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7 **Intermittent aeration to improve wastewater treatment**
8 **efficiency in pilot-scale constructed wetland**

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27

28 **Abstract**

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

47 **Keywords:** Intermittent aeration, Ammonium removal, Nitrification/Denitrification,
48 Constructed Wetlands, Horizontal sub-surface flow.
49

50 **Introduction**

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

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

66 Forced or active aeration, originally developed by Wallace (2001), has received
67 increasing attention in the recent years as an efficient technique to improve removal of
68 organic matter and reduce nitrogen species in HSSF CWs (Nivala et al. 2007; Wu et al.
69 2014). This technology has been employed for industrial waste streams, including
70 contaminated groundwater (Wallace and Kadlec, 2005), coffee processing wastewater
71 (Rossmann et al., 2013), landfill leachate (Nivala et al., 2007), airstrip deicing runoff
72 (Higgins, 2003; Murphy et al. 2015), aquaculture (Webb et al., 2013) and livestock
73 wastewater (Zhu et al., 2012). Recent studies highlight the efficiency of aerated systems
74 in reducing nitrogen (Li et al., 2014), emerging contaminants (Avila et al., 2014) and

75 greenhouse gas emissions (Maltais-Landry et al., 2009). Besides this Labella et al.
76 (2015) showed that the reduction of the surface required by aerated systems
77 counterbalances the investment and power consumption of aeration, resulting in similar
78 costs for both aerated and conventional systems.

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

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

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

97 **Materials and Methods**

98 *Pilot plant*

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

121

122 *Aeration system*

123 Aeration was provided in each bed by means of six aeration pipes (outer diameter of 15
124 mm) pierced with 3 mm holes at a 305 mm separation. These parameters were selected
125 based on typical values used in industrial settings. The system of pipes covering the
126 bottom of the beds was connected to a compressor injecting air at a flow rate of 12.1
127 m³/h (Josval Serie Cierzo NK 50, Zaragoza, Spain).

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

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

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

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

145

146 *Physical and Chemical analysis*

147 Water quality was monitored during twelve weeks (between May and July 2015)
148 collecting 27 samples from CWs influent (effluent of the stirred plastic tank) and 27
149 samples from the CWs effluent. The surveyed water quality parameters were the total
150 chemical oxygen demand (COD), total Kjeldahl nitrogen (TKN), ammonium nitrogen
151 ($\text{NH}_4^+\text{-N}$), nitrite (NO_2^-N) and nitrate (NO_3^-N) nitrogen. Analyses were carried out
152 according to Standard Methods (APHA-AWWA-WEF, 2005) 5220 for COD and 4500
153 for TKN. Ammonium was measured according to the Solorzano method (Solorzano,
154 1969), while nitrites and nitrates were determined by a DIONEX ICS-1000 ion
155 chromatograph (limit of detection 0.5 ppm NO_x). COD and ammonium nitrogen were
156 monitored two or three times per week, while the others parameters were analyzed
157 weekly.

158 For each configuration, the removal efficiencies were calculated for nitrogen species
159 and COD according to Eq. 1.

$$160 \quad \text{Removal efficiency (\%)} = \left(1 - \frac{C_e * V_e}{C_i * V_i}\right) * 100 \quad \text{Eq. 1}$$

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

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

173 **Results and discussion**

174 The dissolved oxygen concentration recorded in the three beds clearly displays the
175 effect of aeration (Figure 2). In the bed without air injection, oxygen concentrations
176 were always near to zero, while in the bed with intermittent air injection (0.5mg/L)
177 oxygen concentrations ranged between 0.5 and 2 mg/L (due to the excess power of the
178 compressor used for air injection). The oxygen concentration in the fully aerated bed
179 was significantly higher, fluctuating between 7 and 8 mg/L.

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

184 The average COD influent concentration was 118±62 mg/L, with spot peaks of 300
185 mg/L (Table 2). COD removal was clearly observed in the three beds, where the
186 average discharge concentrations were 68±14 mg/L and 53±12 mg/L in the
187 intermittently and fully aerated beds respectively; and 61±14 mg/L in the control bed.
188 Such values correspond to similar mass removals in all the beds (61-65%). Indeed, no
189 significant differences were recorded between the systems. The small differences
190 observed are likely due to the low loading of the beds. Similar results were found by
191 Butterworth et al. (2013) where the authors did not find differences between fully
192 aerated beds and control beds without aeration. Concerning intermittent aeration, Fan et

193 al. (2013) and Zhang et al. (2010) show scarce or a slightly positive effect on organic
194 matter removal in synthetic and domestic wastewater.

195 Total Kjeldhal Nitrogen (TKN) (data not shown) saw influent concentrations ranging
196 between 10 and 40 mg/L. Such low concentrations are attributed to the limited use of
197 the office building, only frequented during working hours. Outlet concentrations of 10-
198 15 mg/L were found in the bed without aeration. On the other hand, when air was
199 injected, concentrations were reduced to about 3 mg/L. Results indicated significantly
200 higher removals in the beds with full (76%) and intermittent aeration (77%) with respect
201 to the control bed, in which only 54% of the TKN was removed. This suggested that the
202 set point of 0.5 mgO₂/L was sufficient for optimal TKN removal and even more
203 efficient than the use of full aeration.

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

218 The concentrations of nitrogen oxides (NO_x) provide a useful assessment of the efficacy
219 of the nitrification and denitrification processes (Figure 4). The influent presented with
220 relatively high concentrations of nitrate (8 ± 4 mg/L). Concentrations found in the
221 effluent of the fully aerated bed (24 ± 6 mg/L) were higher than those of the
222 intermittently aerated one (14 ± 6 mg/L), while lower concentrations were found in the
223 control (5 ± 3 mg/L). Such results indicated the high contribution of aeration to the
224 nitrification process, which results in high nitrate concentration. The results of this
225 study are in accordance with the pattern previously showed by Maltais-Landry et al.
226 (2009) comparing fully aerated and non-aerated beds. The authors detected net NO_x
227 productions of about 4 mg/L in the fully aerated systems, while no NO_x was produced
228 in the control. Further investigation of this effect will need to be undertaken on a system
229 with less variation around the 0.5mg/l set point to maximise the creation of zones of
230 varying oxygen concentration.

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

240 It is important to highlight that, even if TN removals were similar for the aerated and
241 non-aerated systems, in the control bed the nitrogen was mainly present in form of
242 ammonium. In this sense, aeration is important for ammonium removal, which is much

243 more harmful for the environment than the other nitrogen forms. Indeed, ammonium
244 toxic effect on zooplankton community has been widely reported (Ankeley et al., 1995;
245 Monda et al., 1995; Puigagut et al., 2005). According to the results found in this study,
246 partial aeration is the most useful option to remove ammonium, nitrate and nitrite due to
247 reduced energy costs over the proven benefits of continuous aeration. This is most
248 likely due to the fact that intermittent aeration provided the coexistence of aerobic and
249 anoxic conditions, stimulating the simultaneous occurrence of nitrification and
250 denitrification. Indeed, a previous study found high removals of ammonium and total
251 nitrogen, demonstrating that the intermittent aeration enhanced the growth of both
252 ammonia-oxidizing bacteria (AOB) and nitrite-oxidizing bacteria (NOB) (Fan et al.
253 2013). From an environmental and economical point of view, it is significant to
254 highlight the extreme benefit provided by intermittent aeration in terms of the energy
255 consumption of the system. In this study, considering 24h energy consumption of the
256 compressor (1.5 kWh), 13.6 kWh/m²·d were consumed by full aeration. On the other
257 hand, the intermittent aeration only needs 8 pulse per day, corresponding to around 20
258 minutes of aeration (Supplementary Material, Figure 1), thus only 0.18 kWh/m²·d were
259 required, resulting in seventy-fold reduction in power usage. Such a short aeration time
260 can be attributed to the fact that the air pump was probably oversized for the treatment
261 bed being aerated, altogether with the low concentration of both COD and ammonia in
262 the influent.

263 Besides this it should be taken into account that this is a preliminary study, conducted
264 during 3 months along the start-up phase of the system. During this period macrophytes
265 were not well established due to the fact that the experiment was performed during the
266 first growing season. Therefore, the effect of the aeration strategy under well-developed
267 macrophytes remains unknown and shall be further addressed. A longer study would be

268 required to confirm the results collected in this study and to better characterize the
269 observed behavior. A year-round study would be recommended in order to test the
270 seasonal effect of aeration on nitrogen removal.

271

272 **Conclusions**

273 In this study we have tested different forced aeration regimes in a three bed pilot plant
274 in order to improve wastewater treatment in HSSF CWs. The three beds were fully
275 aerated, intermittently aerated to a set point of 0.5mg/l and unaerated all of which
276 reached satisfactory performance in term of wastewater treatment. Due to the
277 coexistence of aerobic and anoxic conditions, the intermittent aeration was the most
278 effective solution, reaching the highest COD (66%), ammonium (99%) and total
279 nitrogen (79%) removals. Continuous aeration promotes almost complete ammonium
280 removal, but resulted in nitrate concentrations in the effluent up to 27 mg/L. The
281 intermittent aeration strategy represents an effective and energy efficient means to
282 reduce ammonium concentration and indeed for general wastewater quality
283 improvement over both fully aerated and unaerated systems.

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374 **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

375

376

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

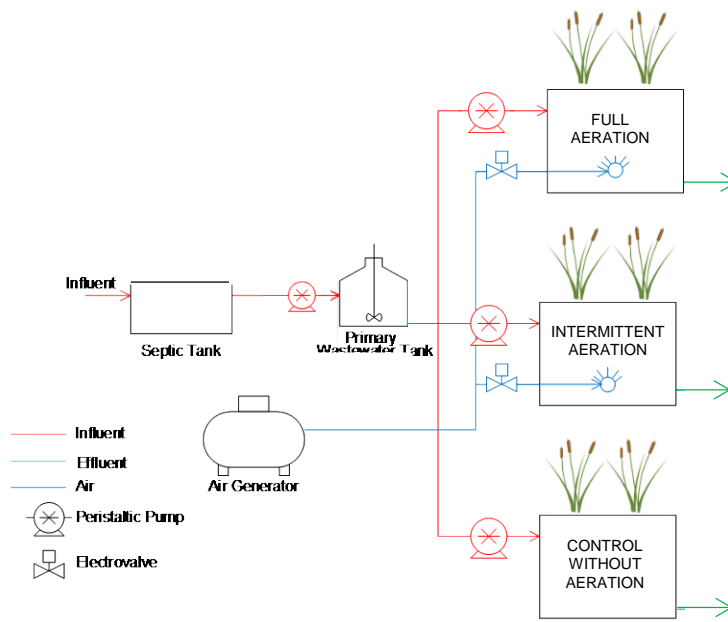
Day of experiment	COD concentration (mgO ₂ /L)			
	Influent	Full	Intermittent	Control
0	58 ± 10	63 ± 23	-	58 ± 6
5	42 ± 6	70 ± 19	-	36 ± 9
7	189 ± 12	77 ± 7	-	61 ± 7
12	118 ± 26	89 ± 10	65 ± 1	57 ± 21
14	162 ± 3	58 ± 11	53 ± 9	62 ± 8
19	145 ± 10	53 ± 10	65 ± 3	35 ± 10
21	122 ± 13	76 ± 18	61 ± 10	93 ± 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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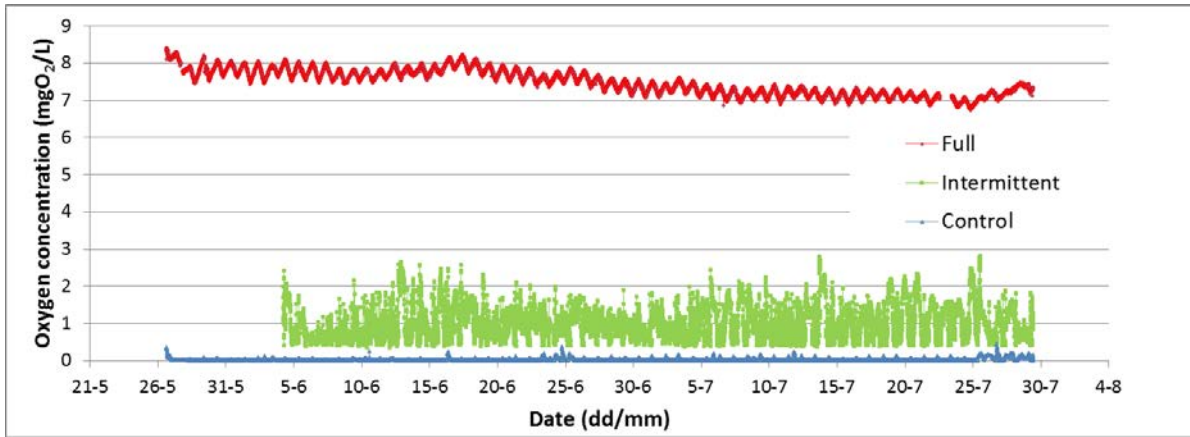
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Figure 1. Diagram of the experimental plant. From the septic tank, wastewater was pumped into a storage tank and conveyed to the three wetlands beds.



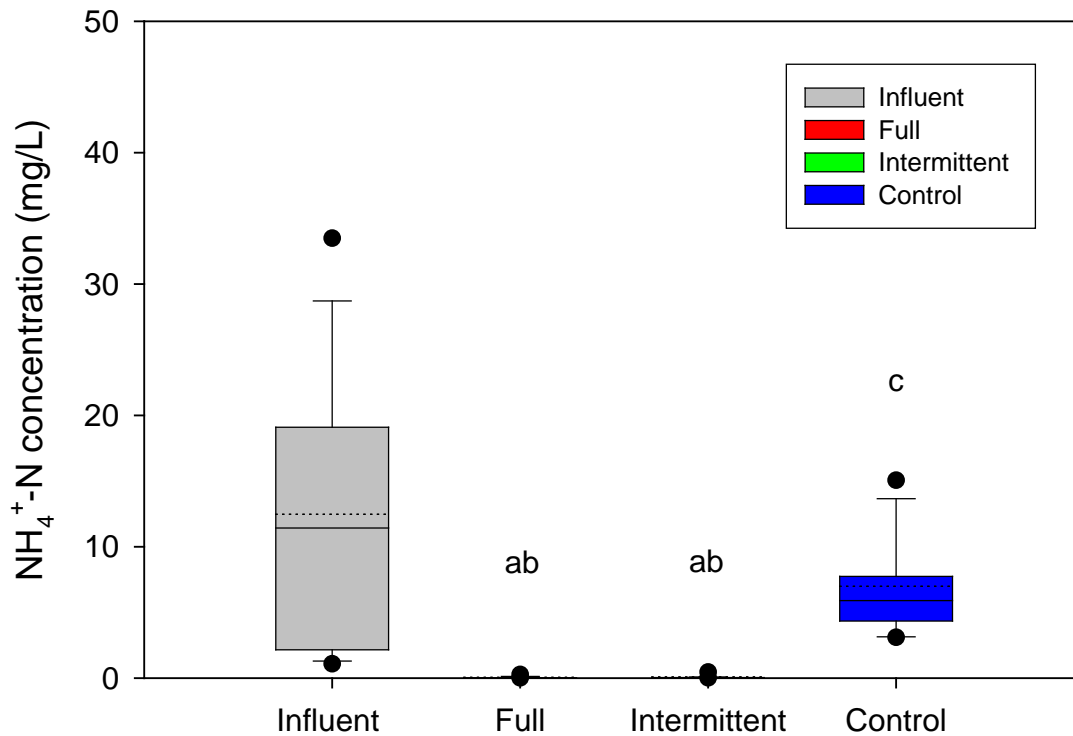
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388 Figure 2. Dissolved oxygen concentration in the three beds used to treat domestic
389 wastewater over the course of the experiment.

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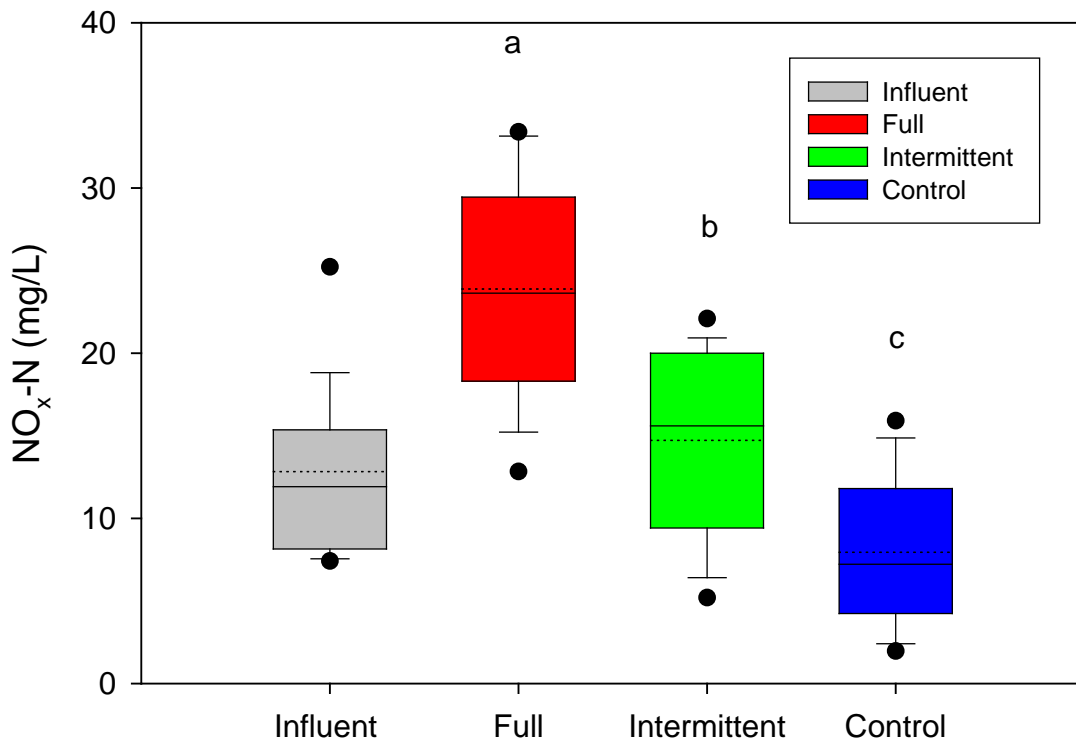


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394 Figure 3. Influent and effluents concentrations of ammonium in the three wetlands
395 (n=27). The lower boundary of the box indicates the 25th percentile, the lines within the
396 box mark the median (solid line) and the average (dotted line), 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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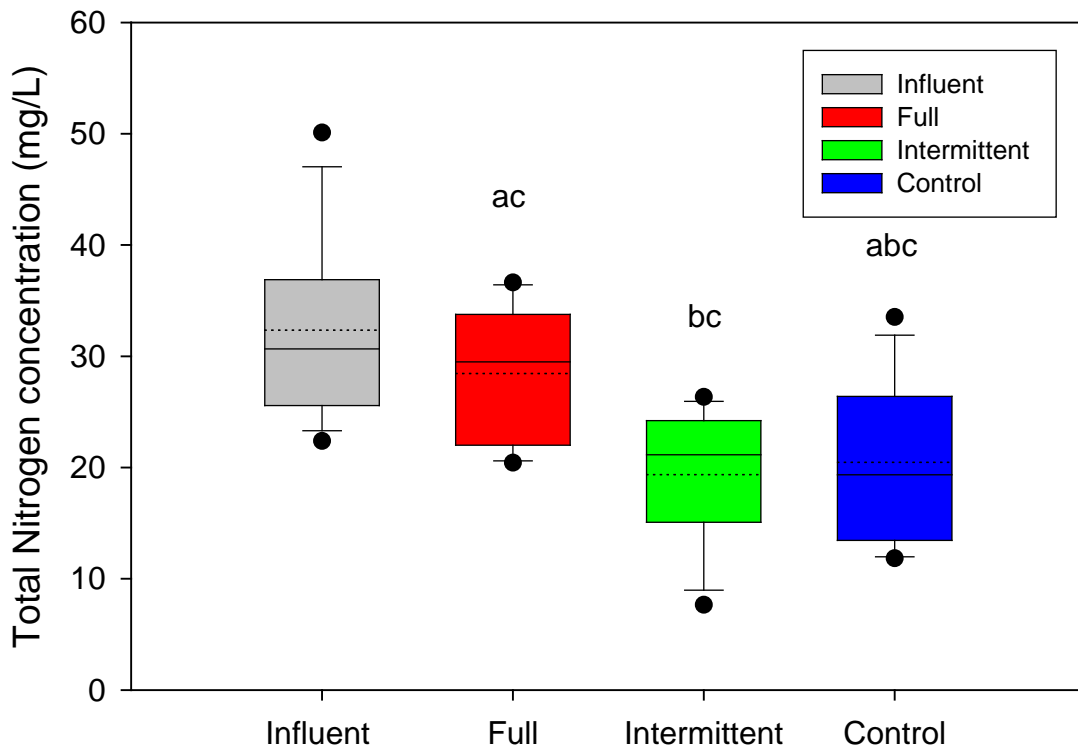
405 Figure 4. Influent and effluents concentrations of nitrite and nitrate in the three wetlands
 406 (n=27). The lower boundary of the box indicates the 25th percentile, the lines within the
 407 box mark the median (solid line) and the average (dotted line), and the upper boundary
 408 of the box indicates the 75th percentile. Whiskers (error bars) above and below the box
 409 indicate the 90th and 10th percentiles, respectively. Upper and bottom dots represent the
 410 95th and 5th percentile, respectively. Letters indicate which groups of data differ with
 411 significance, $p < 0.01$.

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417

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

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

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

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

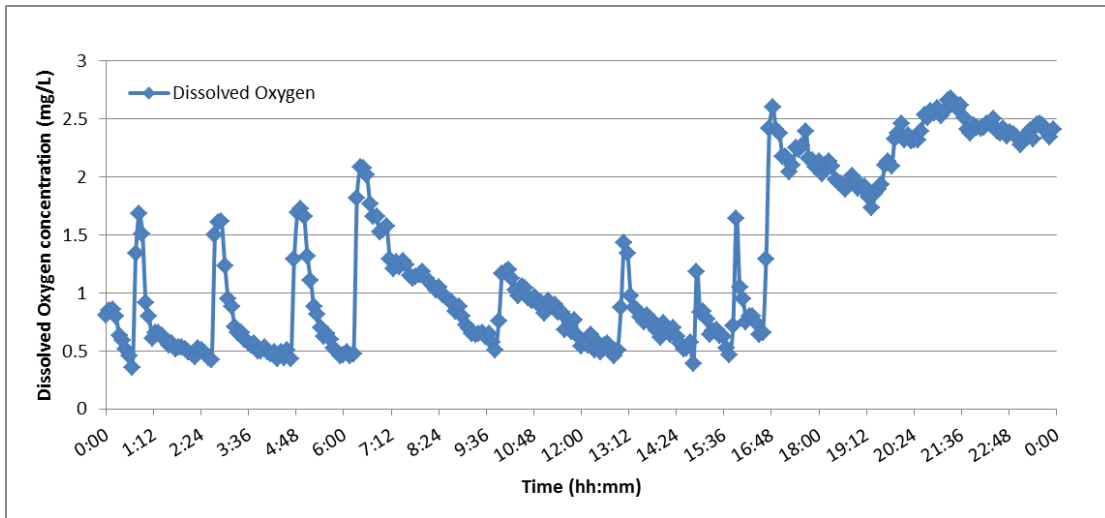
422 75th percentile. Whiskers (error bars) above and below the box indicate the 90th and

423 10th percentiles. Upper and bottom dots represent the 95th and 5th percentile,

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

425

427 **Supplementary Material**



428

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