

Original Research Article

## The Efficacy of Anaerobic Biofilter and Pre-Aeration with Microbubble Generator for Tofu Wastewater Treatment

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### Abstract

The majority of tofu manufacturers lack the necessary infrastructure to treat wastewater, resulting in the direct discharge of wastewater into water bodies. Such practices have the potential to results in environmental pollution. This study examines the efficacy of combining anaerobic biofilter technology and pre-aeration with a microbubble generator for the treatment of wastewater generated by the tofu industry. The research focused on the impact of hydraulic retention time (HRT) on the reduction of pollutants, specifically chemical oxygen demand (COD), biological oxygen demand (BOD), total suspended solids (TSS), and pH levels. The results indicated that prolonged HRT in the anaerobic biofilter significantly reduces pollutant concentrations. 48-hour HRT achieved reductions of 80.78% for COD, 78.53% for BOD, and 89.25% for TSS, respectively. The integration of a microbubble generator further enhanced treatment efficiency. The combination of a 48-hour anaerobic biofilter and a 180-minute microbubble generator achieved reductions of 93.82% for COD, 93.11% for BOD, and 97.5% for TSS. The effluent consistently met the pH quality standards set by the Indonesian government. The findings suggest the potential for optimizing retention times and combining anaerobic and aerobic treatments to address wastewater challenges in the tofu industry.

**Keywords:** wastewater treatment; tofu industry; hydraulic retention time; anaerobic biofilter; microbubble generator

### 1. Introduction

Tofu is a traditional food that is widely consumed (Pangestu et al., 2021) and (Irawan et al., 2020). It is a low-cost food that is produced using conventional methods (Pradana et al., 2018). However, the production process produced wastewater as byproduct. It is estimated that the tofu industry generates approximately 20 million m<sup>3</sup> of wastewater annually. The processing of 1 kg of soybeans in tofu production requires 45 liters of clean water per process, resulting the generation of an average of 43.5 liters of wastewater (Kholif et al., 2021). Wastewater from the tofu industry contains high levels of organic compounds, including protein (40-50%), carbohydrates (25-50%), and fat (10%) (Sari et al., 2022). The activities of the tofu industry are dominated by small-scale enterprises, the majority of which do not have waste treatment facilities. Consequently, the wastewater produced is discharged directly into water bodies (Laksono Putro et al., 2021) and (Pratiwi et al., 2022).

The disposal of wastewater from the tofu industry without treatment efforts can result in the degradation of environmental quality, the pollution of water bodies, and the disruption of aquatic lifeforms (Pagoray et al., 2021). Wastewater discharged from the tofu industry is subject to mandatory treatment before discharge into the aquatic ecosystem. The specific values in question are 200 mg/L for biological oxygen demand (BOD), 150 mg/L for chemical oxygen demand (COD), and pH values between 6 and 9, as specified in the Minister of Environment Regulation No. 5 of 2014, Annex XVII on Wastewater Quality Standards. Meanwhile, wastewater from the tofu industry is typically characterized by high concentrations of COD, BOD, and total suspended solids (TSS). In a study by Hardyanti et al. (2023), it was found that wastewater from the tofu industry exhibits different characteristics depending on the type of soybean raw material used. The results of their study demonstrated that wastewater from tofu production using soybean seeds from Wonogiri generated 444 mg/L, 4583.33 mg/L, 13.86 mg/L, and 3,481 mg/L of TSS, COD, ammonia, and BOD concentrations, respectively.

The treatment and reduction of the concentration of pollutants in wastewater from the tofu industry are achieved using various technologies. These include physical methods (sedimentation and filtration), chemical methods (chemical additives) and biological methods (using microbes). The latter can be carried out in anaerobic conditions or anaerobic-aerobic combinations. In the anaerobic biofilter process, biofilter media is used as a growth medium for microorganisms to form biofilms. The carbon source for microbial growth is derived from the wastewater flow (Pramita & Puspita, 2019) and (Ojha et al., 2021). Following the wastewater treatment can also be followed, an aerobic process utilizing a microbubble generator can be employed. The small-sized air bubble size has higher retention time, leading to an enhanced the oxygen transfer process (Mahdariza et al., 2023). This oxygen is used by aerobic microorganisms to decompose the organic materials present in the wastewater (Afisna & Juwana, 2020).

The anaerobic treatment employs the anaerobic fluidized bed reactor (AFBR) method, while the aerobic process utilizes microbubble generator technology. In a study conducted by Budhijanto et al., (2020), it was demonstrated that the 30% and 60% removal rates of organic compounds could be achieved for the treatment process with AFBR and its combination with the microbubble generator, respectively, with a hydraulic retention time (HRT) of 24 hours. These results indicate that using a microbubble generator could be a solution for wastewater treatment. Furthermore, Zunidra et al., (2022) reported the performance of anaerobic and aerobic biofilters in reducing pollutant levels in wastewater from the tofu industry. The filter media employed in this study was bio-ball. The seeding time for biofilm growth was 14 days, while the HRT of anaerobic and aerobic biofilters in the experiment was nine days. The results demonstrated the efficacy of COD removal at 49.6%, BOD at 64.4%, ammonia at 79.8%, and the capacity to elevate pH levels between 6 and 8. Other study have also provided comparable results. Ratnawati & Kholif, (2018) demonstrated the removal rates for 58.02% of BOD and 63% of COD, with an HRT of 24 hours. Moreover, the utilization of a microbubble generator to treat synthetic wastewater was found to reduce the concentration of COD from 500 mg/L to 100 mg/L and to increase the concentration of dissolved oxygen (DO) from 0 mg/L to 3.1 mg/L over five days, as reported by Afisna & Juwana, (2020).

In biological wastewater treatment processes, especially for industries like tofu production, the process duration can be long, particularly in anaerobic systems. Hydraulic Retention Time (HRT) is a critical factor for improving the efficiency of biological processes. The variation of HRT's duration from each anaerobic and aerobic process will be able to determinate the best conditions for the operational time of the reactor, because the reactor HRT has a significant influence on biological nutrient removal (Brown et al., 2011). As such, this study explores the effectiveness of combining anaerobic biofilter technology with pre-aeration using a microbubble generator. Microbubble technology plays a vital role in enhancing treatment time and improving process efficiency in terms of pollutant removal quality. By increasing the aeration time using microbubbles, there is a potential for reducing the overall treatment time. However, this increase also impacts the operational costs of wastewater management, particularly in terms of energy consumption. Anaerobic filter technology, while efficient in certain scenarios, faces

limitations in terms of biofilm surface area, which can reduce its effectiveness. To address this, the study aims to optimize the treatment time and improve pollutant degradation in a combined anaerobic-aerobic process. This is particularly relevant for industries like tofu production, which generates high organic load wastewater. The study will focus on optimizing both process time and treatment load by targeting specific wastewater parameters from the tofu industry. It will also address the limitations of anaerobic filter systems, examining how these can be enhanced in combination with microbubble-based aeration for improved treatment performance.

## 2. Material and Methods

### 2.1. The Wastewater Characteristics of Tofu Industry

The materials employed in this study comprise wastewater derived from the tofu industry, which was utilized as a sample of 35 L collected in a tank and will be transferred manually to an anaerobic process with a volume of 5 L. The wastewater was collected from the traditional tofu industry in Banda Aceh. Based on analysis of the laboratories, the wastewater characteristic of tofu industry is shown in Table 1. The result indicates that the BOD/COD ratio of tofu industry wastewater has a range of 0.27-0.649, with COD parameter values exhibiting high values. High values or strength wastewater typically containing of COD, BOD and TSS ranging from 1.000-5.000 mg/L, 800-1.500 mg/L and 200-800 mg/L, respectively (Fudala-Książek et al., 2018; Fudala-Ksiazek et al., 2014; and Rajab et al., 2017). This condition is crucial in determining the type of treatment technology to be employed, particularly when COD is above 1,000 mg/L. Anaerobic and aerobic processes can effectively treat wastewater whose BOD/COD ratio is greater than 0.5. However, aerobic treatment works best when COD levels are less than 1,000 mg/L (Campos et al., 2014). When COD levels are higher than 4,000 mg/L (Luo et al., 2016), anaerobic processes must be used as a first line of treatment before an aerobic process can be implemented. The anaerobic process is the optimal choice for the initial stage of treatment due to its compatibility with the conditions described above (COD approaching a value of 4,000 and a low BOD/COD ratio of 0.27), supported by a relatively acidic pH of 4,0-5,0. A comparison of the BOD/COD ratio of the test wastewater with that of previous studies by (Ida Zahrina et al., 2022 and Putri & Kartohardjono, 2018). For DO concentration in this study is relatively suitable for the aeration process, therefore, aeration is another feasible option for the technological process in tofu wastewater treatment.

**Table 1.** The characteristic of wastewater from tofu industry

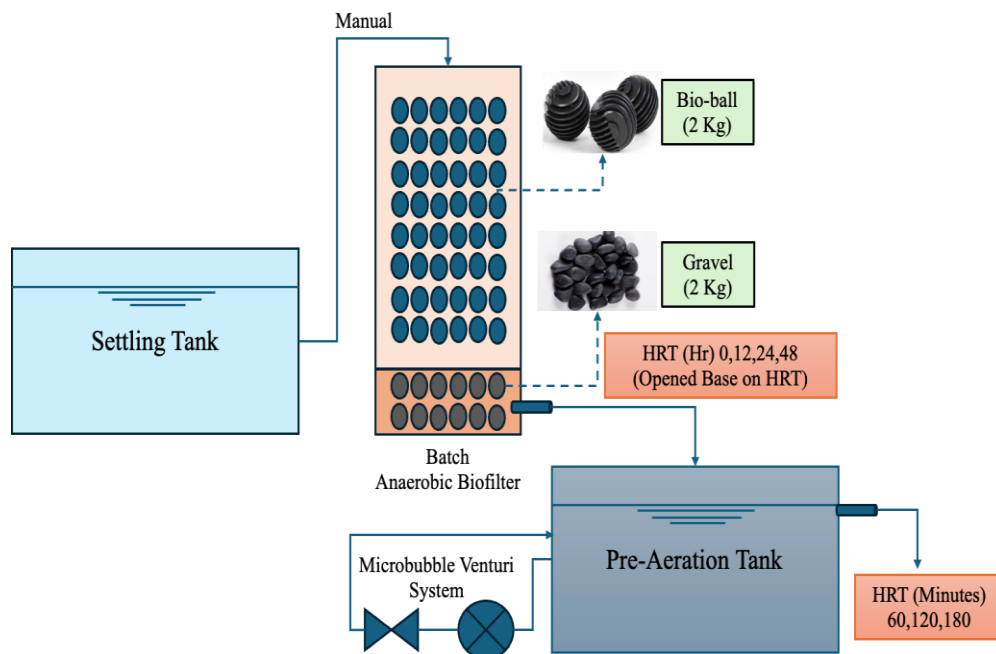
No	Parameter	Units	Value (this study)	(Ida Zahrina et al., 2022)	(Putri & Kartohardjono, 2018)	Standard Discharge*
1	pH	-	5,00	4,50	4,90	6,0-9,0
2	BOD	mg/L	1.020,00	3.246,00	3.575,50	150,00
3	COD	mg/L	3.773,00	5.000,00	6.500,00	300,00
4	TSS	mg/L	400,00	20,00	-	20,00
5	Ammonia	mg/L	11,47	-	36,10	-
6	DO	mg/L	3,70	-	-	-
7	BOD/COD	-	0,270	0,649	0,55	-

\*source: Minister of Environment Regulation No. 5 of 2014, Annex XVII on Wastewater Quality Standards.

### 2.2. Set Up an Anaerobic Biofilter Reactor and Microbubble Generator

The process is comprised of three distinct stages: the settling stage, the anaerobic stage, and the pre-aeration stage with microbubble water. The settling basin is constructed from plastic and has the following dimensions: 40 cm in length, 20 cm in width and 25 cm in height. The capacity of the tub is 20 L, which is sufficient for the wastewater produced by the tofu industry. The tank is equipped with a faucet,

which allows for the regulation of the flow to the anaerobic tank. The anaerobic filter reactor is constructed from glass with a thickness of 6 mm, a length of 15 cm, a width of 15 cm, and a height of 70 cm, with an effective volume of 5 L. In the arrangement of the anaerobic biofilter media, from the bottom to the top, the gravel with a diameter of 6-12 mm is positioned up to 10 cm high, and the bio ball with a diameter of 3-4 cm are positioned up to 45 cm high. Bio ball media filter has a specific area 200-235 m<sup>2</sup>/m<sup>3</sup> (Said, 2005). At the top of the media, 10 cm of space is provided, maintaining a water level 5 cm above the filter media. Following the anaerobic biofilter process, the pre-aeration reactor with a microbubble generator is constructed from glass with a thickness of 5 mm, a length of 30 cm, a width of 15 cm, and a height of 20 cm. The pre-aeration reactor has an effective volume of 5 L. The series of tools employed in this study can be observed in **Fig 1**. Subsequently, the tofu wastewater is directed to an anaerobic biofilter and pre aeration process for treatment in a variable of HRT. The reactor anaerobic bio-filter and pre-aeration tank and the system for this research is a batch mode based on HRT. The microbubble generator was self-constructed using PVC pipe fittings, an air hose, and PVC pipe. Amara AA-103 water pump (China) and an Amara AA-350 aerator (China) were integrated to the reactor. In addition, the equipment utilized in this research includes measuring cups, measuring pipettes, drop pipettes, pH meters, COD meters, test tubes, vacuum filtration apparatus, ovens, analytical scales, water pumps, aerators, saws, electric drills, and a variety of other laboratory aids.

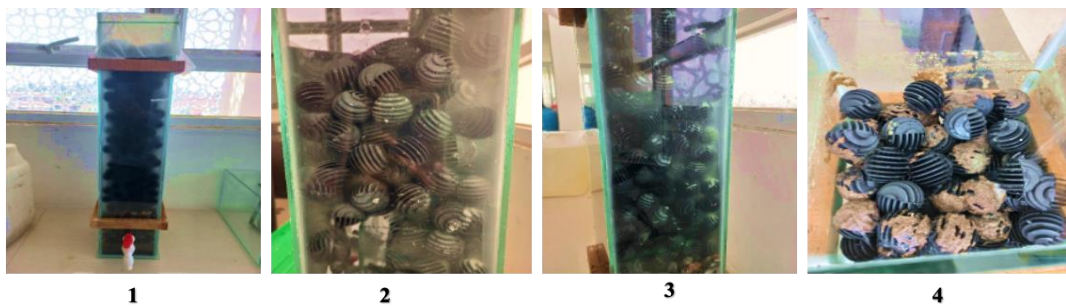


**Figure 2.** The configuration system of the anaerobic biofilter and pre-aeration process with microbubble generator

### 2.3. Seedings Anaerobic Biofilter

The seeding of microorganisms is achieved through the introduction of a wastewater sample, which is the subject of the treatment, into a reactor equipped with filtration media. In this study, the filtration media takes the form of gravel and bio balls. The seeding process is considered completed once a biofilm layer has formed on the filter media, which typically occurs within 14 days. **Fig.2(1)** illustrates that on day 1, the biofilm had not developed, and the wastewater exhibited a cloudy appearance. On the fourth day, a thin cream-colored membrane was observed on the surface of the bio balls. On the eighth day, the biofilm layer on some bio balls had become visibly thicker, while on the fourteenth day, a brownish-beige biofilm had formed on the bio balls. The seeding process in this study indicated that the visual biofilm formation was more pronounced on the fourteenth day. This finding is corroborated by research conducted by (Kahar et al., 2017), which posited that the seeding process requires 10-14 working days to facilitate the growth phase of microorganisms in the receiving new wastewater phase. In addition

to the completion of the biofilm layer on the filter media, the stationary condition must be achieved in the seeding process (Ardhianto et al., 2014). By continuously providing nutrients with tofu industry wastewater, it will facilitate seeding and help achieving the steady-state condition. Moreover, seeding allows microorganisms to adapt and flourish in the new environmental conditions inside the biofilter reactor. The addition of 5% EM<sub>4</sub> as a nutrient is employed to accelerate biofilm growth, as evidenced by the findings of Ratnawati & Kholif (2018) and Badrah et al. (2021). Once the seeding process has been completed and there are indications of microbial growth on the surface of the bio ball media, the anaerobic biofilter reactor can be initiated, and after 14 days, biofilms in bio ball starts to grow (Fig.2). Furthermore, prior to the commencement of the treatment process, a series of parameter measurements were conducted, including pH, COD, BOD, TSS, DO, and ammonia. The experiments employing batch system were conducted by the planned HRT of 12, 24 and 48 hours. Following the completion of each test run, the parameters of pH, COD, BOD, TSS, DO, and ammonia were measured at the outlet of the anaerobic biofilter reactor.



**Figure 2.** Process of seeding in an anaerobic biofilter system of tofu wastewater

(1) first-day seeding, (2) fourth days of seeding, (3) eighth days of seeding, (4) fourteen days of seeding

#### 2.4. The Process using Anaerobic Biofilter and Pre-Aeration with Microbubble Generator - Anaerobic Biofilter

The combination of an anaerobic biofilter and microbubble generator as the pre-aeration process is achieved by re-treating wastewater that has undergone the anaerobic biofilter process and then flowing into the microbubble generator system for pre-aeration. Experiments were conducted with the planned HRT (60, 120, 180 minutes) (Deendarlianto et al., 2015). The microbubble generator process has the potential to elevate the concentration of dissolved oxygen at regular intervals of 60 minutes. To ascertain the impact of this periodicity, a series of time points has been established, spanning 60, 120, and 180 minutes. The microbubble generator used an ejector system to compress the incoming air at high pressure. The employment of a microbubble generator for pre-aeration allows for the observation of the effect of DO concentration on the aeration process, which were monitored in every HRT unit. This can determine the effect of the microbubble generator as a source of oxygen. Subsequently, the pH, COD, BOD, TSS, DO, and ammonia were tested at the outlet of the pre-aeration process with a microbubble generator.

#### 2.5. Analysis Methods

The data obtained from laboratory testing results were tabulated to determine the effectiveness of the anaerobic biofilter method, and the combination of the anaerobic biofilter with microbubble generator for each variation of HRT. This was done using the wastewater effectiveness formula, and the results were analyzed descriptively. In this study, the analysis of pH using, the measurement of COD, BOD, TSS, DO and  $NH_3$  refer to SNI 06-6989.11-2004, SNI 6989.2-2009, SNI 6989.72-2009, SNI 06-6989.3-2004, SNI 06-6898.14-2004 and SNI 06-6989.30-2005, respectively. Following the laboratory analysis process, a qualitative analysis of the results of organic content removal was carried out. The removal

efficiency of organic parameters and the reduction efficiency of wastewater were calculated using Eq. 1 (Li et al., 2018).

$$RE (\%) = \frac{C_{in} - C_{eff}}{C_{in}} \times 100\% \quad (i)$$

where RE represents the removal efficiency of organic parameters (COD, BOD, TSS),  $C_{in}$  represents the initial concentration of organic parameters in the feed tank solution, expressed in mg/L.  $C_{eff}$  represents the concentration of organic parameters at the outlet of the anaerobic biofilter and pre-wind process, based on the HRT sampling process, which is also expressed in mg/L. The value of the parameters at the time of the HRT is indicated by  $C_{eff}$ , which refers to both the anaerobic filter and the pre-aeration processes. Positive values of RE indicate a decreased concentration after wastewater treatment, while negative values indicate an increased concentration.

### 3. Results and Discussions

#### 3.1. The Effect of HRT on the Reduction of Organic Parameters in Anaerobic Biofilter and Pre-Aeration with Microbubble Generator

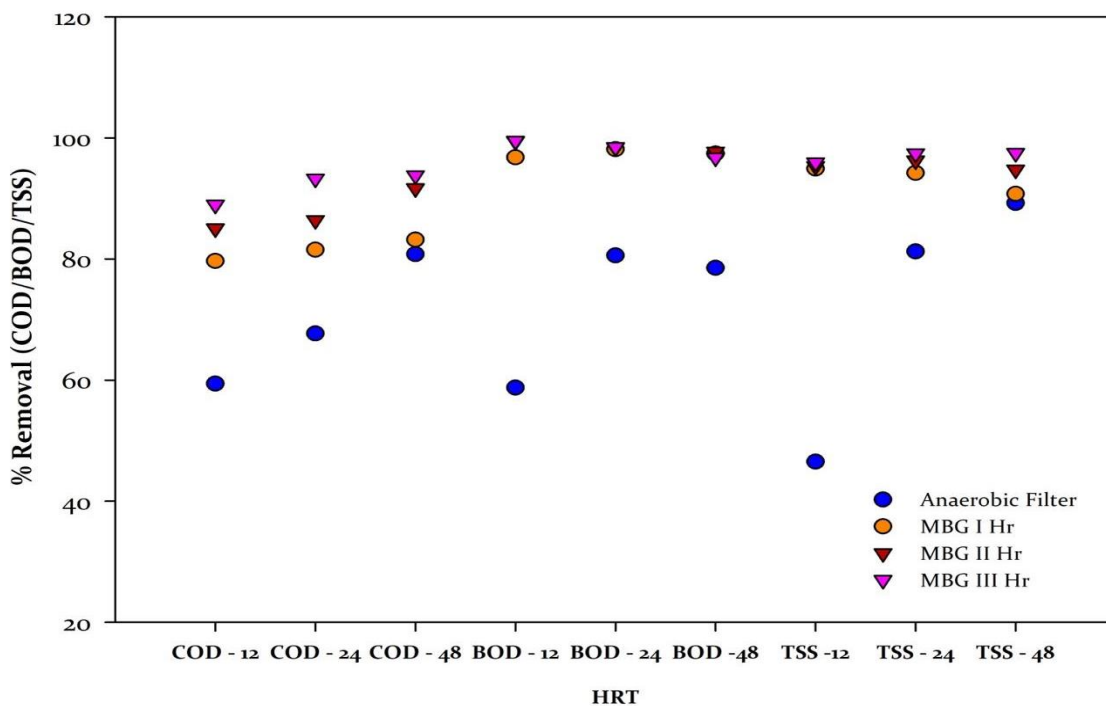
The results obtained indicate that the anaerobic biofilter's HRT process has significantly reduced the levels of TSS, COD, BOD, and ammonia. Figure 3 shows that once HRT was implemented, the COD in the anaerobic biofilter decreased. Additionally, the COD concentration dropped from 3773 mg/L to 1532 mg/L after the 12-hour HRT was implemented. In the meantime, the COD was reduced to 1219 mg/L and 725 mg/L (Table 2), with efficacious rates of 67,69% and 80,78% (Fig. 3) for the 24- and 48-hour HRT, respectively. The outcomes showed that increased efficiency is the consequence of a longer retention period. Microorganisms in the biofilm adhering to the bio ball are assumed to be responsible for the reduction of COD in the anaerobic biofilter processing process. This allows the organic components in wastewater from the tofu industry to be used as nutrients, which can subsequently decompose into simpler organic substances. The findings of this study are consistent with a study by (Widyaningrum, (2020) which discovered that, for HRT of 16, 24, and 40 hours, respectively, the effectiveness of COD degradation in anaerobic horizontal roughing filter processing was at 40%, 55%, and 80%.

**Table 2.** Removal of COD, BOD, and TSS as the effect of different HRT

Process	Variable	Parameters		
		COD	BOD	TSS
Anaerobic Filter	HRT (Hr)	mg/L	mg/L	mg/L
	0	3773	1020	400
	12	1532	421	214
	24	1219	198	75
	48	725	219	43
Pre Aeration (MBG) - Input (AF HRT 12 Hr)	0	1532	421	214
	1	767	121	191
	2	564	20,6	176
	3	417	18,3	152
Pre Aeration (MBG) - Input (AF HRT 24 Hr)	0	1219	198	75
	1	697	19	59
	2	513	16,2	38
	3	254	15,7	26

Pre Aeration (MBG) - Input (AF HRT 48 Hr)	0	725	219	43
	1	634	26,4	37
	2	314	23,1	21
	3	233	33	10
<b>Standard Discharge</b>	-	<b>300</b>	<b>150</b>	<b>200</b>

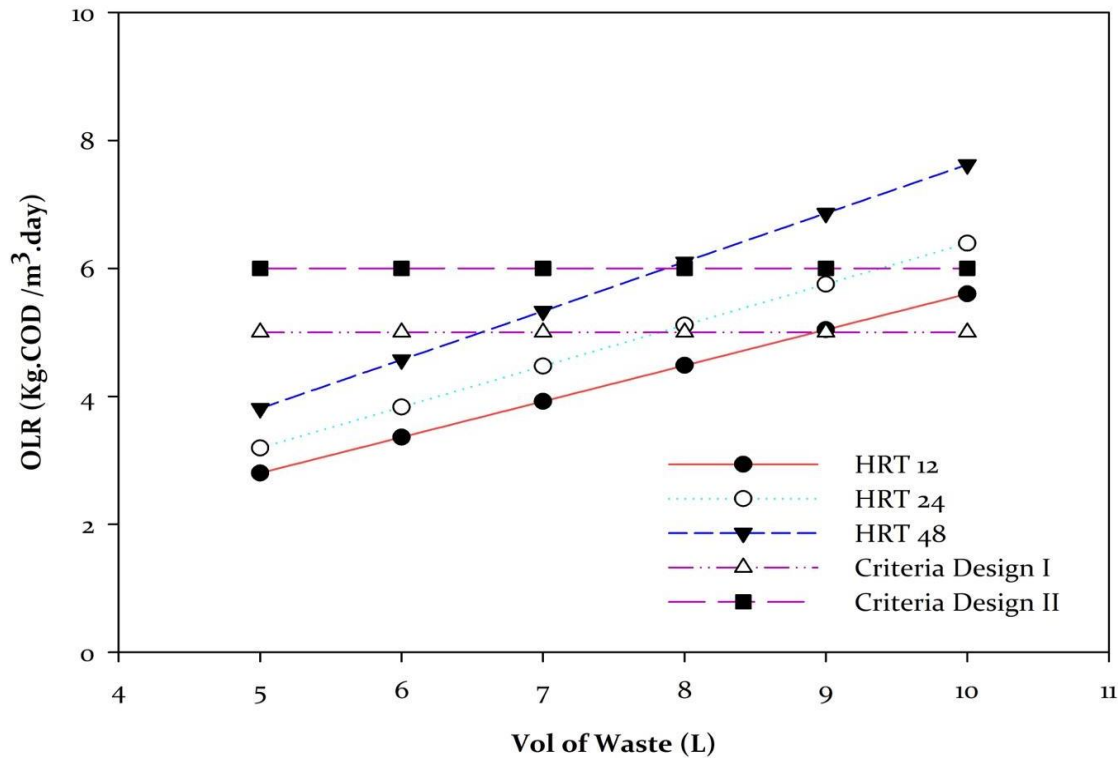
The high population of microorganisms in the bio ball media, which effectively degrades organic matter, is the primary factor responsible for the observed reduction of BOD levels. The process is facilitated by the bio balls, which are coated in biofilms and allow microorganisms and wastewater to come into close contact. The abundance of organic matter optimizes the degradation process by increasing microbial activity. A decline in BOD concentration indicates an optimal microbial activity in effluent from the tofu business, as reported by (Kholif et al., 2021). In a similar vein, (Ratnawati & Kholif, 2018) observed that during the initial phase, microbial growth and BOD reduction are closely related. The results of this investigation, however, indicate that the BOD content rose to 219 mg/L at a 48-hour HRT and that the treatment's effectiveness decreased to 78.53%, falling short of Indonesian requirements. The results from (Diny et al, 2015), which showed a fall in BOD removal efficiency from 77.31% at 12 hours HRT to 27.15% at 18 hours HRT, are following this decline. Using anaerobic biofilters, Ratnawati & Kholif (2018) likewise showed a decline in BOD removal efficacy to less than 80% by the sixth sampling day. According to (Diny et al, 2015), biofilm thickness and stabilization may lead to decreased BOD removal effectiveness by limiting optimum activity to the outermost bacteria. Lower nutrient levels cause inner-layer microorganisms to die and accumulate, raising BOD levels. Through substrate oxidation, which turns organic matter into carbon dioxide, methane, and bacterial mass, COD reduction is accomplished.



**Figure 3.** The measured COD, BOD and TSS removal from the combination of anaerobic biofilter and microbubble generator (MBG)

Longer retention periods improve COD reduction by allowing more organic matter to decompose, according to (Herlina et al., 2020). Unfortunately, the results employing HRTs of 12, 24, and 48 hours in this study did not meet the Indonesian quality standards due to the elevated baseline COD concentrations. The efficacy of COD degradation may be constrained by non-simultaneous biofilm formation and nutrient

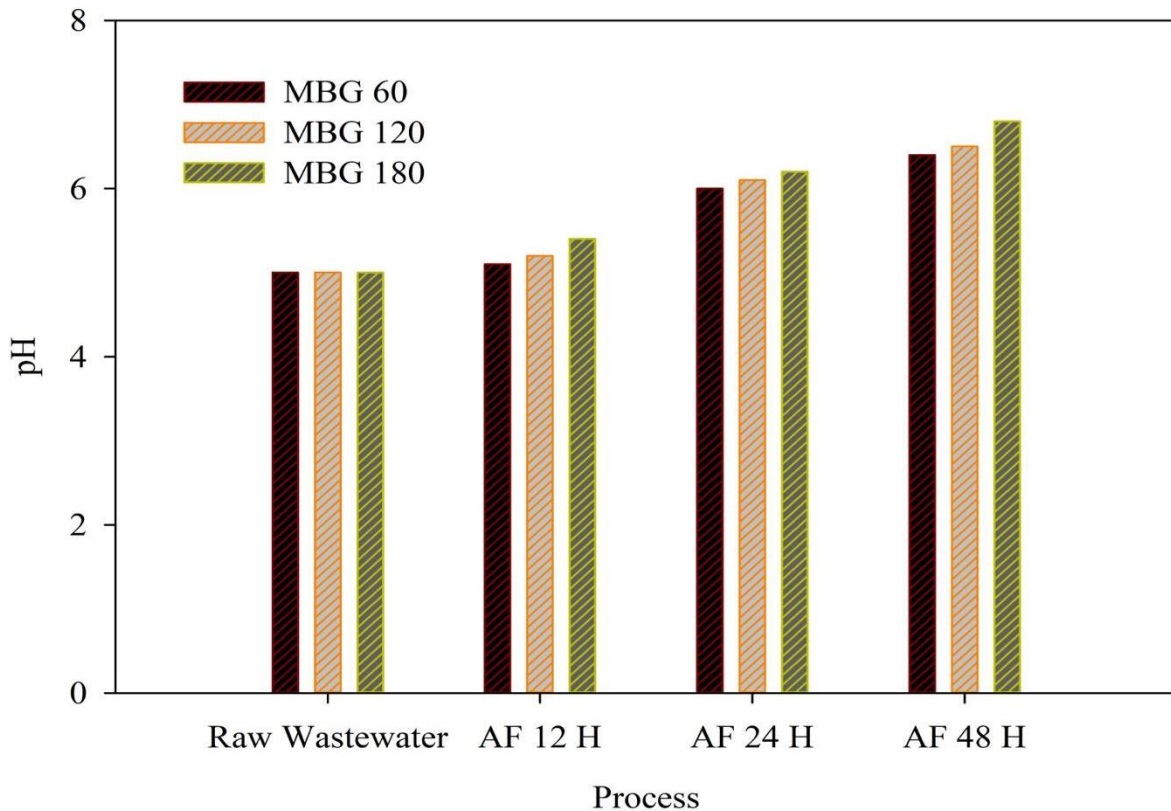
availability, as noted by (Anggarini et al., 2015) High COD concentrations in raw wastewater require effective pretreatment, such as electrocoagulation (Ardhianto & Bagastyo, 2019; Oktiawan et al., 2020; and Ardhianto et al., 2024). A 48-hour anaerobic biofilter in conjunction with a 60-minute microbubble generator reduced COD by 634 mg/L (83.20% effectiveness), according to the study, although the degree of reduction varied according on HRT. With a 120-minute HRT, COD decreased by 314 mg/L (91.68% effectiveness), and at 180 minutes, COD dropped to 233 mg/L (93.82% effectiveness) (Fig.3), meeting quality standards and achieving the highest effectiveness.



**Figure 4.** Saturation of the OLR value (COD) of the combination anaerobic biofilter and a micro-bubble generator (MBG)

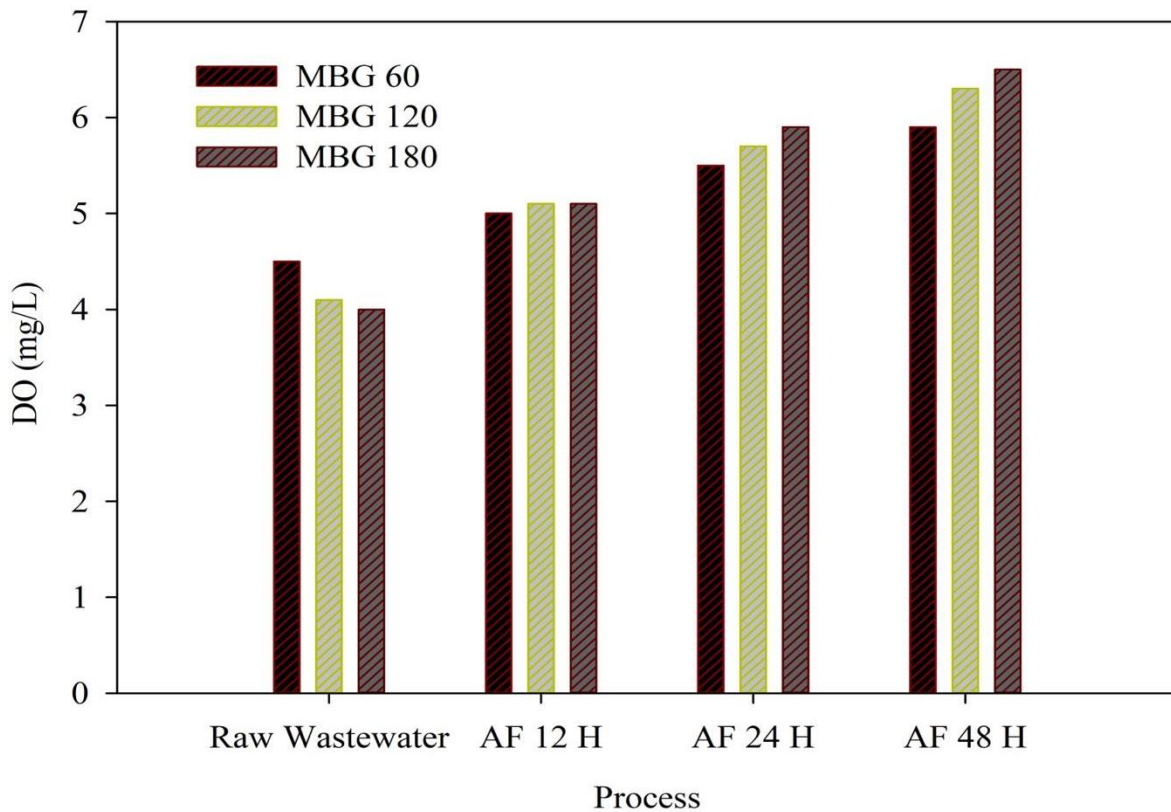
This reduction is linked to the decomposition of organic substances by biofilm microorganisms, producing acetic acid and methane under anaerobic conditions. (Budhijanto et al., 2020) found that the combination of an anaerobic treatment process with an aerobic biological reactor (AFBR) and a microbubble generator resulted in a reduction of COD by 60%, compared to 30% with anaerobic treatment alone. The microbubble generator provides oxygen for aerobic bacteria, further degrading organic compounds (Kholif et al., 2021, Afisna & Juwana, 2020)). The study showed that longer retention times enhance pollutant reduction, as more contact between wastewater, biofilm, and aeration improves effectiveness. The combination of 48-hour anaerobic biofilter and 180-minute microbubble generator achieved a BOD reduction of 93.11%, meeting quality standards. After 60 minutes, microbial growth increases but the organic material remains incompletely degraded. By the 120 minutes mark, the oxygen supply is sufficient, and the slower BOD decline at 180 minutes is due to fewer remaining pollutants. This effective reduction in BOD is attributed to the biofilm on bio ball media and pre-aeration by the microbubble generator, which aids aerobic microbes in breaking down organic matter into simpler compounds. Studies by Pramita & Puspita, (2019) and Afisna & Juwana (2020) support that anaerobic biofilters effectively reduce BOD by breaking down complex compounds, with the microbubble generator enhancing DO and aiding in the removal of residual pollutants.





**Figure 5.** The measured pH on the combination of anaerobic biofilter and microbubble generator

The anaerobic biofilter process is constrained by a limitation on the COD organic loading rate (in kg COD/m<sup>3</sup>.day) in terms of the level of utilization of its organic matter removal capability. According to Omil et al., (2003), the organic loading rate in the anaerobic biofilter process has a limit of 5.0–6.0 kg. COD/m<sup>3</sup>.day. This investigation revealed that the OLR value is nearly at the upper limit of the specified range when a batch anaerobic filter reactor with a treatment capacity of 5 L and 2 kg of bio-ball medium is used. The OLR values for HRT of 12, 24, and 48 hours were 2.80, 3.19, and 3.81 kg COD/m<sup>3</sup>.day, respectively (Fig. 4). Nevertheless, the OLR value remained within the specified range when the treatment load was increased to 6 L. The HRT of 48 hours then exhibited an OLR value above that for the 7 L treatment load, which will result in system saturation. Following the addition of 9 and 10 L treatment loads to the simulation, HRT 12 hours enters the limitation of 5 kg COD/m<sup>3</sup>.day and 6 kg COD/m<sup>3</sup>.day. However, the HRT of 24 and 48 hours was unable to maintain OLR below the threshold when the treatment load was increased to 8 L. The simulation results demonstrate that, even with 100% load value-added, HRT 12 hours in process balancing can accommodate a high treatment load. The ability of this technology to accommodate a higher treatment load with a 12-hour retention duration is undoubtedly a beneficial and robust feature, as evidenced by the saturation levels of the parameters. Subsequently, this is reinforced by the biofilters that are employed, one of which is bio ball, a medium with a surface area of 200 m<sup>2</sup>/m<sup>3</sup>. According to the Surface Load Area Rate (SLAR) value, the surface capacity of this media for HRT 12, 24, and 48 hours is 14.00, 15.96, and 19.05 g COD/m<sup>2</sup>.day, respectively. The maximum processing load of 10 L with a 12-hour HRT will result in an effective surface area of 28.01 g COD/m<sup>2</sup>.day, which represents the upper limit of the media's capacity.



**Figure 6.** Results of DO Testing on the Combination of Anaerobic Biofilter and Microbubble Generator as Pre-Aeration

Furthermore, the capacity of the media to control the BOD load is 3.74, 5.14, and 5.01 g BOD/m<sup>2</sup>/day at the HRT of 12, 24, and 48 hours, respectively, with a 5 L treatment load, based on the BOD load rate value. Concurrently, at the HRT of 12, 24, and 48 hours, the media's ability based on area is 7.49, 10.28, and 10.01 g BOD/m<sup>2</sup>/day, respectively, when the load is increased by 100%. Because there is no pre-treatment procedure in place, the medium is subjected to an extremely high load. In the absence of a pre-treatment procedure, the medium is subjected to an exceedingly high load. The use of high surface area media, such as Kaldness (1000–1200 m<sup>2</sup>/m<sup>3</sup>) and Biochip (2500–5000 m<sup>2</sup>/m<sup>3</sup>), has been demonstrated to achieve optimal results, including a significant reduction in anaerobic filtering residence time and effective load control. The efficient breakdown of TSS is achieved through the combined action of an anaerobic biofilter and a microbubble generator, with the aerobic process serving to further reduce TSS levels. As illustrated in Fig. 3, the treatment period was extended to 180 minutes, resulting in a 97.5% reduction in TSS to 10 mg/L and compliance with Indonesian quality standards. TSS, which is composed of organic matter and minute particles, increases water turbidity due to its difficulty in dissolving or settling (Agung Rahmanto & Salamah, 2022). Moreover, the findings of this study indicate that longer HRT is associated with a reduction in TSS concentration. The elimination of 214 mg/L of TSS (46.5% efficiency) was achieved through the application of HRT over 12 hours, with the process initiated at a concentration of 400 mg/L. However, the threshold of 200 mg/L was not reached. A reduction of 75 mg/L (81.25% efficacy) was observed at the 24-hour mark, with a further reduction of 43 mg/L (89.25% efficacy) evident at the 48-hour mark. These findings indicate that longer treatment durations result in greater TSS reductions due to the decomposition of organic material and retention of particles within biofilms on bio balls and gravel. The study's TSS removal rates are higher than those reported by Pramita & Puspita (2019), who achieved 81.39% removal at 48 hours HRT. Longer contact times and microbubble aeration further break down clumped sediments, enhancing TSS removal, as supported by Timpua & Pianaung, (2019) and

Sisnayati et al., (2021). The presence of microbubbles supplies oxygen, which in turn facilitates sediment separation and further TSS reduction.

Anaerobic biofilter with 24 HRT with a 60-minute microbubble generator increased the pH of tofu industry wastewater to 6 (Fig. 5). Longer HRTs of 120 and 180 minutes further raised the pH to 6.1 and 6.2, respectively. After 48 hours of anaerobic treatment followed by 60, 120, and 180 minutes of microbubble generation, pH values increased to 6.4, 6.5, and 6.8, meeting Indonesian standards (Fig. 4). Microbubbles enhance dissolved oxygen (DO) availability, crucial for aerobic nitrification. (Said & Firly, 2005) reported that aerobic processes break down organic compounds not fully degraded anaerobically, including via nitrification. The sustained rise in DO, due to the prolonged operation of the microbubble generator, supports bacterial decomposition. (Baihaqi et al., 2022) and (Nair et al., 2022) confirm that increased oxygen supply accelerates aerobic bacteria activity in pollutant decomposition. Afisna & Juwana (2020) demonstrated that microbubbles, with a 200 µm diameter, slow their ascent, extending their presence in the wastewater and enhancing DO levels (Fig. 5). This prolonged air exposure improves the effectiveness of aerobic microorganisms in breaking down organic pollutants. In summary, the combination of an anaerobic biofilter and microbubble generator significantly increases DO concentration, enhancing the breakdown of organic pollutants by aerobic bacteria in tofu industry wastewater.

#### 4. Conclusion

This study demonstrates the efficacy of integrating anaerobic biofilter technology with pre-aeration via a microbubble generator for treating wastewater from the tofu industry. Extending the HRT in all systems greatly enhanced pollutant removal, with a 48-hour HRT in the anaerobic biofilter resulting in COD, BOD, and TSS reductions of 80.78%, 78.53%, and 89.25%. The best combination of a 48-hour anaerobic biofilter and 180-minute microbubble treatment resulted in COD, BOD, and TSS reductions of 93.82%, 93.11%, and 97.5%, respectively, while keeping effluent pH within Indonesian norms. When the processing load increases by 8 and 10 L, organic loading in bioball media with HRT 24 and 48 hours is limited. Conditions in which the media's surface area cannot support the organic loading load. The study highlighted the importance of optimising retention durations in treatment procedures and proposes that this combination strategy is a possible answer to the environmental concerns posed by wastewater from the tofu sector. Future study should refine these processes and investigate new pretreatment approaches to improve overall treatment performance.

#### Acknowledgements

The authors would like to express their gratitude to the Department of Environmental Engineering of Universitas Islam Negeri Ar-Raniry and the Department of Environmental Engineering of Institut Teknologi PLN for providing the supporting data. In particular, the authors also acknowledge Tofu Industry for providing access to the application of research in this study.

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