



## Effect of effluent circulation and hydraulic retention time (HRT) on the performance of a modified anaerobic baffled reactor (MABR) during start-up period

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### ABSTRACT

Effluent circulation may affect the phase separation ability of a compartmentalized anaerobic reactor such as anaerobic baffled reactor. In this study, it is proven that this is not always the case. Moreover, the effect of circulation to this type of reactor under different hydraulic retention time (HRT) is still unclear. The present study investigates the start-up performance of a novel modified anaerobic baffled reactor (MABR) at various effluent circulation ratios ( $R$ ). Results showed that tremendous increase of the treatment efficiency and stable performance was achieved by the MABR system when effluent circulation was employed (e.g. 95.7% COD removal during  $R$  of 2 at HRT of 2 d and an OLR of  $0.75 \text{ kg COD m}^{-3} \text{ d}^{-1}$ ). The pH profiles, volatile acids (HOAc) occurrence and biogas production ( $L_{\text{biogas}} \text{ g COD}_{\text{destroyed}}^{-1}$ ) during the start-up period showed favourable conditions in the reactor. In addition, the effect of HRT variations (4, 3, 2 and 1 d) to the MABR with circulation operation  $R$  of 2 (optimum circulation) showed the HRT of 2 and 3 caused the MABR to start-up rapidly and efficiently with a chemical oxygen demand removal efficiency of more than 90%. It was concluded that rapid start-up can be achieved by applying effluent circulation to the MABR.

*Keywords:* Anaerobic reactor start-up; Effluent circulation; Modified anaerobic baffled reactor (MABR); Hydraulic retention time (HRT)

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## 1. Introduction

Among the high-rate anaerobic reactors, anaerobic baffled reactor (ABR) can be considered as one of the most convenient anaerobic treatment system. Its design ensures contact of biomass with substrates without the need to use any mechanical mixing. This is done by the narrow down-flow and the wide up-flow inside each compartment of the ABR. The reactor is better than other bioreactors in terms of long sludge retention time, good confrontation to organic shock loading, and its exclusive capability to separate phases of anaerobic microbial activity [1]. Starting up an anaerobic reactor is a very essential yet delicate procedure. During this process, vigorous and steady biomass were cultivated, accumulated, and maintained in high concentration to make the reactor ready to perform treatment of wastewater [2]. Therefore, this process requires appropriate planning. The sooner an anaerobic reactor achieved start-up the better.

Past investigations have reported comparable anaerobic reactor start-up strategy, duration and performance. Ferraz et al. [3] reported the treatment of a cassava wastewater using ABR with continuous feeding of low OLR ( $0.5 \text{ kg COD m}^{-3} \text{ d}^{-1}$ ) with a slow HRT of 4 d. The start-up was carried out for 30 d with 82–92% COD removal. On the contrary, Bodkhe [4] successfully start-up their ABR system without inoculums with a very low OLR ( $0.067 \text{ kg COD m}^{-3} \text{ d}^{-1}$ ) and HRT (6 d) resulting in the treatment efficiency of more than 90% COD removal in 90 d. Ji et al. [5] demonstrated that only 65% COD removal could be achieved in an ABR treating heavy oil produced water during start-up when the reactor was operated for 164 d at an OLR of  $0.2 \text{ kg COD m}^{-3} \text{ d}^{-1}$  and HRT of 5 d. Recently, Zwain et al. [6] studied the start-up performance of a modified anaerobic baffled reactor (MABR) treating recycled paper mill wastewater and reported a chemical oxygen demand (COD) removal of 71% on day 30 at an HRT of 5 d.

It is known that circulation from methanogenic to acidogenic phases enhances the hydrolysis in compartmentalized reactor such ABR [7]. It is also reported that circulation could accelerate the rate of organic degradation in two-stage anaerobic digestion of vegetable waste [8,9]. In addition, Lee et al. [10] examined the thermophilic two-stage anaerobic digestion of high solid food waste for  $\text{H}_2$  and  $\text{CH}_4$  production at three different OLRs and the results implicated the function of circulation on pH adjustment and dilution. Saritpongteeraka and Chaiprapat [11] investigated performance of an anaerobic baffled reactors treating rubber latex wastewater at different recycling ratios

(0, 0.3 and 0.5). Considering the dependence of circulation effect on a variety of factors such as characteristics of wastewater, OLR and HRT, however, the effects of circulation on the performance of ABR under different HRT and OLR are still unclear. Accordingly, the main objective of this research was to investigate the effect of circulation on a MABR performance treating synthetic wastewater during start-up period. In addition, the effect of HRT during effluent circulation to the reactor performance was also examined.

## 2. Materials and methods

### 2.1. Modified anaerobic baffled reactor (MABR)

The MABR was a rectangular box consists of four identical Plexiglas compartments with a total active volume of 28 L (4 compartments of 7 L). The operational set-up, the flow diagram and the reactor design are presented in Fig. 1. Each compartment was further divided by slanted baffles to encourage mixing within each compartment (the detail of the baffle modification not provided), and within each compartment down-comer and up-comer regions were created. This produced efficient mixing and contact between the wastewater and biomass at the base of each up-comer. During up-flow, the waste flow contact with the active biomass and it is retained within the reactor providing the homogenous distribution of wastewater. The passage of liquid from one compartment to another was through an opening measuring  $10 \text{ mm} \times 50 \text{ mm}$  located about 23 mm from the top of each compartment. The influent wastewater entered through the first compartment. In the first compartment, wastewater will first go through a down flow area before being in contact with biomass at the bottom of the reactor and channelled to the up flow area. A series of vertical baffles forces the wastewater to flow under and over them as it passes from an inlet to outlet. The outlet of MABR was connected to a plastic U-tube for controlling the level of wastewater and to trap the solids. Gas production was monitored separately for each compartment using an optical gas-bubble counter having a measurement range of  $0\text{--}1.5 \text{ L h}^{-1}$  and precision within  $\pm 1\%$ . Each compartment was installed with a heater and the temperature was maintained at  $37^\circ\text{C}$ . A digital temperature probe located in each compartment provided the constant operation temperature. Peristaltic pump (Longer Pump BT100-2J) was used to control the influent feed rate to the first compartment of the MABR.

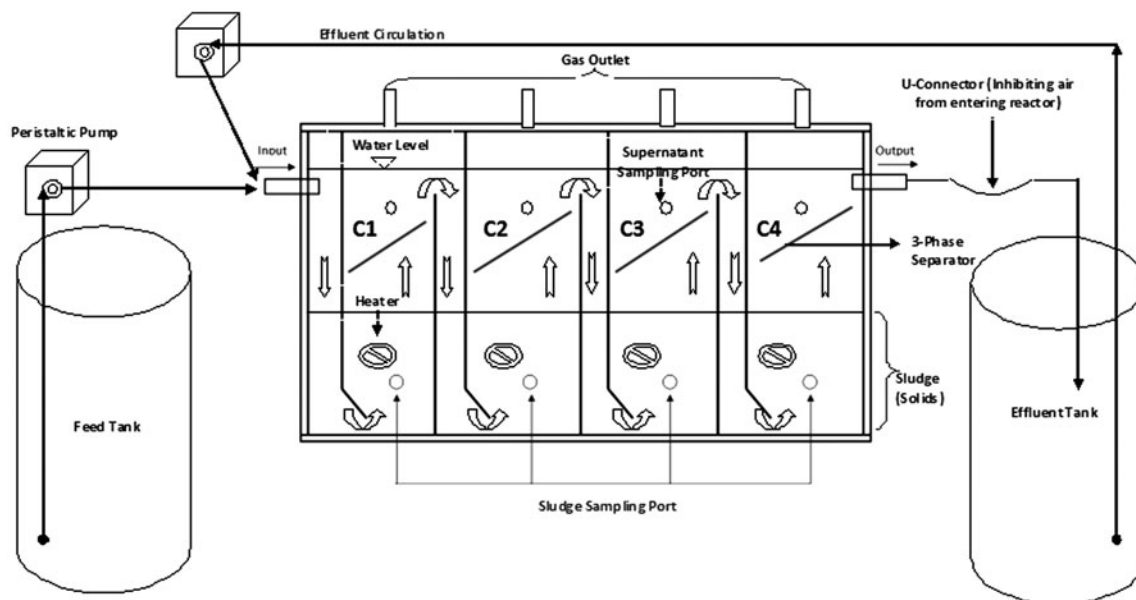


Fig. 1. Design and flow diagram of MABR system (C1, C2, C3 and C4 denotes compartment 1, 2, 3 and 4).

## 2.2. Seed sludge and substrate

The reactor was seeded with anaerobic digested sewage sludge (Bunus Sewage Treatment Plant, Kuala Lumpur). Twelve litre of sieved sludge (using 2.0 mm mesh) was added equally to each compartment (3 L in each compartment), the remaining volume being filled with tap water. This amount of sludge contributed substantially to the solid requirement in the reactor system after settling. The sieved sludge contains total solids of  $30,100 \text{ mg L}^{-1}$  and total volatile solids of  $9,525 \text{ mg L}^{-1}$ . After seeding, the head plates were attached and the headspace above each compartment was flushed with nitrogen gas to displace residual air in the system before introducing the feed. The reactor was allowed to stabilize at  $37^\circ\text{C}$  for 7 d without further modification.

As for the substrate, glucose was used in this study because of its degradation simplicity and high COD value. Glucose is a sugar with the molecular formula of  $\text{C}_6\text{H}_{12}\text{O}_6$ . The pH of the substrate is neutral (pH 7.0) and the COD was according to the desired OLR during the treatment process. Noike et al. [12] stated that the glucose is a soluble carbohydrate that is readily degradable and it will not limit itself from anaerobic biodegradation rate. It generates simply measurable intermediary metabolites in anaerobic digestion and is widely used in experimental studies as carbonaceous substrate [13]. The ratio to correct macronutrient deficiency was selected as COD:N:  $p = 250:5:1$ . The nutrient deficiency was corrected using macronutrients, Nutrient (N) 100 (Table 1) [14].

The alkalinity was maintained in all the reactor compartments at  $1,000\text{--}2,000 \text{ mg L}^{-1}$  as  $\text{CaCO}_3$  using sodium hydroxide (NaOH).

## 2.3. Reactor operation

This study was carried out in four (4) main steps; (1) start-up of MABR, (2) effect of effluent circulation ratio ( $R$ ), (3) start-up with effluent circulation and (4) effect of HRT with effluent circulation. In the beginning, the MABR was initiated with OLR of  $0.25 \text{ kg COD m}^{-3} \text{ d}^{-1}$  and HRT of 4 d for a total of 82 d. After that, the MABR underwent the effect of effluent circulation ratio study of 26 d. During this period, three different effluents ( $R = 1.0, 2.0$  and  $3.0$ ) to feed ratio were tested with HRT of 2 d and OLR of  $0.75 \text{ kg COD m}^{-3} \text{ d}^{-1}$ . This was followed with the start-up with effluent circulation for 49 d. Using circulation ratio of 2, another start-up phase was investigated with COD feed concentration of  $1,000 \text{ mg L}^{-1}$  and HRT of 4 d (OLR of  $0.25 \text{ kg COD m}^{-3} \text{ d}^{-1}$ ). Finally, the effect of HRT (after HRT 4 d and OLR of  $0.25 \text{ kg COD m}^{-3} \text{ d}^{-1}$ ) with effluent circulation was executed for 29 d at an HRT of 3, 2 and 1 d (at an OLR of 0.33, 0.5 and  $1.0 \text{ kg COD m}^{-3} \text{ d}^{-1}$ ) (Table 2).

## 2.4. Sampling and analysis

Supernatant liquor, gas and sludge samples were taken separately for each compartment. In addition, gas production rate was determined separately for

Table 1  
Composition of macronutrient, N100

Parameter	Concentration
Crude protein	(min) 5%
Crude fat	(min) 2%
Crude fibre	(max) 8%
N.free extract	45%
Calcium	2%
Phosphorus	1%
Magnesium	0.50%
Sulphur	2%
Potassium	2%
Salt	2%
Iron	0.08%
Iodine	0.03%
Boron	0.018%
Cobalt	0.0008%
Copper	0.0005%
Fluorine	0.015%
Riboflavin	8.00 mg
Manganese	0.09%
Molybdenum	0.0012%
Selenium	0.00002%
Zinc	0.005%
Vitamin A	50,000 IU
Vitamin D	3,000 IU
Vitamin E	150 IU
Vitamin K	1.00 mg
Vitamin B12	0.04 mg
Ascorbic acid	1,500.00 mg
Biotin	0.30 mg
Choline	50.00 mg
Folic acid	0.30 mg
Niacin	25.00 mg
Panthenic acid	0.20 mg
Thiamin	3.00 mg

each compartment using an optical bubble counter. Samples were analysed for every end of feed cycle (each feed cycle followed the operational HRT). Sample analysis included COD, pH, alkalinity, volatile acids (VA), suspended solids (SS) and volatile suspended solids (VSS). The measurement of SS and VSS was adapted from the procedures described in section 2450-D and 2450-E of standard methods [15].

Table 2  
Summary of reactor operating conditions in MABR

Phase	OLR (kg COD m <sup>-3</sup> d <sup>-1</sup> )	COD (mg L <sup>-1</sup> )	HRT (d)	Operational time (d)
(1) Start-up of MABR	0.25	1,000	4.0	82
(2) Effect of effluent circulation ratio	0.75	1,500	2.0	26
(3) Start-up with effluent circulation	0.25	1,000	4.0	49
(4) Effect of HRT with effluent circulation	0.33–1.0	1,000	3.0–1.0	29

Spectrophotometer (DR-2800) was used to measure the COD (as referred to the reactor digestion method adapted from Jirka and Carter [16]) and VA (as referred to esterification method adapted from Montgomery et al. [17]).

### 3. Results and discussion

#### 3.1. Preliminary MABR start-up performance

The MABR start-up was performed by maintaining a constant feed concentration (1,000 mg L<sup>-1</sup>) at an HRT of 4 d and an OLR of 0.25 kg COD m<sup>-3</sup> d<sup>-1</sup>. By maintaining a steady feed concentration with a long detention time, the reactor would achieve enhancement of solids build-up, encouragement of methanogenic populations and also rapid recuperation to hydraulic shock [18]. During the initial start-up period (until 28th day, Fig. 2(a)), the COD removal efficiency declined tremendously from 86 to 44%, while the pH profile in compartment 1 (C1) and 4 (C4) was in the range of 4.01–6.37 (Fig. 2(b)). To reduce the stressful condition in the reactor, the MABR has been left idle without feeding. This idle period gives ample time for the microbes to consume excess organic acids in the reactor. Similar trend was also observed when the MABR system operated again until 52nd day (Fig. 2(a)). Major fall of pH in the reactor could damage the anaerobic treatment process [19]; therefore, the reactor was then flushed with water and let idle again for 10 d. The highest COD removal (91%) was achieved on 66th day at pH of 6.7 (Fig. 2(b)). Despite this, the reactor performance tends to decline again on 82nd day. It was concluded that steady-state conditions were not achieved and effluent circulation was employed to speed up the start-up process.

#### 3.2. Effect of circulation ratio

During the degradation process, substrate underwent external mass transfer before successive intra-granule mass transfer and biochemical reaction within the granule [20]. Consequently, low hydraulic loading would restrict the degradation operation and

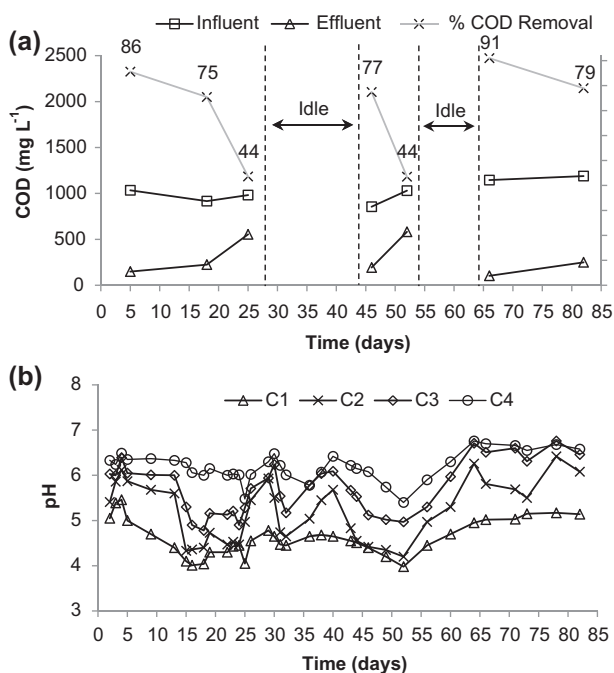


Fig. 2. MABR performance during initial start-up; (a) COD profiles and removal efficiencies and (b) pH profiles of compartment 1 (C1) and compartment 4 (C4).

microbial growth [20]. Accordingly, the MABR system was operated with effluent circulation on 82nd day. Different effluent circulation ratios ( $R$ ) were tested on the MABR performance. Circulation ratio is the ratio of effluent circulation flow to the feed flow. Effluent from MABR was recycled in three different ratios ( $R = 1$ ,  $R = 2$  and  $R = 3$ ) to the first compartment and the results are shown in Fig. 3. The  $R = 0$ , when the reactor was operated at normal operation (no effluent circulation). Tremendous increase of treatment efficiency and stability achieved by the MABR system

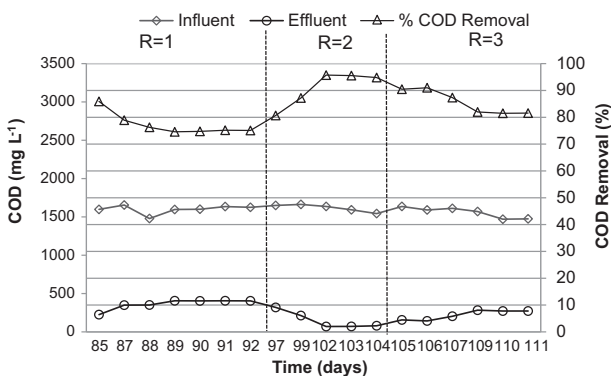


Fig. 3. Influence of effluent circulation ratio ( $R$ ) to COD profile and removal efficiencies.

when effluent circulation was employed, where the COD removal efficiency increased up to 95.7% during  $R$  of 2. According to Zhang et al. [19], circulations enhance start-up procedure by encouraging mass transfer between the substrate and biomass. The COD removal profile during the circulation study differs for each  $R$ . Highest COD removal efficiency was observed at  $R$  of 2, followed by  $R$  of 3 and 1. It should be mentioned here that  $R$  may not give better removal efficiency of COD. Therefore, finding the optimum  $R$  for the reactor was necessary. In the present study, circulating the effluent by two times the feed proves to be the most excellent way for circulation in MABR. Furthermore, the stability of the COD removal percentage during the effect of circulation ratio study approximately reflects that the MABR achieved a steady-state condition.

### 3.3. Start-up with circulation

The MABR impressively shows no lower COD removal efficiency than 70.9% since being started in 120th day (Fig. 4). The COD removal efficiency increased to 98.5% on 156th day. The reactor demonstrates a merely s-shaped acclimatization curve for start-up. Initially, interception of substrates and organics in the sludge bed caused lower COD removal; however, adaptation of organics in sludge to the substrates increased and this acclimatization phase tends to improve the COD removal efficiency bringing it to a sufficient aggregation of biosolids in the reactor, hence reaching its steady state [21]. Circulation of effluent to the feed increased the interception and mass transfer rate between the substrate and organics.

The pH profiles, VA occurrence and biogas production during start-up show favourable condition in the reactor. The observed distribution of pH values was in the range between 6.0 and 7.5, which gives advantageous environment for methanogenic activity in the reactor [14]. As for VA, its high removal efficiency throughout the MABR confirms that the reactor was in stable condition. There are two distinct patterns of pH, VA and biogas yield profiles throughout the start-up operation (Fig. 4).

Throughout the first 30 d of operation, the pH profile was in the order of  $C4 > C3 > C2 > C1$ , with distinctive average values of 7.23, 7.15, 6.95 and 6.62, respectively. According to Cysneiros et al. [22], there are various stages of anaerobic catabolism in compartmentalized anaerobic reactor. The early compartment in the reactor is dominated by promptly emerging microbes that can withstand high substrates level. Conversely, the near last compartment is dominated

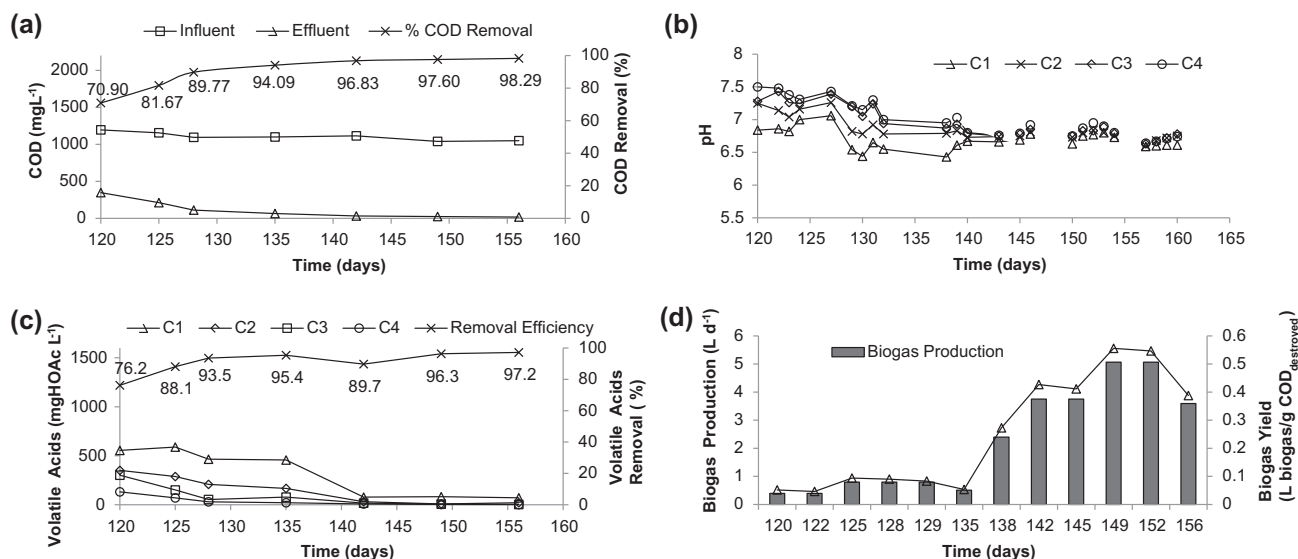


Fig. 4. Start-up of MABR with circulation; (a) COD profile and removal efficiencies, (b) pH profile, (c) VA profile and removal efficiencies and (d) biogas production and yield.

with gradually emerging forage microbes that favour high pH for growing.

In all compartments of MABR, pH initiates at the lowest level and hike rapidly after 10 d of operation. During this time, the COD removal was also highly increased. This is in contrast of what usually happened in the initial operation of start-up without circulation. Usually, the pH would fluctuate signifying that adjustment of the micro-organisms takes place to preserve the desired pH levels (6.6–7.7) for an anaerobic reactor [6]. In the present study, a fast growing acidogens were not outnumbering the slower growing methanogens and certain acetogens. This is probably because the circulation has provided more methanogens to the first compartment, thus maintaining the desired pH. This confirms that the circulation helps increase front pH of the reactor. Subsequently, this also reduces the abundance appearance of VAs and dissolved hydrogen [6].

Meanwhile at this point of the present study, the fact that circulation brings methanogens to the initial compartment of MABR did not affect the phase separation ability of the MABR. VA occurrences in MABR reduced towards the end compartment, which was with the order of C1 > C2 > C3 > C4. Higher VA in the initial MABR compartments (average 516.5 mg HOAc L<sup>-1</sup>) as compared to the final compartment (63.3 mg HOAc L<sup>-1</sup>) may be caused by excessive activity of hydrolytic and acidogenic micro-organism as contrast to methanogenic micro-organism [6]. Phase separation in the MABR was confirmed in this period of operation when the VA in the next compartment

decreased while the pH increased. This illustrates that diverse microenvironments in the MABR select the dominant population of microorganism. The VA removal efficiency increased from 76.2 to 95.4% during this operation period proves that methanogenic activity was not restricting the MABR performance and this could be operated at higher OLR [23]. However, the observed biogas productions during this operation period were quite low if compared to its COD destroyed, and VA removal in the reactor. During these early 30 d of operation period, only less than a litre of biogas was observed per day. This was probably due to technical condition with the bubble counting device during the experiment where the biogas might be released from MABR without accurately countered.

On the other hand, the end 20 d of start-up operation gives completely different scenario for the pH, VA and biogas yield. Although the pH order throughout the MABR followed C4 > C3 > C2 > C1, there were no distinctive variations amongst the average amounts of each compartment (C4: 6.79, C3: 6.77, C2: 6.75, and C1: 6.68). It can be concluded that all pH values across the MABR for this period of operation was in average of 6.8 which is favourable to methanogens bacteria activity. As for the VA, even though the removal efficiencies tend to be higher than the previous period with maximum removal of 97.2%, the observed occurrences were extremely low throughout the reactor (less than 85 mg HOAc L<sup>-1</sup>). This was in contrast with the biogas observations. Initially, the biogas yield was 0.3 L<sub>biogas</sub> g COD<sub>destroyed</sub><sup>-1</sup> and increased gradually to

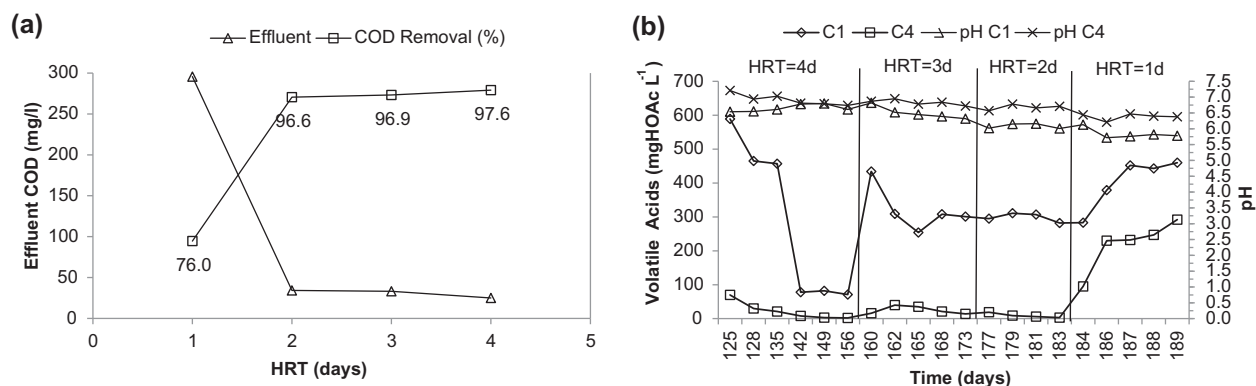


Fig. 5. Effect of HRT on MABR operated with circulation ratio ( $R$ ) of 2; (a) effluent COD profile and removal efficiencies and (b) VA and pH profiles.

reach double the value towards the end of the operation period ( $0.6 \text{ L}_{\text{biogas}} \text{ g COD}_{\text{destroyed}}^{-1}$ ). Other than showing that the initial technical condition has been fixed, it suggests that the reactor was working fine.

Solera et al. [24] reported that acidogenic bacteria have the optimum pH from 5.2–6.5 with over 2 d of specific growth rate; in addition, acetate, propionate, butyrate,  $\text{H}_2$  and  $\text{CO}_2$  are conversions products of organic substance by acidogenic bacteria. Favourably, selected of these products are capable of being metabolized by methanogenic bacteria which are acetate and  $\text{H}_2$  [24]. Therefore, during the last 20 d of the start-up experiment, the very low of acetate occurrences and relatively high pH of 6.8 suggest that methanogenic bacteria populated in the reactor but did not have the opportunity to produce massive methane ( $\text{CH}_4$ ) by metabolizing the few amount of acetate. This was also documented by Badiei et al. [25] where immediate drop in acetate concentration coupled with an increase in the  $\text{H}_2$  production observed. Relatively, all of the discussed condition of pH, VA and biogas during the final 20 d of the start-up experiment confirms the setback of circulation mentioned by [18]. However, this might be due to very low HRT caused the methanogens from the recycled effluent (which is two times the feed flow) to overcome the rapid growth of acidogens in the first compartment of MABR. This situation might be resolved by increasing the OLR. During the start-up experiment, the maximum VSS observed was only  $40 \text{ mg L}^{-1}$  (data not provided). This might be due to long HRT and low OLR. Therefore, sludge washout was neglected [26].

### 3.4. Effect of HRT

The performances of MABR with circulation at various HRT were investigated. It is crucial to know

the behaviour of MABR performance at various HRT because cost-effective design in practical application of any anaerobic treatment depends on the minimum HRT possible. The OLR was slowly increased throughout this operation by gradually reducing the HRT from 4 to 1 d. The effect of HRT variations to the MABR with circulation operation was presented in Fig. 5. Reducing the HRT to be less than 2 d was observed to decrease the COD removal efficiency excessively from 96.6 to 76% (Fig. 5(a)). However, when the HRT was increased to 3 and 4 d, the COD removal efficiency increased to 96.9 and 97.6%, respectively. Therefore, increasing the MABR HRT of more than 2 d significantly increases its COD removal efficiency. However, COD removal efficiency is not solely referred to in determining the optimum HRT for the MABR.

The VA and pH profiles in C1 and C4 of the MABR are illustrated in Fig. 5(b). In relative to COD removal, the pH profile in the reactor system was stable when the HRT was increased from 1 to 4 d. The average pH in C1 and C4 for HRT 1–4 d was 5.84, 6.09, 6.50, 6.65 and 6.38, 6.68, 6.83, 6.92, respectively. This establishes the fact that circulation helps to overcome the low pH problem in MABR.

In the meantime, interesting observations have been shown by the VA occurrences. Since operating the MABR continuously at HRT of 4 d resulted in a single phase, increasing the OLR by reducing the HRT helps to counter the adverse effect. This is demonstrated by the VA in the reactor. During the transition of HRT from 4 to 3 d, a spike in VA in C1 was observed (from 71 to  $434 \text{ mg HOAc L}^{-1}$ ). Thereafter, the VA in C1 maintained at an average value of  $321 \text{ mg HOAc L}^{-1}$  until the end of HRT 3 d. The average VA in C1 during HRT of 2 d was  $299 \text{ mg HOAc L}^{-1}$ . HRT 3 and 2 d showed high VA

removal efficiency with 92.1 and 96.9% making them optimum HRT for the MABR. As for HRT 1 d, the average VA occurrence increased to 403 mg HOAc L<sup>-1</sup> with a low VA removal efficiency (45.7%). Relatively to the removal efficiency, the average VA in C4 for HRT 1 d was high with 219 mg HOAc L<sup>-1</sup>. Damasceno et al. [27] reported that the earliest sign of instability for an ABR is when the effluent VFA contains more than 150 mg L<sup>-1</sup>, thus the ABR performance started to depreciate. This is caused by the lowered amount of VFA converted to biogas. Zhang et al. [19] reported that as OLR increased (HRT decreased), gas production rate increased as well as VSS concentration in the effluent (solid washout). They suggest that the sludge also need to be recycled in the reactor. In the present study, the effluents recycled were pumped to the first compartment from the deep bottom of the effluent tank. Since no scouring takes place in the effluent tank, circulation the effluent also provide additional solids to the reactor. The most minimum HRT for an anaerobic reactor increases the compactness and cost effectiveness of the operation [4]. Apparently, HRT of 2 and 3 d is sufficient to give high COD removal, stable pH and also VA thus expected to produce biogas well.

#### 4. Conclusions

From the above results, it can be concluded that the circulation had effected positively to the MABR performance during the start-up period. Tremendous increase of treatment efficiency and stability achieved by MABR system when effluent circulation was employed, where the COD removal efficiency increased up to 95.7% during *R* of 2. The observed distribution of pH values was in the range between 6.0 and 7.5, which gives advantageous environment for methanogenic activity in the reactor. For the HRT study, reducing the HRT less than 2 d was observed to decrease the COD removal efficiency excessively from 96.6 to 76%. However, when the HRT was increased to 3 and 4 d, the COD removal efficiency increased to 96.9 and 97.6%, respectively.

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