Nitrifying trickling filters and denitrifying bioreactors for nitrogen management of high-strength anaerobic digestion effluent

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The treatment of high-strength anaerobic digester effluent in laboratory-scale trickling filters for nitrification and then anaerobic filters for denitrification is reported. Five media types were investigated in the trickling filters: biochar, granular activated carbon (GAC), zeolite, Pall rings, and gravel. Three media were tested in five denitrifying filters: sand (S), bamboo wood chips (B), eucalyptus wood chips (E), bamboo with sand (B+S), and eucalyptus with sand (E+S). The different wood chips served as a supplemental electron donor for denitrification. From six months of operation, biochar, GAC, zeolite, Pall rings, and gravel media had turbidity (NTU) removal efficiencies of 90, 91, 77, 74, and 74%, respectively, and ammonia removal efficiencies of 83, 87, 85, 30, and 80%, respectively, which was primarily by nitrification to nitrate. For the anaerobic filters, S, B, B+S, E, and E+S had nitrate removal efficiencies of 30, 66, 53, 35, and 35%, and turbidity removal efficiencies of 88, 89, 84, 89, and 88%, respectively. Biochar and bamboo were selected as the best combination of media for trickling filter and anaerobic filter sequential treatment. Based on an average initial influent of 600 mg NH₃-N L⁻¹, 50 mg NO₃-N L⁻¹, and 980 NTU, the biochar filter’s effluent would be 97 mg NH₃-N L⁻¹, 475 mg NO₃-N L⁻¹, and 120 NTU. The bamboo filter’s final effluent would be 82 mg NH₃-N L⁻¹, 157 mg NO₃-N L⁻¹, and 13 NTU, which corresponds to 63% removal of total N and 99% removal of turbidity. These filter media thus present a simple option for sustainable post-treatment for nitrogen management and effluent polishing in low-resources settings.

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1. Introduction

The need for onsite sanitation and fecal sludge management is considerable as 2.7 billion people worldwide currently rely on these technologies, and the number is expected to grow to 5 billion by 2030 (Strande, 2014). Onsite sanitation is desirable as the centralized sewer-based collection and treatment systems existing in developed nations are too costly, too complex, and use too much energy to implement in poor and less developed countries (Lalander et al., 2013; Mara, 2013). While influent concentrations at conventional sewage treatment plants are generally in the range of $300-2000 \text{mcg L}^{-1}$, $<1$% TSS, and $50-250 \text{mg LN L}^{-1}$, fecal sludge reaching onsite treatment systems can have as much as $30,000-100,000 \text{mcg L}^{-1}$, 3–7% TSS, and $3000-7000 \text{mcg LN L}^{-1}$ (where COD is for chemical oxygen demand and TN is total nitrogen) (Strande et al., 2014). Further, most existing onsite sanitation solutions (e.g. anaerobic digestion, stabilization ponds, drying beds, etc.) only consider organic removal and pathogen destruction but do not address nutrients. In conventional sewage treatment, nutrient removal is a cumbersome, expensive step. Therefore, there is a need for alternative methods for efficient onsite nutrient recovery or removal which require minimal, if any, external resources.

Application of trickling filters (TF) for the post-treatment of anaerobic reactors handling sewage is a relatively recent interest (Chernicharo and Nascimento, 2001; Tandukar et al., 2006; von Sperling and Chernicharo, 2005). Trickling filters can achieve biochemical oxygen demand (BOD) and solids removal as well as nitrification, the biological oxidation of ammonia to nitrite and nitrate (Daigger and Boltz, 2011). Trickling filters are therefore worth consideration for onsite treatment.

Studies on trickling filter for post-an aerobic digestion treatment are limited (Khan et al., 2011), Chernicharo and Nascimento (2001) operated a trickling filter filled with 4–6 cm furnace slag at organic loading rates of 0.3–3.9 \text{kgBOD m}^{-3} \text{d}^{-1}. This filter treated effluent from a UASB and reached BOD removals in the range of 50–60% but authors did not monitor NH3 removal. Using pebbles with an effective size of 5.6 mm for the treatment of anaerobically digested dairy manure, Xia et al. (2012) achieved average removal of 71% of soluble BOD (sBOD) and 75% NH3-N for influent containing 692 \text{mgBOD} m^{-3} and 1210 mg NH3-N L^{-1}. Vieira et al. (2013) used a novel TF design to achieve 32% BOD and 31% NH3-N removal from UASB effluent starting with 72 mgBOD L^{-1} and 29 mg NH3-N L^{-1} under OLR and HLR of 0.08 kgBOD m^{-3} d^{-1} and 1.1 m^{3} m^{-2} d^{-1}, respectively. In a downstream hanging sponge (DHS) reactor, a setup similar to TF, Tandukar et al. (2006) found 83–87% BOD removal and 70–73% NH3 removal (inlet concentration up to 40 mg NH4-N L^{-1}) while operating at 0.67 to 1.24 kgBOD m^{-3} d^{-1}, respectively. Most recent work with this reactor type achieved removals of 87% and 83% of BOD and NH3-N, respectively at a loading of 1.11 kgBOD m^{-3} d^{-1} with influent concentrations of 93 mgBOD L^{-1} and 24.6 mg NH4-N L^{-1} (Onodera et al., 2014). It should be noted that most of these previous studies dealt with domestic sewage where influents contained less than 50 mg NH4-N L^{-1} whereas treating anaerobic digester effluent from onsite fecal sludge treatment will require handling NH3-N concentrations on the order of 1–5 g L^{-1}.

Ideal media characteristics for TF are high specific surface area for biofilm attachment, high porosity to prevent clogging and allow for easy air flow, low cost, and long durability (Tchobanoglous et al., 2003; Daigger and Boltz, 2011). Rocks have been traditionally used though today, plastic media have become more common allowing high organic loadings, and effective biofilm wetting and aeration. Zeolite is a medium of interest in trickling filters due to its adsorption properties. Aiyuk et al. (2004) used a zeolite column following a UASB reactor treating domestic wastewater and achieved 99% removal of NH4. The proposed ammonium removal mechanism was ion exchange as interstitial spaces in the zeolite framework allow replacement of Na+, K+, and Ba2+ cations with NH4. One interesting finding was the ability to recharge the zeolite's ion exchange capabilities through nitrification and that the recharged zeolite displayed similar performance as fresh zeolite (Aiyuk et al., 2004). The process of adsorption, biological degradation, and nitrification was later explored by various investigators (Luo et al., 2014; Masunga et al., 2007; Yidong et al., 2012). In particular, Luo et al. (2014) used zeolite in a trickling filter with influent concentrations of 265 mgCOD L^{-1} and 41.1 mg NH4-N L^{-1} and achieved 94.7% and 90.4% removal, respectively.

The use of granular activated carbon (GAC) as a biofilm carrier was introduced in drinking water treatment and showed improvements over adsorption-only processes in that media lifespan was extended such that it did not require regeneration (Servais et al., 1994). In greywater trickling filter applications, Dalahmeh et al. (2012) achieved 97% BOD and 98% total nitrogen reduction of an artificial greywater containing 425 mgBOD L^{-1} and 75 mg total N (TN) L^{-1}. There are no known studies of the use of GAC as a trickling filter media for domestic wastewater.

Biochar is a medium of interest due to its physical characteristics, sustainable production, and global availability. Biochar presents a low cost alternative to GAC. Its physical properties are highly variable depending on substrate and pyrolysis temperature with ranges for surface area and bulk density of 50–500 m^{2} g^{-1} and 1.5–2.0 g cm^{-3}, respectively (Downie et al., 2012). In the treatment of artificial greywater with 1390 mgBOD L and 95 mg TN L^{-1}, Berger (2012) found 99.2% and 99.1% COD and 96.6% and 90.9% TN removal efficiencies with GAC and biochar, respectively. Li et al. observed similar performance with biochar and achieved up to 91.4% removal of NH4-N under loading concentrations of 100–150 mg NH4-N L^{-1} while operating at a 72 h retention time (Li et al., 2016). Like GAC, there are no known studies of the use of biochar as a trickling filter media for domestic wastewater.

The above review suggests that treatment of digester effluent treatment in trickling filters shows great promise for reducing organic and solids content while oxidizing ammonia into nitrate. This process should improve the effluent quality to make it more likely to be reused as a fertilizer (cleaner, and nitrate being more desirable than ammonia). Additionally, the trickling filter effluent could be denitrified if total nitrogen removal is necessary, making it possible to consider various water reuse scenarios. Thus, potential low-cost means for denitrification of nitrified digester effluent are discussed next.

The nitrate and nitrite produced from nitrification of anaerobically digested fecal sludge can be removed by denitrification, the anaerobic biological reduction of nitrate and nitrite to N2 gas. Submerged fixed beds have been successfully used for denitrification. The denitrification process requires an electron donor which can be residual BOD or provided extraneously. Generally the electron donor is the rate-limiting substrate, and a requirement of 4 g BOD/g NO3-N is normally assumed for complete denitrification to nitrogen gas (Rittmann and McCarty, 2001). Effective trickling filter operation is expected to remove COD; however, particulate and slowly biodegradable COD that passes through the trickling filter may be utilized for denitrification (Nakhla and Farooq, 2003). An exogenous electron donor may be necessary if not enough BOD remains in the stream undergoing treatment. Methanol and acetate are commonly supplied as electron donors in conventional wastewater treatment plants, but this practice would probably be cost- and resource-prohibitive for onsite fecal sludge treatment.

An alternative method of providing an electron donor is by using a carbon-based filter medium. The use of wood chips as a denitrifying filter media has gained attention recently, typically for...
stormwater and agricultural drainage (Addy et al., 2016; Bock et al., 2015, 2016; Cameron and Schipper, 2010; Christianson et al., 2017; Lynn et al., 2015a, 2015b; Pluer et al., 2016; Schipper et al., 2010c; Schipper et al., 2010a). These studies have relied on woodchips to support denitrification of waste streams with up to 30 mg NO₃-N L⁻¹ and at rates of 2–22 g N m⁻³ d⁻¹ (Schipper et al., 2010b). The highest nitrate loading concentration and removal rate of these studies was found by Lepine et al. (2016) at 80 mg NO₃-N L⁻¹ and 39 g N m⁻³ d⁻¹, respectively. Two studies evaluated the use of wood chips in denitrifying onsite sewage effluent (Rambags et al., 2016; Robertson et al., 2005) with similar influent nitrate loading (~30 mg NO₃-N L⁻¹) and removal (87–99%). In an evaluation of alternative nitrogen management technologies for onsite sewage treatment, a single simple pass sand filter followed by a wood chip biofilter outperformed more complicated technologies (Oakley et al., 2010).

Still, few studies have evaluated wood chips for nitrogen treatment of onsite sewage. These studies have experimented with different types of wood chips from hardwoods or softwoods such as eucalyptus, pine, willow, or mixed, but none so far has used bamboo. Bamboo is a wood source that is readily available in tropical regions of the world and can be sustainably grown. Study on three types of woodchips (tropical hardwood, eucalyptus, bamboo with sand (B+S), and eucalyptus with sand (E+S)), respectively, to a depth of 50 cm (total volume of 4.0 L). Mixed wood chip and sand columns were filled in 10 cm increments until reaching the full height of 50 cm. Wood chips were added first, and then sand was added while shaking the column to minimize heterogeneous void space.

Media physical characteristics were determined through a sieve analysis on a mechanical shaker at 30 rpm for 10 min. Results are shown in Table 1 and in Supplementary Information Table A1. Each medium was passed through 6.3, 4.75, 3.35, 2.36, and 1.18 mm meshes to determine effective grain size (Dₑ) and uniformity coefficient (D₉₀/D₁₀). Bulk density, particle density, and porosity were determined for each column. Bulk density (ρₑ) was calculated as the mass of media filling the column volume, and particle density (ρₚ) was determined using the volume of deionized water displaced by 25 g of each sample. Porosity (p) was calculated with equation (1).

\[
p = 100 \times \left(1 - \frac{\rho_p}{\rho_e}\right)
\]

2.2. Fecal waste treated

Minimally diluted anaerobically digested human fecal sludge was not available in sufficient quantities for these experiments. Thus, a suitable simulant was sought. Expected BOD and total nitrogen concentrations are 1–5 gBOD L⁻¹ and 1–3 g N L⁻¹. The effluent from an intermittently aerated basin treating anaerobically digested swine manure system was selected as a mimic for human fecal digestate (Xu et al., 2016). Previous monitoring of this system had shown effluent characteristics to be in the target range for COD and TN; however, further analysis revealed that most of the COD was recalcitrant (COD:BOD = 5:1) which may have resulted in low heterotrophic activity in the trickling filters and low availability of electron donor for denitrification in the anaerobic filters. The effluent was known to contain both nitrifying and denitrifying bacteria and was used to inoculate the trickling and submerged filters. During the experiment, the swine waste was added directly to the trickling filters. The effluent from all trickling filters was pooled together and collected in a 5 L flask that was continuously mixed, and this liquid was used as the influent for submerged
Each trickling and anaerobic filter was fed 1.35 L d\(^{-1}\) using peristaltic pumps connected to timers. Feeding was applied at 27 mL min\(^{-1}\) for 5 min at the beginning of each chosen hour from 6:00–8:05, 11:00–13:05, and 17:00–20:05. This feeding pattern was an approximation of the expected daily flow pattern in an on-site fecal sludge treatment system. The daily average hydraulic loading rate (HLR) on each filter was 0.17 m\(^3\) m\(^{-2}\) section d\(^{-1}\), the organic loading rate (OLR) was 0.38 and 0.23 kg COD m\(^{-2}\) media d\(^{-1}\) for the trickling filters and the anaerobic filters, respectively and nitrogen loading rate was 0.3–1.0 g m\(^{-2}\) media d\(^{-1}\). Empty bed residence time (EBRT) was 6 days for the trickling filters and 3 days for the submerged filters. However, the trickling filters have a liquid hold-up of about 5–10% and thus the true residence time in the TFs is more in the range of 7–15 h. These design characteristics were chosen so that a conservative estimate for performance (assuming a media surface area of 500 m\(^2\) m\(^{-3}\) would remain under previously established maximum operational parameters for simultaneous nitrification and BOD removal: 80 m\(^2\) m\(^{-3}\) d\(^{-1}\), 0.5 kg BOD m\(^{-3}\) d\(^{-1}\), and 1 g N m\(^{-2}\) media d\(^{-1}\) (Tchobanoglous et al., 2003). Due to the low flow rate, a uniform distribution of the substrate at the top of the filter was not achieved. However, uniform liquid distribution was achieved within the depth of the media and was confirmed in hydraulic tests conducted with water. Filters were wetted with tap water for two weeks before substrate addition. Filters were operated at ambient laboratory temperatures (20 ± 2°C).

Swine farm effluent used as substrate was undiluted for the first 8 weeks of operation (Period I). The high concentration of suspended solids (approximately 2000 mg L\(^{-1}\)) contributed to operational issues (plugging and issues with pumping thick slurries), thus the substrate was diluted 1:1 with tap water for the remainder of the study (Periods II and III). Average COD and TN concentrations during these periods were 3040 and 1830 mg COD L\(^{-1}\) and 1790 and 790 mg N L\(^{-1}\), respectively. The denitrifying filter study began after the trickling filter study (day 225, Period II) and ran for 120 days. Operating conditions did not change for the denitrifying filters, so this study is not divided into periods as in the trickling filter study.

### 2.3. Analytical and experimental protocols

The true hydraulic retention time was determined in both reactor types from tracer studies using conductivity probes (Vernier, Beaverton, OR). A step change from water to trickling filter substrate was used due to high conductivity of substrate (~20,000 µS/cm). Conductivity probes were placed in the low volume vessels (~50 mL) that collected the TF effluent before it overflowed into the secondary container. Substrate for submerged filters had a lower conductivity, and thus a NaCl solution (20,000 µS/cm) was pumped into these filters for one day. Conductivity was measured in the low volume vessels (~50 mL) that collected filter effluent before it left the system. The retention time in the trickling filters was determined as the time at which 50% of the peak conductivity was measured. In the submerged filters, retention time was determined as the time in which 50% of the tracer input was recovered in the effluent.

Samples were taken weekly for the study duration from the outlet of each filter and at the mid-depth of the submerged filter. Total NH\(_3\)-N, NO\(_2\)-N, NO\(_3\)-N, and COD were measured using Hach kits. Dissolved oxygen (DO) and pH were measured using digital meters (Hach HQD portable meter with Intellitcal LDO101 DO sensor, Loveland, CO; and Oakton Instruments Ion 510 series pH meter, Vernon Hills, IL; respectively), and turbidity was measured using EPA Method 180.1. Periodic tests of BOD\(_5\) and total suspended solids were performed according to EPA Methods 405.1 and 160.2, respectively.

Filters were not backwashed during this study. After 260 days (end of Period II), plugging at the top of the trickling filters had become a regular issue. Feeding was stopped at this time, and media was removed from each filter and washed with tap water. Period III began at this time (day 274). In Period III fresh biochar (BC-new) and fresh GAC (GAC-new) replaced Zeo and PR filters while washed BC, GAC, and G media were reused. Operating conditions in Period III were the same as in Period II.

COD release from wood chips was evaluated in both a batch and in a column experiment. For the batch experiment, 10 g of either bamboo or eucalyptus wood chips were added into 250 mL flasks filled with 100 mL DI water, 100 mL trickling filter effluent, and 1 mL of sludge for inoculation. Separate flasks were filled with the same solution without wood chips as a control. All flasks were prepared in triplicate and incubated on a shaker table at room temperature (22 °C) without exposure to light. Liquid samples were taken with a syringe (2 mL) to measure COD and NO\(_3\)-N concentrations in the solution. A manometer was used to measure the change of pressure in the headspace. Samples were taken at 12 h, 1, 2, 3, and 6 days and then weekly after the start of the experiment. A COD mass balance was performed to determine how the COD released from wood chips was dissolved into solution, consumed by denitrifiers, or used for CH\(_4\) production as measured in the headspace. The total COD release was calculated using the following equations:

\[
\text{COD}_{\text{solution}}[\text{g}] = V_{\text{solution}}[L] \times \text{COD}_{\text{measured}}[\text{g L}^{-1}] 
\]

\[
\text{COD}_{\text{NO3}}[\text{g}] = V_{\text{solution}}[L] \times (\text{NO3}_{\text{initial}} - \text{NO3}_{\text{final}}) [\text{g L}^{-1}] \times 4 \times \text{g COD g NO3}^{-1}
\]
\[
\text{COD}_{\text{CH}_4}(g) = \%\text{CH}_4 \times \frac{P[Pa] + V_{\text{headspace}}[m^3]}{R[m^3 Pa][K/mol] + T[K]} \times 64 \left[ \frac{g \text{ COD}}{\text{mol CH}_4} \right]
\]  (4)

\[
\text{COD}_{\text{total}}(g) = \text{COD}_{\text{solution}}(g) + \text{COD}_{\text{NO}_3}(g) + \text{COD}_{\text{CH}_4}(g)
\]  (5)

where \(V_{\text{headspace}}\) is the volume of solution on the day of sampling, \(P\) is pressure of gas in headspace, \(\%\text{CH}_4\) the fraction of methane in the headspace, \(V_{\text{headspace}}\) is the volume of gas headspace on the day of sampling, \(R\) is the ideal gas law constant \(8.314 [m^3 Pa]/[K/mol]\), and \(T\) is temperature in Kelvin \((295 \text{ K})\). The conversion of \(g \text{ NO}_3-N\) to \(g \text{ COD}\) is based on the relationship of \(g \text{ COD}\) expected to denitrify \(\text{NO}_3-N\) to \(\text{NO}_3\) (Rittmann and McCarty, 2001). The \(\%\text{CH}_4\) was determined by GC-TCD analysis (SRI 8610C, California). Only gauge pressure was considered to calculate number of moles of \(\text{CH}_4\) produced, assuming atmospheric pressure remained constant during the experiment. The conversion of \(\text{CH}_4\) to \(\text{COD}\) is based on the empirical COD of \(\text{CH}_4\), 4 mol \(\text{O}_2\) to oxidize 1 mol \(\text{CH}_4\) and 16 g/mol \(\text{CH}_4\) resulting in 64 g COD/mol \(\text{CH}_4\).

A similar experiment was performed to determine COD released by operating columns with fresh bamboo and eucalyptus. Filters were fed trickling solution of DI water and \(\text{KNO}_3\) (0 mg COD L\(^{-1}\)) for the first three days. The feed was then switched to a solution of DI water and \(\text{KNO}_3\) (0 mg COD L\(^{-1}\) and 400 mg \(\text{NO}_3-N\) L\(^{-1}\)) so that all denitrification would be due to COD released from wood chips. COD and \(\text{NO}_3-N\) were determined in the inlet, mid-depth, and outlet of the columns at the start and then 1, 2, 3, 6, 9, and 12 days after the start and then weekly until the outlet concentration of nitrate equaled the inlet concentration. The COD released by wood chips was determined by a mass balance, assuming steady-state conditions after the first three days (no COD accumulation) where \(C_{\text{in}}\) and \(C_{\text{out}}\) were COD concentrations measured in inlet and outlet, respectively, \(Q\) was substrate flow rate \((1.35 \text{ L d}^{-1})\), and \(t\) was time step between sampling days \([\text{d}]\). The following mass balance was solved for wood chip release, \(W_{\text{release}}\) in g COD.

\[
0 [g \text{ COD}] = (C_{\text{in}} - C_{\text{out}}) \times Q \times t + W_{\text{release}} - (\text{NO}_3,_{\text{in}} - \text{NO}_3,_{\text{out}}) \times Q \times t + \frac{4[g \text{ COD}]}{[g \text{ NO}_3]} \]  (6)

\[
W_{\text{release}}[g \text{ COD}] = -(C_{\text{in}} - C_{\text{out}}) \times Q \times t + (\text{NO}_3,_{\text{in}} - \text{NO}_3,_{\text{out}}) \times Q \times t + \frac{4[g \text{ COD}]}{[g \text{ NO}_3]} \]  (7)

3. Results

3.1. Nitrifying filters

Tracer study results found that the actual retention time in BC, GAC, Zeo, PR, and Gr filters was 66.1, 88.1, 61.0, 50.5, and 65.2 h, respectively. The ratio of the actual retention time to EBRT (6 days) for these filters were 0.46, 0.61, 0.42, 0.35, and 0.45, respectively. Table 2 shows average values for COD, pH, and turbidity for filter substrate (influent) and concentrations for each filter’s effluent during Periods II & III. Table 3 shows the average concentrations and rates of removal of \(\text{NH}_3-N\) and production for \(\text{NO}_3-N\) for each filter during Periods II & III. The full nitrogen speciation concentrations can be seen in Table A2 in SI. The difference between \(\text{NH}_3-N\) removal and \(\text{NO}_3-N\) production is the amount of \(\text{NH}_3-N\) that was either lost from the system through adsorption or was not fully nitrified and exited as nitrite. The greatest difference between these values was in the GAC and Zeo filters. The GAC \(\text{NO}_3-N\) concentration was 38 mg N L\(^{-1}\) during Period II, and the fresh GAC and washed GAC in Period III both released less than 8 mg N L\(^{-1}\). The Zeo \(\text{NO}_3-N\) concentration was 171 mg N L\(^{-1}\). The summation of \(N\) species from these filters does not account for all \(\text{NH}_3-N\) removed from the influent; therefore, some total \(N\) was removed from solution. Zeolite had the most removal of total \(N\), but also released the greatest concentration of \(\text{NO}_3-N\), indicating incomplete nitrification.

The filters had stable performance from the start of Period II (day 63) until day 239. During this time, average turbidity removal for BC, GAC, Zeo, PR, and Gr was 90, 91, 77, 74, and 74%, respectively, and \(\text{NH}_3-N\) removal was 83, 87, 85, 30, and 80%, respectively.

Figs. 1 and 2 show the \(\text{NH}_3-N\) concentration and pH in the influent and effluent of each filter while turbidity data are shown in Figure A1 in SI. The decrease in \(\text{NH}_3-N\) and pH in the majority of filters during Period I (first 60 days) is indicative of the build-up of the nitrifying community. The filters had stable performance from the start of Period II (day 63) until day 239. During this time, average turbidity removal for BC, GAC, Zeo, PR, and Gr was 90, 91, 77, 74, and 74%, respectively, and \(\text{NH}_3-N\) removal was 83, 87, 85, 30, and 80%, respectively.
Table 2
Average influent and effluent COD, pH, and turbidity, and changes (Δ) from inlet to outlet during Period II (days 56–260) and Period III (days 274 onwards).

| COD (mg/L) | pH (–) | Turbidity (NTU) |
|-----------|--------|-----------------|
| Avg. | St. Dev. | Δ (%) | Avg. | St. Dev. | Δ (pH units) | Avg. | St. Dev. | Δ (%) |
| Influent – II | 1863 | 904 | – | 8.2 | 0.2 | – | 1146 | 770 | – |
| Influent – III | 1919 | 803 | – | 7.5 | 0.6 | – | 814 | 357 | – |
| BC – II | 818 | 367 | –56% | 5.4 | 1.2 | –2.8 | 239 | 154 | –87% |
| BC – III | 398 | 229 | –79% | 6.1 | 1.0 | –0.6 | 53 | 75 | –94% |
| BC new - III | 747 | 468 | –61% | 5.5 | 1.5 | –2.0 | 141 | 185 | –83% |
| GAC – II | 716 | 486 | –62% | 6.0 | 1.2 | –2.2 | 217 | 214 | –88% |
| GAC – III | 441 | 379 | –77% | 6.8 | 1.0 | –0.7 | 135 | 134 | –83% |
| GAC new – III | 483 | 397 | –75% | 6.2 | 1.1 | –1.3 | 129 | 157 | –84% |
| Zeo – II | 1445 | 578 | –22% | 6.7 | 0.7 | –1.5 | 493 | 266 | –73% |
| PR – II | 1535 | 570 | –18% | 8.4 | 0.4 | 0.2 | 707 | 484 | –61% |
| Gr – II | 1388 | 746 | –25% | 7.4 | 0.3 | –0.8 | 491 | 208 | –73% |
| Gr – III | 921 | 467 | –52% | 7.8 | 0.4 | 0.3 | 221 | 128 | –73% |

Table 3
Average influent and effluent NH₃-N, and NO₃-N in trickling filters over Period II and III. The NH₃ Δ is the removal from influent to effluent (reduction in concentration yields a negative Δ). The %NH₃ value for nitrate is the percent of influent NH₃-N recovered as NO₃-N produced.

| NH₃-N (mg/L) | NO₃-N (mg/L) |
|-------------|--------------|
| Avg. | St. Dev. | Δ (%) | Removal rate (kg N/m³-d) | Avg. | St. Dev. | (% NH₃) | Production rate (kg N/m³-d) |
| Influent – II | 728 | 311 | – | 56 | 50 | – | – |
| Influent – III | 502 | 85 | – | 51 | 85 | – | – |
| BC – II | 138 | 47 | –81% | 0.100 | 569 | 171 | 70% | 0.087 |
| BC – III | 56 | 70 | –89% | 0.075 | 381 | 184 | 66% | 0.056 |
| BC new - III | 107 | 114 | –79% | 0.067 | 359 | 184 | 61% | 0.052 |
| GAC – II | 125 | 81 | –83% | 0.102 | 530 | 113 | 65% | 0.080 |
| GAC – III | 50 | 57 | –90% | 0.076 | 223 | 137 | 50% | 0.029 |
| GAC new – III | 123 | 113 | –76% | 0.064 | 304 | 160 | 34% | 0.043 |
| Zeo – II | 125 | 47 | –83% | 0.102 | 366 | 215 | 43% | 0.052 |
| PR – II | 473 | 360 | –35% | 0.043 | 136 | 105 | 11% | 0.014 |
| Gr – II | 172 | 85 | –76% | 0.094 | 472 | 176 | 57% | 0.070 |
| Gr – III | 258 | 132 | –49% | 0.041 | 110 | 55 | 12% | 0.010 |

Fig. 1. Total ammonia-nitrogen concentrations of influent and effluent of each trickling filter. Days 0–56 were Period I (start-up), days 63–252 were Period II, and days 281–351 were Period III (washed and new media).

3.3. Wood chip COD release

The amount of COD released by bamboo and eucalyptus during the batch experiment is shown in Fig. 4. One of the flasks for bamboo exploded on day 18 due to high gas pressure. COD release from bamboo was analyzed in duplicate after this time. Greater scattering of the results was noticed for the tests with eucalyptus after day 21. Slow denitrification rates were seen until this time, but two of the three bottles exhibited rapidly increased COD release during this time while the third remained at a slow rate. Some dips in average cumulative COD release can be seen in Fig. 4; they are the result of experimental uncertainties. In general, the error bars overlap for points before and after dips. Because of these dips, the maximum value instead of value per gram TS (bamboo 94% TS, eucalyptus 36%), the maximum releases were 0.076 gCOD g⁻¹, while the maximum COD released for eucalyptus was 0.043 gCOD g⁻¹. If COD release was normalized by gram TS (bamboo 94% TS, eucalyptus 36%), the maximum releases were 0.080 gCOD g⁻¹ and 0.119 gCOD g⁻¹. In terms of future design, however, COD release per wet mass may be the more practical parameter than dry mass.

The total COD release and rate of release from bamboo were
both significantly greater than for eucalyptus. This is consistent with the observation that the rate of nitrate removal was much faster in bamboo flasks than in eucalyptus flasks. No nitrate remained in bamboo flasks after 8 days, but 73 days were required to remove nitrate in eucalyptus flasks (one of the three flasks still had 87 mg NO₃-N L⁻¹ on this day) (see additional results in Figure A.3).

Fig. 4 also displays the COD released during the continuously-fed column experiment. Similar to the batch experiment, the bamboo wood chips released more COD per gram of wood. The total amount released was 0.092 gCOD gwood⁻¹ and 0.020 gCOD gwood⁻¹. In terms of release per gram of TS, the values were 0.098 gCOD gTS,bamboo⁻¹ and 0.083 gCOD gTS,eucalyptus⁻¹. The amounts of COD released during the continuously-fed column were comparable to those of the batch experiment.

4. Discussion

4.1. Trickling filters

4.1.1. Start up

An indicator of nitrification is change in pH as 2 mol of H⁺ are released per mole of NH₄ oxidized to nitrite, and the decline of pH over time can be interpreted as the time for the nitrifying community to build up. As seen in Fig. 2, the pH dropped the most for eucalyptus flasks still had 87 mg NO₃-N L⁻¹ on this day) (see additional results in Figure A.3).

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Table 4
Average influent and effluent COD, pH, and turbidity for the submerged filters.

|       | COD (mg/L) | pH (–) | Turbidity (NTU) |
|-------|------------|--------|-----------------|
|       | Avg.       | St. Dev. | Δ (%) | Avg. | St. Dev. | Δ (%) | Avg. | St. Dev. | Δ (%) |
| Influent | 949 | 541 | 7.4 | 0.6 | 364 | 284 |
| Sand-mid | 683 | 480 | 7.8 | –28% | 47 | 36 | –84% |
| Sand-final | 479 | 290 | 7.5 | –6% | 57 | 48 |
| Bamboo-mid | 587 | 286 | 7.7 | –3% | 44 | 37 |
| Bamboo-final | 477 | 257 | 7.4 | 0% | 40 | 35 |
| B-S-mid | 641 | 329 | 7.7 | 0% | 59 | 47 |
| B-S-final | 505 | 296 | 7.6 | 0% | 59 | 51 |
| Eucalyptus-mid | 533 | 279 | 7.4 | 3% | 54 | 43 |
| Eucalyptus-final | 404 | 287 | 7.6 | 3% | 40 | 37 |
| E-S-mid | 542 | 404 | 7.6 | 4% | 55 | 48 |
| E-S-final | 434 | 235 | 7.5 | 0% | 44 | 41 |

Table 5
Average influent and effluent TN and NH₄-N for the submerged filters with normalized rates of removal.

|       | TN (mg/L) | NO₃-N (mg/L) |
|-------|-----------|-------------|
|       | Avg. | St. Dev. | Δ (%) | Avg. | St. Dev. | Δ (%) |
| Influent | 405 | 134 | – | – | 299 | 79 |
| Sand-mid | 384 | 111 | –5% | 0.015 | 250 | 74 |
| Sand-final | 334 | 133 | 0% | 0.024 | 223 | 70 |
| Bamboo-mid | 278 | 75 | –32% | 0.086 | 167 | 64 |
| Bamboo-final | 223 | 85 | –45% | 0.062 | 103 | 61 |
| B-S-mid | 324 | 104 | –20% | 0.055 | 217 | 61 |
| B-S-final | 278 | 140 | –31% | 0.043 | 141 | 63 |
| Eucalyptus-mid | 318 | 96 | –22% | 0.059 | 237 | 72 |
| Eucalyptus-final | 277 | 94 | –32% | 0.043 | 195 | 64 |
| E-S-mid | 334 | 84 | –18% | 0.048 | 219 | 67 |
| E-S-final | 304 | 105 | –25% | 0.034 | 183 | 72 |
exhibited the greatest reduction of NH$_3$ by nitrification. Another observation during the start-up period is the low concentrations of NH$_3$ in the effluent from the beginning for the BC, GAC, and Zeolite filters, even before the nitrifying community developed as indicated by pH (Fig. 1). This decrease of NH$_3$ concentration can be interpreted by adsorption to these media, which was expected based on their properties. In contrast, NH$_3$ effluent concentrations from the gravel (which has no N sorption capacity) filter slowly decreased, consistent with the need for the nitrifying community to build up for NH$_3$ removal.

4.1.2. Normal operation

The BC and GAC filters showed the best performance in reducing NH$_3$. The performance of these two filters were similar for the duration of the experiment (see Fig. 1) with NH$_3$ removal in the GAC filter being generally slightly better, possibly because of slightly better adsorption capacity. The filter packed with gravel showed an opposite trend, improving in performance with time; the smooth surface of the gravel certainly caused the biofilm to form more slowly than onto the more porous supports. Additionally, gravel has no absorption or adsorption capacity like BC, GAC, and zeolite. The filter packed with Pall rings performed the worst, most likely due to the short liquid retention time and limited biofilm growth on the plastic packing.

Biochar and GAC filters provided similar ammonium removal performance over both Period II and III. The primary difference between the two was that the filter packed with GAC removed more TN than the filter packed with biochar. This removal is hypothesized to be due to adsorption in the GAC which is made up of micropores (more so than biochar) that are effective in adsorbing dissolved contaminants, such as ammonium (Simpson, 2008). These micropores, however, become blocked by suspended solids and biofilm growth, limiting adsorptive capacity when treating high-strength waste (Huggins et al., 2016). Reductions in NH$_3$-N concentrations were similar in both periods; however, increases in NO$_3$-N concentrations were greater in the BC filter than in the GAC one. Decreased absorptive capacity was displayed as the nitrate production was greater in GAC filters used for a longer duration (Table 3, GAC-II). Further, the pH decrease shown in Fig. 2 for GAC-II indicates increased nitrification over time while ammonium removal remained relatively constant (Fig. 1). Additionally, a significant difference was displayed in nitrate production in Phase III between the washed GAC and fresh GAC. This can be explained by the fact that washed GAC would not reactivate micropores and begin with a lower adsorptive capacity than fresh GAC. Therefore, while nitrification and adsorption both reduce NH$_3$-N concentrations in each filter, adsorption likely played a greater role in the GAC column.

In comparison to filters with BC and GAC, NH$_3$ concentrations in the effluent of the Zeolite filter rapidly increased at the end of the startup phase (days 35–56), followed by a decrease of effluent NH$_3$ concentrations during Period II to reach levels similar to that of
filters packed with BC & GAC. This increase and then decrease in effluent ammonium concentration is attributed to filling adsorption sites during startup and a longer time to build up the nitrification community. Later, from days 133–252, the filter packed with Zeo had similar effluent NH3 but much lower NO3 concentrations than the BC and GAC filters. This signifies a shift in NH3 removal mechanism from ion exchange (during Period I) to nitrification and back to ion exchange (during Period II). This dynamic is likely related to the ability for zeolite to be regenerated as nitrifying bacteria consume NH4+ as was found by Aiyuk et al. (2004).

Overall, most of the trickling filters showed high NH3-N removal rates. With the exception of the Pall ring filter, all filters showed the ability to achieve nearly 0.10 kg N m⁻² d⁻¹. The rates achieved in this study are significantly higher than found in a previous study testing both biochar and GAC (approximately 0.03 kg NH3-N m⁻³ d⁻¹ each) (Berger, 2012). The high removal rate values could be due to the high inlet concentrations (800 mg N L⁻¹ vs 95 mg N L⁻¹ for the Berger study), which also show that these filters can treat high strength waste.

Nitrification has a significant oxygen demand (4.57 g O₂/g NH4-N (Rittmann and McCarty, 2001)). Our trickling filters were operated with passive aeration using a natural draft which was sufficient as seen by high nitrification rates. Monitoring DO (Figure A2) confirmed adequate oxygenation as the DO concentration increased from 0.8 mgDO₂ L⁻¹ in the substrate influent to 5 mgDO₂ L⁻¹ in BC and GAC filter effluent. Proper aeration would have to be reassessed in full-scale application, however, as air draft is size and geometry dependent. The narrow columns (1 m length x 10 cm dia.) in this experiment are more conducive to natural draft than a full-scale system which is a much larger diameter.

4.2. Submerged filters

Denitrification performance was much greater over the first two months than for the next two months. From day 1–62, the average NO3-N concentration in the influent of the anaerobic filters was 265 mg-N L⁻¹, and outlet concentrations for S, B, B+S, E, and E+S were 187, 70, 100, 171, and 171 mg NO3-N L⁻¹ with corresponding removals of 29, 73, 64, 35, and 35%, respectively. The NO3-N loading did not significantly change from the first half to the second half of the study, however, the inlet COD concentration decreased from first half to second half (1150 to 730 mgCOD L⁻¹) which may have affected the denitrification performance. Another possible reason for decreased performance could be a partial exhaustion of the high nitrate concentrations in the influent consumed for denitrification by the bamboo and eucalyptus filters for denitrification can be predicted. B, B+S, E, and E+S filters removed 265, 215, 143, and 156 mg NO3-N d⁻¹, respectively. The COD:NO3 ratio in the sand filter was 5.2:1. Using that same ratio for NO3-N removal in other filters yields 1,380, 1,120, 740, and 810 mg COD d⁻¹ consumed for denitrification in the B, B+S, E, and E+S filters, respectively of which 749, 487, 110, and 179 mg COD d⁻¹ came from the wood chips. The B filter had 978 g bamboo, resulting in a release rate of 0.765 mgCOD g bamboo d⁻¹, and the E filter had 1070 g eucalyptus, resulting in 0.103 mgCOD g eucalyptus d⁻¹.

The calculated rate of COD release from bamboo wood chips was less in the treatment study (0.765 mgCOD g bamboo d⁻¹) than found in the column study (1.26 mgCOD g bamboo d⁻¹) and batch studies (1.04 mgCOD g bamboo d⁻¹). However, COD release plateaued in column and batch studies by day 73, while the COD release rate in the treatment study had still not plateaued (see Fig. 4) by the end of the treatment study period (123 d). The total release in the treatment, column, and batch studies according to study duration was 0.094 gCOD g bamboo d⁻¹ and 0.092 gCOD g bamboo d⁻¹, and 0.076 gCOD g bamboo d⁻¹, respectively. These findings show that the bamboo wood chips in the column study for COD release produced a greater total COD release as well as release rate than in the batch study while the greatest total COD release for bamboo wood chips was in the actual treatment study. These results may be that the continuous feed of substrate (nitrate) in the column study enhanced microbial activity, either directly extracting more COD out of the wood chips or promoting hydrolysis of the wood chips.

Eucalyptus wood chips showed high NH3-N removal linked to COD release by the bamboo only can be found by difference and was 0.056 and 0.036 kg N m⁻³ d⁻¹, respectively. This rate is higher than a previous study using eucalyptus wood chips that found a rate of 0.014 kg N m⁻³ d⁻¹ (Lynn et al., 2015a). Eucalyptus wood chips in our study, however, did not perform as well. The nitrate removal rates for the eucalyptus-only filter calculated from mid-depth and final outlet concentrations were 0.042 and 0.036 kg N m⁻³ d⁻¹, respectively. While this is higher than the previous study by Lynn et al., 2015b the removal rate in the sand-only filter was similar (0.030 kg N m⁻³ d⁻¹ across the whole filter), showing that eucalyptus did not significantly contribute to denitrification in terms of supplementing electron donors. Removal rates in all filters were greater in the first half of the total study. The higher rate in the first half of the filter supports the finding of Aslan (2008) that the majority of nitrate removal occurs in the top portion of the filter bed in a downflow reactor. If the bamboo- and eucalyptus-only reactors were to be resized based on their overall removal rates to completely denitrify an inflowing load of 300 mg NO3-N L⁻¹ at 1.35 L d⁻¹, the resulting volumes would be 6.1 L for bamboo and 11.3 L for eucalyptus.

At a flow of 1.35 L d⁻¹, the sand filter removed 635 mg COD d⁻¹ and 122 mg NO3-N d⁻¹. By using sand as the control for the amount of COD from the influential consumed for denitrification, the COD provided by the bamboo and eucalyptus filters for denitrification can be predicted. B, B+S, E, and E+S filters removed 265, 215, 143, and 156 mg NO3-N d⁻¹, respectively. The COD:NO3 ratio in the sand filter was 5.2:1. Using that same ratio for NO3-N removal in other filters yields 1,380, 1,120, 740, and 810 mg COD d⁻¹ consumed for denitrification in the B, B+S, E, and E+S filters, respectively of which 749, 487, 110, and 179 mg COD d⁻¹ came from the wood chips. The B filter had 978 g bamboo, resulting in a release rate of 0.765 mgCOD g bamboo d⁻¹, and the E filter had 1070 g eucalyptus, resulting in 0.103 mgCOD g eucalyptus d⁻¹.

The calculated rate of COD release from bamboo wood chips was lower in the treatment study (0.765 mgCOD g bamboo d⁻¹) than found in the column (1.26 mgCOD g bamboo d⁻¹) and batch studies (1.04 mgCOD g bamboo d⁻¹). However, COD release plateaued in column and batch studies by day 73, while the COD release rate in the treatment study had still not plateaued (see Fig. 4) by the end of the treatment study period (123 d). The total release in the treatment, column, and batch studies according to study duration was 0.094 gCOD g bamboo d⁻¹ and 0.092 gCOD g bamboo d⁻¹, and 0.076 gCOD g bamboo d⁻¹, respectively. These findings show that the bamboo wood chips in the column study for COD release produced a greater total COD release as well as release rate than in the batch study while the greatest total COD release for bamboo wood chips was in the actual treatment study. These results may be that the continuous feed of substrate (nitrate) in the column study enhanced microbial activity, either directly extracting more COD out of the wood chips or promoting hydrolysis of the wood chips.

Total eucalyptus COD release as well as release rate was lower in the treatment study (0.013 gCOD g eucalyptus d⁻¹ and 0.103 mgCOD g eucalyptus d⁻¹) than in the column study (0.030 gCOD g eucalyptus d⁻¹ and 0.411 mgCOD g eucalyptus d⁻¹) and batch study (0.043 gCOD g eucalyptus d⁻¹ and 0.589 mgCOD g eucalyptus d⁻¹). This opposite relationship for
eucalyptus is unclear, but the greater release in the batch experiment than in the column experiment may be due to the shorter retention time compared to column with bamboo, and due to slow hydrolysis.

Turbidity removal performance was nearly identical across filters (84–88%). Bamboo and eucalyptus wood chip filters showed less turbidity removal which was attributed to higher void space compared to sand, which is why two filters were tested using a combination of sand and wood chips. However, wood chips plus sand filters did not show any significant improvement in turbidity removal. The use of sand resulted in decreased performance over time as clogging was experienced in sand and B+S filters by day 49 and day 62 for E+S. From this time on, the top layer of the filter was stirred weekly to disturb buildup and induce free flow. With this scenario, bamboo wood chips alone were the overall best performing filters by providing the greatest nitrate removal without losing turbidity removal performance while having greater longevity. Biochar could also be used as an alternative medium to improve filtration and increase nitrate removal as has been suggested by Bock et al. (2015, 2016). These studies found increased nitrate removal by amending wood chips with 10% biochar by volume when influent nitrate concentrations are high.

4.3. Combined filter performance

A desired outcome of this study was to inform a possible future implementation for effluent treatment of onsite sanitation systems such as those described by Forbis-Stokes et al. (2016). Based on the current study, the chosen media would be biochar for the trickling filter and bamboo for the submerged denitrifying filter because of their high performance as well as widely available and sustainable procurement. By keeping the design and loading conditions the same with average influent concentrations of 600 mg NH₃-N L⁻¹, 50 mg NO₃-N L⁻¹, and 980 NTU and 6 d EBRT, the biochar filter's expected effluent would be 97 mg NH₃-N L⁻¹, 475 mg NO₃-N L⁻¹, and 120 NTU. The bamboo filter (3 d EBRT) would then produce an effluent of 82 mg NH₃-N L⁻¹, 157 mg NO₃-N L⁻¹, and 13 NTU. This theoretical combined performance would thus result in 63% removal of total N and 99% removal of turbidity. The limiting step to greater total N reduction in this setup would be denitrification. If more N removal is required, increasing the volume of the bamboo denitrifying filter is likely to improve performance as the electron donor was demonstrated to limit denitrification. Based on the observed nitrate removal rates, the bamboo filter volume would need to be increased to 7.2 L (or a volumetric loading of 0.19 m³ m⁻³ d⁻¹) to accommodate the nitrate loading. In this scenario, total nitrogen removal efficiency of 80–90% will likely be achieved. Another approach to improve denitrification is to pre-treat wood chips to promote hydrolysis of the cellulosic structure. Particle size reduction, thermal treatment, acidic treatment, and alkali treatment are all methods of improving the biodegradability of lignocellulosic materials (Montgomery and Bochmann, 2014) that may be beneficial in this application.

The filter systems that were investigated required minimal maintenance. Trickling filters were in operation for about 6 months before clogging became an issue. When media was removed and washed with tap water, full treatment performance was recovered almost instantly and required less start-up time than in the initial start-up period. Submerged filters with sand clogged within 2 months of operation while filters without sand did not experience clogging in the 4-month study period. Additionally, no increasing nitrate effluent concentration trend was seen at the end of the study period, showing that the wood chips were still supplementing electrons for denitrification at that time. Based on these findings, a 6-month maintenance interval for trickling filters, and a 4- or up to 6-month maintenance interval for bamboo denitrifying filters are recommended.

Another consideration for filter performance is phosphorus removal which is not included in the previous discussion as the focus of this work was on nitrogen removal. The phosphorus removal efficiencies (measured as PO₄³⁻) in the trickling filters with BC, GAC, and Zeo were similar and ranged from 31 to 42%. Removal in the denitrifying filters ranged from 27% for the eucalyptus filter to 41% in the all-sand filter. The primary mechanism of phosphorus removal in this experiment is hypothesized to be precipitation and filtration and not biological removal which is consistent with the observation that the sand denitrifying filters performed better than those without sand. Additionally, the operational condition for these filters was unfavorable for biological phosphorus removal. For phosphorus treatment, two methods of interest are (1) incorporating media into the denitrifying filters such as iron (Erickson et al., 2012) or biochar amended wood chips (Bock et al., 2015; Pluer et al., 2016) for combined physical and chemical phosphate removal, or (2) phosphorus precipitation downstream of the denitrifying filter by adding magnesium to produce struvite (Etter et al., 2011) or flowing through steel slag (Christianson et al., 2017). Further research is required to quantify and optimize phosphorus removal.

5. Conclusion

This study evaluated the use of various packing materials as nitrification and denitrification media for the treatment of high-strength anaerobic digester effluent. Maximum ammonia-nitrogen removal (either nitrification or total N removal) in filters with biochar and GAC reached 81–90% (at an influent concentration of 600 mg NH₃-N L⁻¹). These same filters achieved 83–94% removal of turbidity while treating a waste stream with approximately 1000 NTU. Investigations with submerged attached growth filters evaluated the use of sand, bamboo, bamboo with sand, eucalyptus, and eucalyptus with sand as denitrification systems for treating the effluent of the nitrification trickling filters. The maximum nitrate removal was 66% of NO₃-N and 89% removal of turbidity in the filter packed with bamboo. The trickling and submerged filters showed high removal rates while treating high concentrations of ammonia and nitrogen and, thus, offer promising post-treatment options for digestor effluent for simultaneous organic and nitrogen removal. These systems required minimal attention as estimated time between required maintenance was 4–6 months. Both filter stages provided dramatic improvement in effluent clarity as well as odor, as there was little to no detectable offensive odor in the effluent of either filter. The improved aesthetics of the treated effluent may allow for water reuse (e.g., for toilet flushing). The trickling filter produces an effluent with high total nitrogen concentrations which could be reused as fertilizer if only this first stage of treatment was used. If local fertilizer reuse is not desired, the second stage denitrification filter can be used. If adequately sterilized, the second stage denitrification filter effluent could potentially be reused as flushing water or wash water, or be safely discharged in the environment, depending on local regulations.

Author disclosure statement

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