Start-up of a “zero-discharge” recirculating aquaculture system using woodchip denitrification, constructed wetland, and sand infiltration

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ABSTRACT

Recirculating aquaculture systems (RAS) discharge management limits the development of the aquaculture sector, because RAS do not automatically result in low nutrient emissions. Research has helped develop discharge management systems such as wetlands and woodchip bioreactors that have been adopted by Danish commercial model trout farms. To further develop the Danish concept, we have modelled and built a novel “zero-discharge” recirculating aquaculture system with an annual capacity of approximately 14 tonnes. The aim of this paper is to describe the entire concept and present the results from the start-up phase of the whole system. The concept includes the treatment of RAS effluent (overflow and sludge supernatant) using a hybrid solution of a woodchip bioreactor, constructed vertical wetland, and sand infiltration. Using this three-step process, the nitrate, phosphorus, and organic matter effluent are decreased to acceptable levels to reuse the water in the RAS process reducing the need for new raw water. In the first nine months of operation, a water treatment field was used as an end-of-pipe treatment to ensure the water was safe to recirculate for fish. During the winter, the water temperature dropped to 2.7 degrees in the sand filter, but the frost did not reach the water levels in any of the treatment processes. It therefore appears that a hybrid solution can operate sufficiently even in winter conditions. In the first year of operation, a woodchip bioreactor can remove 97 % of the nitrate, although the slow start-up of the RAS caused the bioreactor to be N-limited. On average, 79 % and 92 % of the inflow phosphate concentration was removed in the woodchip bioreactor and the entire hybrid treatment field respectively. The wetland and sand filter removed organic matter sufficiently (35 %), but because of the longer than designed actual water residence, it leached from the bioreactor more than was expected. Further experimentation is needed to identify the financial applicability and performance during higher feeding rates.

1. Introduction

Most of the aquaculture production in the Nordic countries takes place in sea cages and on flow-through farms, where it has not been economically feasible to treat phosphorus, nitrogen, and organic matter discharges (Silvenius et al., 2017; Pelletier et al., 2009; Grønroos et al., 2006). Land-based recirculating aquaculture systems (RAS) have been developed, in which a lower rate of water usage enables treatment of effluents and a lower fish farming environmental load (Piedrahita, 2003). However, designing well-functioning but cost-effective and low-maintenance treatment units is challenging for low-carbon wastewaters such as RAS effluents. This means that in many cases, the net removal of nutrients and organic matter has not been achieved as effectively as desired (e.g. Dalsgaard et al., 2013). High-technology RAS have been investigated and constructed as a solution, to which more water treatment steps have been added to achieve near-zero water exchange rates and diminish environmental impacts (Krom et al., 2014; Tal et al., 2009), but they have yet to be widely adopted commercially. In recent years, low-technology solutions such as woodchip denitrifying bioreactors (WB) and constructed wetlands have been applied.
successfully to the aquaculture industry, especially in treating the outflow effluent of RAS farms in Denmark (Dalsgaard et al., 2018; von Ahnen et al., 2018). Since cold climatic conditions challenge outdoor wastewater treatment, more studies and pilot testing are needed to obtain design parameters for cold climate regions.

Woodchip bioreactors have been widely used for nitrogen (N) removal in agricultural (Christianson et al., 2012; Greenan et al., 2009), forestry (Homýak et al., 2008), mining (Nordstöm and Herbert, 2018), and aquaculture effluents (von Ahnen et al., 2018). Nitrate (NO\textsubscript{3}) removal rates in WBs are generally low, being around 5–25 g NO\textsubscript{3} m\textsuperscript{-3} d\textsuperscript{-1} (Lepine et al., 2020; von Ahnen et al., 2018), compared to external carbon-fed bioreactors, where much higher nitrate removal rates (1.2–2.8 kg NO\textsubscript{3} m\textsuperscript{-3} d\textsuperscript{-1}) are reported (Letelier-Gordo et al., 2020; Dupla et al., 2006). However, the total cost of N removed in WBs has been estimated to be $2.4–15.2 per kg N (Lepine et al., 2018; Christianson et al., 2012), which is similar to the cost of substrate alone in external carbon-fed denitrifying bioreactors, $1.3–14.4 kg\textsuperscript{-1} N (Wang and Chu, 2016; Gutierrez-Wing et al., 2012). In addition, the use of WBs does not require potentially toxic chemicals, indoor facilities, or electricity. Because of their low cost and maintenance, woodchip bioreactors are an excellent choice for passive water treatment for the aquaculture sector.

To control carbon leaching and low dissolved oxygen concentration in the outflow of the WB, further treatment steps are needed to recirculate the water back to the fish culture tanks. A properly designed constructed wetland (CW) as one of the natural-based solutions can serve efficient physical, chemical, and biological nutrient and carbon removal processes, including the aeration of the treated water (e.g., Kadlec and Wallace, 2009; Vymazal, 2007). CWs have also generally been reported to be suitable in cold climatic conditions (Postila and Heiderscheidt, 2020; Heikkinen et al., 2018). Among the many different types of CW, the vertical flow CW remains one of the most effective at aerating the treated water (e.g., Cooper, 1999), especially using the step feeding method (Wang et al., 2020).

The managed artificial recharge of groundwater (MAR) is a method in which water is infiltrated through a layer of sand and gravel. During the process, dissolved and particulate organic matter is removed from the water (Kolehmainen et al., 2009; Lindroos et al., 2002). The process is widely used for drinking water treatment in the Nordic countries, and it has also been shown to remove organic matter from recirculating aquaculture effluents (Lindholm-Lehto et al., 2020; Lindroos et al., 2020).

To test these low-technology/passive water treatment methods for aquaculture in a cold climate, we have designed and constructed an RAS farm capable of feeding 14 tonnes per year, accompanied by an external hybrid water treatment field that includes a WB, a CW, and a sand infiltration unit (SF). By using this three-step approach, we aimed to remove acceptable levels of effluent nitrate, phosphorus, and organic matter to re-use the water in the RAS process and reduce the need for fresh intake water. When optimising the water treatment field, we hypothesised that sludge (total solids content approx. 1%) was the only outflow discharged from the system, which might further be recycled for other applications such as agricultural fertiliser. This paper presents the construction details and start-up phase of the whole system, and the first year of operation of the water treatment field. To avoid any damages, the start-up phase of the system was carried out carefully, and the full capacity of the water treatment field was not used.

2. Materials & methods

2.1. Recirculating aquaculture system

The RAS (FREA Aquaculture solutions, Denmark) was built in the research hall at the Laukaa fish farm of Natural Resources Institute Finland (Lake), where the climate is classified as Dfc (a snowy climate characterised by cold moist winters) in the Köppen climate classification system (Chen and Chen, 2013). Based on the 1981–2010 norm period, the region’s mean annual temperature is 4 °C, and the annual precipitation is 600 mm (Finnish Meteorological Institute, 2020). The typical snow cover duration is 147 days, whereas the length of the growing season is 165–175 days. Regularly occurring ground frost challenges year-round water treatment in outdoor conditions.

The RAS consists of two separate identical units (Fig. 1) that can be operated individually with or without an outdoor hybrid water treatment field (Fig. 2). Units can also be used as a joint system in which water circulates through a common pump sump, providing equal water quality for both units. Make-up water is taken from the oligotrophic Lake Peurunkajärvi (62.44886, 25.85201) using a peristaltic pump (Watson Marlow 630, Spirax-Sarco Engineering, UK). A similar type of pump is used to pump the RAS water into the hybrid water treatment field. From the water treatment field, the water is pumped into the research hall to an aerated reservoir tank with a volume of 3.2 m\textsuperscript{3}, from which the water can be further shared with the individual RAS units.

One RAS unit has two aluminium 5 m\textsuperscript{3} raceway fish tanks, which include a 1 m\textsuperscript{3} space for sludge cones collecting uneaten feed and settleable solids. After the fish tank, the water flows through a 60 μm mesh size drum filter (Hydrotech HDF800, Veolia, France), followed by two parallel 2.5 m\textsuperscript{3} fixed bed bioreactors (each filled with 1.5 m\textsuperscript{3} Saddle-Chips, KSK Aqua, Denmark), a 2.24 m\textsuperscript{2} degassing unit, and a 0.74 m\textsuperscript{3} pump sump. From the pump sump, the water is pumped (Flygt 3085, Xylem, USA) through a low-head oxygenator (FREA Aquaculture Solutions, Denmark) to the fish tanks. The total volume of one unit is approximately 25 m\textsuperscript{3}. The waterflow and degassing rates can be adjusted with frequency converters (Vacon 100 Flow, Grundfos, Denmark). An emergency oxygen diffuser located at the bottom of each fish tank is controlled by a monitoring system (Atlantic, OxyGuard, Denmark). A storing video surveillance system is installed to monitor the system’s operation. Energy consumption is measured by circulating pumps, aeration pumps, and drum filters using an energy meter (DSZ12E, Eltako, Germany). The system’s hydraulic head is 1.4 m. The RAS water temperature is adjusted by controlling the hall air temperature, and pH is adjusted by dosing a dissolved sodium bicarbonate mixture to the pump sumps (EJ-R, Iwaki, Japan). Fish are fed with a commercial feeding system (T Drum 2000, Arvo-Tec, Finland). Rainbow trout (Oncorhynchus mykiss) and European whitefish (Coregonus lavaretus) were reared in the system during the first year.

2.2. Sludge management

Sludge from the sludge cones (manually operated valve, controlled...
once per working day), drum filters (water level-based backwash), and the backwash of the four parallel fixed bed bioreactors (backwash approx. 3-week interval) are directed to a storage tank in a sludge container (Supplementary Fig. 1, Clewer Technology Oy, Finland). Sludge treatment is operated automatically in batch mode. The treatment process starts by pumping water from the storage tank to the coagulation tank. During the pumping, a coagulant agent is dosed. A flocculation chemical may be added, and the pH is adjusted in the coagulation tank. Sludge is separated by sedimentation and pumped to an additional peat bed or municipal water treatment plant, and the supernatant is directed to a storage tank in a sludge container (Supplementary Fig. 1, Clewer Technology Oy, Finland).

### Table 1

<table>
<thead>
<tr>
<th>Operating month</th>
<th>Average RAS feeding rate (kg d⁻¹)</th>
<th>Fresh water flow to the RAS (m² d⁻¹)</th>
<th>Water flow to the treatment field (m² d⁻¹)</th>
<th>Treatment field water flow to the RAS (m² d⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
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<td>10.8</td>
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</tr>
<tr>
<td>August ’19</td>
<td>1.3</td>
<td>7.3</td>
<td>Lake 40.0</td>
<td></td>
</tr>
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<tr>
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<td>3.8</td>
<td>RAS 4.3</td>
<td></td>
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<td></td>
</tr>
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<td>6.6</td>
<td>5.6</td>
<td>RAS 3.7</td>
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<td>7.4</td>
<td>3.6</td>
<td>RAS 7.0</td>
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</tr>
<tr>
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<td>6.7</td>
<td>RAS 5.1</td>
<td>1.2</td>
</tr>
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<td>8.3</td>
<td>4.7</td>
<td>RAS 5.6</td>
<td>3.1</td>
</tr>
</tbody>
</table>

### 2.3. Passive water treatment outdoor solution

In the first nine months of operation, the water treatment field was used as an end-of-pipe treatment to ensure the water was safe to recirculate for fishes. Gradually, the water-flow to the field was increased, and after nine months, some of the water was fed to the RAS (Table 1).

### 2.3.1. Woodchip bioreactor

A horizontal subsurface flow WB was constructed as the first unit in the hybrid passive treatment, followed by a vertical flow constructed wetland (CW) and a sand filter (SF) (Fig. 2, Supplementary Fig. 2). A trapezoidal WB with total dimensions of 14 (length) × 9 (width) × 1.5 m (depth) was designed based on the results of pre-testing in a column experiment at the Laukaa RAS research platform. Based on the pre-tests, a maximum nitrate removal rate of 33 g NO₃-N·m⁻³·d⁻¹ was achieved at a hydraulic retention time (HRT) of approximately 48 h (Kiani et al., 2020), which was further used to size and design the pilot-scale bioreactor unit as part of the hybrid solution. Inlet water was loaded from one side of the WB by placing two inlet perforated pipes (Ø 5 mm) with a length of 3.3 m and 4.6 m at the base, and 75 cm above the base. The maximum capacity of the bioreactor was designed to provide a 50 m³ saturated woodchip layer corresponding to a 1 m depth of active media and 0.5 m of dry woodchips on top. The purpose of the two-level inlet structures and the dry woodchip layer on top was to provide enough dry layer of woodchips to act as an isolator and avoid deep frost during the harsh Arctic winter. The outlet structure mainly regulates the water level in the woodchip bioreactor. However, the location of the inlet pipe has a significant effect on the flow path and HRT, which improves removal efficiency (Suliman et al., 2006). In winter, when more insulation is required, the use of a lower inlet pipe provides a deeper layer of insulation in the inlet zone along with a low water level in the outlet structure. Placing the inlet pipe at the bottom of the woodchips provides a longer flow path (higher HRT), which enhances the effect of low temperature on low removal efficiency (Suliman et al., 2006). In addition, in the summer, when the maximum capacity of the WB can be reached, two inlet pipes are used to distribute a high flowrate through the pore media by providing a longer HRT (Suliman et al., 2006). Setting the two inlet pipes at different heights forces most of the flow through the filter bed using the largest inlet zone. In its first year of operation, the volume of the WB used was 26.3 m³. This means that the average HRT was 5.0 days with a range of 2.3–8.5. Because of the slow start-up phase, this was longer than aimed for at full capacity (about 2 days). The treated water was collected with a perforated pipe (diameter 110 mm, length 3.3 m) at the end of the bioreactor and discharged into an adjustable outlet well, which controlled the water level inside the WB by adjusting the height of the PVC outlet pipe (Supplementary Fig. 3). A controlled outlet structure was designed to cope with the variation in the inflow rate, load, and desired HRT as important design criteria in the system. It was hypothesised that alkalinity increased during the denitrification process, further improving RAS performance and reducing pH.
control costs.

2.3.2. Constructed wetland

The constructed wetland was designed to remove reactive P and N, as well as to control biological oxygen demand (BOD) and the dissolved organic carbon (DOC) that can be leached from the WB. Furthermore, the purpose was to aerate water, because the water exiting the WB had a low dissolved oxygen concentration (DO). Based on the literature, the common reed (Phragmites australis) is one of the most commonly used plant species in different types of CW and is found to be efficient at removing nutrients (e.g. Tanner, 1996; Vymazal, 2011). In a gravel-based vertical flow, the common reed CW, oxygen-rich rhizosphere, and aerobic microbial degradation processes can also enhance BOD removal. It was hypothesized that a low BOD concentration in the outflow of the CW unit would improve the last SF unit, in which clogging and metal leaching at a very low DO level would be a problem.

The CW with a length/width ratio of 1.25 (7.5m × 6m) contained three gravel layers, with different particle sizes (Fig. 3) placed on the bentonite mat liner. The water was distributed to the CW using 50-mm perforated pipes located in the root zone of plants, and it was collected using perforated pipes with a 110-mm diameter in the drainage layer (20 cm) from the bases of the unit. The CW was designed using parameters selected from the literature: the BOD removal rate was 7 g BOD7 m⁻² d⁻¹, and the hydraulic loading rate of 278 mm d⁻¹ and water level were adjustable due to the outlet structure. In the first year of operation, the CW water-saturated volume was 41.3 m³ (average HRT 4.9 days, range 2.2–8.3 days). A dry mulch layer (20 cm) was used for soil-frost protection and placed only in the winter as part of the annual maintenance procedure required in cold climatic conditions.

2.3.3. Sand infiltration

Sand infiltration was designed to operate as slow sand filtration. The objective of this step was to achieve an infiltration rate of water corresponding to those used in Finnish artificial groundwater plants (ARG). In ARG plants, soil infiltration through gravel- and sand-containing sediments is used to remove dissolved organic carbon and suspended solids (Lindroos et al., 2020). The target water velocity was set at 1–4 m d⁻¹, and the retention time in the SF to ca. 10 d, when the passive water treatment field operated at an infiltration rate of 12.5 m³ d⁻¹. Based on RAS-related pre-studies (Lindroos et al., 2020) and the results from the ARG plants (Lindroos et al., 2002), a retention time of at least 10 days would mean a significant TOC reduction. The SF was designed to operate as a saturated water-flow, because this was assumed to result in a stronger DOM reduction than in an unsaturated vertical water-flow (Lindroos et al., 2020, 2002). Based on the retention time criteria, the optimal dimensions of the sand infiltration unit were determined by adopting a fluid flow and solute transport modelling approach (e.g. Muniruzzaman and Rolle, 2015, 2016; 2017; 2019). The latter was performed by considering the SF a water-saturated porous medium, and the transport of a conservative tracer was simulated to identify the optimal design that allowed a minimum residence time of 10 days (e.g. Rolle et al., 2013, 2018; Muniruzzaman et al., 2014, 2020; Sprocati et al., 2019).

The sand material for the SF was collected from the nearby esker area, which is composed of glaciofluvial sand and gravel sediments. The sand was relatively well sorted and classified as coarse sand (0.6–2.0

![Fig. 3. Cross-section of the constructed wetland. Mulch is only spread during the winter. The substrate includes sand and 10 % peat.](Image)
mm). The mean effective porosity (n_e) was determined to be 0.35 (0.32–0.37). The sand material was composed mainly of quartz and feldspars, and the proportion of dark mafic minerals was low.

The dimensions of the SF were: width 16 m, length 31 m, and depth 2 m. The total volume was 650 m³. During the starting period, the SF was operated at 165 m³ (average HRT 17 days, range 7.8–28.9).

All the water treatment field unit bottoms were lined with a bentonite mat, to prevent water leaching out of the system, and to enable accurate water balance estimation and purification for research purposes. The average HRT for the whole water treatment field was 26.8 days (ranging from 12.3–45.7 days).

2.4. Water quality analyses

Carbon dioxide (Franatech, Germany), dissolved oxygen concentration (Oxylsyr, s:can, Austria), and the water-flow rate (Fluxus FS01, Flexim, Germany) were measured online in the four fish tanks. The water pH (ProMinent, Germany), and the inflow water rate (Watson Marlow 630, Spirax-Sarco Engineering, UK) from Lake Peurunka and the water treatment field (Flexim) were measured online from both RAS units. The online measurement data were stored on an industrial computer (con:cube, s:can, Austria).

The total ammonia nitrogen (TAN), nitrite, nitrate, and phosphate were analysed on site using a spectrophotometer (Procedure 8038 Nessler, LCK341/342, LCK340, and LCK349 respectively, DS 3900, Hach, USA). Alkalinity was analysed on site using a standard titration method (ISO 9963-1:1994) (TitraLab AT1000, Hach, Loveland, USA). The total and dissolved organic carbon were analysed in a commercial laboratory in fresh samples (SFS-EN 1484). Oxidation-reduction potential (ORP), fluorescent dissolved organic matter (fDOM), pH, conductivity, and turbidity (FNU) were measured using an YSI EXO probe (Xylem, USA). The temperature was measured using a YSI EXO probe, and online using a HOBO Pendant (MX2201, Onset Computer Corporation, USA). Oxygen was measured using YSI EXO, YSI ProODO, and Ponsel OPTOD optical probes (Aqualabo Servises SA, France). The measurement intervals for the different water quality parameters are shown in Table 2.

Water quality from the sludge dewatering process was analysed in a commercial laboratory, and samples were frozen (-22 °C) before analysis (TSS and ash, SFS-EN 872:2005; COD, SFS 5504:1988; total nitrogen, EF2021; phosphate, EF2087; total phosphorus, SFS-EN ISO 17294-2; dry matter and ash, SFS 3008:1990).

3. Results

3.1. Removal rates and efficiencies

Due to the slow start-up phase and low inflow concentrations, most of the volumetric removal rates were lower than expected (Table 3). At their highest, the nitrate removal rate in the WB was 11.6 g N m⁻³ d⁻¹.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>VRR (g m⁻³ d⁻¹)</th>
<th>RE (%)</th>
<th>Inflow (mg l⁻¹)</th>
<th>Outflow (mg l⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Woodchip bioreactor</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NO₂⁻-N (n = 35)</td>
<td>4.1 (0.5–11.6)</td>
<td>97% (74–99%)</td>
<td>28.1</td>
<td>28.1</td>
</tr>
<tr>
<td>NO₃- (n = 35)</td>
<td>0.03</td>
<td>63%</td>
<td>0.21</td>
<td>0.21</td>
</tr>
<tr>
<td>TAN (n = 35)</td>
<td>-0.07</td>
<td>-225%</td>
<td>0.36</td>
<td>0.36</td>
</tr>
<tr>
<td>PO₄-P (n = 10)</td>
<td>0.12–0.40</td>
<td>79% (60–96%)</td>
<td>2.07</td>
<td>2.07</td>
</tr>
<tr>
<td>DOC (n = 5)</td>
<td>-9.39</td>
<td>-1186%</td>
<td>6.4 (4.7–8.8)</td>
<td>77 (37–110)</td>
</tr>
<tr>
<td>TOC (n = 5)</td>
<td>-9.79</td>
<td>-1030%</td>
<td>7.42</td>
<td>81.2</td>
</tr>
</tbody>
</table>

Concentrated wetland

| NO₂⁻-N (n = 34) | 0.02 | 29% (-13–100) | 0.65 | 0.47 |
| NO₃- (n = 32) | 0.00 | 28% | 0.02 | 0.01 |
| TAN (n = 35) | 0.03 | 32% (3–64%) | 0.89 | 0.61 |
| PO₄-P (n = 10) | 0.02 | 14% (140–49) | 0.42 | 0.24 |
| DOC (n = 5) | 4.32 | 65% (45–81%) | 37 (77–110) | 26 (12–41) |
| TOC (n = 5) | 3.39 | 52% (2–84%) | 81.2 | 37.8 (12–82) |

Sand infiltration

| NO₂⁻-N (n = 32) | 0.00 | 20% (-38–100) | 0.47 | 0.39 |
| NO₃- (n = 30) | 0.00 | -68% | 0.01 | 0.02 |
| TAN (n = 33) | 0.00 | 9% (-140–100) | 0.61 | 0.52 |
| PO₄-P (n = 10) | 0.00 | 34% (-67–94) | 0.24 | 0.12 |
| DOC (n = 5) | 0.32 | 57% (37–76%) | 26 (12–41) | 10.9 |
| TOC (n = 5) | 0.58 | 59% (31–78%) | 37.8 (12–82) | 13.3 (5.1–22) |

Total water treatment field

| NO₂⁻-N (n = 34) | 0.48 | 98% (91–100) | 28.1 | 3.9 |
| NO₃- (n = 35) | 0.00 | 82% (17–99%) | 0.23 | 0.12 |
| TAN (n = 33) | 0.00 | -84% | 0.04 | 0.02 |
| PO₄-P (n = 10) | 0.03 | 92% (78–99%) | 2.07 | 0.12 |
| DOC (n = 5) | -0.07 | -72% | 6.4 (4.7–8.8) | 10.9 |
| TOC (n = 5) | -0.09 | -80% | 7.42 | 13.3 |

Table 3: Average volumetric removal rates (VRR), removal efficiencies (RE), inflow, and outflow concentrations, and their ranges from the woodchip bioreactor, constructed wetland, sand infiltration, and total water treatment field.

Table 2: Measurement intervals from the water treatment field (W = weekly, B = biweekly, M = monthly, O = online). TAN = total ammonia nitrogen, TOC = total organic carbon, DOC = dissolved organic carbon, ORP = oxidation-reduction potential, fDOM = fluorescent dissolved organic matter.

<table>
<thead>
<tr>
<th>Operating month</th>
<th>TAN, Nitrite, Nitrate</th>
<th>Alkalinity</th>
<th>PO₄-P</th>
<th>TOC, DOC</th>
<th>Temperature, Oxygen</th>
<th>ORP, pH, fDOM, Conductivity, Turbidity</th>
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<tbody>
<tr>
<td>July '19</td>
<td>W</td>
<td>W</td>
<td></td>
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<td>M</td>
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<td>O</td>
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(HRT = 2.3 d), and the phosphate removal rate was 0.4 g P m⁻³ d⁻¹ (HRT = 3.8 d). The WB was the most efficient for N removal, while the CW was the most efficient for carbon, as planned. The total ammonia nitrogen (TAN) was released from the WB, but the CW could remove 32 % and SF 9%.

3.2. Water quality changes

The total ammonia nitrogen concentration increased during the first 10 months of operation in the outflow of the SF (Fig. 4), and the concentration was higher in the outflow of the entire hybrid water treatment field than in the inflow. An unexpected nitrite peak was observed in the RAS, but it was almost completely removed during the water treatment process (Fig. 4).

During the coldest part of the winter (January–March), phosphate leaching was observed (average inflow 0.22 mg l⁻¹ and outflow 0.28 mg l⁻¹, Fig. 5A). During the initial start-up period with lake water, conductivity increased after all treatment units (Fig. 5B). After the WB, pH decreased substantially: it was below 6 during the first five months of operation, but it increased in the CW and in the SF (Fig. 5C). Due to denitrification and possible sulphate reduction, alkalinity increased in the WB by 89 %, and 19 % in the CW (Fig. 5D). Some turbidity peaks were observed in all treatment units, but in general, turbidity increased in the WB, and decreased in the CW and SF (Fig. 5E). Between July and February, the WB released some dissolved organic matter (DOM), but an average of 35 % of it was removed in the CW and SF (Fig. 5F).

Inflow water was saturated with oxygen, and it was consumed in the WB. However, oxygen was also depleted in the CW and SF, which had reducing conditions (Fig. 6). The water temperature in the treatment system showed some seasonal trends, with few buffer effects against the local air temperature, and below-zero temperature values were not recorded at all (Fig. 7). During the first year of operation, the lowest water temperature was 4.2 °C in the wetland unit (CW) and 2.6 °C in the SF, although the average air temperature was -0.8 °C between November and March (Fig. 7). The winter was unusually mild but long compared to the average Finnish winter. The highest frost depth was 27 cm in the WB, whereas it was somewhat less in the CW (15 cm) (Table 4). In the SF, the frost depth was recorded as deeper (62 cm) than in the control sampling point (46 cm) outside the water treatment area (Fig. 2). This was expected, because it is common that soil frost is typically deeper in sandy soils than in e.g. clay due to its higher heat conductivity and lower heat capacity characteristics (e.g. Rajaei and Baladi, 2015).

3.3. Sludge management

An average of 328 l of sludge from the sludge cones, drum filter backwash, and fixed bed bioreactors was produced per kg of feed. On average, 99 % of the total phosphorus and phosphate, and 86 % of total nitrogen, were removed in the coagulation process (Table 5). Thirty per cent of the water fed to the water treatment field was from the sludge supernatant.

4. Discussion

4.1. N balance

The water treatment field was able to remove nitrogen, phosphorus, and organic matter efficiently throughout the year. The RAS start-up phase took longer than expected, which led to low loading to the water treatment field, and the water treatment field did not operate at its full purification capacity. The fish feeding rate was 10.1 kg d⁻¹ at its peak, and the woodchip bioreactor (WB) operated at its lowest capacity of 21 kg feed per day (53 % of the designed full capacity). Due to the low feed rate, the WB was N-limited, because the effluent nitrate concentration was mostly below 1 mg l⁻¹ (Robertson, 2010; von Ahnen et al., 2018v). The highest measured nitrate removal rate of 11.6 g NO₃-N m⁻³ d⁻¹ is similar to previously reported rates (5.3–25 g N m⁻³ d⁻¹) in pilot or full-scale WBs (Lepine et al., 2020; von Ahnen et al., 2018v; 2016). However, higher removal rates can be expected when the nitrate load increases, because the removal rate has been found to be positively correlated to the inflow concentration in the pre-testing phase for this full system (Kiani et al., 2020). There are also previous studies in which such a correlation was not observed (Robertson, 2010). During the coldest part of the winter (February–March), the removal rate was somewhat lower than during other months, being an average of 4.0 g N m⁻³ d⁻¹, but the removal efficiency remained high (99 %). The results proved that the WB also had a high capacity to remove nitrate during winter conditions. It should be noted that the studied winter was mild, and further studies are needed to investigate how colder air temperatures such as those below -10 °C affect nitrogen removal efficiency.

Some ammonia was released from the WB, and this was probably caused by dissimilatory nitrate reduction to ammonium (DNRA), which is possible in conditions in which nitrogen limits denitrification instead of carbon (Ju et al., 2013; Zhao et al., 2018; Kiani et al., 2020). Some of this ammonia was removed in the CW, and the removal efficiency is expected to be higher when vegetation is fully grown and the microbial communities have matured, because they have been found to improve N

Fig. 4. A) nitrate, B) total ammonia nitrogen, and C) nitrite concentrations from the inflow to the woodchip bioreactor, outflow from the woodchip bioreactor, outflow from the constructed vertical wetland, and outflow from the sand infiltration unit (DD.MM.YY).
4.1. N removal processes in CWs (e.g. Zhang et al., 2016). Overall, TAN concentration was higher after the treatment field than in the inflow to the water treatment field. This was caused by the DNRA in the WB, as well as the potential mineralisation of organic matter trapped in the different water treatment compartments. In the RAS context, this will cause elevated nitrification demand in indoor treatment steps if oxygen depletion continues, and nitrification is disabled in the field.

4.2. P balance

A high removal efficiency of phosphate (80 %) was observed in the WB, which was not observed in previous studies of the WB treatment of aquaculture effluent. Lepine et al. (2020) reported a minor production of dissolved reactive phosphorus from the WB, and von Ahnen et al. (2016) did not observe a net release or net production of phosphate from the start-up period of the woodchip bioreactor. When using agricultural drainage, phosphate reduction was observed (Hua et al., 2016), which might be explained by the intake of phosphorus by the growing bacterial biomass, as well as extracellular polymeric substances that can absorb phosphate (Li et al., 2015). This may be related to the high hydraulic retention time in the WB (between 2.5 and 5.7 days), which enhanced the woodchip/wastewater contact time (Sharrer et al., 2016). In addition, the inflow phosphate concentration in the WB decreased in the first six months of operation, which might be explained by the biofilm growth and maturation in the RAS, which assimilated some of the phosphorus released from the fish faeces.

The sludge dewatering process effectively removed phosphorus and nitrogen. However, the dewatering process requires further optimisation, because the relative amount of sludge leaving the process was still fairly high at 114 l kg\(^{-1}\) feed. There was a large variation in the
properties of the sludge, which affected the quantity of solids settling in the coagulation tank. The gradient between clear supernatant water and thick sludge was not automatically detected, so as a precaution, more water was fed to the municipal water treatment plant, because sludge escaping to the water treatment field might have caused the WB to be clogged.

### 4.3. Organic matter and turbidity

The main purpose of the slow sand infiltration was the removal of organic carbon and suspended solids, as well as the decrease in turbidity. During the first seven months of operation, it was able to remove 36% of the organic matter (fDOM) when it was operated at an HRT of 19 days. After the flow rate was increased to an HRT of 10 days (7.8–10.3), the organic matter removal efficiency dropped to 6%. It has been reported that the removal of organic matter from the infiltration water is based on the physical filtering and chemical retention processes in the soil particles and biological degradation of natural organic matter in the artificial recharge of ground water, which is widely used in Finland for household water production (Kolehmainen et al., 2009; Lindroos et al., 2002). In ARG, lake water is infiltrated through sand- and gravel-containing soil layers to remove TOC from the infiltration water, and the sand filtration has also been shown to remove TOC from the RAS effluents in small-scale experiments (Lindroos et al., 2020). The molecular size distribution of TOC is also reflected in the TOC removal; small molecular TOC fractions are not removed as effectively as large molecular fractions (Kolehmainen et al., 2009; Lindroos et al., 2002). All these processes are undoubtedly partly responsible for the organic matter removal in the SF.

There are several possible reasons for fDOM removal being weaker during the spring and summer of 2020 than 2019. The fDOM values in wetland outflow dropped in the spring and summer of 2020 on two occasions, and the SF outflow did not respond to this drop as strongly. After the decline, the fDOM values soon increased again in the wetland outflow. Generally, the oxygen concentration was low in the wetland outflow and the SF. Oxygen is consumed by the biodegradation of organic matter, and it is likely that the higher oxygen content in the SF in 2019 contributed to the better organic matter removal during that period. The possible accumulation of organic matter in the SF may also have affected the removal efficiency of organic matter from the infiltration water. Turbidity decreased significantly in the SF, as has also been reported in small-scale RAS experiments (Lindroos et al., 2020).

### 4.4. Future perspectives and challenges

The low oxygen concentration in the water entering the CW and SF appeared to be the main challenge when treating RAS effluent. Winter conditions mean that the distribution pipes need to be placed underground to prevent freezing when the oxygenation of water is limited. In addition, snow and soil frost decrease the transport of oxygen from the atmosphere, posing challenges for water oxidation. However, when vegetation and microbial communities in the CW are fully grown, it is expected that the water oxygen level will improve, because the common reed has a good capacity to enhance microbiological activity (e.g. Gagnon et al., 2007) and increase the oxygen level in the media (e.g. Nivala et al., 2013). In addition, the high reducing conditions in the SF caused the dissolution of potentially toxic metals such as arsenic (As), cobalt (Co), and manganese (Mn) in the water (manuscript in progress). However, these metals can be converted back to solid form by vigorous aeration and recovered in the fixed bed bioreactor. Artificial aeration or recirculation of water within the CW unit has been found to be efficient at improving removal processes (Poldvere et al., 2005; Ji et al., 2020), and they may be solutions for increasing the oxygen level in the outflow of the CW and preventing metal leaching in the SF.

In the start-up phase, waterflow rates and the N load on the WB were limited, which led to much longer HRT than planned. Due to this, a high amount of organic matter leached from the WB, further aggravating the oxygen depletion in the CW. To fully control the removal rates of nitrogen in the WB, the water level adjustment wells need to be redesigned, because the outlet wells were operated already operating at their lowest level. This means that there was no option to shorten the HRT with the available flow-rate. Experience has shown the importance of proper HRT and the adjustability of the system hydraulics for efficient removal rates and the prevention of carbon leaching from the woodchips.

When designing a full-scale hybrid water treatment field, it would be
useful to look for sites where water can be distributed between units by gravity. In addition, a sand filter requires hundreds of cubic meters of sand, so it should be built in areas with natural sand deposits. It must also be carefully examined whether the CW could achieve sufficient P and OM removal, making the SF unnecessary.

Author statement


Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

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References


