useful option to remove ammonium, nitrate and nitrite due to reduced energy
237 costs over the proven benefits of continuous aeration. This is most likely due to the fact
238 that intermittent aeration provided the coexistence of aerobic and anoxic conditions,
239 stimulating the simultaneous occurrence of nitrification and denitrification. Indeed, a
240 previous study found high removals of ammonium and total nitrogen, demonstrating that

241 the intermittent aeration enhanced the growth of both ammonia-oxidizing bacteria (AOB)
242 and nitrite-oxidizing bacteria (NOB) (Fan et al. 2013). From an environmental and
243 economical point of view, it is significant to highlight the extreme benefit provided by
244 intermittent aeration in terms of the energy consumption of the system. In this study,
245 considering 24h energy consumption of the compressor (1.5 kWh), 13.6 kWh/m²·d were
246 consumed by full aeration. On the other hand, the intermittent aeration only needs 8 pulse
247 per day, corresponding to around 20 minutes of aeration (Supplementary Material, Figure
248 1), thus only 0.18 kWh/m²·d were required, resulting in seventy-fold reduction in power
249 usage. Such a short aeration time can be attributed to the fact that the air pump was
250 probably oversized for the treatment bed being aerated, altogether with the low
251 concentration of both COD and ammonia in the influent.

252 Besides this it should be taken into account that this is a preliminary study, conducted
253 during 3 months along the start-up phase of the system. During this period macrophytes
254 were not well established due to the fact that the experiment was performed during the first
255 growing season. Therefore, the effect of the aeration strategy under well-developed
256 macrophytes remains unknown and shall be further addressed. A longer study would be
257 required to confirm the results collected in this study and to better characterize the
258 observed behavior. A year-round study would be recommended in order to test the
259 seasonal effect of aeration on nitrogen removal.

260

261 **Conclusions**

262 In this study we have tested different forced aeration regimes in a three bed pilot plant in
263 order to improve wastewater treatment in HSSF CWs. The three beds were fully aerated,

264 intermittently aerated to a set point of 0.5mg/l and unaerated all of which reached
265 satisfactory performance in term of wastewater treatment. Due to the coexistence of
266 aerobic and anoxic conditions, the intermittent aeration was the most effective solution,
267 reaching the highest COD (66%), ammonium (99%) and total nitrogen (79%) removals.
268 Continuous aeration promotes almost complete ammonium removal, but resulted in
269 nitrate concentrations in the effluent up to 27 mg/L. The intermittent aeration strategy
270 represents an effective and energy efficient means to reduce ammonium concentration
271 and indeed for general wastewater quality improvement over both fully aerated and
272 unaerated systems.

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282 **References**

283 Ankeley, G.T., Schubauer-Berigan, M.K., Monson P.D. (1995). Influence of pH and
284 hardness on toxicity of ammonia to the amphipod *Hyalella Azteca*. *Can. J. Fish Aquat.*
285 *Sci.* 52, 2078–2083.

286 Ávila, C., Nivala, J., Olsson, L., Kassa, K., Headley, T., Mueller, R.A., Bayona, J.M.,
287 García, J. (2014). Emerging organic contaminants in vertical subsurface flow constructed
288 wetlands: Influence of media size, loading frequency and use of active aeration. *Science*
289 *of the Total Environment* 494-495, 211-217.

290 Butterworth, E., Dotro, G., Jones, M., Richards, A., Onunkwo, P., Narroway, Y.,
291 Jefferson, B. (2013). Effect of artificial aeration on tertiary nitrification in a full-scale
292 subsurface horizontal flow constructed wetland. *Ecological Engineering* 54, 236-244.

293 Boog, J, Nivala, J, Aubron, T, Wallace, S, Afferden, M, Müller, RA. (2014). Hydraulic
294 characterization and optimization of total nitrogen removal in an aerated vertical
295 subsurface flow treatment wetland. *Bioresource Technology* 162, 166–174.

296 Brix, H., Schierup, H-H. (1990). Soil oxygenation in constructed reed beds: the role of
297 macrophyte and soil–atmosphere interface oxygen transport. In: Findlater BC, Cooper
298 PF, editors. *Constructed wetlands in water pollution control*. Oxford, United Kingdom:
299 Pergamon Press; 1990. p. 53–66.

300 Fan, J., Zhang, B., Zhang, J., Ngo, H.H., Guo, W., Liu, F., Guo, Y., Wu, H. (2013).
301 Intermittent aeration strategy to enhance organics and nitrogen removal in subsurface
302 flow constructed wetlands. *Bioresource Technology* 141, 117-122.

303 García, J., Aguirre, P., Mujeriego, R., Huang, Y., Ortiz, L., Bayona, JM. (2004). Initial
304 contaminant removal performance factors in horizontal flow reed beds used for treating
305 urban wastewater. *Water Reserach* 38 (7), 1669–78.

306 García, J., Rousseau, DPL., Morató, J., Lesage, E., Matamoros, V., Bayona, JM. (2010).
307 Contaminant removal processes in subsurface-flow constructed wetlands: a review. *Crit*
308 *Rev Environmental Science Technology* 40 (7), 561–661.

309 Higgins, J. (2003). The use of engineered wetlands to treat recalcitrant waters. In: Mander
310 Ü, Jenssen P, editors. Constructed wetlands for wastewater treatment in cold climates.
311 Southampton, United Kingdom: WIT Press; 137–59.

312 Kadlec, R., Wallace, S. (2009). Treatment Wetlands, second ed. CRC Press, Boca Raton,
313 Florida.

314 Labella, A., Caniani, D., Hughes-Riley, T., Morris, R.H., Newton, M.I., Hawes, P.,
315 Puigagut, J., García, J., Uggetti, E. (2015). Assessing the economic suitability of aeration
316 and the influence of bed heating on constructed wetlands treatment efficiency and life-
317 span. *Ecological Engineering* 83,184-190.

318 Li, F., Lu, L., Zheng, X., Ngo, H.H., Liang, S., Guo, W., Zhang, X. (2014). Enhanced
319 nitrogen removal in constructed wetlands: Effects of dissolved oxygen and step-feeding.
320 *Bioresource Technology* 169, 395-402.

321 Maltais-Landry, G., Maranger, R., Brisson, J. (2009). Effect of artificial aeration and
322 macrophyte species on nitrogen cycling and gas flux in constructed wetlands. *Ecological*
323 *Engineering* 35, 221-229.

324 Monda, D.P., Galat, D.L., Finger S.E. (1995). Evaluating ammonia toxicity in sewage
325 effluent to stream macroinvertebrates: I. A multi-level approach. *Arch. Environ. Contam.*
326 *Toxicol.* 28, 378–384.

327 Murphy, C, Wallace, S., Knight, R., Cooper, D., Sellers, T. (2015). Treatment
328 performance of an aerated constructed wetland treating glycol from de-icing operations
329 at a UK airport. *Ecological Engineering* 80, 117-124.

330 Nivala, J., Hoos, M.B., Cross, C., Wallace, S., Parkin, G. (2007). Treatment of landfill
331 leachate using an aerated, horizontal subsurface-flow constructed wetland. *Science Total*
332 *Environment* 380, 19–27.

333 Puigagut, J., Salvadó, H., García, J. (2005). Short-term harmful effects of ammonia
334 nitrogen on activated sludge microfauna. *Water Research* 39 (18), 4397-4404.

335 Rossmann, M., Teixeira Matos A., Carneiro Abreu, E., Fonseca Silva, F., Carraro Borges,
336 A. (2013). Effect of influent aeration on removal of organic matter from coffee processing
337 wastewater in constructed wetlands. *Journal of Environmental Management* 128, 912-
338 919.

339 Solorzano, L. (1969). Determination of ammonia in natural waters by the
340 phenolhypochlorite method, *Limnol. Oceanogr.* 14: 799.

341 Tanner, C.C., Kadlec, R.H., Gibbs, M.M., Sukias, J.P.S., Nguyen, M.L. (2002). Nitrogen
342 processing gradients in subsurface-flow treatment wetlands – influence of wastewater
343 characteristics. *Ecological Engineering* 18, 499–520.

344 Vymazal, J. (2011) Constructed wetlands for wastewater treatment: five decades of
345 experience. *Environmental Science and Technology* 45, 61–69.

346 Wallace, S.D., 2001. Patent: system for removing pollutants from water. United States:
347 US 6,200,469 B1.

348 Wallace, S.D., Knight R.L. (2006). Small-scale Constructed Wetland Treatment systems:
349 Feasibility, Design Criteria, and O&M Requirements Water Environment Research
350 Foundation (WERF), Alexandria, Virginia.

351 Webb, JM., Quintã, R., Papadimitriou, S., Norman, L., Rigby, M., Thomas, DN., Le Vay,
352 L. (2013). The effect of halophyte planting density on the efficiency of constructed
353 wetlands for the treatment of wastewater from marine aquaculture. *Ecological*
354 *Engineering* 61, 145-153. Wu S., Kusch P., Brix H., Vymazal J., Dong R. (2014).
355 Development of constructed wetlands in performance intensification for wastewater
356 treatment: A nitrogen and organic matter target review. *Water Research* 57, 40-55.

357 Zhang, L-Y., Zhang, L., Liu, Y-D., Shen, Y-W., Liu, H., Xiong, Y. (2010) Effect of
358 limited artificial aeration on constructed wetland treatment of domestic wastewater.
359 Desalination 250, 915-920.

360 Zhu, D., Sun, C., Zhang, H., Wu, Z., Jia, B., Zhang, Y. (2012). Roles of vegetation, flow
361 type and filled depth on livestock wastewater treatment through multi-level mineralized
362 refuse-based constructed wetlands. Ecological Engineering 39, 7-15.

363 **Tables and figures***Table 1. Technical data of the domestic wastewater wetlands*

Parameter	Value
Dimensions (WxLxH) (cm)	120 x220x130
Water level (cm)	100
Surface area (m ²)	2.64
Flow (L/d)	190
Surface hydraulic loading rate (cm/d)	7.2
Hydraulic retention time (d)	5.5
Cross-sectional organic loading rate (gCOD/m ² ·d)	7.8
Surface organic loading rate (gCOD/m ² ·d)	8.5

364

365

Table 2. Influent and effluents concentrations of Chemical Oxygen Demand (COD) in the three wetlands along the experiment (\pm s.d.).

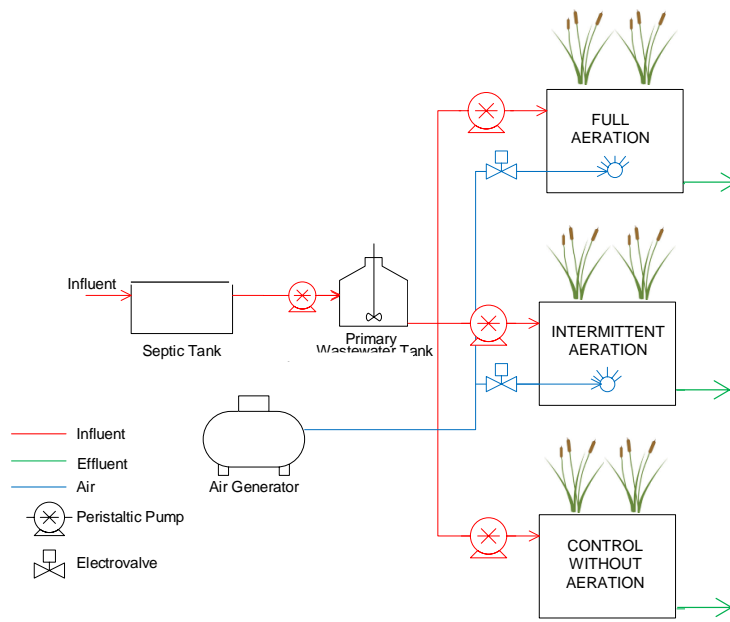
Day of experiment	COD concentration (mgO ₂ /L)			
	Influent	Full	Intermittent	Control
0	58 ± 10	63 ± 23	-	58 ± 6
5	42 ± 6	70 ± 19	-	36 ± 9
7	189 ± 12	77 ± 7	-	61 ± 7
12	118 ± 26	89 ± 10	65 ± 1	57 ± 21
14	162 ± 3	58 ± 11	53 ± 9	62 ± 8
19	145 ± 10	53 ± 10	65 ± 3	35 ± 10
21	122 ± 13	76 ± 18	61 ± 10	93 ± 7
26	115 ± 19	102 ± 6	64 ± 17	54 ± 4
28	102 ± 6	66 ± 4	74 ± 6	73 ± 7
33	100 ± 5	72 ± 4	59 ± 3	51 ± 3
35	68 ± 10	58 ± 17	44 ± 12	52 ± 13
38	72 ± 22	48 ± 5	57 ± 11	53 ± 14
42	121 ± 12	100 ± 9	73 ± 6	81 ± 11
45	102 ± 7	64 ± 5	53 ± 5	57 ± 3
47	99 ± 18	71 ± 4	43 ± 5	55 ± 9
49	136 ± 8	82 ± 14	48 ± 23	65 ± 6
52	87 ± 13	61 ± 4	55 ± 5	63 ± 3
54	125 ± 9	60 ± 13	72 ± 7	94 ± 6
56	124 ± 7	55 ± 8	55 ± 7	61 ± 6
59	71 ± 6	48 ± 3	38 ± 0	49 ± 2
61	89 ± 6	49 ± 9	47 ± 9	51 ± 8
63	71 ± 7	55 ± 7	53 ± 4	82 ± 2
66	286 ± 111	85 ± 4	29 ± 5	66 ± 13
68	83 ± 1	-	29 ± 15	47 ± 9
70	78 ± 5	67 ± 4	47 ± 8	53 ± 13
73	319 ± 9	67 ± 8	40 ± 6	59 ± 20
75	108 ± 13	69 ± 15	51 ± 12	67 ± 8

Table 3. Influent and effluents concentrations of ammonium, Total Kjeldhal Nitrogen and organic nitrogen in the three wetlands.

Day of experiment	NH ₄ ⁺ -N (mg/L)				TNK (mg/L)				N org (mg/L)			
	Influent	Full	Intermittent	Control	Influent	Full	Intermittent	Control	Influent	Full	Intermittent	Control
19	19.45	0.02	0.03	7.67	32.30	2.80	2.80	11.20	12.85	2.78	2.77	3.53
26	14.19	0.03	0.03	13.33	17.20	2.80	-	14.90	3.01	2.77	-	1.57
33	5.80	0.01	0.02	5.68	11.20	3.50	4.20	9.80	5.40	3.49	4.18	4.12
47	7.40	0.08	0.08	3.11	25.30	9.10	3.50	9.80	17.90	9.02	3.42	6.69
54	28.46	0.52	1.06	6.78	38.60	2.80	7.00	14.80	10.14	2.28	5.94	8.02
61	3.07	0.01	0.02	7.31	9.80	2.80	2.80	16.90	6.73	2.79	2.78	9.59
68	0.78	0.01	0.01	4.89	9.80	3.50	2.80	13.30	9.02	3.49	2.79	8.41
75	3.40	0.00	0.01	3.08	9.80	2.80	2.80	7.00	6.40	2.80	2.79	3.92

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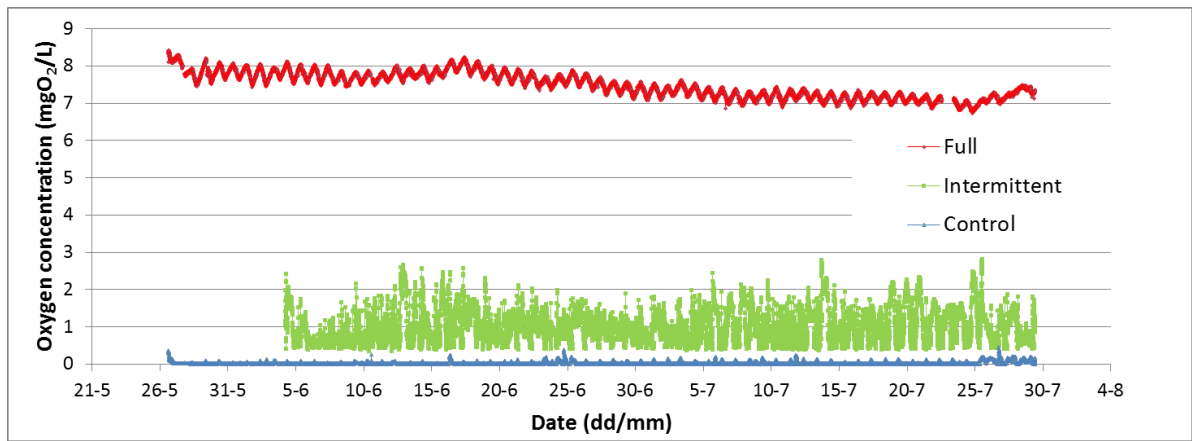
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Figure 1. Diagram of the experimental plant. From the septic tank, wastewater was pumped into a storage tank and conveyed to the three wetlands beds.



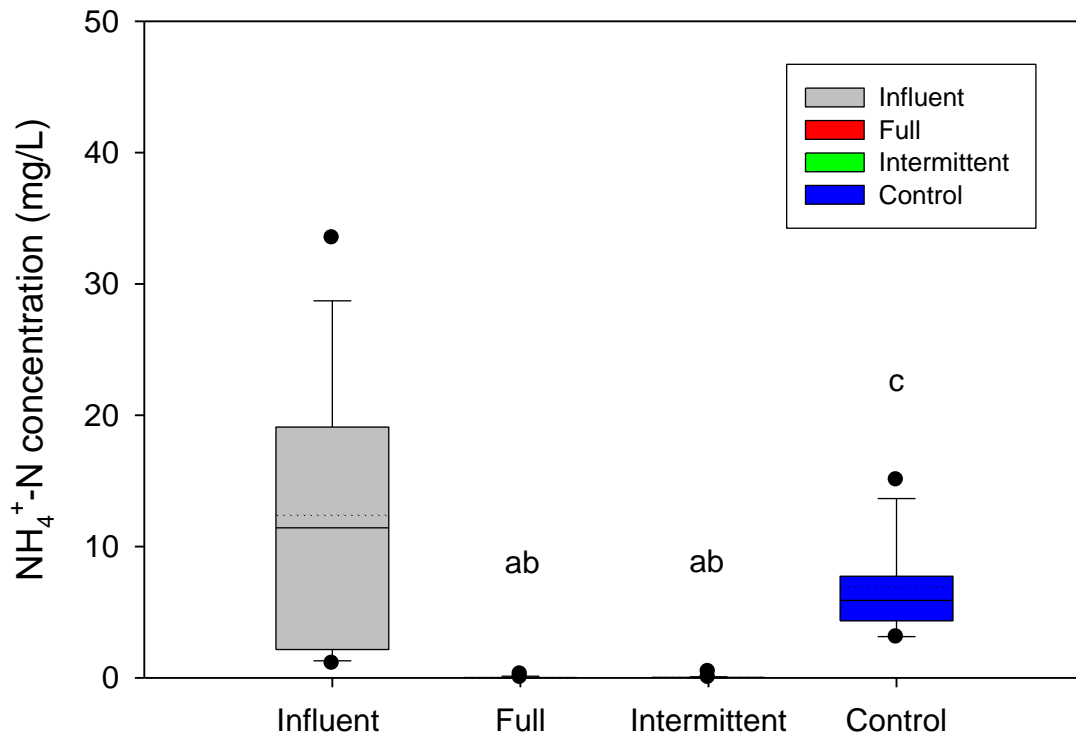
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377 Figure 2. Dissolved oxygen concentration in the three beds used to treat domestic
378 wastewater over the course of the experiment.

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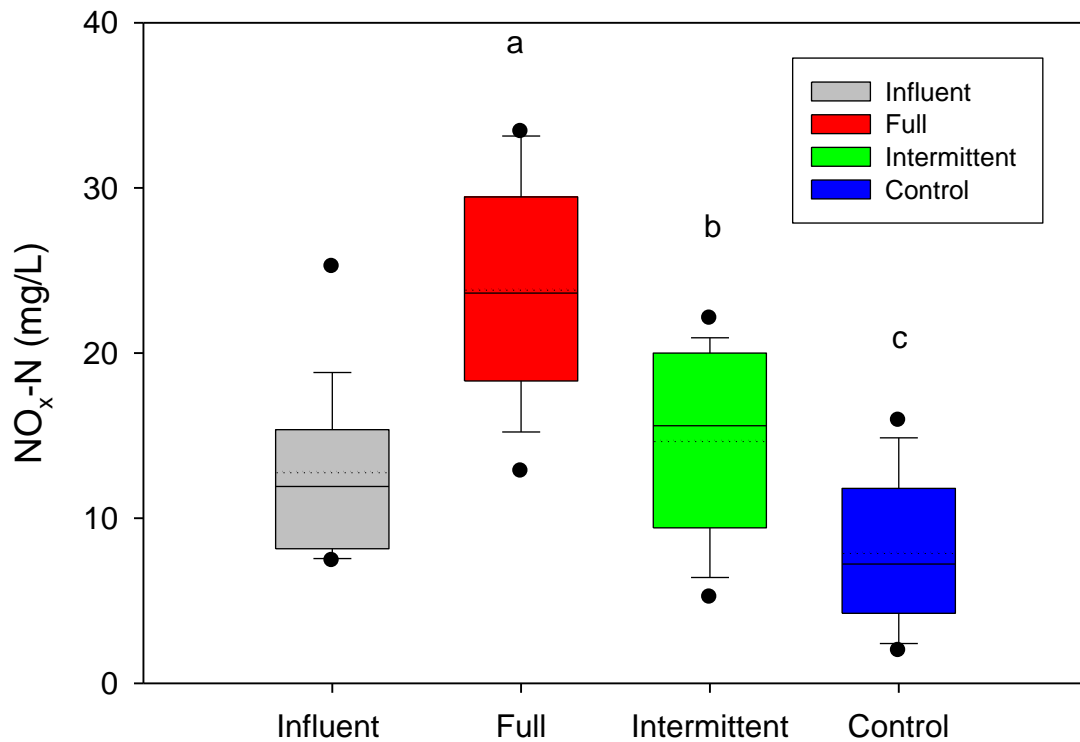


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383 Figure 3. Influent and effluents concentrations of ammonium in the three wetlands
384 (n=27). The lower boundary of the box indicates the 25th percentile, the lines within the
385 box mark the median (solid line) and the average (dotted line), and the upper boundary
386 of the box indicates the 75th percentile. Whiskers (error bars) above and below the box
387 indicate the 90th and 10th percentiles, respectively. Upper and bottom dots represent the
388 95th and 5th percentile, respectively. Letters indicate which groups of data differ with
389 significance, $p < 0.01$.

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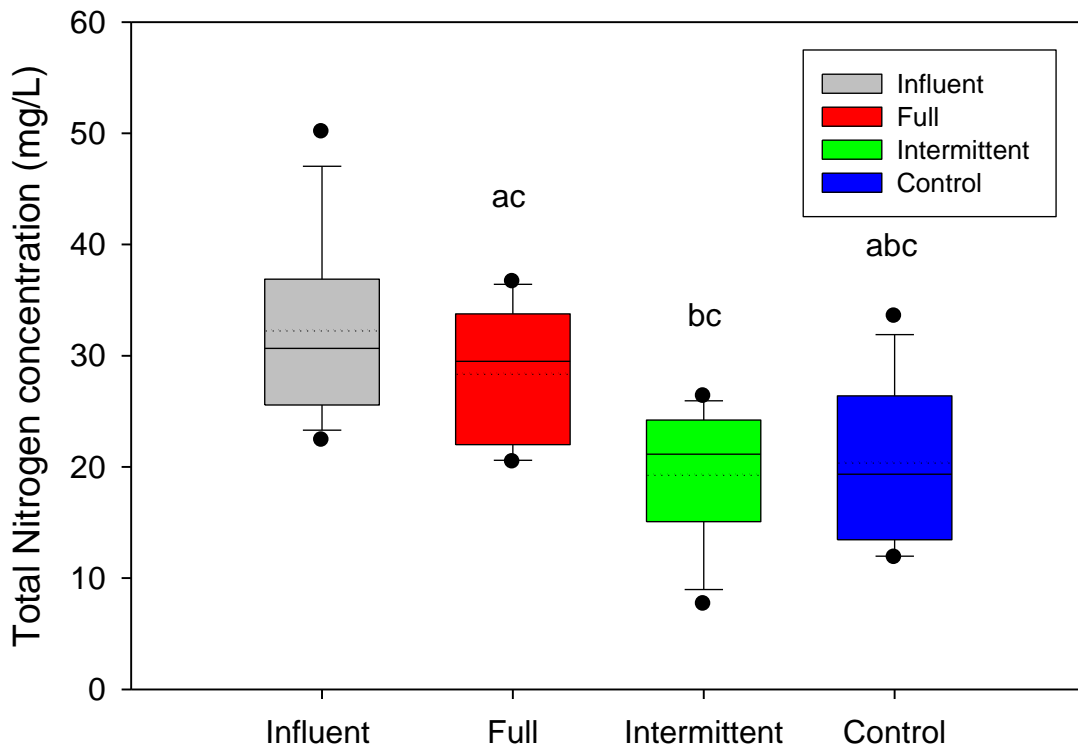
394 Figure 4. Influent and effluents concentrations of nitrite and nitrate in the three wetlands
 395 (n=27). The lower boundary of the box indicates the 25th percentile, the lines within the
 396 box mark the median (solid line) and the average (dotted line), and the upper boundary
 397 of the box indicates the 75th percentile. Whiskers (error bars) above and below the box
 398 indicate the 90th and 10th percentiles, respectively. Upper and bottom dots represent the
 399 95th and 5th percentile, respectively. Letters indicate which groups of data differ with
 400 significance, $p < 0.01$.

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406

407 Figure 5. Influent and effluents concentrations of total nitrogen in the three wetlands

408 (n=27). Total nitrogen is calculated as the sum of TKN, nitrite and nitrate. The lower

409 boundary of the box indicates the 25th percentile, the lines within the box mark the

410 median and the average (dotted line), and the upper boundary of the box indicates the

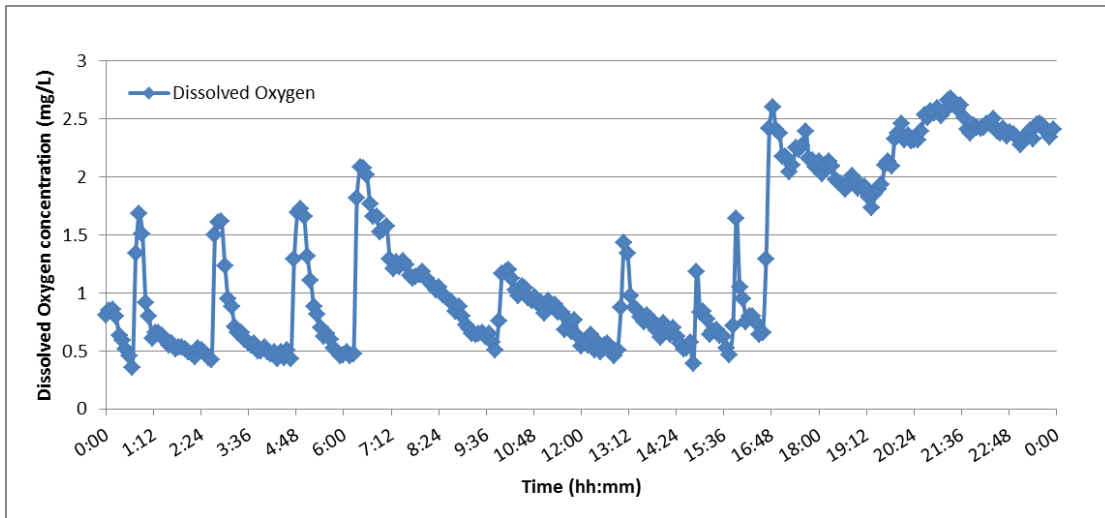
411 75th percentile. Whiskers (error bars) above and below the box indicate the 90th and 10th

412 percentiles. Upper and bottom dots represent the 95th and 5th percentile, respectively.

413 Letters indicate which groups of data differ with significance, $p=0.012$.

414

416 **Supplementary Material**



417

418 Figure 1. Data of dissolved oxygen collected in the bed with intermittent aeration along
419 one day.