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Influence of mixed liquor suspended solids on the removal efficiency of a hybrid membrane bioreactor

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Abstract. The characterization of treatment performance with respect to mixed liquor suspended solids (MLSS) concentration enables greater control over system performance and contaminant removal efficiency. Hybrid membrane bioreactors (HMBRs) have yet to be well characterized in this regard, particularly in the context of greywater treatment. The aim of this study, therefore, was to determine the optimal MLSS concentration for a decentralized HMBR greywater reclamation system under typical loading conditions. Treatment performance was measured at MLSS concentrations ranging from 1000 to 4000 mg/L. The treated effluent was characterized in terms of biochemical oxygen demand (BOD_5), chemical oxygen demand (COD), turbidity, ammonia (NH_3), total phosphorus (TP), total kjeldahl nitrogen (TKN), and total nitrogen (TN). An MLSS concentration ranging from 3000 to 4000 mg/L yielded optimal results, with BOD_5, COD, turbidity, NH_3, TP, TKN, and TN removals reaching 99.2%, 97.8%, 99.8%, 99.9%, 97.9%, 95.1%, and 44.8%, respectively. The corresponding food-to-microorganism ratio during these trials was approximately 0.23 to 0.28. Operation at an MLSS concentration of 1000 mg/L resulted in an irrecoverable loss of floc, and contaminant residuals exceeded typical guideline values for reuse in non-potable water applications. Therefore, it is suggested that operation at or below this threshold be avoided.

Keywords: greywater reclamation; hybrid membrane bioreactor; membrane bioreactor; mixed liquor suspended solids

1. Introduction

The utilization of treated greywater as a supplementary water supply occurs in many arid and semi-arid regions or in regions of high population density (Laaffat *et al*. 2015, Ammari *et al*. 2014, Oron *et al*. 2014, Mandal *et al*. 2011, Mourad *et al*. 2011, Wintgens 2005, Al-Jayyousi 2003). Increasing water demand, coupled with limited infrastructure and stringent regulatory pressure, will likely necessitate continual use of this water management strategy. The implementation of greywater reclamation can be achieved through the use of large-scale centralized development, or through the use of small-scale decentralized treatment systems. Small-scale greywater reclamation typically involves the collection of greywater from domestic sources which is then treated onsite and reused in various applications, such as toilet flushing and irrigation. Decentralization has increasingly been recognized as a suitable approach to service areas with supply or sanitation constraints (Libralato *et al*. 2012, Bieker *et al*. 2010), and it may offer more opportunities to

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recover recyclable wastewater than through centralized treatment (Al-Jayyousi 2003). The continual development of effective decentralized treatment systems is therefore of great importance to present day greywater reclamation schemes.

The combination of aerobic biological processes, physical filtration, and disinfection has been presented as a feasible approach for greywater recycling (Li *et al*. 2009). Thus far, membrane bioreactors (MBRs) and their related processes have satisfied many of the design requirements for decentralized treatment, making them well-suited for greywater reclamation. A relatively new technology that combines both suspended and attached growth processes with membrane separation – a hybrid membrane bioreactor (HMBR) – has attracted widespread attention as an alternative to conventional MBR configurations (Zhang *et al*. 2014, Young and Munoz 2012). An HMBR incorporates moving bed media directly into a membrane bioreactor, thereby enabling a greater total biomass concentration without increasing the concentration of the mixed liquor suspended solids (MLSS). This enables HMBRs to operate at a lower MLSS range compared to conventional MBRs while maintaining a similar total biomass concentration and treatment capacity.

The MLSS that comprise an activated sludge process form the basis of biological treatment, and it is a key operational parameter for MBR processes (Lousada-Ferreira *et al*. 2010). The characterization of treatment performance in terms of MLSS affords greater predictability of effluent quality and is therefore useful to system operators responsible for maintaining optimal operation. In general, MBRs operate within an MLSS range three to five times greater than a conventional activated sludge reactor. This greatly reduces the size of the reactor, which is advantageous over conventional systems when space is limited. It has been generally observed that increases in MLSS concentration correlate to an increase in the removal of organics (Ren *et al*. 2005, Kumar *et al*. 2014), suspended solids, and turbidity (Katayon *et al*. 2004). However, increases in MLSS concentration also correlate to an increase in sludge viscosity (Delrue *et al*. 2011), a decrease in oxygen transfer efficiency (Rodríguez *et al*. 2012), and an increase in membrane fouling (Lee and Kim 2013, Wu and Huang 2009, Trussell *et al*. 2007). These latter relationships are problematic because they reduce system efficiency. The relatively lower MLSS concentration of HMBRs compared to MBRs may therefore make them favourable in this regard.

The objective of this study was to determine the optimal MLSS concentration and food-to-microorganism (F/M) ratio for a decentralized HMBR greywater reclamation system. The reclamation system was designed to treat approximately 160 L/d and was subjected to an organic load of 247 mg/L BOD/d. Treatment performance was measured in terms of biochemical oxygen demand (BOD_5), chemical oxygen demand (COD), turbidity, ammonia (NH_3), total phosphorus (TP), total kjeldahl nitrogen (TKN), and total nitrogen (TN). MLSS concentrations of 1000, 2000, 3000, and 4000 mg/L were tested at a hydraulic retention time (HRT) of 8 h. Food-to-microorganism ratios ranged from 0.23 to 0.95. The results of this study aimed to improve the operation of HMBR greywater reclamation systems designed to meet the requirements for non-potable water reuse.

2. Experimental methods

2.1 Materials

The bioreactor was constructed from an acrylic cylinder with an inner diameter of 290 mm, an outer diameter of 305 mm, and a height of 915 mm. Considering displacement caused by the

moving bed media, module, and other interior components, the effective volume of the bioreactor was 53 L. The biocarriers consisted of high-density polyethylene (HDPE) cylinders 15 mm high and 18 mm in diameter. Each biocarrier consisted of 12 interior chambers which provided a specific surface area of 600 to 700 m^2/m^3. A custom-built microfiltration module (Young and Munoz 2012) was used for solid-liquid separation. This module consisted of a hollow fibre polyvinylidene fluoride (PVDF) microfiltration membrane with a nominal pore size of 0.1 μm. The effective surface area of this membrane module was 1.32 m^2. Two air stones, one embedded into the base of the module, and the other at the bottom of the reactor, provided air scouring, aeration, and hydraulic mixing. A continuous airflow was supplied to both stones by a single blower and regulated by flow control valves to maintain a dissolved oxygen (DO) concentration of approximately 2.5 ± 0.5 mg/L. Permeate and backflush cycles were controlled by means of a programmable peristaltic Masterflex L/S pump.

2.2 Synthetic greywater

Synthetic greywater was used to provide consistent influent characteristics during the entire experimental period. The composition of the synthetic greywater is provided in Table 1. The synthetic greywater was formulated to be representative of domestic greywater generated from mixed sources (Eriksson *et al*. 2002). Greywater sources generally include sinks, showers, bathtubs, dishwashers, and washing machines. Characterization of the synthetic greywater is summarized in Table 2.

Table 1 Composition of the synthetic greywater

Chemical	Molecular Formula	Amount	Unit
Starch	$C_6H_{10}O_5$	75.0	g
D-glucose	$C_6H_{12}O_6$	35.0	g
Peptone	n/a	36.0	g
Beef extract	n/a	25.5	g
Sodium carbonate	Na_2CO_3	90.0	g
Sodium bicarbonate	$NaHCO_3$	46.5	g
Urea	NH_2CONH_2	12.0	g
Ammonium sulphate	$(NH_4)_2SO_4$	13.0	g
Tap water	H_2O	500	L

Table 2 Characteristics of the synthetic greywater

Parameter	Synthetic Greywater	Eriksson *et al*. (2002)	Unit
COD	423 ± 40	13 to 549	mg/L
BOD_5	247 ± 25	90 to 360	mg/L
NH_3	40.9 ± 6.1	0.03 to 25.4	mg/L
Turbidity	33.1 ± 2.8	15.3 to > 200	NTU
TP	4.03 ± 0.62	0.16 to 27.3	mg/L
TKN	54.2 ± 6.7	2.1 to 31.5	mg/L
TN	54.5 ± 6.9	0.54 to 5.2	mg/L

2.3 Biofilm culture

Biofilm was cultured by first soaking the media to an activated sludge sample before submerging the media in a tank of synthetic greywater. This initial exposure period was done to initiate attachment of bacteria to the biofilm substrate. The tank was treated as a batch reactor with old greywater being replaced with fresh greywater every 24 h. Aeration was provided to the tank to supply a DO concentration of 1.5 ± 0.5 mg/L. Vigorous mixing of the media was avoided. Sessile colonies began to establish on the media within 2 to 3 days and a mature biofilm developed within the following 15 to 20 days. The process was completed within approximately three weeks. The media was then transferred to the HMBR and acclimated for an additional 14 days under continuous flow conditions.

2.4 Experimental setup

The 53 L bioreactor was filled with Bioportz^{TM} moving bed media at a filling ratio of 20%. The media filled the upper compartment of the bioreactor and were supported by a plastic mesh to prevent collisions between the media and the membrane module. The mesh had openings of approximately 10 mm which permitted ample hydraulic mixing and aeration to the biocarriers. Influent greywater was collected within a 450 L storage tank which was elevated above the HMBR to allow gravity to feed the greywater directly into the reactor. A float valve situated at the top of the reactor was used to maintain a constant water level and pressure head within the HMBR. The module was orientated vertically in the centre of the reactor. The flow cycle consisted of permeate extraction for 9 min 30 s followed by relaxation for 15 s followed by backflush for 15 s. A schematic diagram of the HMBR system is depicted in Fig. 1.

Contaminant removal efficiencies were evaluated at MLSS concentrations of 1000, 2000, 3000, and 4000 mg/L at an HRT of 8 h. The MLSS, HRT, solids retention time (SRT), applied flux, and

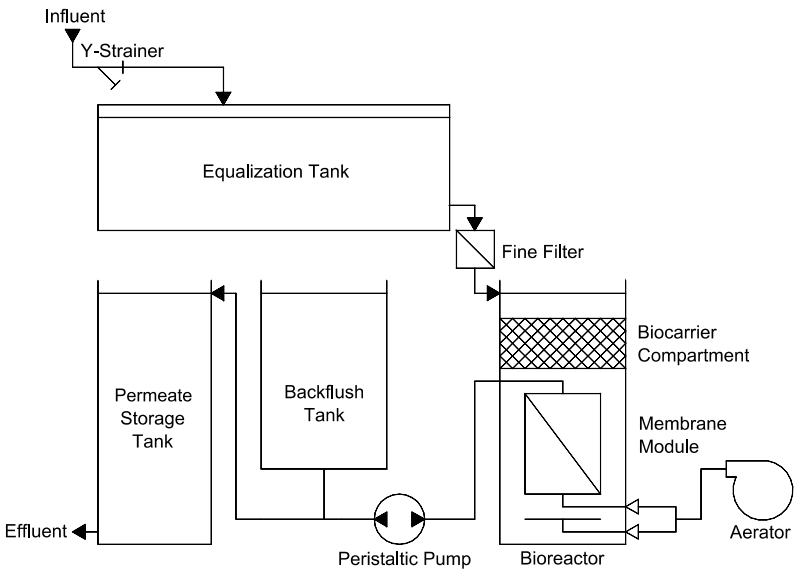

Fig. 1 A schematic diagram of the HMBR greywater reclamation system

Table 3 Experimental variables

Trial Number	MLSS (mg/L)	HRT (h)	SRT (d)	Flux (L/d)	F/M Ratio
1	1000	8	7	159	0.95
2	2000	8	9	159	0.40
3	3000	8	11	159	0.28
4	4000	8	14	159	0.23

F/M ratio per trial are summarized in Table 3. Before each trial, the mixed liquor was gradually diluted to the desired concentration by removing a fraction of sludge and replacing it with tap water. The desired concentration was then maintained by wasting a fraction of sludge daily. This method yielded a relatively constant MLSS concentration throughout the duration of each trial. MLSS concentrations did not vary by more than 300 mg/L of the desired value. Solids retention times ranged from 7 to 14 days depending on the MLSS concentration being tested. Following each adjustment in MLSS concentration, the reactor was subjected to a 10 day period of acclimation before water quality analysis began.

2.5 Sample collection

The reactor contained two sampling ports, one located at the midpoint of the reactor and the other near the base, which were used for mixed liquor sample collection and sludge removal, respectively. The mixed liquor was thoroughly mixed before a sample was drawn so that the concentration of MLSS was approximately isotropic with respect to depth. Mixed liquor samples of 250 mL were then drawn from the midpoint sampling port. Dissolved oxygen measurements were taken immediately following extraction using a CellOx 325 DO probe. Temperature measurements and pH measurements using an InoLab pH 730 pH meter were also taken on the extracted sample. A separate mixed liquor sample of 250 to 500 mg/L was taken for MLSS and mixed liquor volatile suspended solids (MLVSS) analysis. Influent greywater samples were taken from a sampling port located near the base of the equalization basin. The stored greywater was thoroughly mixed before samples were drawn. Effluent samples were taken from a sampling port between the HMBR and permeate storage tank.

2.6 Sample analysis

Temperature, DO, pH, MLSS, and MLVSS were monitored daily to ensure consistent biological conditions during each trial period. Temperature, DO, and pH were maintained at 20.4 ± 0.2℃, 2.67 ± 0.29 mg/L, and 7.36 ± 0.09, respectively. Influent and effluent samples were characterized in terms of 5-day biochemical oxygen demand (mg/L BOD_5), chemical oxygen demand (mg/L COD), turbidity (NTU), ammonia (mg/L NH_3-N), total phosphorus (mg/L PO_4-P), total kjeldahl nitrogen (mg/L N), total nitrogen (mg/L N), nitrite (mg/L NO_2-N), and nitrate (mg/L NO_3-N). Treatment performance was measured in terms of removal efficiency and contaminant residual concentration. MLSS and MLVSS were measured according to Standard Methods (APHA 2005). Chemical oxygen demand, NH_3, TP, TN, nitrite, and nitrate were measured using Hach test kits in conjunction with two Hach DRB 200 ovens and a Hach DR 2800 spectrophotometer. Total kjeldahl nitrogen was determined using the nitrogen relationship shown in Eq. (1).

$$TKN = TN - (NO_3^- + NO_2^-) \quad (1)$$

Protocols for each measurement using the Hach test kits are described within each kit. Turbidity was measured with a Hach 2100N turbidometer. Biochemical oxygen demand was measured using a series of 12 Oxitop-C measuring heads in conjunction with an OxiTop OC100 controller. Analysis procedures were followed as supplied by the manufacturer. Duplicate results were collected for each sample and sampling was repeated daily over three days for a total of six samples per parameter per trial, with the exception of BOD_5, which was repeated over the course of two days for a total of four samples per trial. This was due to the limited number of BOD_5 bottles available at the time. The total duration of each trial was approximately two weeks. Food-to-microorganism ratio was determined using the relationship shown in Eq. (2), where Q = flow rate and V = reactor volume.

$$\frac{F}{M} = \frac{BOD_5\left(\frac{mg}{L}\right) \times Q\left(\frac{L}{d}\right)}{MLVSS\left(\frac{mg}{L}\right) \times V(L)} \quad (2)$$

The removal efficiency of each parameter was calculated using the relationship shown in Eq. (3), where m_1 and m_2 were the initial and final concentrations of each parameter, respectively. Turbidity was measured in nephelometric turbidity units.

$$Per\ cent\ removal = \left(1 - \frac{m_2(mg/L)}{m_1(mg/L)}\right) \times 100\% \quad (3)$$

For reuse in non-potable water applications, the residual concentrations of BOD_5 and turbidity were compared to the Canadian guideline values of ≤ 10 mg/L and ≤ 2 NTU, respectively (Health Canada 2010).

3. Results and discussion

3.1 Removal efficiency versus mixed liquor suspended solids

Trials 1, 2, 3, and 4 were operated at a fixed HRT of 8 h with MLSS concentrations ranging from 1000, 2000, 3000, and 4000 mg/L, respectively. The removal efficiencies of various contaminants during each of these trials are summarized in Fig. 2. Contaminant residual concentrations are summarized in Fig. 3. Biochemical oxygen demand removals of 93.7%, 96.3%, 99.2%, and 98.8%, and COD removals of 93.6%, 96.1%, 97.7%, and 97.8% were observed during each trial. As expected, the removal efficiency of BOD_5 and COD increased with increasing MLSS concentration owing to the direct increase in biodegradation capacity. This trend corresponds with observations made by other researchers (Liu *et al*. 2010, Banaei *et al*. 2013). Both BOD_5 and COD showed a positive correlation with MLSS concentration up to 3000 mg/L. However, little improvement was observed following an MLSS increase from 3000 to 4000 mg/L. This suggests that the organic materials entering the bioreactor were sufficiently removed via biological degradation or adsorption to the biomass at these MLSS concentrations. Further increases in MLSS are therefore unlikely to yield further organics removal at the given organic load and retention time. Mixed liquor suspended solids concentrations less than 3000 mg/L were insufficient to achieve optimal organics removal.

Turbidity removals of 99.5%, 99.7%, 99.8%, and 99.8% were observed during each trial.

Consistent turbidity removal was expected since most constituents contributing to turbidity were excluded from the permeate by the membrane module. The removal efficiency for turbidity was therefore confirmed to be largely independent of MLSS concentration.

Ammonia removals of 35.3%, 98.3%, 99.9%, and 99.8% were observed during each trial. Likewise, total kjeldahl nitrogen removals of 43.7%, 89.4%, 88.1%, and 95.1% were observed during each trial. At an MLSS concentration ≥ 2000 mg/L, ammonia removal was nearly complete, with approximately 1.6% increase when MLSS was adjusted from 2000 to 3000 mg/L. Significantly lower ammonia removal was observed at an MLSS concentration of 1000 mg/L. Ammonia removal is largely dependent on nitrifying bacteria which produce nitrite and nitrate during biological oxidation. In order to maintain a constant MLSS concentration at 1000 mg/L, regular sludge wasting was required. The average SRT during this time was seven days, though values as low as four days were observed. Nitrifying bacteria possess a relatively slower growth rate compared to other bacteria within the activated sludge. In the HMBR, the presence of media provides a stable environment for these bacteria, which protects them from washout during sludge wasting. However, the nitrifiers within the mixed liquor cannot benefit from this effect. Thus, washout may have been a contributing factor to the observed loss of ammonia oxidizing bacteria, in addition to the reduced biomass concentration. Persistent nitrification within the biofilm may have been primarily responsible for the observed 35.3% ammonia removal. It has been reported that up to 50 to 60% of the nitrification capacity of an HMBR can be attributed to the biofilm (Artiga *et al*. 2005). The greater retention time of the biofilm occupying the moving bed media permits a greater fraction of nitrifying bacteria within the bioreactor than would otherwise be present with suspended biomass alone (Artiga *et al*. 2005). Within the biofilm, nitrate produced during ammonia oxidation would have been quickly reduced to elemental nitrogen by simultaneous nitrification denitrification (SND) processes. At an MLSS of 1000 mg/L, nitrite and nitrate residuals were in fact undetected within the treated effluent in support of this explanation. Residual nitrite and nitrate concentrations are shown in Fig. 3.

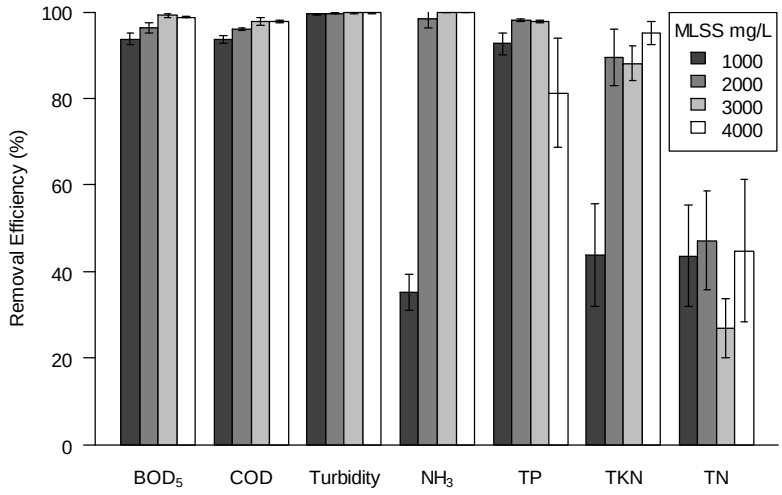

Fig. 2 Removal efficiency versus mixed liquor suspended solids concentration

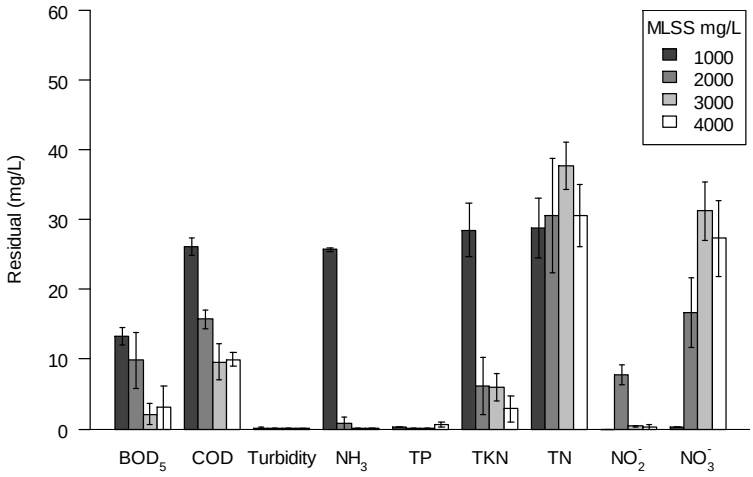

Fig. 3 Contaminant residuals versus mixed liquor suspended solids concentration

Total nitrogen removals of 43.6%, 47.2%, 26.9%, and 44.8% were observed during each trial. No discernable trend was observed in correlation to MLSS concentration. Poor TN removal within HMBRs has been attributed to the limited development of anoxic processes (Yang *et al*. 2012). This was partly unavoidable due to the aerobic environment of the HMBR. Limited anoxic zones within the bioreactor resulted in poor denitrification. This was evidenced by the generally high effluent nitrate concentrations. At an MLSS of 3000 mg/L, nitrate concentrations were measured to be 31.2 ± 4.2 mg/L. Within the aerated bioreactor, the interior regions of the biofilm comprise the main oxygen poor zone in which denitrification may take place. The inclusion of biofilm may therefore increase the rates of denitrification, and increased ammonia and TN removal following the addition of bio-media has indeed been observed (Yang *et al*. 2009). Therefore, it was assumed that denitrification was mainly limited to the SND processes within the biofilm and larger floc, which substantially limited the reduction of nitrate. Trials run at MLSS concentrations ≤ 2000 mg/L required SRTs of < 10 d, which is shorter than the regeneration time of denitrifying bacteria, and would have further diminished any denitrification within the suspended biomass. The result of this effect is a dependency of TN removal on biofilm concentration, not MLSS concentration. Since the biocarrier filling ratio remained constant throughout each trial, TN removal remained relatively unchanged. Excessive nitrate residuals in trials run as MLSS concentrations > 1000 mg/L indicate that a filling ratio of 20% was not optimal for nitrogen removal and that increasing the amount of biofilm, and hence denitrification potential, would yield greater TN removal.

Total phosphorus removals of 92.6%, 98.1%, 97.9%, and 81.2% were observed during each trial. No discernable trend was observed in correlation to MLSS concentration. Considering the low influent concentrations of phosphorus, it was likely a limiting nutrient within the reactor. Therefore, available amounts would have been readily consumed by the biomass during regular cell development. Little to no residual phosphorus was detected in the treated effluent as shown in Fig. 3.

3.2 Removal efficiency versus food-to-microorganism ratio

Food-to-microorganism ratio directly affects the metabolism and growth processes of the biomass, which in turn affects treatment performance. Low F/M ratios limit cell growth and

therefore promote the synthesis and utilization of storage products while high F/M ratios favour biomass replication (Lobos *et al*. 2008). High F/M ratios can be used advantageously for the fast development of large granular sludge particles with excellent sludge settling properties (Li *et al*. 2011). However, higher F/M ratios increase sludge production and the concentration of solubilized lysis by-products (Lobos *et al*. 2008). This corresponding increase in fine particles and soluble microbial products may account for the increased fouling observed in systems operated at a high F/M ratio compared to a low F/M ratio (Liu *et al*. 2012). In an HMBR, the settleability of sludge is of little importance since solid-liquid separation is achieved using a membrane module. Thus, the impact of F/M ratio on sludge settleability can be neglected. However the impact of F/M ratio on treatment performance is still a matter of interest as F/M ratio is a critical parameter in the design SRT. Trials 1, 2, 3, and 4 examined food-to-microorganism ratios of 0.95, 0.40, 0.28, and 0.23, respectively. The BOD_5, COD, NH_3, and TKN parameters were observed to be negatively correlated with F/M ratio, and the optimal removal efficiencies for these parameters occurred at an F/M ratio of 0.23 to 0.28. Turbidity, TP, and TN removals showed no significant correlation.

3.3 Mixed liquor suspended solids = 1000 mg/L

Several operational problems were encountered when the system was operated at an MLSS concentration of 1000 mg/L. At an MLSS concentration of 1000 mg/L, the MLSS appeared to consist primarily of planktonic bacteria and minimal flocculation was observed. Biofilm therefore made up the dominate portion of biomass within the reactor. Ammonia removal showed a marked decline at this concentration. The decline in ammonia oxidation was likely caused by a corresponding reduction in nitrifying bacteria due to the considerable loss of floc under these conditions. Nitrate concentrations were undetected at an HRT of 8 h. As such, the observed nitrate values were approximately 25 ± 7 mg/L NO_3-N lower than the values observed at an MLSS concentration of 2000, 3000, and 4000 mg/L. The severe loss of floc could not be easily recovered following the completion of the trials operated at 1000 mg/L MLSS and the reactor needed to be reseeded in order to reinitiate floc formation. Thus, it was determined that operation at or below this threshold yielded poor biomass quality and general system instability. It was further noted that the frequency of required membrane cleaning due to flux decline was greater at this concentration than during previous trials. This observation supports those made by other researchers who observed a sevenfold increase in the rate of membrane fouling in an attached growth system compared to a suspended growth system (Lee *et al*. 2001). Residual COD concentrations were notably higher when operating at 1000 mg/L MLSS, and BOD_5 residuals exceeded typical guideline values for non-potable greywater reuse. Incomplete degradation of organic matter at very low MLSS in the range of 1000 to 1500 mg/L has also been noted by other researchers (Kawasaki *et al*. 2011). Therefore, it is suggested that operation at or near 1000 mg/L MLSS be avoided.

4. Conclusions

The performance of an HMBR greywater reclamation system was characterized with respect to MLSS concentration and F/M ratio. It was found that the removal efficiency of BOD_5, COD, NH_3, and TKN increased with increasing MLSS, while turbidity, TP, and TN showed no significant change. The presence of attached biomass may have contributed to the overall removal efficiency of TN. Optimal BOD_5 and COD removals were attained at an MLSS concentration of 3000 to

4000 mg/L which corresponded to an F/M ratio of approximately 0.28 to 0.23, respectively. Reusable effluent was produced during operation at an MLSS concentration between 2000 to 4000 mg/L at an HRT of 8 h. At an MLSS of 1000 mg/L, the quality of the suspended biomass sharply declined leading to a severe loss in floc. As such, concentrations at or below this threshold were considered untenable for prolonged periods of operation. Overall, optimal performance was observed at an MLSS concentration of 3000 to 4000 mg/L. Turbidity and BOD_5 residuals were recorded at 0.076 ± 0.023 NTU and 2.1 ± 1.5 mg/L during this time, indicating that the HMBR could generate reusable effluent for non-potable greywater reuse.

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