© (2015) Trans Tech Publications, Switzerland
doi:10.4028/www.scientific.net/AMM.773-774.1210
Revised: 2014-12-08
Accepted: 2015-01-10

Removal of Nutrients and Selected Heavy Metals in Wet Market Wastewater by Using Microalgae *Scenedesmus* sp.

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Keywords: Microalgae *Scenedesmus* sp., wet market wastewater, nutrient and heavy metal removals

Abstract. The wet market wastewater may lead to pollution, odour and aesthetic problems to the environment and society if not treating properly. Thus, this research was conducted to culture microalgae *Scenedesmus* sp. and to identify the optimal concentration of microalgae *Scenedesmus* sp. due to the nutrient and heavy metal removals from wet market wastewater based on laboratory scale. The samples were taken from Public Market Parit Raja, Batu Pahat at 9a.m. in the morning as a peak hour operating stall using grab sampling technique. The characteristics of raw wastewater and microalgae *Scenedesmus* sp. were determined. There are five sample wastewater (used 625ml for each five sample wastewater) with five different samples concentration of microalgae which are $6.50x10^5$, $49.88x10^4$, $34.75x10^4$, $19.63x10^4$ and $49.88x10^4$ cell/ml with 16 days as duration for period study. The microalgae were cultured by BBM for eight days and another eight days for treatment with replicates three times for each sample. The analysis were measured due to the nutrient and heavy metal removals which are TN, TP, TOC, Fe and Zn during eight days treatment process. Based on experimental result, the optimum removal efficiency for each concentration were achieved 45.6-86.4% of nutrients and heavy metals. The highest amount of nutrient and heavy metal removals after wastewater treatment by microalgae are TN 74.77%, TP 82.17%, TOC 86.36%, Fe 65.76% and Zn 84.14%. As conclusion for this experiment, Sample 2 (concentration $49.88x10^4$ cell/ml of microalgae *Scenedesmus* sp.) is the optimum concentration due to the highest percentage of nutrients and heavy metals removal which achieved 65.3-82.1% which TN 65.32%, TP 76.77%, TOC 80.34%, Fe 65.76% and Zn 82.12%.

Introduction

In Malaysia, the wet market activities contribute to unpleasant odour and dirty environment because the discharge were directly to the drainage without any treatment. This problem may lead to uncomfortable condition to buyers, sellers and residents at wet market area [1]. Moreover, the wastewater flow directly into drainage in that area may cause the drain clogged and the problem getting worst if no action taken by responsible person [2].

Microalgae had been choose as new technology to treat wastewater because it cheap and easily to handle because of their capabilities during photosynthesis process and eutrophication if wastewater contains excessive amount of nitrogen and phosphorus which able to incorporate with some nutrients such nitrogen and phosphorus [3]. From previous studies, there still less information about wet market wastewater treatment using microalgae cause the researches were focused on special media from laboratory, municipal wastewater, industrial wastewater and animal wastewater [4,5]. From analysis findings, the best result analysis is from Korea where this research had been using microalgae *Scenedesmus sp.* to treat swine wastewater by fermentation process which involved aeration system for 7 days and from the results, the removal efficiency nutrient for TN was 95%, TP was 86%, SS was 88% and pH was 13% [4]. The study of treatment of drainage solution

from hydroponic greenhouse production with microalgae *Chlorella vulgaris* had done by Hultberg (2012) from Sweden with the large reduction in nutrients which phosphorus was 99.7% while nitrogen was 20.7% removal efficiency was achieved [6]. Microalgae *Scenedesmus* sp. and *Chlorella* sp. have good efficiency for heavy metals removal such as Iron (Fe) and Zinc (Zn) [3].

Therefore, the objectives of this study were to culture the microalgae *Scenedesmus* sp. and to identify the optimal concentration of microalgae *Scenedesmus* sp. due to the nutrient and heavy metal removals from wastewater by this type of treatment.

Materials and Methods

Sampling of Wet Market Wastewater

10 liters of wastewater was collected for the sampling. The duration of this sampling is on peak hour operating stall which around 9.00a.m. in the morning and the samples were analyzed in UTHM Environmental Laboratory.

Characterization of Wet Market Wastewater

In this study, the wastewater characteristics that were determined are biochemical oxygen demand (BOD), chemical oxygen demand (COD), total suspended solid (TSS), sulfate, total chlorine, pH, turbidity, temperature and oil and grease. Table 1 below shows the parameters were monitored during the wastewater characteristic operations and methods that were used by using Hach DR 5000 Spectrophotometer.

Table 1. Characteristics of Wastewater and Methods

Wastewater Characteristics	Methods
Biochemical Oxygen Demand (BOD)	Standard Method APHA 5210 -B
Chemical Oxygen Demand (COD)	Standard Method APHA 5220 -D
Total Suspended Solid (TSS)	Standard Method APHA 2540 -D
Sulfate	Sulfa Ver4 Method (Method 8051, DR5000)
Total Chlorine	Mercuric Thiocyanate Method (Method 8113, DR 5000)
pH	Standard Method APHA 4500 -H B
Turbidity	Attenuated Radiation Method (Method 10047, DR5000)
Temperature	Standard Method APHA 5220 -D
Oil and Grease	USEPA[1] Hexane Extractable Gravimetric Method (Method 10056)

(Source: Department of Civil and Environmental Engineering, UTHM, 2013)

Characterizations of Microalgae *Scenedesmus* sp.

Scenedesmus sp. are green algae which commonly found in the plankton of freshwater rivers, ponds, lakes, and sometimes in brackish habitats [7]. The advantages of using microalgae *Scenedesmus* sp. over the other available feedstock such as inorganic nutrients which preferred to the nitrogen, phosphorus and carbon are essential the following: their rapid growth rate, productivity and these microalgae are able to grow in a variety of environmental conditions [8].

Experimental Design

1) Culturing Microalgae and Concentration of Microalgae *Scenedesmus* sp. for Treatment Process

The culturing for this study had done by using Bolts Basal Medium (BBM) which additional of some stock solutions consisted of $NaNO_3$ 25g/l, $CaCl_2.2H_2O$ 2.5g/l, $MgSO_4.7H_2O$ 7.5g/l, K_2HPO_4 7.5g/l, $KH_{2PO}4$ 17.5g/l, NaCl 2.5g/l, EDTA 50.0 g/l, KOH 31.0g/l, $FeSO_4.7H_2O$ 4.98g/l, H_2SO_4 1ml, H_3BO_3 11.42g/l and 1ml of trace elements solution ($ZnSO_4.7H_2O$ 8.82 g/l, $MnCl_2.4H_2O$ 1.44 g/l, MoO_3 0.71 g/l, $CuSO_4.5H_2O$ 1.57 g/l and $Co(NO_3)_2.6H_2O$ 0.49 g/l) to the distilled water and autoclave the microalgae *Scenedesmus* sp. at 121°C (15PSI for 15 minutes) [9].

2) Lab Batch Reactor Preparation for Treatment Process

These microalgae were cultured within 8days before started the wet market wastewater treatment process. The five sets of sample wastewater were prepared and the concentration of microalgae *Scenedesmus* sp. that were used are 6.50 x 10^5, 49.88 x 10^4, 34.75 x 10^4,19.63 x 10^4 and 4.50 x 10^4 cell/ml. These five different concentrations were repeated three times to get accurate readings. Each sample was used to measure the amount of nutrients and elements removal on 0, 2nd, 4th, 6th and 8th day. There are three different apparatus were used to measure the nutrients and heavy metals removal due to the laboratory analysis parameter which Total Organic Carbon Analyzer (TOC): TOC-VCSH used for TN and TOC, Ion Chromatography System (ICS-2000) used for TP while Flame Atomic Absorption Spectrometry (AAS) used for heavy metals Fe and Zn.

3) Calculation of Removal Efficiency

The percentage removal of nutrients and heavy metals was used to determine how efficient the treatment process by microalgae. In wastewater, the percentage removal of TN, TP, TOC, Fe and Zn can be determined by the following equation:

$$\frac{(C_0 - C_i)}{C_0} x100\% = \%\text{Removal} \quad (1)$$

where C_0 represents nutrient concentration at the beginning experiment while C_i represents the nutrients concentration on the day which reading taken during the wet market wastewater treatment process [10].

Result and Discussions

Wet Market Wastewater Characteristics

Table 2 shows the characteristics of raw wastewater from Public Market Parit Raja compared to previous research by Zulkifli [2].

Table 2. Characteristics Wet Market Wastewater

Parameter	Public Market Parit Raja	Pasar Ampangan (Zulkifli, 2011)
pH	6.0±0.1	5.6 – 5.8
Turbidity (NTU)	66.0±8.9	-
Biochemical Oxygen Demand, BOD (mg/l)	89±3.61	71 - 122
Chemical Oxygen Demand, COD (mg/l)	456±8.19	381 - 560
Total Suspended Solid, TSS (mg/l)	132.3±1.7	60 - 122
Sulfate, SO_4^{2-} (mg/l)	32.3±0.78	-
Total Chlorine, Cl^- (mg/l)	32±0.69	-
Total Nitrogen, TN (mg/l)	36.9±0.5	30.3 – 37.3
Total Phosphorus, TP (mg/l)	1.61±0.13	ND – 22.2
Total Organic Carbon, TOC (mg/l)	118.67±2.89	-
Oil and Grease	5.22±0.07	13 - 43
Zinc, Zn (mg/l)	0.312±0.0021	-
Ferum, Fe (mg/l)	1.071±0.0010	-

Based on the Table 3, the value for COD and BOD were 456±8.19 mg/l and 89±3.61 mg/l for BOD in Public Market Parit Raja while COD and BOD from Pasar Ampangan were 381 to 560 mg/l and BOD in Pasar Ampangan is 71 to 122 mg/l which shows that both value for COD and BOD from Public Market Parit Raja wastewater is in range by the Pasar Ampangan wastewater. For pH value comparison, it shows that the range of pH is 5.6 to 5.8 in Pasar Ampangan compared to 6.0±0.1 in Public Market in Parit Raja where the wastewater from Parit Raja is less acidic than Pasar Ampangan because of less usage of detergents for washing floor. The value for TSS is 132.3±1.7 mg/l for Public Market Parit Raja wastewater which is higher than TSS in Pasar Ampangan where the value is 60 to 122 mg/l. The total nitrogen, TN for Public Market Parit Raja wastewater is 36.9±0.5 mg/l and this value is similar compared to total nitrogen in Pasar Ampangan where the value is 30.3 to 37.3 mg/l.

Efficiency Removal of Wet Market Wastewater Treatment by Microalgae *Scenedesmus* sp.

The nutrients such as nitrogen and phosphorus are the primary concerns in wastewater treatment. According to previous studies, microalgae are known to sequester heavy metal [3]. Table 3 below shows the highest percentage removal of nutrients and heavy metals for each five different concentration while Fig. 1 shows that the highest percentage nutrient (TN, TP and TOC) and heavy metal (Fe and Zn) removals for each concentration which these highest efficiency removal results within 8 days treatment process.

Table 3. Highest Percentage Removal of Nutrients and Heavy Metals

Nutrient	Highest Percentage Removal (%)				
	Sample 1 (6.50×10^5 cell/ml)	Sample 2 (49.88×10^4 cell/ml)	Sample 3 (34.75×10^4 cell/ml)	Sample 4 (19.63×10^4 cell/ml)	Sample 5 (4.50×10^4 cell/ml)
TN	74.77	65.32	46.03	51.52	50.71
TP	79.75	76.77	82.17	82.07	79.29
TOC	68.90	80.34	86.36	78.10	73.72
Fe	55.77	65.76	59.79	61.21	45.60
Zn	84.14	82.12	79.67	81.33	71.94

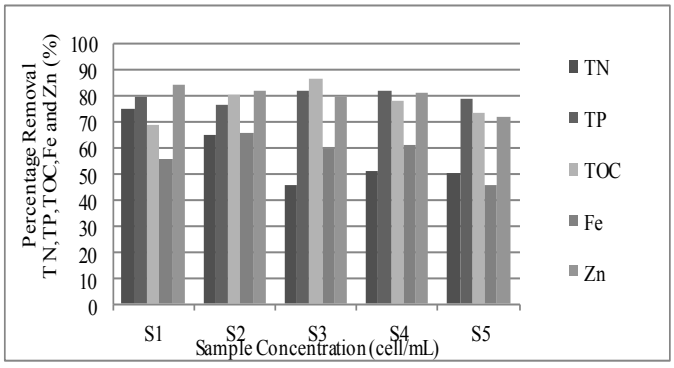

Fig. 1. Percentage nutrients and heavy metal removals against concentrations

From the Table 3, the maximum percentage removal for TN is 74.77% from Sample 1 (6.50×10^5 cell/ml) while the lowest percentage removal is 46.03% from Sample 3 (34.75×10^4 cell/ml). Nitrogen reduction by microalgae which performed in wastewater with high concentration of TN is commonly from 60% to 90% [9]. The less concentration of microalgae cause the amount of TN cannot be removed perfectly from the wastewater. The maximum percentage removal for TP is 82.17% from Sample 3 (34.75×10^4 cell/ml) and the minimum percentage removal for TP is 76.77% from Sample 2 (49.88×10^4 cell/ml). For this study, the microalgae used lot of phosphorus amount as nutrients in this wastewater sample because the PO_4^{3-} are not present in the atmosphere [11]. The third parameter, TOC from Sample 3 (34.75×10^4 cell/ml) is 86.36% as the highest percentage removal and 68.90% from Sample 1 (6.50×10^5 cell/ml) is the lowest percentage removal.

Based on Table 3 and Fig. 1, the highest removal efficiency for Fe for treated wastewater is 65.76% for Sample 2 ($49.88x10^4$cell/ml) and 84.14% from Sample 1(6.50×10^5 cell/ml) for Zn. Generally, heavy metal able to be removed in wastewater treatment with application of microalgae *Scenedesmus* sp.. The result as percentage maximum removal for Zn in treated wastewater in Public Market Parit Raja was lower when compared to previous research by Travieso where the maximum removal of Zn using *Scenedesmus acutus* was 91% [12].

The highest of TN and Zn removal which 74.77% and 84.14% were obtained in Sample 1 ($6.50x10^5$ cell/ml), TP and TOC removal which 82.17% and 86.36% were obtained in Sample 3 ($34.75x10^4$ cell/ml) and Fe removal which 65.76% were obtained in Sample 2 ($49.88x10^4$ cell/ml). It clearly shows each concentration will give different value of percentage removal. Overally, the growth time on day 8 give good removal efficiency.

Conclusion

As conclusion, Sample 2 (concentration $49.88x10^4$ cell/ml of microalgae *Scenedesmus* sp.) is the most efficient concentration due to the highest percentage of nutrients and heavy metals removal were achieved 65.3-82.1% which TN 65.32%, TP 76.77%, TOC 80.34%, Fe 65.76% and Zn 82.12%.

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