Treatment of freshwater fish farm effluent using constructed wetlands: the role of plants and substrate

S. Naylor*, J. Brisson*, M.A. Labelle**, A. Drizo** and Y. Comeau**

* Institut de Recherche en Biologie Végétale, 4101 Sherbrooke St. East, Montreal (Quebec), Canada H1X 2B2 (E-mail: jacques.brisson@umontreal.ca)
** Department of Civil, Geological and Mining Engineering, Ecole Polytechnique, Montreal (Quebec), Canada H3C 3A7

Abstract Freshwater fish farm effluents have low nutrient concentrations but high flow rates, resulting in a pollutant load, especially phosphorus (P), causing eutrophication. The feasibility was tested of a treatment combining, within a single constructed wetland, the contribution of macrophytes for reducing organic matter and nitrogen (N), with the high efficiency of steel slag and limestone for P removal. Twenty subsurface flow (SSF) basins of 280 L with different combinations of plants (Phragmites communis or Typha latifolia) and substrates (steel slag, limestone, gravel, peat) were fed with a reconstituted fish farm effluent in a greenhouse experiment. Pollutant removal was generally very good under all treatments. N and organic matter removal were correlated with plant biomass while P removal was better in substrates with steel slag and limestone. However, the high pH of the P-adsorbing substrate was detrimental to plant growth so that no combination of plants and substrates could maximise in one step the simultaneous removal of all evaluated pollutants. Therefore, the use of two sequential units is recommended, a first one consisting of a macrophyte planted basin using a neutral substrate to remove organic matter and N, followed by a second unplanted basin containing only a P-adsorbing substrate.

Keywords Constructed wetland; nitrogen; phosphorus; plants; steel slag

Introduction The expansion of the freshwater fish farm industry in the United States and Canada raises concerns regarding its environmental impacts on water use and quality. The effluent from a typical fresh-water fish farm contains small concentrations of pollutants, 20 to 25 times more diluted than typical municipal wastewater, but has a relatively high flow rate. Phosphorus is of particular concern despite its low concentration (total P of 0.30 mg P/L) because of its contribution to the eutrophication of freshwaters. In Quebec, stringent freshwater fish farm effluent discharge criteria as low as water quality objectives of 0.02 or 0.03 mg P/L are being considered (Ouellet, 1999). Without achieving such a low level, a significant pollutant reduction could be achieved by treating the microscreened washwater of a fish farm effluent with constructed wetland systems (CWS; Comeau et al., 2001). CWSs for wastewater treatment are cost effective, can cope well with load variations as well as cold climate and are simple to operate. Macrophytes, by providing a hospitable habitat for many decomposing microorganisms in the rhizosphere, play an indirect but important role in reducing organic matter and nitrogen from various types of wastewaters. Phosphorus removal is mostly related to physical and chemical processes with the substrate. Some substrates such as limestone and electric arc furnace (EAF) steel slag were shown to have a high P-retaining capacity (Yamada et al., 1986; Forget, 2001; Drizo et al., 2002). We tested the feasibility of combining macrophytes, to reduce organic matter and nitrogen, and slag and limestone, efficient for P removal in a CWS for fish farm effluent treatment.
Materials and methods

Wetland units

Twenty plastic basins of 280 L and 1 m², filled up to 3 cm from the edge with a 5–10 mm substrate, and planted or not with macrophytes, were used as SSF wetland units. Three types of substrate were used (Table 1). The slag-limestone-granite (SLG) substrate was composed of 25% EAF-steel slag, 20% limestone and 55% granite gravel, on a weight basis. Steel slag induces a rapid adsorption reaction and limestone a slower precipitation. The two-speed reaction of adsorption and precipitation was expected to retain a maximum amount of phosphorus. Granite was used as a neutral material in order to avoid filling the basins with only high pH material unfavourable to plant growth. The slag-limestone-granite-peat (SLG-P) substrate was composed of 50% SLG and 50% of peat on a volume basis. Peat further reduced the high pH and provided an organic soil for plants. Finally, the slag-limestone (SL) substrate was composed of 50% EAF steel slag and 50% limestone on a weight basis. Because the pH of the SL substrate was too high for plant growth, units with this substrate were kept unplanted and exclusively used as a polishing step for the planted units to test a design criterion.

We tested the efficiency of two macrophytes, *Phragmites australis* and *Typha latifolia*. Both are locally available and widely used in CWSs. *Phragmites* is the most commonly used macrophyte in SSF wetlands, notably because of its high biomass production, good resistance, strong and deep root system and ability to adapt to a variety of loading rates and water regimes. *Typha* was tested because of its similar competency in wastewater treatment but particularly to propose an alternative macrophyte for some regions where *Phragmites* is not naturally present and may become invasive (Grandtner, 1999). Planting was done one year prior to the experiment, during the summer of 2000, to allow adequate establishment. The *Phragmites* seedlings (20 cm tall) were produced in a plant nursery and were supplied in 100 mL cells filled up with organic soil. *Typha* of 1.0–1.5 m tall were taken from a natural wetland. Both species were planted at a high density of 10 plants/m² to allow rapid maximum cover. During the first growing season, a complete nutrient solution (N:P:K of 15:30:15) was applied to all treatments for the establishment of the young plants. During the winter months, the system was rinsed several times with tap water. Also, temperature and light period were reduced in the greenhouse to force plant hibernation and photosynthetates translocation in the underground reserve organs.

Reconstituted fish farm effluent

We used diluted sludge from a freshwater fish farm anaerobic sludge digester as a substitute for a typical effluent. The sludge was collected at the Alleghany’s Fish Farm in Saint-Damien (Quebec), transferred into plastic pots, and stored in a freezer at −12°C. The dry matter content of the collected sludge was about 1.5%. Twice a week, 24 L of sludge was first melted then filtered with a 2 mm openings diameter strainer to remove the filamentous

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Substrate</th>
<th>Substrate mass (kg)</th>
<th>Macrophyte</th>
<th>Replicates</th>
</tr>
</thead>
<tbody>
<tr>
<td>T1 m</td>
<td>Slag-limestone-granite (SLG)</td>
<td>103 82 226 0 410</td>
<td>Unplanted</td>
<td>2</td>
</tr>
<tr>
<td>T1 p</td>
<td>Slag-limestone-granite and peat (SLG-P)</td>
<td>51 42 113 20 225</td>
<td>Unplanted</td>
<td>2</td>
</tr>
<tr>
<td>T2</td>
<td>SLG</td>
<td>103 82 226 0 410</td>
<td><em>Phragmites</em></td>
<td>4</td>
</tr>
<tr>
<td>T3</td>
<td>SLG</td>
<td>103 82 226 0 410</td>
<td><em>Typha</em></td>
<td>4</td>
</tr>
<tr>
<td>T4</td>
<td>SLG-P</td>
<td>51 42 113 20 225</td>
<td><em>Phragmites</em></td>
<td>4</td>
</tr>
<tr>
<td>T4'</td>
<td>Slag and limestone (SL)</td>
<td>205 205 0 0 410</td>
<td>Unplanted</td>
<td>4</td>
</tr>
</tbody>
</table>
or large particles likely to clog the pumping and tubing system. The filtered sludge was diluted in 2,000 L of water in two containers (1,000 L each) to obtain a feeding solution of about 200 mg/L of suspended solids. The stainless steel refrigerated containers kept the solution at around 12°C with constant stirring to minimise fermentation and settling. Even though the reconstituted effluent prepared in this way is a product of a real fish farm effluent, there are differences between this effluent and a real fresh fish farm wastewater. In particular, freezing the sludge before use may have modified its structure. Also, depending on the duration of stay in the refrigerated container before pumping to the wetland units, some fermentation may have occurred, leading to some mineralization of the effluent.

**Experimental setup**

The twenty wetland units were disposed in two rows, each one supporting the same sets of randomly positioned treatments: one SLG unplanted control basin (T1 m), one SLG-P unplanted control basin (T1 p), two basins with *Phragmites* in an SLG substrate (T2), two basins with *Typha* in an SLG substrate (T3) and two basins planted with *Phragmites* in an SLG-P substrate (T4) followed by two basins filled with SL substrate (T4’), providing two identical blocks to the experiment (Table 1). Irrigation of the wetland units with the reconstituted fish farm effluent occurred between May and November 2001. The units were irrigated using a peristaltic pumping system connected to timers. Two batches of 15 L per unit, lasting 45 minutes, occurred every 12 h, resulting in a vertical hydraulic loading rate (vHLR) of 0.03 m/d and a theoretical void hydraulic retention time (HRTv) of 4 days. The T4’ units were irrigated with the T4 effluent overflow.

**Analytical and statistical methods**

The volume of the effluent from each basin was measured daily for closing the hydraulic mass balance. A sampling point was located at the end of each basin to allow direct collection of the overflow for pollutant analysis. Parameters were analysed according to Standard Methods (1998). Sampling frequency and analyses varied from twice a week to once or twice over the monitored period, depending on the parameter. At the end of the experiment, during the fall of 2001, the entire above ground portion of the macrophytes was harvested, oven dried (72 h at 60°C) and weighed. Carrots of soil were taken in T2 and T4 in 6 and 4 sampling points, respectively, along the basins to estimate belowground biomass and distribution of *Phragmites* within the SLG and SLG-P substrates. Analyses of variance (two-way-ANOVA) followed by multiple comparisons of means according to Tukey’s method were performed to test differences between treatments (combinations of plants and substrates).

**Results and discussion**

**Plant biomass production in relation to substrate pH**

*Typha* biomass was low in SLG substrate and *Phragmites* growth was twice more important in the SLG-P substrate than in the SLG substrate (Table 2). This difference could be attributed to the difference in pH between the substrates (Table 3).

In natural systems, *Phragmites* have been reported in soils with pH ranging from 3.8 to 8.5 and *Typha* from 4.7 to 11 (Grandtner, 1999; Lakshman, 1986). Despite the high proportion of granite in the substrate and more than one year of rinsing with an effluent of neutral pH, the pH in the T2 and T3 basins remained above 10 during the second growing season. This high pH was detrimental to *Phragmites* and *Typha* growth.

Above ground biomass production in the SLG-P was two fold greater than in the SLG (Table 2). Below ground biomass of *Phragmites* was also three fold greater in the SLG-P substrate than in the SLG substrate, confirming the importance of substrate quality on plant
productivity. Due to the presence of peat (pH of 4.2) in the unplanted SLG-P substrate, the soil pH rapidly decreased to 8.5 and then stabilised at 9.43 (average pH for summer 2001). In the planted basins with the SLG-P substrate, the pH fell further and stabilised at 7.45 (Table 3), apparently due to the presence of macrophytes. Plant effects on soil pH involve proton release by roots during nutrition, exudation of organic acids and production of carbohydrates, and CO2 release by roots. The bacterial and fungi life within the root zone may also affect pH. Finally, enhanced nitrification due to the presence of plants that bring oxygen under the water level may also contribute to a decrease in pH.

*Phragmites* biomass production reported in the literature ranges from 400 to 3,500 g m–2 yr–1 for above ground and 230 to 8,900 g m –2 yr–1 for below ground tissues (Hoffman, 1997; DeBusk and Ryther, 1986; Brix, 1993). While biomass production of *Phragmites* in the SLG-P substrate falls within this range, it was still below the maximum productivity even after the second growing season because of low nutrient concentration in the feeding solution. Nutrient deficiency was also reflected in the relatively high below ground to above ground biomass ratio (T2 B/A of 0.91 and T4 B/A of 1.34), which should be lower (0.6 to 0.7) in hypertrophic conditions, as is usually the case in CWSs (Hoffman, 1997). N and P content in the above ground tissues were lower than in the below ground tissues in both SLG and SLG-P due to nutrient translocation at the end of the growing season (Table 2). The lower plant nutrient content in the SLG-P unit may be due to a greater proportion of support or non-photosynthetic tissue.

### Table 2

<table>
<thead>
<tr>
<th>Treatment</th>
<th>AG Bm (g dw/m²)</th>
<th>AG Tot. N (%N/dw)</th>
<th>AG Tot. P (%P/dw)</th>
<th>BG Bm (g dw/m²)</th>
<th>BG Tot. N (%N/dw)</th>
<th>BG Tot. P (%P/dw)</th>
</tr>
</thead>
<tbody>
<tr>
<td>T2</td>
<td>947</td>
<td>0.80</td>
<td>0.068</td>
<td>858</td>
<td>1.47</td>
<td>0.18</td>
</tr>
<tr>
<td>T3</td>
<td>399</td>
<td>n.a.</td>
<td>n.a.</td>
<td>n.a</td>
<td>n.a.</td>
<td>n.a.</td>
</tr>
<tr>
<td>T4</td>
<td>1930</td>
<td>0.53</td>
<td>0.043</td>
<td>2583</td>
<td>0.93</td>
<td>0.21</td>
</tr>
</tbody>
</table>

### Table 3

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Temp °C</th>
<th>pH</th>
<th>TSS mg/L</th>
<th>COD mg/L</th>
<th>BOD₅ mg/L</th>
<th>TKN mg N/L</th>
<th>NH₄⁻ mg N/L</th>
<th>NO₃⁻ mg N/L</th>
<th>TP mg P/L</th>
<th>o-PO₄³⁻ mg P/L</th>
</tr>
</thead>
<tbody>
<tr>
<td>Inflow</td>
<td>12</td>
<td>6.6</td>
<td>187</td>
<td>373</td>
<td>104</td>
<td>12.41</td>
<td>1.39</td>
<td>0.99</td>
<td>2.69</td>
<td>1.78</td>
</tr>
<tr>
<td>T1 m</td>
<td>23</td>
<td>10.8</td>
<td>1</td>
<td>64</td>
<td>25</td>
<td>6.20</td>
<td>2.99</td>
<td>0.55</td>
<td>0.24</td>
<td>0.08</td>
</tr>
<tr>
<td>T1 p</td>
<td>23</td>
<td>9.4</td>
<td>2</td>
<td>176</td>
<td>33</td>
<td>7.43</td>
<td>5.38</td>
<td>0.42</td>
<td>0.38</td>
<td>0.16</td>
</tr>
<tr>
<td>T2</td>
<td>23</td>
<td>10.6</td>
<td>1</td>
<td>37</td>
<td>7</td>
<td>3.75</td>
<td>1.23</td>
<td>0.45</td>
<td>0.27</td>
<td>0.10</td>
</tr>
<tr>
<td>T3</td>
<td>23</td>
<td>10.7</td>
<td>1</td>
<td>45</td>
<td>8</td>
<td>4.48</td>
<td>2.34</td>
<td>0.50</td>
<td>0.25</td>
<td>0.09</td>
</tr>
<tr>
<td>T4</td>
<td>23</td>
<td>7.5</td>
<td>6</td>
<td>57</td>
<td>5</td>
<td>1.40</td>
<td>0.81</td>
<td>0.30</td>
<td>3.07</td>
<td>1.69</td>
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<tr>
<td>T4+4'</td>
<td>23</td>
<td>10.7</td>
<td>0</td>
<td>33</td>
<td>1</td>
<td>1.29</td>
<td>0.26</td>
<td>0.36</td>
<td>0.30</td>
<td>0.12</td>
</tr>
</tbody>
</table>

**Water balance and evapotranspiration (Et)**

Comparison between planted and unplanted units with identical substrate showed that between 87 and 92% of the water loss is due to plant transpiration (Figure 1). The control basins showed very little evaporation, mostly because subsurface flow reduces considerably the water-atmosphere exchanges. The SLG-P control (T1p) had a higher evaporation than the SLG control (T1m), probably because water-atmosphere contact was enhanced by the capillarity of the peat material. Within planted units, transpiration was related to plant biomass. As much as 56% of the influent water was lost by Et in T4+4’ (Figure 1). Given that evaporation in the unplanted T4’ basin was probably negligible, as in T1m, transpir-
tion in the first step (T4), where plant biomass was high corresponds to an average of 15 L m\(^{-2}\)d\(^{-1}\) or 1.9 m over the monitored period. Pollutant removal efficiency for systems with little water loss or gain is often expressed in terms of concentration ratio between inflow and outflow. The large proportion of water loss due to plant transpiration increases artificially the concentration of the effluent. Removal efficiency is thus better illustrated through mass balance of the pollutant, which is not dependent on water loss.

**Total suspended solids (TSS)**

TSS in the effluent were highly organic, 90% of them being volatile. With a loading of 505 g/m\(^2\) over the monitored period, TSS removal in all treatments ranged between 98 and 100% removal, on a mass balance basis (Figure 2). TSS removal was significantly higher in SLG substrates, planted or not, with an effluent TSS concentration barely detectable (Table 3). The slightly lower TSS removal in SLG-P substrates was probably due to peat degradation and the release of particles in the effluent.

Plants are reported to have a positive effect on TSS removal by reducing water velocity and by encouraging settling and filtration in the root network (Brix, 1997). After 1.5 years of operation, both *Typha* and *Phragmites* in SLG substrate did not show a positive effect on TSS removal compared to the SLG control probably due to the already high efficiency of the substrates for TSS removal or the young age or low growth of the macrophytes.

**Chemical and biochemical oxygen demand (COD/BOD\(_5\))**

The COD loading for the monitored period was 1,010 g/m\(^2\). Removal efficiency in all treatments varied from 55% to 96%. T1p was the least efficient treatment, highlighting the input of oxidisable molecules like humic acids and various organic compounds brought by peat degradation (Figure 2). Peat affects the COD removal efficiency enhanced by the productive plants in T4, resulting in a moderate efficiency. *Phragmites* (T2) were slightly more efficient than *Typha* (T3) in the SLG substrate although the difference was not significant. Once again, T4+4' gave the highest performance, probably because of its longer HRT resulting from having two beds in series.

With a loading of 281 g/m\(^2\) over the monitored period, BOD\(_5\) summer removal rates ranged from 76.5% to 99.5%. BOD\(_5\) removal in wetlands is due to physical and biological processes that involve sedimentation and microbial degradation, principally by aerobic bacteria attached to plant roots. As expected, the performance of the planted units was higher than that of the unplanted controls (T1m and T1p; Figure 2). The best result in BOD\(_5\) removal was achieved by T4+4’. This treatment had healthier macrophytes and a longer HRT, two factors contributing to improve BOD\(_5\) removal.

**Nitrogen**

During the monitored period, 33.5 g/m\(^2\) of TKN was added (TN load of 36.2 g/m\(^2\), or 0.366 g TN/m\(^2\)/d), resulting in a nitrogen removal ranging from 48% for T1p to 95% for T4.
(Figure 3). Nitrogen removal in constructed wetland systems is largely dependent on input loading rates. In field scale systems, efficiencies of up to 70% were reported (Faulkner and Richardson, 1989) for input rates ranging between 20–30 g TN/m²/y (0.055–0.082 g TN/m²/d), which is 4.4–6.7 fold lower than the loading rates in our study. On the other hand, Hammer and Knight (1994) reported that N removal rates of up to 79% could be achieved for loading rates up to 1,600 g TN/m²/y (4.383 g TN m⁻² d⁻¹), which is 12 fold higher than the rates in the present study.

As expected, nitrogen removal was highly correlated with the presence and productivity of plants. Plants provide favourable nitrification and denitrification conditions. There was a high proportion of nitrogen incorporated in the above ground biomass (21% in T2 and 28% in T4) because of the relatively low nitrogen loading. We found no difference in nitrogen removal between the two species in the SLG substrate.

All basins produced about the same amount of NO₃⁻ (Table 3), without any significant difference between the treatments, suggesting that anaerobic zones became established in all units and that denitrification was faster than nitrification, as is usually the case in SSF CWSs. Ammonification produced by the breakdown of organic nitrogen took place even in the SLG and SLG-P (T1p) controls for which the effluent NH₄⁺ concentration remained high (Table 3). We suppose that ammonia volatilisation may have been more significant in the SLG substrate due to its high pH.

**Phosphorus**

With a TP load of 7.3 g P/m² over the monitored period, the removal efficiency was above 86% for all treatments except T4 that gave a 38% removal on a mass balance basis (Figure

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**Figure 2** Removal efficiency on a mass balance basis for total suspended solids (TSS) and organic matter (COD and BOD₅). Treatments with the same letter are not significantly different, based on multiple comparisons of means using Tukey’s method.

**Figure 3** Percentage of removal on a mass balance basis for total Kjeldahl nitrogen (TKN). Treatments with the same letter are not significantly different.
4). The general pattern was similar with o-PO₄, with a load of 4.8 g P/m², and an efficiency above 91% for all units but T4 which gave 48%. Units containing steel slag but no peat were very efficient in P removal. The series (T4+4') gave the highest efficiency for o-PO₄ and TP mainly because of the T4' unit, given that T4 removed less than 50% of the added P.

The high phosphorus removal observed in this study was due to the high affinity of the slag and limestone substrates, which are more efficient under high pH conditions (Forget, 2001; Drizo et al., 2002). The lower removal efficiency in T4, especially compared to T1p, is due to the decrease in pH related to the presence of plants. The slightly lower pH in T1p (9.4) also affected the efficiency, at least for o-PO₄, compared to T1m, but to a much lesser degree than T4. There was no difference in TP or o-PO₄ removal in the SLG treatments, planted or not planted all being very efficient. Nonetheless, there was 8.9% and 11.4% of the P incorporated in the above ground biomass in T2 and T4 respectively. As for nitrogen, this high contribution of plant uptake in these treatments was attributed to the low phosphorus load.

**Conclusion**

This experiment showed that CWSs are effective in treating freshwater fish farm effluent despite its relatively low nutrients content. Pollutant removal was generally good under all different treatments. However, no specific combination of plant and substrate could maximise a one-step simultaneous removal of all evaluated pollutants. On one hand, organic matter (BOD₅, COD) and nitrogen removal was superior under strong plant cover. On the other hand, removal of phosphorus was better achieved by the substrates containing the highest proportion of EAF steel slag and limestone. Because of slag’s high pH, plant growth of both *Typha* and *Phragmites* was inhibited enough to reduce significantly their pollutant removal efficiency. Adding granite gravel to the SL substrate had no effect on its pH over the 1.5 years of monitoring. Adding peat to the SLG substrate lowered the pH enough to stimulate *Phragmites* growth but also affected P removal and TSS removal. Therefore, to treat fish farm effluent, we recommend that a horizontal subsurface flow CWS should be in two sequential steps, the first one being planted in a typical gravel bed (neutral pH) to remove SS, organic matter and nitrogen, followed by a second basin filled with steel slag and limestone without plants, as a polishing step for maximal P removal.

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References


