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Application of Sequencing Batch Biofilm Reactor (SBBR) Using Microalgae *Chorella* sp. to Removal Nutrient in Grey Water

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ABSTRACT

Grey water contains organic matter that is directly disposed to the environment without any treatment previously, will cause pollution and impacting life in the water. Treatment that can be done is using microorganisms. One of its kind is the microalgae Chlorella sp. which utilizes organic matter as a source of nutrients for its growth. In this study, the Kaldness 1 (K1) bio carrier was added as a medium for attaching microorganisms using the Sequencing Batch Biofilm Reactor (SBBR) process. The research objectives were (1) to know the maximum number of *Chlorella* sp. both attached and suspended in the Sequencing Batch Biofilm Reactor (SBBR), (2) to obtain the best cycle time and stabilization time in the removal of COD, Ammonia, and MLSS in grey water. The research was conducted by varying the stabilization time (1.5; 2 and 2.5 hours) in each cycle for four cycles with a constant variation of charging time 30 minutes, reaction 120 minutes, 45 minutes, separation 45 minutes, and carried out with four cycles, stirring speed at 60 rpm, the concentration of algae suspension in SBBR was 25% and the volume of Kaldness K1 medium was 20%. The results showed the number of microalgae cells *Chlorella* sp. was suspended and attached to 1.85 x 106 and 1.46 x 106 cells/ml. The best removal of COD, ammonia, and MLSS was found in the stabilization time variation of 1.5 hours in 4 cycles with a removal efficiency of 84% and 76%, respectively, and an increase in the concentration of suspended and attached MLSS by 4780 mg/l and 4720 mg/l. It can be concluded that the faster stabilization time, the more removal efficient will be.

***Keywords*:** Chlorella sp., grey water, sequencing batch biofilm reactor, stabilization time.

1. INTRODUCTION

Population growth, which keeps increasing every year will be directly proportional to human activities every day. The increase in human activities will cause positive and negative impacts on the environment. One of the negative impacts caused is the increase in the amount of waste produced generated. Domestic wastewater tends to contain high organic compounds and nitrogen, which if not treated as well will cause pollution to water receiving bodies [1]. According to the characteristics, there are two types of domestic wastewater, namely black water which comes from the lavatory and is usually collected in the septic tank, while the other type is grey water which comes from washing, bathing, and cooking activities that are generally directly discharged into drainage channels and receiving water bodies [2]. Characteristics of gray water namely TSS of 121-127 mg/L; COD 79-700 mg/L; BOD 121-151 mg/L; pH 6.2-8.5; oils and fats 6-95 mg/L and ammonia 12-50 mg/L [3]. In the wastewater treatment process, especially those containing organic compound pollutants, the most used technology utilized the activity of microorganisms to decompose these organic pollutant compounds. The process of treating wastewater with microorganism activity is commonly referred to as a biological process [4].

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One of the microorganisms that can be used to treat wastewater biologically is microalgae *Chlorella* sp. The use of microalgae has the advantage that the treatment process runs naturally like the principle of a natural ecosystem, so it is very environmentally friendly, does not produce secondary waste, has low energy requirements, and has microalgae biomass production [5]. In this study, microalgae will be grown in suspension and attached to the media bio carrier which is known as a *biofilm*. Currently, a combination system of microalgae and bacteria has been developed for wastewater treatment.

This research uses a Sequencing Batch Biofilm Reactor (SBBR) as one of the biological wastewater treatments. This research uses a Sequencing Batch Biofilm Reactor (SBBR) is one of the biological wastewater treatment systems. This technology uses a combination of microorganisms and a buffer medium that is used as an attachment area. Technical advantages Sequencing Batch Biofilm Reactor (SBBR) is a biological treatment process, and the deposition process occurs in one tank, so it does not require a separate system of settling unit. The operation of this reactor consists of 5 stages, namely: fill, react, settle, draw, and idle [6].

This research will be carried out by adding bio-carrier plastic as a place to attach microorganisms. The media used in this research is Kaldness K1 [7]. This media is made of High-Density Polyethylene (HDPE) with a specific gravity of ± 0.95 g/cm^3 which has a length of 7 mm and a diameter of 10 mm with a cross shape inside the cylinder and elongated fins on the outside. The specific surface area of the media is about 500 m^2/m^3, large enough for the attachment of microorganisms. In general, to treat domestic wastewater, four cycles (six hours per cycle) to six cycles (four hours per cycle) are used per day [8].

He et al. [9] researched to remove carbon in artificial waste by Sequencing Batch Reactor aerobics. The variations used were stabilization time and reaction time. The results obtained at the variation of reaction time and stabilization 1: 6 gave the optimum COD removal efficiency of 95.23%.

Mareai et al. [10] reported a carbon removal in synthetic wastewater using the Sequencing Batch Reactor (SBR). Variations used are reaction time and stabilization time. The best carbon removal occurred at a reaction time of 1 hour, and a stabilization time of 6 hours was 97.02%.

Chan et al. [11] carried out the removal of organic compounds in the wastewater of palm oil mills using a *Sequencing Batch Reactor* (SBR) This research was conducted with varying reaction times against time stabilization, namely: 4:4 hours/hour, 6:4 hours/ hour, 4:6 hours/hour, and 6:6 hours/hour. The best result obtained is the reaction time to the stabilization time (r:s) of 6:4 hours/hour of 86.60%.

Based on the explanation above, this study aims to study the effect of the stabilization time of the process *Sequencing Batch Biofilm Reactor* (SBBR) using microalgae *Chlorella* sp. grown in suspension and attached to Kaldness K1 on the removal of COD and ammonia content in the grey water. To obtain the optimum removal efficiency, various stabilization times, namely 1.5, 2, and 2.5 hours, and COD and ammonia reduction tests were carried out in each cycle for four cycles.

2. RESEARCH METHODS

This research uses materials, namely: grey water taken from the Communal IPAL of the Widya Graha II Housing, Delima village, Pekanbaru, Riau, Indonesia. The microalgae used are *Chlorella* sp. from the Algae Research Center, Faculty of Fisheries and Marine Sciences, University of Riau. The tools used are Sequencing Batch Biofilm Reactor (SBBR) is cylindrical with a height of 50 cm and a diameter of 20 cm and equipped with a paddle wheel with a diameter of 12 cm and a width of 1.2 cm. Media *kaldness* K1 as biocarrier, thomacytometer, pH meter, and thermometer were also used in this study.

2.1. EQUIPMENT PREPARATION

The tool used is SBBR consisting of three cylindrical reactors made of acrylic material with a height of 50 cm and a diameter of 20 cm. SBBR is planned to be able to treat domestic liquid waste with an effective volume of 5 liters. Each reactor is equipped with a stirrer. The stirrer is used in the form of a paddle wheel with a diameter of 12 cm and a width of 1.2 cm. Following the installation design *Sequencing Batch Biofilm Reactor* (SBBR) can be seen in Figure 1.

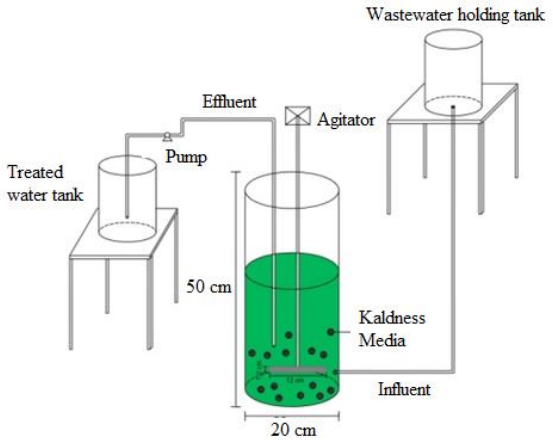

Figure 1. Installation S*equencing Batch Biofilm Reactor* (SBBR)

The biocarrier media used was Kaldness K1. The biofilter media used was media made of non-corrosive materials, resistant to chemical decomposition and destruction. The Kaldness bio carrier K1 used in the study can be seen in Figure 2.

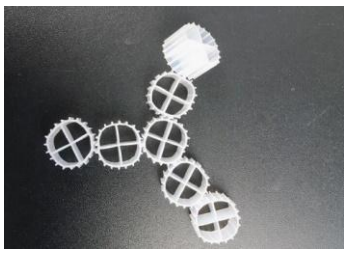

Figure 2. Media biocarrier *Kaldness* 1 (K1)

2.2. PREPARATION OF GREY WATER

The suspension medium used is domestic liquid waste grey water taken from the Communal IPAL of the Widya Graha II Housing, Delima village, Tampan District, Pekanbaru City. Riau Province. Domestic wastewater grey water takes as much as 30 liters according to SNI 6989-59-2008 with one time of collection. Furthermore, the initial characteristics of the content of COD, ammonia, pH, and temperature were tested. Sample pick-up point domestic wastewater sample grey water can be seen in Figure 3.

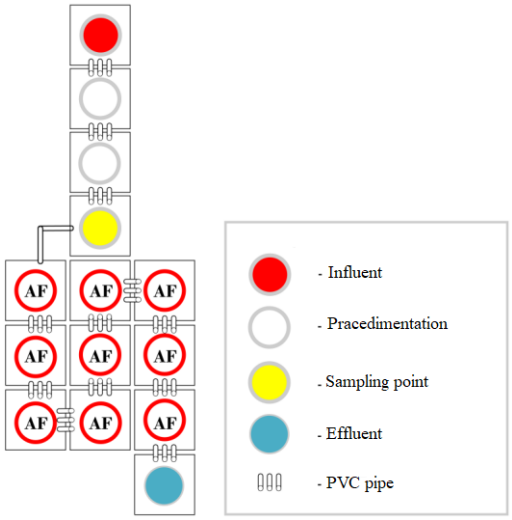

Figure 3. Examples of domestic wastewater sampling point to grey water

2.3. CULTIVATION MICROALGAE

Chlorella sp. was obtained from the Central Algae Research Laboratory, Faculty of Fisheries and Marine Sciences, Riau University, cultivated in medium Dahril Solution. Microalgae cultivation of *Chlorella* sp. is performed with the addition of 100 ml microalgae *Chlorella* sp. and 400 ml medium Dahril *Solution* into 3.5 liters of distilled water with the light source coming from sunlight. Microalgae propagation *Chlorella* sp. is carried out until the growth of microalgae cells *Chlorella* sp. is on the exponential phase and sufficient 1 × 106 cells/ml. Calculation of the number of microalgae cells *of Chlorella* sp. performed every 24 hours using thomacytometer with the help of cover glass and hand counter observed under a light microscope.

2.4. MICROALGAE ACCLIMATIZATION

Acclimatization was carried out in two stages with a working volume of 4 liters. The first stage is done by mixing 50% (2L) of cultivated algae and 50% (2L) of domestic liquid waste grey water with added

biocarrier media Kaldness K1 which will be used in the main experiment in the acclimatization process. If the microalgae in the first stage had reached the exponential growth stage or the microalgae cell density reaches 1×10^6 cells/ml then the second stage of acclimatization will be carried out. The second stage is carried out by mixing 75% (3L) of the culture from the first acclimatization stage and 25% (1L) of domestic liquid waste grey water. Measurements of pH and temperature were carried out during acclimatization to determine whether these conditions could be optimal support for microalgae growth.

Table 1. Calculation of Each Reactor

Reactor 1	
Wastewater Grey Water	3750 ml
Algae Suspension	1000 ml
Media Kaldness K1	250 ml
fill	30 minutes
react	120 minutes
settle	45 minutes
decant	45 minutes
idle	90 minutes
Reactor 2	
Wastewater Grey Water	3750 ml
Algae Suspension	1000 ml
Media Kaldness K1	250 ml
fill	30 minutes
react	120 minutes
settle	45 minutes
decant	45 minutes
idle	120 minutes
Reactor 3	
Wastewater Grey Water	3750 ml
Algae Suspension	1000 ml
Media *Kaldness* K1	250 ml
fill	30 minutes
react	120 minutes
settle	45 minutes
decant	45 minutes
idle	150 minutes

2.5. MAIN EXPERIMENT

The working volume in the main experiment is 5 liters. Liquid waste grey water, algae, and media suspension *Kaldness* K1 were added to the SBBR as much as 20% of the algae suspension (250 ml). The concentration of the algae suspension included in this experiment was constant at 25% of the working volume (1250 ml). The calculation description of each reactor in this experiment can be seen in Table 1.

Calculation of the number of media Kaldness K1 contained in each reactor is done by using a 1000 ml measuring cup. The measuring cup is filled with 750 ml of water, then added Kaldness K1 until the water volume reaches 1000 ml. Many media Kaldness K1 used to increase 250 ml of water is 893 pieces. Treatment will be carried out with a filling time of 30 minutes, reaction of 120 minutes, precipitation of 45 minutes, separation of 45 minutes, and stabilization time with variations of 1.5, 2, and 2.5 hours observed in each cycle for four cycles and stirring speed of 60 rpm. Influent for the second cycle is the processed product of the first cycle.

2.6. DATA ANALYSIS AND CALCULATIONS

The purpose of using parameter analysis is to determine the level of parameters after treatment which refers to the applicable SNI. Parameter analysis can be seen in Table 2.

Table 2. Pollutant Parameter Analysis

Analysis	Method/Tool
Number of cells	Thomacytometer
pH	SNI 06-6989.11-2004
Temperature	Thermometer
COD	SNI 06-6989.73-2009
Ammonia	SNI 06-6989.30-2005
MLSS	Gravimetry

The efficiency of decreasing test parameters used the Equation (1).

$$\text{Efficiency (\%)} = \frac{\text{Cin - Cef}}{Cin} \times 100 \qquad (1)$$

Where C_{in} is the concentration influent (mg/L) and C_{ef} is the concentration effluent (mg/L).

3. RESULTS AND DISCUSSION

3.1. INITIAL CHARACTERISTICS OF GREY WATER

Initial concentrations of COD and ammonia in grey water from the Communal IPAL is 198 mg/L and 9.87 mg/L. Based on the test results of the COD and ammonia parameters, showed that the COD level was still above the quality standard, which was 100 mg/l [12]. Meanwhile, the ammonia level is close to the quality standard threshold of 10 mg/L. Therefore, treatment is needed to reduce pollutant concentrations before being discharged into water bodies. Pollutants in liquid waste contain organic compounds, which are needed as nutrients for the growth of microalgae *Chlorella* sp.

3.2. MICROALGAE CULTIVATION AND ACCLIMATIZATION ON *GREY WATER* AND MEDIA BIOCARRIER *KALDNESS* 1 (K1)

The cultivation process aims to make microalgae *Chlorella* sp. run into cell growth until it enters the exponential phase with the number cell density reaching 1 x 10^6 cells/ml for further acclimatization. Medium Dahril Solution is a source of nutrients that are needed for the process of cell growth and microalgae metabolism *Chlorella* sp. and sunlight which is used as a light source for the process of cell division and photosynthesis of microalgae *Chlorella* sp. The culture in this process is continuously aerated using an aerator to avoid the deposition of algal cells *of Chlorella* sp. and maximize algal cell contact [13]. To determine the density of the number of microalgae cells, the number of cells is calculated every 24 hours. A graph of the average number of microalgae cells *of Chlorella* sp. at the cultivation stage can be seen in Figure 4.

The cultivation process was completed on the 8th day, and the number of microalgae cells was obtained at 1.61 x 10^6 cells/ml. Microalgae cells *Chlorella* sp. can be seen in Figure 5.

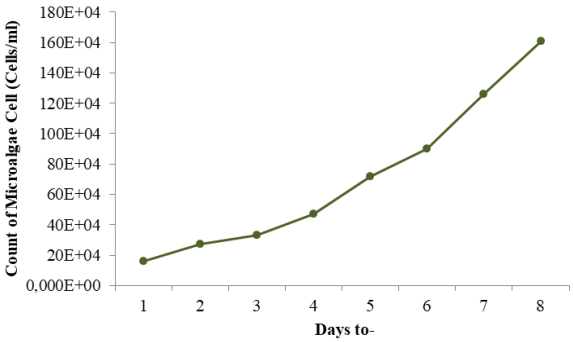

Figure 4. The average number of microalgae cells *Chlorella* sp. cultivation stage

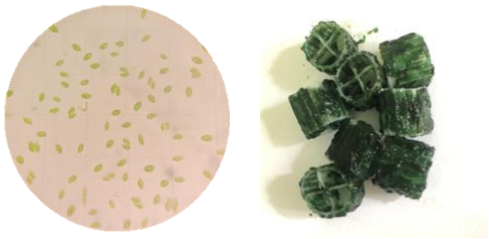

Figure 5. Microalgae cell shape *Chlorella* sp. microscopically and *biofilm*

After the microalgae cultivation process *Chlorella* sp. is already in the exponential phase, followed by the acclimatization phase. The acclimatization stage was carried out to adapt the microalgae *Chlorella* sp. for suspension growth on grey water and growth attached to the media bio carrier *Kaldness* 1 (K1). The acclimatization process is carried out in two stages to avoid the occurrence of shock loading so that microalgae can adapt to the new medium and grow attached to the media bio carrier *Kaldness* 1 (K1). Microalgae can divide up to three times per day [14]. Cell division occurs because microalgae can take advantage of the pollutants present in the cell's grey water. This process was carried out for 16 days for acclimatization stage one and two. The average number of microalgae cells in the first and second acclimatization stage based on suspension and *biofilm* can be seen in Figure 6.

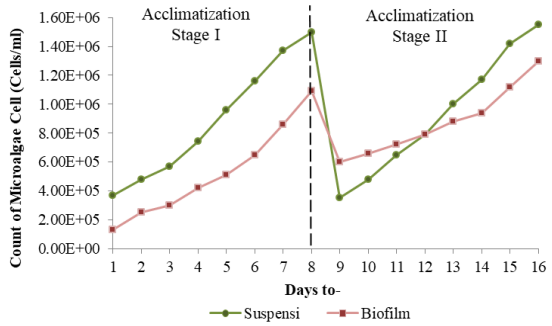

Figure 6. The average number of microalgae cells *Chlorella* sp. acclimatization stage

Figure 6 shows that the number of microalgae cells *Chlorella* sp. suspension-based growth at stage one acclimatization has reached an exponential phase with a cell density of 1.50×10^6 cells/ml on the eighth day, while for the -based cell count *biofilm* acclimatization stage one is 1.09×10^6 cells/ml. The second stage of the acclimatization process was continued for eight days so that the microalgae could adapt to the medium and increase the contact time between the microalgae and the microalgae grey water which can increase the thickness *biofilm* microalgae *Chlorella* sp. on media biocarrier *Kaldness* 1 (K1) as well as suspension on grey water. On the first day of the second stage of acclimatization, there was a decrease in the number of suspension-based cells. The decrease in the number of cells is caused by the dilution that occurs in the process of acclimatization. Acclimatization stage two is considered a complete stage when the number of microalgae cells *Chlorella* sp. suspension-based and *biofilm* already in the exponential phase with successive cell density is 1.55×10^6 and 1.30×10^6 cells/ml on day 16. According to Lines et al. [15], the increased growth pattern and the number of cells were already at a cell density of 1×10^6 cells/ml indicates that the microalgae at the second acclimatization stage can be used for treatment.

3.3. MEASUREMENT FACTOR ENVIRONMENT DURING TREATMENT

Measurement of pH and temperature was carried out in this study where pH and temperature were environmental factors for the growth of microalgae. Microalgae growth pH and temperature range *Chlorella* sp. during the treatment depicted in Table 3.

Table 3. Microalgae pH and Temperature Range During Treatment

Environmental Factor	Value Range
pH	7.6 – 8.60
Temperature	29 – 30.5^{o}C

According to Torres-Tiji et al. [16], The optimum pH for microalgae growth is 6-9. The pH of microalgae growth during the treatment ranged from 7.6 - 8.60 so the pH in this study was still in the range of microalgae growth. *Chlorella* sp. increasing the pH value is directly proportional to the total number of microalgae cells *Chlorella* sp. suspension-based and *biofilm* [17]. This is following the data obtained that the total number of microalgae cells *Chlorella* sp. Suspension-based and *biofilm* and the pH value at the 1.5 hour stabilization time variation in cycle 4 was the highest compared to other variations. This indicates that at the 1.5 hours stabilization time variation in cycle 4, the growth of suspension-based and the following microalgae is optimal in absorbing carbon dioxide and bicarbonate as a substrate carbon source so that each microalgae cell can experience growth and metabolism cell be marked with increasing pH [18,19]. In addition to pH, other factors influence the activity metabolism of cell microalgae *Chlorella*. sp is temperature microalgae growth in this study ranged from 29 - 30.5^{o}C. The optimal temperature range for algae growth is in the range of 25 - 35 ^{o}C [20]. Therefore, the temperature range in this study is still included in the growth range *Chlorella* sp.

3.4. INFLUENCE TIME STABILIZATION AND NUMBER OF CYCLES ON CELL GROWTH *Chlorella* sp. SUSPENSION BASED

During the treatment of grey water, microalgae cells *of Chlorella* sp. suspended in the reactor were calculated at the end of the treatment in each cycle for four cycles. The graph of the number of suspension-based microalgae cells can be seen in Figure 7.

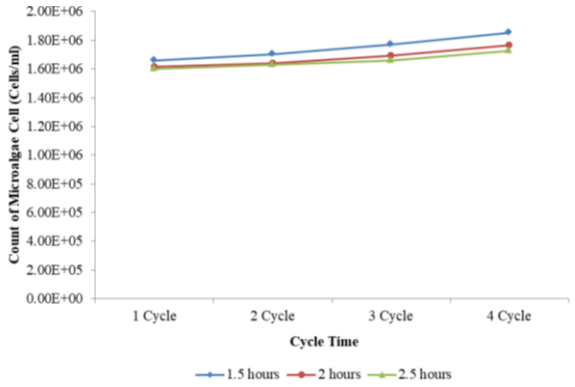

Figure 7. Microalgae cell count *Chlorella sp.* suspension based on variations in stabilization time and number of cycles

Figure 7 shows an increase in the number of microalgae cells in cycles 1, 2, 3, and 4. This increase in cells indicates that *Chlorella* sp. can survive and carry out cell division. Mustafa et al. [21] in their research proved that cell division would occur when *Chlorella* sp. is capable to carry out the metabolism. The stabilization time of 1.5 hours in cycle 4 has the highest number of microalgae cells with a total of 1.85 × 106 cells/ml. This condition occurred because the stabilization time of 1.5 hours received more microalgae suspension than the stabilization time of 2 and 2.5 hours. The number of suspension-based microalgae cells is inversely proportional to the length of stabilization time. The longer the stabilization time, the more microalgae will be lysis, thus affecting the growth of microalgae cells [22,23]. A large number of microalgae cells will increase the removal of pollutants contained in the microalgae grey water. The number of cycles in treatment will be directly proportional to the number of suspension-based microalgae cells. The longer the cycle time, the more contact occurs between microalgae and grey water. This condition causes microalgae to be able to reduce the pollutant content in wastewater and utilize it as a source of nutrients.

3.5. INFLUENCE TIME STABILIZATION AND NUMBER OF CYCLES ON CELL GROWTH *Chlorella* sp. BASED ATTACHED

Microalgae cell growth *Chlorella* sp.-based *biofilm* in this study was started with the formation of the initial layer *biofilm* on the surface *Kaldness* 1 (K1) through the adsorption process that occurs during the acclimatization process, resulting in the growth of microalgae cells *Chlorella* sp.-based *biofilm* marked by an increase in cell number and thickness *biofilm* the media *Kaldness* 1 (K1). based microalgae cell count graph *biofilm* during treatment can be seen in Figure 8.

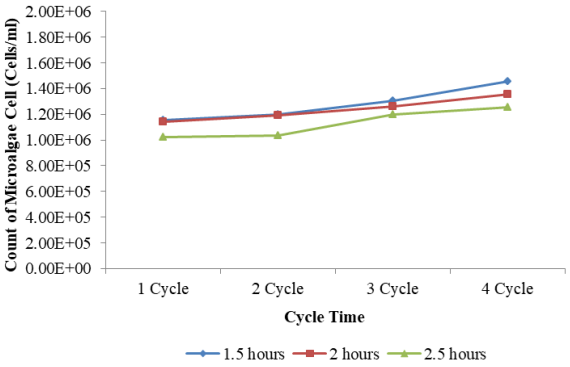

Figure 8. Microalgae cell count *Chlorella* sp.-based *biofilm* on variations in stabilization time and number of cycles

Figure 8 shows that at a stabilization time of 1.5 hours in cycle 4, the number of microalgae cells is higher compared with time stabilization 2 and 2.5 hours. Microalgae cells on the layer of *biofilm* can absorb organic substances and nutrients contained in grey water to carry out the process of metabolism. Formation *biofilm* is influenced by more factors than suspended growth including the effect of substratum, hydrodynamics, medium characteristics, and the resulting EPS. It can be seen in the graph

that the 1.5 hours stabilization time in cycle 4 resulted as the best in the number of -based microalgae cells biofilm is 1.46×10^6 cells/ml. The result shows that microalgae *Chlorella* sp. can utilize organic compounds and pollutants that are present in grey water maximally. Algae cells can be at decreased cell growth at any time and even die if they cannot adapt well [16].

3.6. INFLUENCE OF TIME STABILIZATION ON COD CONCENTRATION AND EFFICIENCY

Testing of COD parameters aims to determine the concentration of organic substances that have been successfully removed in treatment. The results of the COD concentration test during the treatment can be seen in Figure 9.

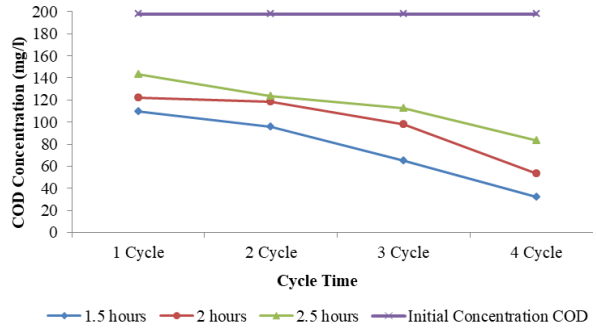

Figure 9. COD concentration during treatment

Figure 9 shows that all treatments in the treatment process experienced a decrease in COD concentration. The best decrease in the COD concentration during the treatment occurred at a stabilization time of 1.5 hours in cycle 4. The COD concentration value in cycle 1 was 110 mg/l. In 2 cycle it was 96 mg/l, in 3 cycle it was 65 mg/l in 4 cycle it was 32 mg/l. That is because the longer the cycle time, the more microorganisms can multiply well. According to Elysti et al. [24] the more microalgae that grew by dividing, the COD concentration decreased. That is because microalgae cells grow by utilizing organic substances as nutrients for their growth. The shorter the stabilization time, the higher the COD removal. Following the high number of microalgae cells, *Chlorella* sp. obtained at a stabilization time of 1.5 hours in 4 cycles resulted in high efficiency in COD removal.

The degradation process of organic matter has started to occur when the charging time starts (at 30 minutes). Then continued with the reaction period until the reaction time (120 minutes) was completed. The removal of organic matter occurs as the result of the phenomenon of absorption of organic matter into the biomass floc during the contact period after the biomass has passed through the stabilization stage. The next phase is deposition which was set for 45 minutes. Then proceed with the pouring phase which was set for 45 minutes. At 1.5 hours of stabilization in cycle 4, there was a fairly high removal of COD. That is due to the maximum biosorption. The variation of the stabilization time between 2 and 2.5 hours resulted in lower COD removal compared to the variation of the stabilization time of 1.5 hours in cycle 4. This was because variations in the stabilization time of 2 and 2.5 hours resulted in the number of dead cells compared to cells that did not grow, but also because biomass undergoes a period of no substrate after the reaction period so that it can be said that the biomass is experiencing a lack of substrate for its growth. COD removal efficiency in each reactor can be seen in Table 4.

In Figure 10 the COD removal efficiency in cycle 4 shows that the highest COD removal efficiency is found at a stabilization time of 1.5 hours, which is 84% with a concentration value of 198 mg/l and a final concentration value of 32 mg/l. The shorter the stabilization time, the higher the COD removal. The longer the stabilization time, the more microorganisms will lyse. Chan et al. [11] states that increasing the stabilization does not affect on increasing the efficiency of organic removal. A good stabilization process plays an important role because the starvation process that occurs

will make microorganisms ready to set aside the organic matter that is included in the next cycle.

Table 4. COD Elimination Efficiency

Time Stabilization (hours)	Amount Cycle	Score COD (mg/l)	Efficiency
1.5	Cycle 1	109.8	45
	Cycle 2	96.0	12
	Cycle 3	64.8	33
	Cycle 4	32.0	51
2	Cycle 1	122.4	38
	Cycle 2	118.4	3
	Cycle 3	97.6	18
	Cycle 4	53.6	45
2.5	Cycle 1	143.2	28
	Cycle 2	123.2	14
	Cycle 3	112.8	8
	Cycle 4	83.2	26

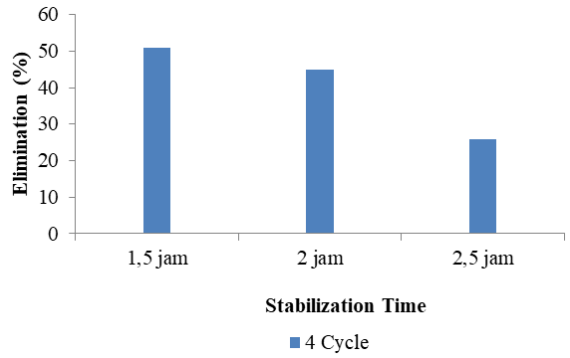

Figure 10. COD removal efficiency in cycle 4

The decrease in COD value also occurs due to the mutualism symbiosis between microalgae and bacteria. In waste grey water generally, there are mixed culture bacteria, and the growth of bacteria will encourage the growth of microalgae. Nutrients contained in grey water The nutrients needed for microalgae growth are in the form of organic complexes, so they must be oxidized to inorganic by decomposing bacteria. Microalgae can photosynthesize in organic waste fluids and produce oxygen (O_2) as the reaction product so that it can provide the required O_2 to aerobic bacteria to speed up the decomposition process. Microalgae can use CO_2 as the main carbon source for the synthesis of new cells and release O_2 through the mechanism of photosynthesis. The carbon dioxide obtained is the result of an overhaul of decomposing bacteria and O_2 The solute produced by microalgae is used by aerobic bacteria for the decomposition process. To accelerate the process of degrading waste, the use of symbiosis between microalgae is carried out by *Chlorella* sp. and decomposing bacteria [25]. The following is a comparison of the efficiency of removal of COD levels with previous research studies that can be seen in Table 5.

Table 5. Research Related to Allowance for COD

Method	Micro-organisms	Waste-water	Effi-ciency (%)	Refe-rence
SBBR	Bacteria	Domestic	80.14	Rajput and Khambete [26]
MBBR	*Chlorella sp.*	Domestic	67.3	Setiyawan et al. [27]
SBBR	*Chlorella sp.*	Domestic	84	This report

Table 5 shows that the COD removal efficiency in this study was higher than in the previous research. That is because the stabilization time used in this study is faster than in previous studies. The shorter the stabilization time, the higher the COD removal, and the longer the stabilization time, the more microorganisms will be lysis which is fewer microalgae cells that will affect the efficiency of pollutant removal in domestic wastewater.

3.7. INFLUENCE ON TIME STABILIZATION AND NUMBER OF CYCLES ON CONCENTRATION AND AMMONIA EFFICIENCY

The purpose of testing the ammonia concentration is to analyze the removal of ammonia levels in the waste grey water by microalgae *Chlorella* sp. the *Sequencing Batch Biofilm Reactor* (SBBR). The graphics concentration of ammonia during treatment can be seen in Figure 11.

Based on Figure 11 shows that all treatments in the treatment process experienced a decrease in ammonia concentration. The best reduction in ammonia concentration during the treatment occurred at a stabilization time of 1.5 hours in cycle 4. The value of ammonia concentration in cycle 1 was 5.36 mg/l. In cycle 2 there was a decrease of 5.02 mg/l, in cycle 3 it was 4.30 mg/l, and in cycle 4 it was 2.37 mg/l. Ammonia concentration at the variation of stabilization time and the number of cycles has met the quality standard that has been set, which is below 10 mg/l. This indicates that there is an absorption of ammonia by microorganisms. The rapid growth of microalgae *Chlorella* sp. causes ammonia content in the waste grey water will experience a decline. The short of stabilization time will influence the optimization of the degradation of ammonia compounds by microorganisms. The efficiency of ammonia removal in each reactor can be seen in Table 6.

Table 6. Ammonia Removal Efficiency

Stabilization Time (hours)	Amount Cycle	Ammonia (mg/l)	Effi-ciency (%)
1.5	Cycle 1	5.36	46
	Cycle 2	5.02	6
	Cycle 3	4.30	14
	Cycle 4	2.37	45
2	Cycle 1	5.62	43
	Cycle 2	5.34	5
	Cycle 3	5.03	6
	Cycle 4	4.33	14
2.5	Cycle 1	7.22	27
	Cycle 2	6.97	3
	Cycle 3	5.96	14
	Cycle 4	5.54	7

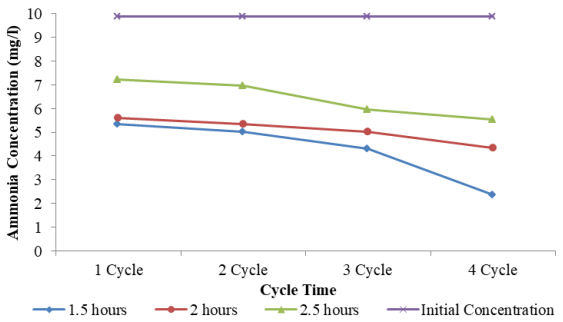

Figure 11. Ammonia concentration during treatment

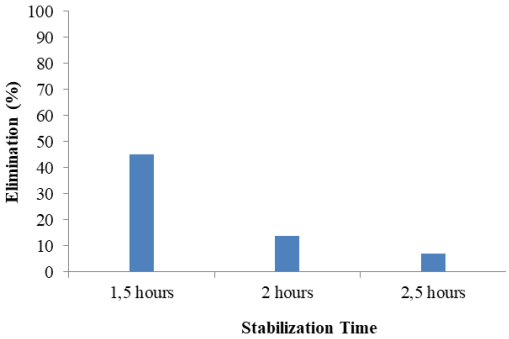

Figure 12. Ammonia removal efficiency in 4 Cycle

In Figure 12 the efficiency of ammonia removal in cycle 4 shows that the highest ammonia removal efficiency is found at a stabilization time of 1.5 hours, which is 76% with an initial concentration value of 9.87 mg/l and a final concentration value of 2.37 mg/l. It can be seen that the removal efficiency has increased. This is because, at longer cycle times, microalgae *Chlorella* sp. can utilize ammonia as a source of nutrients in its growth. According to Mujtaba et al. [28], nitrogen removal in wastewater is more efficient if it utilizes a combination of adhering and suspended growth of microorganisms. In a microalgae culture medium, the presence of ammonia in a certain amount will be advantageous because nitrogen will be more easily absorbed by microalgae in the form of ammonia so ammonia is a better source of nitrogen. Microalgae are preferred because the energy of the metabolic process that takes place to reduce ammonia to organic substances is lower than the reduction of other forms of nitrogen [29]. One of the elements that make up the intracellular component of microalgae is nitrogen. Nitrogen used by microalgae can be from nitrate, nitrite, and ammonia. These macronutrients have a very important role in the metabolism and growth of microalgae. According to Zinatizadeh and Ghaytooli [30], the ammonia compounds contained in the wastewater will be converted into ammonium. The stages of the reaction to change ammonia compounds into ammonium are as follow.

$$NH_3 + H_2O \rightarrow NH_4^+ + OH^- \quad (2)$$

According to Jia and Yunan [31], ammonium can be directly used by microalgae in the process of protein formation from amino acids so that microalgae can be assimilated by consuming less energy because the ammonium assimilation process does not require a redox reaction, while nitrate and nitrite compounds must be converted first.

The increase in the number of microalgae cells from the photosynthesis process resulted in a decrease in the concentration of ammonia. Chlorophyll substances owned by microalgae play a role in the process of photosynthesis, with the help of H_2O, CO_2, and sunlight to produce new energy. This energy is used for cell biosynthesis, cell growth, addition, movement or migration, and reproduction. The oxygen produced from the photosynthesis process can be used by bacteria in the waste grey water to oxidize organic matter into new cells while CO_2 produced by bacteria is used by microalgae as a carbon source to increase the number of cell microalgae indicated by the use of nitrogen as a nutrient for microalgae growth so that the ammonia content in the waste decreases. As for the comparison of the elimination efficiency ammonia levels with previous studies can be seen in Table 7.

Table 7 shows that the ammonia removal efficiency in this study was higher than Rajput and Khambete [26] but lower than Setiyawan et al. [27]. The ammonia removal efficiency in this study was higher than Rajput and Khambete due to the microalgae used in the research. Microalgae use ammonia for photosynthesis with the help of sunlight so that they can increase their biomass [32]. Ammonia removal efficiency in this study was lower than Setiyawan et al. [27] because the MBBR used a contact time of seven days and the bio carrier used had a wider surface than Kaldness 1 (K1).

Table 7. Research Related to Ammonia Provision Comparison

Method	Microorganisms	Provision Efficiency	Reference
SBBR	Bacteria	67.8%	Rajput and Khambete [26]
MBBR	*Chlorella* sp.	95.78%	Setiyawan et al. [27]
SBBR	*Chlorella* sp.	76%	This report

3.8. INFLUENCE TIME STABILIZATION AND NUMBER OF CYCLES ON CONCENTRATION MLSS

The organic materials measured in the MLSS analysis were solid algae, bacteria, and other non-volatile components [33].

Analysis of the MLSS value in the growth process is carried out *Chlorella* sp. suspended and attached to the waste medium grey water obtained during the treatment. The graphs of the MLSS concentration in the suspension growth process and attached to microalgae can be seen in Figure 13 and Figure 14.

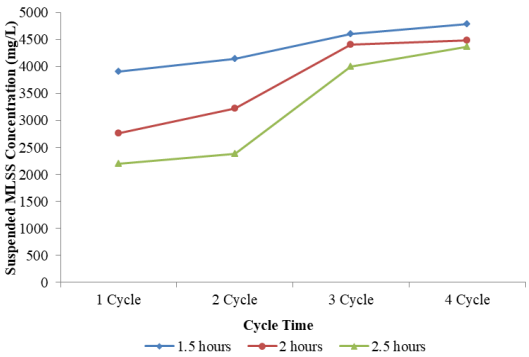

Figure 13. Suspended MLSS concentration

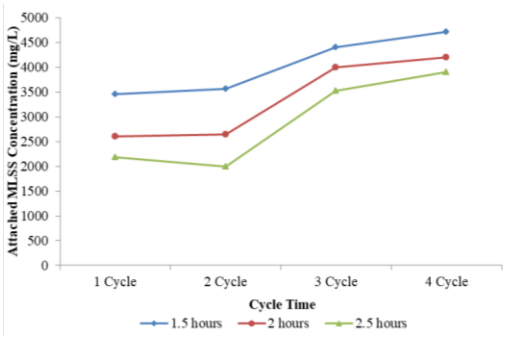

Figure 14. Attached MLSS concentration

The highest MLSS value was found in the growth of microalgae *Chlorella* sp. suspended and attached to the waste concentration grey water at 1.5 hours of stabilization in cycle 4, namely 4780 mg/l and 4720 mg/l. This is because the number of microalgae cells in this variation is high. The longer time cycle causes longer contact between microalgae and waste grey, so the microalgae *Chlorella* sp. optimally utilizes organic matter for its growth. This is in line with the study by Xiao et al. [34] that reported if microorganisms need sufficient time to reproduce and the required nutritional components are met, then microorganisms will grow rapidly. Microorganisms can use organic matter to be converted into energy needed in the growth process so that microorganisms can increase the number of cells. The lowest MLSS value on microalgae growth *Chlorella* sp. suspended and attached to the waste concentration grey water was found at 2.5 hours of stabilization in cycle 1, namely 2200 mg/l and 2180 mg/l. This is because, in this variation, the reduced supply of available nutrients can no longer meet the nutritional needs of microalgae to be able to grow. An increase in the MLSS value indicates the presence of EPS produced by bacteria, thereby accelerating the process of microalgae cell attachment to *Chlorella* sp. shape *biofilm*. The amount of oxidized organic matter can cause the MLSS concentration to increase. MLSS value is influenced by the amount of organic matter oxidized [35]. Organic matter found in waste grey water is oxidized for the growth of microorganisms. So the bigger numbered cell microorganisms indicate the more amount of organic matter that is oxidized and causing an increase in the concentration of MLSS in the reactor

4. CONCLUSION

The results obtained are the growth rate of microalgae cells *Chlorella* sp. The best suspended and attached to the SBBR with variations in stabilization time and the number of cycles in a row is 1.85×10^6 cells/ml and 1.46×10^6 cells/ml. The highest removal efficiency of COD and ammonia as well as MLSS during treatment occurred in SBBR with a stabilization time of 1.5 hours in cycle 4 with a removal efficiency of 84% and 76%, respectively, and an increase in suspended and attached MLSS concentrations of 4780 mg/l and 4720 mg/l. The shorter of stabilization time, the greater of removal efficiency of COD and ammonia will be. The concentration of COD and ammonia present in grey water after treatment using SBBR, the quality standards are met namely COD 100 mg/l and ammonia 10 mg/l. Further research for the treatment of domestic wastewater with the SBBR system with the addition of bacterial culture to increase the efficiency of removing pollutant materials.

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