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## Performance of the three-stages anaerobic tofu wastewater treatment during the second start-up process

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**Abstract.** Tofu industry in Indonesia develops rapidly with the demand of the community. Therefore it is necessary to pay attention to the condition of liquid waste as the by-product of the industry. Traditionally, small- to medium-scale tofu industries produce highly polluted wastewater in compare with the big industries. For this reason, research on the tofu wastewater treatment was carried out to study the stability of anaerobic process in a three-stages packed-bed system after two months of pause or no-feeding process. The experiment used tofu wastewater from one of the factories in Bandung; the untreated wastewater had 15.9 g/L of COD at pH 3.5. Observations were carried out for 26 days. The system consisted of three similar reactors that worked in serial; each has the capacity of 10 litres. Tofu wastewater was introduced from the bottom of the reactor flowing vertically upward to the outlet. The outlet of the first reactor flow to the bottom inlet of the second reactor, and subsequently the outlet of the second reactor fed the third reactor. Each reactor was placed at different altitudes to facilitate gravity flow from one reactor to the next at the same rate. The results of the experiment show that the system overall worked well in a continuous flow rate. The COD value could decrease from 15.9 g/L to 7.4 g/L on average at the third reactor, as well as the pH changed from pH 3.5 to 7.

### 1. Introduction

Tofu industry generates two kinds of waste: solid waste from the residue of the extraction process in the form of soybean fibre containing carbohydrates and proteins, as well as liquid waste from the pressing and washing processes which contains more soluble organic materials. Solid waste usually is sold for animal feed such as the cattle or sheep, but it can be processed into processed materials that can be consumed. Tofu wastewater from the tofu industry, in general, is directly discharged through sewer/drainage system or just simple piping system to the public sewer, river or directly dumped in the backyard of the factory without any further treatment process.

Liquid waste from tofu industry is known to have a high organic matter and COD (Chemical Oxygen Demand) content so that if it is directly discharged into the water body, it will reduce the environmental carrying capacity and produce a bad odour. In tofu production, coagulation is a very important process. Soybean will be ground with water to produce slurry. After boiling the slurry, it will be filtrated to get the soymilk. Using coagulant, the soymilk will be coagulated and form a cheese-like form. The cheese-like clumps will be filtered and pressed to form tofu cake. The filtrate liquid is the whey that will end up as wastewater. Without treatment processes, tofu whey as wastewater can cause a variety of negative impacts such as water pollution, sources of disease, bad odour, increase the growth of microorganisms, insects (e.g. mosquitoes, flies), and reduce the



aesthetics of the surrounding environment. The wastewater has a high organic content and low pH (average pH is 3.5-4.5) [1]. Zhu *et al* in their study found that the main contents of tofu wastewater are sugar reductive, sucrose, starch and volatile fatty acids with total COD concentration of 10–15 g/L [2]. For such wastewater characteristic, aerobic treatment will be very costly due to the high required energy, therefore anaerobic treatment is more recommended.

There is more value that can be obtained during the anaerobic process if the process goes well. It produces less sludge, has a relatively cheaper operational cost, and can produce biogas (methane), which can be used as a source of energy for heating and other applications [3,4]. Anaerobic digestion performance is influenced by several factors including temperature, pH, the ratio of carbon-to-nitrogen, organic loading rate, and hydraulic retention time [5].

Reactor design in an anaerobic system is one of the crucial factors that can determine the whole process cost. The challenge in the engineering aspect is to keep the anaerobic biomass as long as possible in the reactor, whereas at the same time to reduce the hydraulic retention time of the waste. Uncontrollable retention time reduction could jeopardize the system, especially if the important methanogen bacteria fail to grow properly. The simple reactor system for the anaerobic process is a mixed or one-stage reactor, where there is no strict separation between the acidogenesis phase and methanogenesis phase. To increase the performance of the anaerobic process, separate reactors can be used to maximize the performance of each phase. Two-stage anaerobic reactors show a good performance to treat wastewater with high concentration of carbohydrate and protein [6,7].

In a fixed-bed reactor, biomass will be held and fixed in the system longer using media (packed-bed). Packing material can be stone or plastic packing [8,9].

In this study, wastewater from tofu processing industry was treated using a continuous three-stages anaerobic packed-bed system. The purpose of this study was to determine the performance of the three-stages system during the second start-up in processing tofu wastewater. The experiments investigated the stability of the process after a long pause by observing the removal performance in the three-stages system. This research was expected to provide an overview of the stability of such design in treating the tofu wastewater.

## 2. Material and methods

### 2.1. Materials

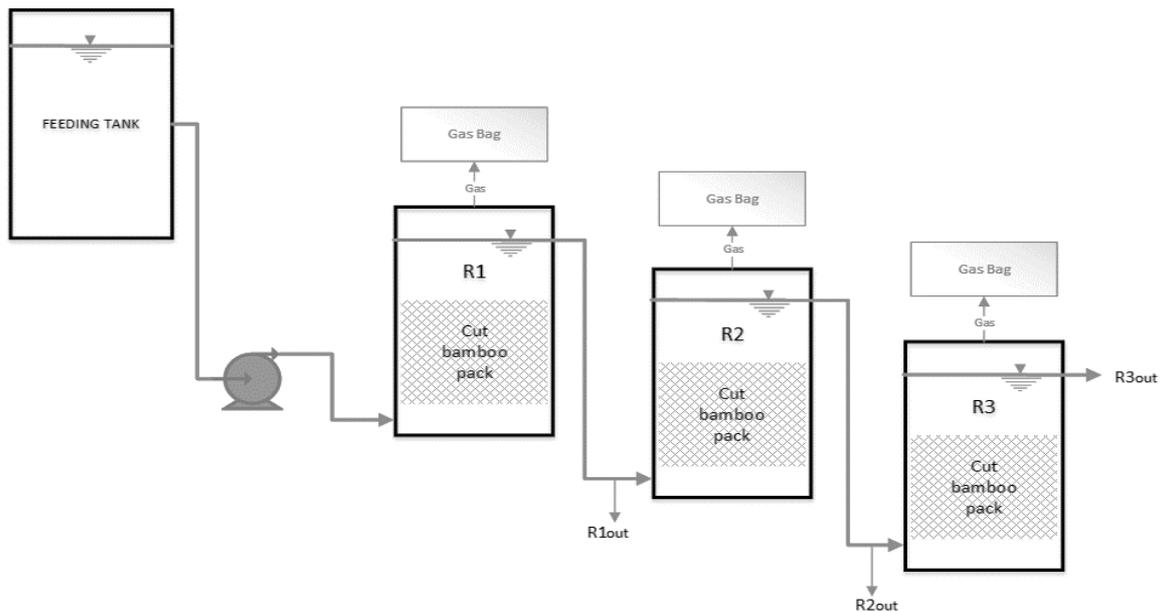
The start-up experiment used tofu wastewater obtained from tofu factories located in Bandung City, West Java, Indonesia. The wastewater used was the liquid from coagulation and pressing processes of soymilk.

The three-stages anaerobic packed-bed reactor was initially seeded with cow rumen obtained from a local slaughterhouse as the microbial source for the reactors. The reactor had previously been used for 3 months in the start-up process and then stopped for 2 months. Without any added microbes, the reactor was directly used in the experiment.

### 2.2. Experimental setup

The experiment was conducted in a three-stages anaerobic packed-bed system at the laboratory scale (Figure 1). The reactors were made from PVC pipes with 8 inches inner diameter. The total volume of each reactor was 12 litres with 10-litres working volume. Each reactor was filled with 91 cutting small bamboo of 5 cm length and the average outer diameter of 3 cm; the bamboo was arranged in a vertical configuration. The substrate flowed from the bottom inlet to the upper outlet (up-flow). The experimental conditions are presented in Table 1.

This setup was already used for treating tofu wastewater and reached a steady state at the substrate flow rate of 8 L/day, but then it was stopped during a repair period and long holiday for about eight weeks. After a long pause, the process was started again at the same flow rate as before, which was 8 L/day. The performance of each reactor to stabilize the pH and convert COD to methane was observed during the start-up period of 26 days.



**Figure 1.** Diagram of the three-stages packed-bed system.

**Table 1.** Characterization of tofu wastewater and the three-stages reactor.

Parameter	Unit	Value
<b>Untreated tofu wastewater:</b>		
- Total COD	g/L	15.9
- Filtrate COD	g/L	13.5
- pH	-	3.5
- C/N ratio [10]	-	5
<b>Three-stages reactor:</b>		
- Total volume	l	3 x 12
- Working volume	l	3 x 10
- Bed porosity	-	0.92
- Hydraulic retention time	day	3 x 1.25
- Temperature [11]	°C	23 ± 1

### 2.3. Analysis

The effluent from the third reactor (R3) was analysed every day for pH and COD concentration. The effluents from the first and second reactors (R1 and R2) were analysed periodically. COD was measured using the closed-reflux spectrophotometric method with a UV-VIS Spectrophotometer (Shimadzu GC14-A, Japan) at 615 nm. The pH of the sample was measured by using a pH-meter (LT Lutron pH-207, United States)

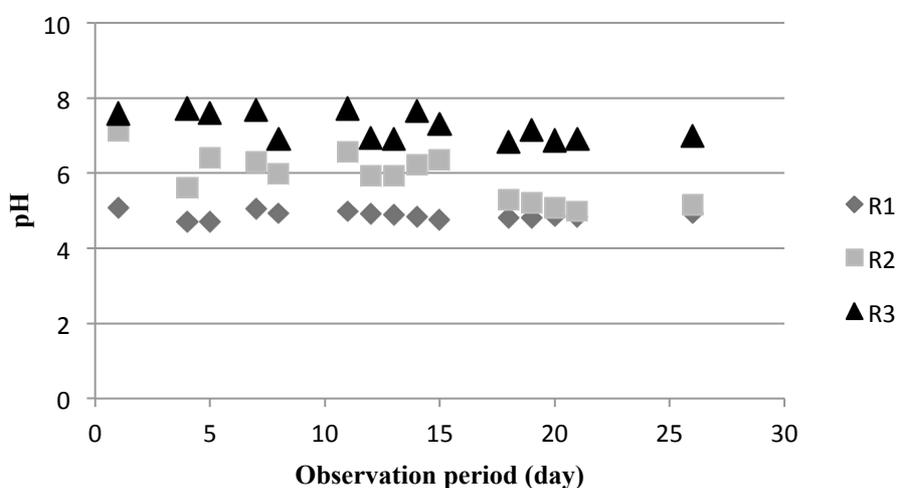
Biogas was collected in gas bags attached to each reactor (Figure 1). Biogas volume was measured using a wet gas meter (Ritter, Germany). Biogas composition was measured using a handheld multigas analyser (Gasboard 3200 plus, China).

### 3. Results

The characteristics of fresh tofu wastewater for feeding were 3.46, 15.9 g/L, and 13.5 g/L for pH, total COD, and filtrate COD, respectively (Table 1). The experiment had a constant flow rate of 8 L/day, the hydraulic retention time in each reactor was 1.25 days.

**Table 2.** The performance of three-stages anaerobic reactors.

Parameter	Unit	R1	R2	R3	Total
<b>Reactor outlet:</b>					
- pH	-	$4.9 \pm 0.1$	$5.9 \pm 0.6$	$7.3 \pm 0.4$	-
- Total COD	g/L	$14.9 \pm 0.8$	$11.5 \pm 2.0$	$7.4 \pm 1.9$	-
- Filtrate COD	g/L	$12.6 \pm 1.3$	$9.1 \pm 1.9$	$5.5 \pm 1.9$	-
<b>Biogas:</b>					
- Biogas production	L/day	$2.4 \pm 1.4$	$7.0 \pm 3.8$	$10.0 \pm 1.4$	$19.5 \pm 8.5$
- CH <sub>4</sub> production	L/day	$0.3 \pm 0.1$	$3.6 \pm 2.0$	$6.5 \pm 3.2$	$10.3 \pm 4.4$
- CH <sub>4</sub> concentration	%	$23 \pm 18$	$55 \pm 7$	$79 \pm 8$	$53 \pm 5$

**Figure 2.** pH value at the outlet of each reactor.

### 3.1. pH analysis

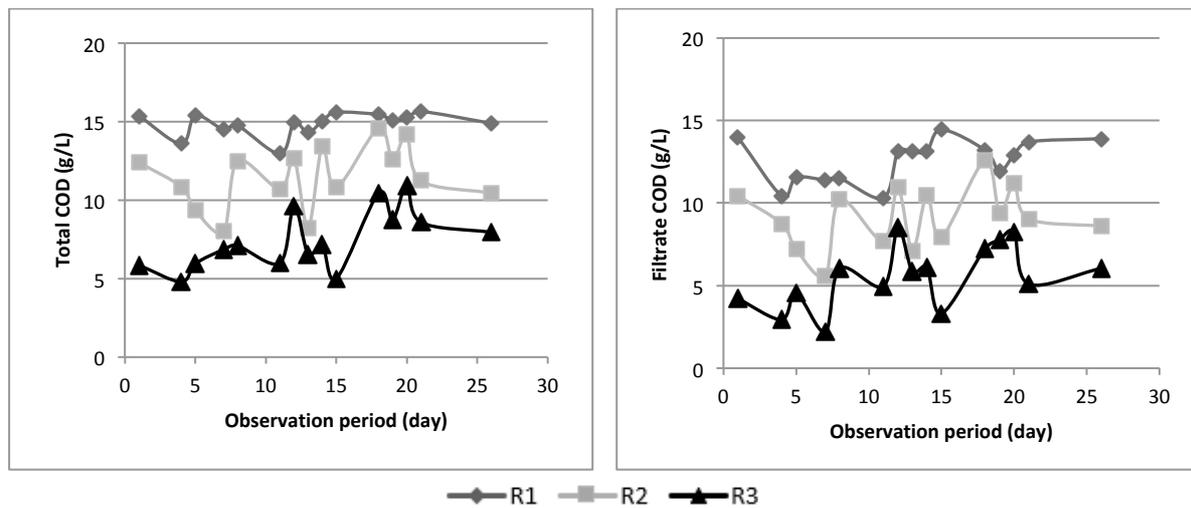
In the first reactor, the pH of the substrate could increase from 3.5 to 5. The pH in the outlet of the first reactor (R1) was relatively constant at 5 on average (Table 2, Figure 2).

After R1, the performance of the second reactor (R2) in the first 15 days showed a higher pH value compared with R1, i.e. 6-7, but then decreased slowly to pH 5 approaching the same pH like R1. The pH was constant at 5 until the end of observation time. In the third reactor (R3), the outlet pH increased from the average of 6 in the R2 to slightly above 7 in R3 in the first 15 days (Table 2), and then it decreased to a constant pH value of 7 until day 26 (Figure 2).

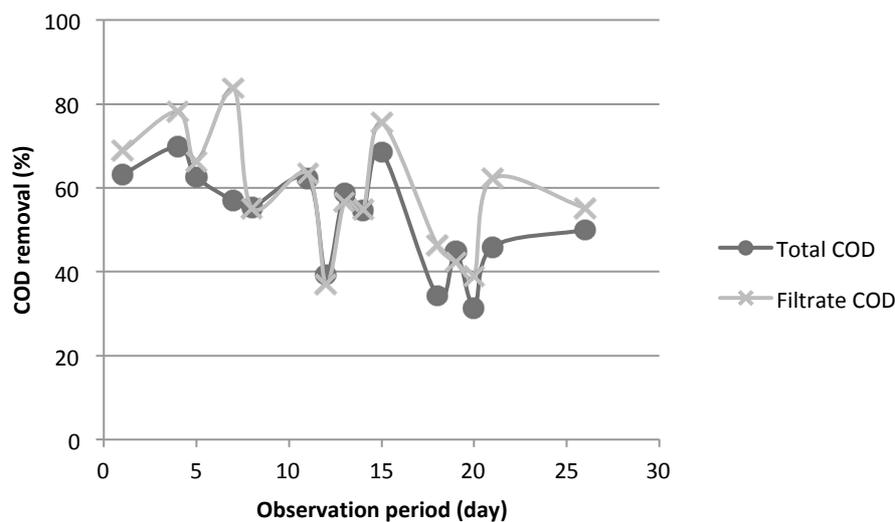
The 3-stages reactor system showed a stable condition and it did not need anything to keep the pH in the system, especially in R3, at the safety level. It is known that the anaerobic process, especially the methanogen consortia, is very sensitive to low pH.

### 3.2. COD concentration

In the 26 days of the experiment, COD concentration in each reactor showed similar patterns between the total and the filtrate concentration (Figure 3). During the observation, the average concentration of total COD in R1, R2, and R3 were 14.9, 11.5, and 7.4 g/L, respectively (Table 2). The concentrations of filtrate COD were lower than the total, on average 12.6, 9.1, and 5.5 g/L in R1, R2, and R3, respectively.



**Figure 3.** Concentration of total COD (a) and filtrate COD (b) at the outlet of each reactor.



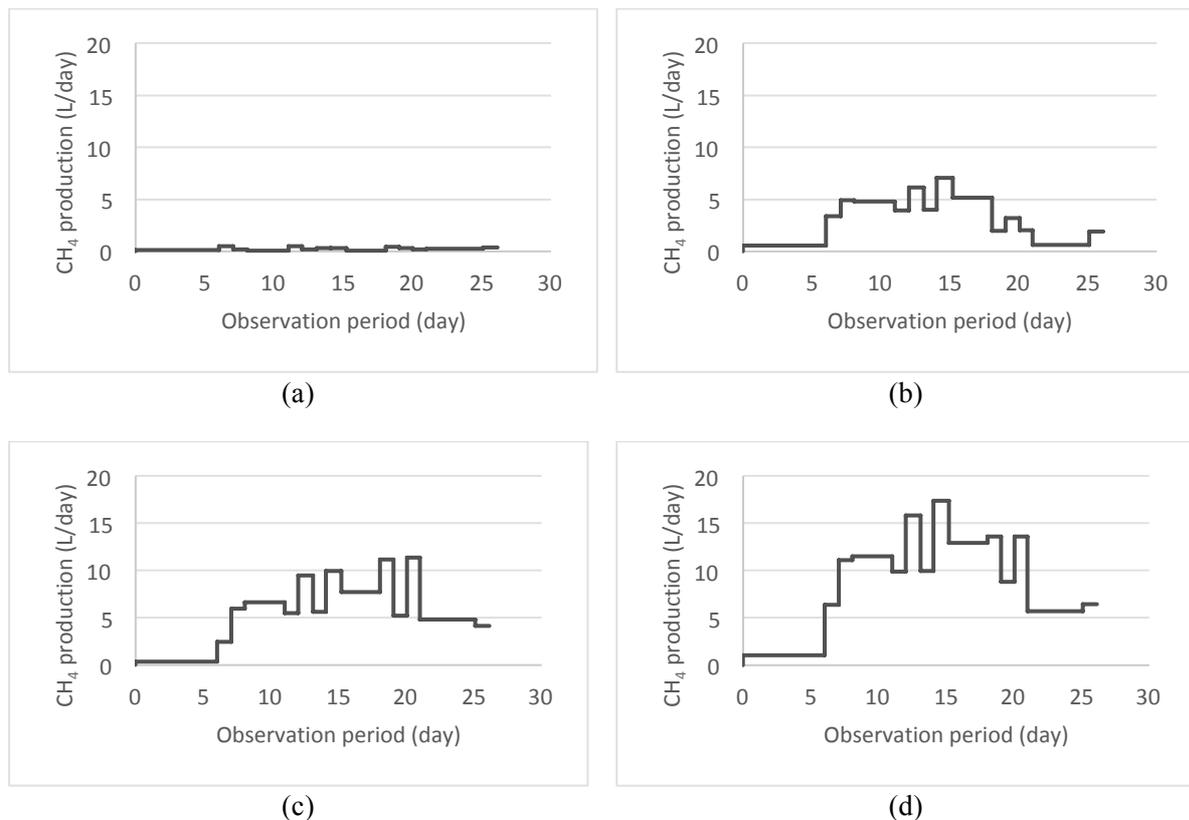
**Figure 4.** COD removal in the three-stages anaerobic reactor.

The removal rate in the three-stages anaerobic reactor is presented in Figure 4. In the first three days, the removal of the total COD was above 60% and then slightly decreased to an average of 50% after 25 days of the experiment. The pattern for filtrate COD was higher than the total.

### 3.3. Biogas production

During the 26 days of the start-up, the average biogas productions were 2 L/day from R1, 7 L/day from R2, and 10 L/day from R3; the total was 20 L-biogas per day on average (Table 2). Figure 5 shows that  $\text{CH}_4$  production started to increase from day-6 of the start-up.

The pattern of biogas production in each reactor fit the COD concentration in each reactor. In the R1, the COD was just slightly degraded and showed a hydrolysis process. The degradation of COD to methane and  $\text{CO}_2$  significantly happened in R2 and R3.



**Figure 5.** CH<sub>4</sub> production in (a) R1, (b) R2, (c) R3, and (d) Total.

#### 4. Discussions

A start-up in relatively short retention time, i.e. 1.25 day in each reactor or total of 3.75 days could be held by the system. The COD concentration fluctuated within 30% of the average value (Figure 3 and 4). The average COD removal compared to the influent was 12% for R1, 30% for R2, and 51% for R3. This indicates that methanogenesis already occurred in R1, but the dominant processes were hydrolysis and acidogenesis. It could be seen in the volume of methane gas in R1. The pH value and the produced biogas in R1 indicated the occurrence of hydrolysis and acidogenic processes. The increase of methane in R2 and R3, followed by the decrease of COD concentration in the outlet of R2 and R3 compared with their inlet, confirmed the role of R2 and R3 in the methanogenic phase.

This result also showed that systems with more stages gave a better performance with a higher COD removal rate. It might be due to the longer hydraulic retention time and more contacts between the substrate and microbial organisms for better biodegradation. In addition, multi-stage systems gave a better environmental condition (pH) for the biodegradation process. As shown in Figure 2, the pH increased gradually from 3.5 to above 7 when tofu wastewater was processed using the three-stages system.

A multiple-stages—normally two-stages—system is applied to avoid the impact or effect of fluctuation or shock loading in the system. For a continuous discharge of waste that needs to be treated immediately, a multiple-stage system could muffle the fluctuation in wastewater concentrations or flows.

To keep the methanogenic bacteria, which played a special role in degrading the organic load from the substrate in the anaerobic process, the pH value of the system should be kept between 6-8. During the anaerobic process, volatile fatty acids, ammonium and H<sub>2</sub>S can be produced as the degradation products of the process. The hampering effects of these products depend on the pH value because it can increase or decrease their toxicity [12,13]. Using a system of three-stages anaerobic packed-bed

reactors to treat tofu wastewater gave an advantage for the overall anaerobic process. The microbial consortium could be optimized at each stage. Judging from the pH value that was still quite low in the first reactor, it could be estimated that in this reactor the hydrolysis process was going well, but at the same time the removal process had occurred as shown by the 51% total COD removal or 57% COD filtrate removal (on average) compared to the influent COD.

The three-stages up-flow anaerobic fixed-bed system was proven to be stable to treat the wastewater that normally has a low pH such as tofu wastewater. As already known that the anaerobic process, especially the methanogenic phase, is very sensitive to pH. They work in the neutral range pH. Tofu wastewater with pH below 5 as input did not hamper the system at all, although we did not add any chemicals or materials to neutralize the system. It can be shown in Figure 2 that the pH in the outlet of R1 was already above 6 without using any additional neutralizing compounds.

## 5. Conclusions

Our results showed that the three-stages anaerobic system was stable enough to overcome the low pH of tofu wastewater. The multi-stage reactor played an important role in increasing pH gradually to an ideal condition for the anaerobic process. Even though the effluent quality was still higher than the maximum allowable effluent quality standard, the organic matter in tofu wastewater can be stably degraded by an anaerobic process with an average total removal rate of 51% using total retention time of 3.75 days. Further research is still needed to obtain good results, which in turn can help provide information regarding the good and right wastewater processing to the user.

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