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Optimization of POME treatment process using microalgae and ultrafiltration

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Abstract. Palm oil mill effluent (POME) was produced in huge amounts in Malaysia, and if it discharged into the environment, it causes a serious problem regarding its high content of nutrients and high levels of COD and BOD concentrations. This study was devoted on POME treatment and purification using an integrated process consisting of microalgae treatment followed by membrane filtration. The main objective was to find the optimum conditions as retention time and pH in the biological treatment of POME. Since after the optimum conditions there is a diverse effect of time and the process become costly. According to our knowledge, there is no existing study optimized the retention time and percentage removal of nutrients for microalgae treatment of POME wastewater. In order to achieve with optimization, a second order polynomial model regression coefficients and goodness of fit results in removal percentages of ammonia nitrogen (NH_3-N), orthophosphorous (PO_4^{-3}), COD, TSS, and turbidity were estimated. WinQSB technique was used to optimize the objective function of the developed model, and the optimum conditions were found. Also, ultrafiltration membrane is useful for purification of POME samples as verified by experiments.

Keywords: POME; microalgae; optimization; membrane filtration

1. Introduction

Raw palm oil mill effluent (POME) wastewater is a colloidal suspension containing 95-96% water, 0.6-0.7% oil, and 4-5% total solids, including 2-4% suspended solids that mainly consist of debris from the palm fruit mesocarp generated from three main sources, namely sterilizer condensate, separator sludge, and hydrocyclon wastewater. Also POME contains high concentrations of proteins, carbohydrate, nitrogenous compounds, lipids and minerals that may be converted into useful materials using microbial processes (Wu *et al*. 2007). If this untreated POME was thrown out to the environment, it has a negative effect on it. Therefore it must be a critical needful solution savings the equilibrium between environmental safety and sustainable energy that can be produced from nutrient materials existing in the POME (Ahmad *et al*. 2005).

Bio-treatment of POME with microalgae is the solution because the algae grows in photosynthesis method that has the ability to use solar energy and transforming it to a profitable biomass take advantages from nitrogen and phosphorous compounds presented already in POME. Microalgae culture becomes an economizing solution to the problem treats the wastewater by

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consuming nutrients by microalgae growth and transform it to a biomass which is in turn a major source of biofuel (Abdel-Raouf *et al*. 2012). Also algae can be cultivated in non- agriculture land using wastewater (Shaikh *et al*. 2013). Microalgae lowering values of the chemical oxygen demand (COD) and biological oxygen demand (BOD) from POME, Colak and Kaya (1988), studied the biological wastewater treatment by algae, they found that in domestic wastewater treatment, reduction of BOD and COD were 68.4% and 67.2% respectively, Kaplan *et al*. (1988), used the microalgae for industrial wastewater treatment. Mohammed (1994), used algae to treat wastewater with high rates of nitrogenous compounds, also, Giorgos and Nerantzis (2013), reviewed the studies available on cultivation of microalgae under stress conditions. The optimization of bio-treatment of POME wastewater is significant for process expansion. It is overwrought and time exhaustion to carry out this process involving classical method of experimentation (Wang and Lu 2005). Furthermore, it has been neglecting the interaction effects between the operating parameters and enclosing the predication of the optimum conditions. In contrast, the statistical optimization technique which satisfies the above function with minimum number of experiments is the experimental design, this statistical optimization techniques have been effectively appointed in optimization of various bioprocesses. In the last two decades, the statistical optimization experimental design for bio-treatment has been reported. For example, Abdel-Fattah and Olama (2002), used factorial design method to evaluate and optimize the culture conditions of solid state fermentation of microalgae, Li *et al*. (2002), optimized the cultural medium for bacteriocin production using response surface methodology, while, Poonsuk *et al*. (2009), optimized the microbial community for production of biohydrogen from POME wastewater by thermophilic fermentation process. Taha and Ibrahim (2014), studied the reduction of COD from anaerobically treated palm oil mill effluent and they used the response surface methodology, they found that 75% of COD was removed at the optimum conditions.

Ultrafiltration technology depends on pore size that can hold the suspended solid materials, and it also hinders bacteria and viruses. Thus it has been used in water purification applications (Ahmad *et al*. 2006). In the last decades, ultrafiltration (UF) membrane was put into application in production of pure water used in different fields as in food or pharmaceutical industries or in industrial wastewater treatment (Jonsson and Tragardh 1990, Girard and Fukumato 2000, Van Reis and Zydney 2001). Therefore UF membrane may act as a good tool in the purification of POME. In this case, chemical cleaning is utilized to enhance the efficiency of membranes for filtration. The potential use of sodium hydroxide (NaOH), sodium chloride (NaCl), hydrochloric acid (HCl), ethylenediaminetetraacetic acid (EDTA), and ultrapure water (UPW) as cleaning agents have been investigated by Mohammad *et al*. (2014), It was found that sodium hydroxide is utmost strong cleaning agent, the optimum conditions that apply were: 3% for the concentration of NaOH, 45°C for temperature solution, 5 bar operating pressure, and solution pH 11.64. This study is devoted to the treatment of POME wastewater using integrated unit, consisting of microalgae treatment of the wastewater followed by an ultrafiltration membrane process, as a treatment and purification of POME wastewater, then the optimization of the parameters affecting the biological treatment process. Because there is no specific literature existed on how the retention time and pH of algal solution would affect along with the treatment of POME in range of applicable experimental variables. Thus the main goal of the present study was to investigate the effect of retention time (0-23 days) and pH (7.5-9.26) on the efficiency of algae treatment of POME and optimize these operating parameters to find the optimum conditions to reach a highly efficient microalgae treatment with highest removal percentages of ammonia nitrogen (NH_3-N), orthophosphorous (PO_4^{-3}), COD, total suspended solids (TSS), and turbidity. Also, this process is

Table 1 Coded and real values of operating variables

Coded variables		Real variables	
pH (-)	Retention time (day)	pH (-)	Retention time (day)
-1	-1	7.5	0
0	-1	9.14	0
+1	-1	9.26	0
-1	0	7.5	12
+1	0	9.26	12
0	0	9.14	12
-1	+1	7.5	23
+1	+1	9.26	23
0	+1	9.14	23
0	0	9.14	12
0	0	9.14	12

considered as a completely green process without using any chemicals that produce an environmental injured by products.

2. Design of experiments

In order to achieve with the SRM of experimental design, we must firstly calculate the number of experiments required by using Eq. (1)

$$N = 2^K + 2K + 1 \tag{1}$$

Where, N = no. of experiments, K = no. of variables.

It means that for two operating variables, we need at least 9 experiments, plus two experiments represent the probability of experimental errors. Then the experiments must be designed in form of real and coded values of operating variables in order to represent three levels of effect as high (+1), medium (0), and low level (-1), as shown in Table 1. The operating range of time is (0-23) day, which is the range applied in industry. 0 day represents the POME before starting the microalgae treatment process, and 23 day is the maximum economic time. The pH range used in this study is (7.5-9.26), which is related to retention time, and it has been obtained from the following empirical correlation (Eq. (2))

$$pH = 8.751 + 0.374\log(t) \tag{2}$$

where, t = time (day), while, pH values between 9 and 11 have been applied by Ting *et al*. (2013).

3. Materials and methods

3.1 Palm oil mill effluent (*POME*)

Palm oil mill effluent (POME) samples were taken directly after the last anaerobic digester

Table 2 Characteristic of anaerobic digested POME after centrifugation

Parameter	Unit	Value
pH	-	7.48
Chemical Oxygen Demand, COD	mg/L	4045.0
Ammonia Nitrogen, NH_3-N	mg/L	334.0
Orthophosphorus, PO_4^{-3}	mg/L	465.0
Total Suspended Solids, TSS	mg/L	607.0
Turbidity	NTU	395.5

from East Mill Sime Darby Plantation, Carey Island, Selangor, Malaysia. The samples were kept in a cool room at a temperature around 4°C before using. They were being centrifuged at 8000 rpm for 5 minutes to obtain the supernatant of the samples. The characteristics after centrifugation are shown in Table 2. The supernatant of the samples will be named anaerobic POME (AnPOME).

3.2 Microalgae

In this research, the microalgae strain selected was *Botryococcus braunii* which was obtained from the Department of Chemical and Process Engineering, Faculty of Engineering and Built Environment in the National University of Malaysia. *B. braunii* was 0.3 g/l cultivated in Bold Basal Medium (BBM) for 10 days before transferring it into a conical flask containing two litters of AnPOME. The medium was aerated using a peristaltic pump with aeration rate of 1 L/h placed on the bottom. The flask was placed under two continuous fluorescent lamps from both sides with an average light intensity of 100 lux and a constant temperature of 21°C.

3.3 Type of membrane

The membrane being used in the research was purchased from the company of Amfor Inc. (China). The membrane used was an ultrafiltration (UF) membrane type PES 10 having the nominal molecular weight cut-off (WMCO) of 10 kDa that made from polyethersulphone with pH resistance range from 1 to 13. The average permeability of UF, was 26.617 ± 4.472 $L.m^{-2}.h^{-1}.bar^{-1}$.

3.4 Analytical method

Chemical oxygen demand (COD) was measured using reactor digestion method with two types of vials for high range (HR) concentration and low range (LR) concentration ranging from 0 to 150 mg/L and 0 to 1500 mg/L respectively. Ammonia nitrogen (NH_3-N) was measured using Nessler method while orthophosphorus (PO_4^{-3}) was measured using Molybdovanadate method. Total suspended solid (TSS) was measured using photometric method. All analysis of parameters COD, NH_3-N, PO_4^{-3} and TSS were analysed with the help of HACH DR/3900 spectrophotometer. As for turbidity, it was measured using HACH turbidimeter (2100AN) with the unit NTU. For pH, a pH meter was used.

3.5 Experimental set-up

After the microalgae strain of *B. braunii* was cultivated for 10 days, 200 ml of *B. braunii* was inoculated into 2 liters of anaerobic digested palm oil mill effluent (AnPOME) that was pretreated

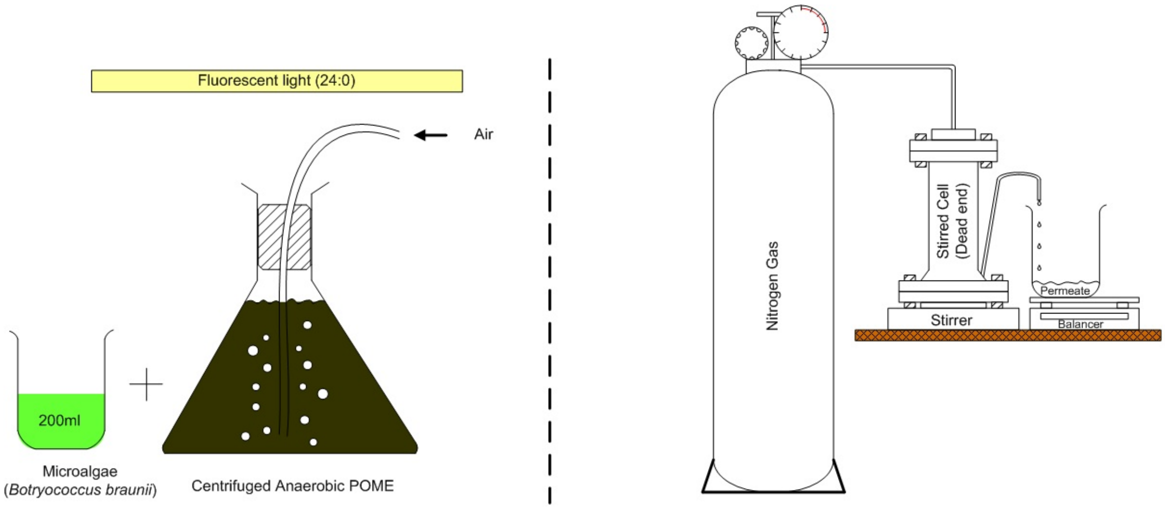

Fig. 1 Schematic diagram of experimental work

with centrifugation for removal of solids. The reactor consists of a 2 liter conical flask with aeration from the bottom using a peristaltic pump. The top of the conical flask was covered with double layer aluminum foils for prevention of contamination with other microalgae strains. Two continuous fluorescent lamps were placed from both sides above the reactor with a constant temperature of 21°C. The samples were taken with an interval of 3 days, from day 0 to day 15 retention time (RT) with the final sample taken on day 23 retention time. In the experiment, retention time of day 0 was the initial sample of AnPOME without being treated with microalgae. Throughout the entire treatment, the pH was not adjusted by any chemicals.

As the samples were collected, another round of centrifugation was done to obtain the supernatant of all the samples. Then analysis of COD, NH_3-N, PO_4^{-3}, TSS, turbidity and pH were done to observe the performance of microalgae treatment with different retention time. The supernatant of the samples after centrifugation from microalgae treatment were used to undergo further treatment using membrane filtration process. The membrane filtration process was carried out using a dead-end stirred cell provided from sterlitech, model HP4750.

Before the samples were filtered using membranes, the newly cut membrane with an effective membrane area of 0.00146 m^2 was soaked in pure water overnight to dissolve any chemical left after manufacture. The next day, compaction of the membrane was done for 30 minutes using pure water to further remove the impurity and to unblock the pores that were previously blocked. Each sample from different retention time was filtered using UF membrane. The transmembrane pressures used with UF membrane was 5 bars. In every sample, new pieces of membrane were cut and used and there was no attempt of any sort of cleaning of membranes to be reused. Analysis of COD, NH_3-H, PO_4^{-3}, TSS, turbidity and pH were done again to identify the removal or changes after membrane filtration. Fig. 1 shows a schematic diagram of the experimental work.

4. Optimization technique

According to surface response methodology of experimental design, the operating variables that need to be optimized such as retention time (X_1), and pH (X_2), were correlated in a non-linear polynomial model equation, which is more sufficient proposed model to show the interaction effects between variables. The coefficients of model equation were estimated using a STATISTICA program (version 8.0). The proposed model was

$$Y = a_0 + a_1X_1 + a_2X_2 + a_3X_1X_2 + a_4X_1^2 + a_5X_2^2 \quad (3)$$

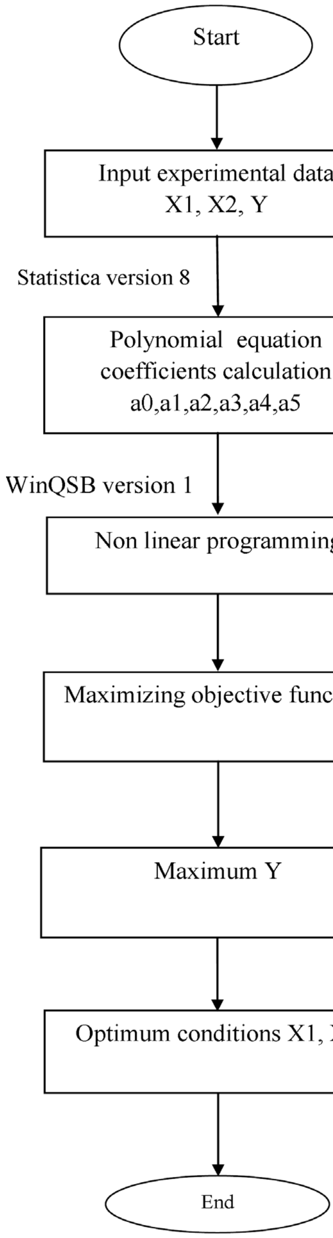

Fig. 2 Flow chart of optimization process

Where: X_1 and X_2 are operating variables representing retention time (day) and pH, while, Y represents the objective function such as percentage removal of nutrients (%), COD, TSS, and turbidity.

The optimization process was done using winQSB technique (winQSB software version 1.0) in order to find the optimum values of operating conditions for the purpose of enhancing the efficiency of POME treatment by microalgae. The system consists of several modules, one of them is a nonlinear programming used in this study. There are three methods to solve the problems with this system: Graphical method, Algebraic method, and Simplex method. The simplex method is highly sensitive with high efficiency; therefore it was used to solve the nonlinear optimization problem dealing with the present study.

5. Solution procedure

After an experimental data were prepared and input to the system, a problem, solve is made by choosing the nonlinear programming. A seven constrains are made according to each experiment operating conditions, and the constraints represented in a matrix form. The program was running for maximizing the objective function (% removal of nutrients), iteration made until the error reaches a minimum value to fit the suggested nonlinear model equation. Fig. 2, shows the flow chart of the solution procedure, starting from finding the polynomial model equation coefficients by a multiple regression method with the aid of STATISTICA software, then the model equations which represent the removal efficiencies of pollutants were optimized using WINQSB technique to maximize or minimize the objective function and finding the optimum conditions.

6. Results and discussion

6.1 Multiple regression analysis results

The objective is to find the optimum retention time in the microalgae treatment of POME wastewater considering as a most important parameter in algae treatment because that below the optimum time the treatment process is not wholly complete and water still containing polluted materials, while above optimum value, there is a diverse effect of time and the process become costly, therefore retention time was considered as a very important parameter must be estimated carefully. Consequently pH is dependent on time, but it has a considerable effect on the removal efficiency of nutrients. According to our knowledge, there is no study optimized the retention time and percentage removal of nutrients, COD, TSS, and turbidity for microalgae treatment of POME wastewater. In order to achieve the above objective, a polynomial model regression coefficients and goodness of fit results in removal percentages of nutrients and other properties such as COD, TSS, and turbidity were estimated and illustrated in Table 3-4. The observed (experimental) and predicted (model) values for each property, demonstrated a good agreement between experimental and model values which is approved by a goodness of fit analysis through measurements of correlation coefficient, variance, mean absolute error, and mean square error values.

6.2 Optimum condition results and effect of operating parameters

The optimum conditions that give optimum objective functions are illustrated in Table 5, and explained as follows:

Table 3 Results of multiple regression analysis

Property	Regression coefficients					
	a0	a1	a2	a3	a4	a5
COD (mg/L)	289399.3	3653.1	-66992.3	-424.46	9.194	3902.43
NH_3-N Removal (%)	3389.118	124.819	-856.201	-13.451	0.00714	53.8917
PO_4^{-3} Removal (%)	-6494.96	-105.02	1522.328	11.997	-0.194	-88.506
TSS (mg/L)	57270.05	528.913	-13712.9	-64.041	1.573	821.049
Turbidity (NTU)	41809.66	804.827	-9663.57	-90.292	0.946	558.771

Table 4 Goodness of fit analysis results

Property	Goodness of fit analysis			
	Correction coefficient (R)	Variance explained (%)	Mean absolute error	Mean square error
COD (mg/L)	0.9998	99.97	10.056	169.26
NH_3-N Removal (%)	0.9995	99.91	0.7115	0.849
PO_4^{-3} Removal (%)	0.9992	99.84	0.5535	0.5128
TSS (mg/L)	0.9993	99.87	4.746	42.082
Turbidity (NTU)	0.9995	99.90	2.815	13.267

Table 5 Optimum condition results

Property	Optimum conditions	
	X_1 (time, Day)	X_2 (PH)
COD (mg/L)	15	9.2
NH_3-N Removal (%)	18	9.22
PO_4^{-3} Removal (%)	15	9.2
TSS (mg/L)	15	9.2
Turbidity (NTU)	15	9.2

The optimum conditions for percentage removal of NH_3-N, are:

Time = 18 days, and pH = 9.22 gives maximum removal of 92.857%. Fig. 3 shows the optimum retention time, and Fig. 4 shows the effect of time and pH on % removal of NH_3-N as a response surface. It was clearly obvious that 18 days is the best time for maximum removal of NH_3-N, since before day 18, the microalgae still grown by feeding with nitrogen and phosphorous compounds in the POME wastewater, and after day 18, the values of removal percentages of NH_3-N were lowered because that microalgae capacity to eliminate NH_3-N was reduced and the microalgae became older. Also, when the microalgae removed NH_3-N, it was simultaneously transferred it to the biomass as a result of concentration equilibrium and mass conservation, that's clarified why the removal percentage was decreased after optimum retention time. But at the optimum it has a maximum removal and maximum conversion of NH_3-N into biomass, which is more useful in biofuel production.

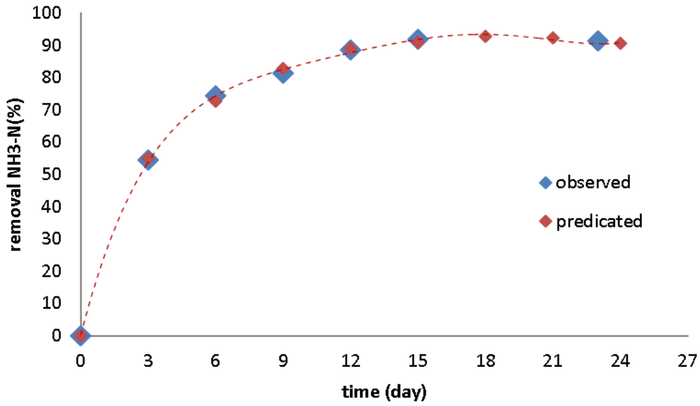

Fig. 3 Effect on retention time on % removal of NH_3-N

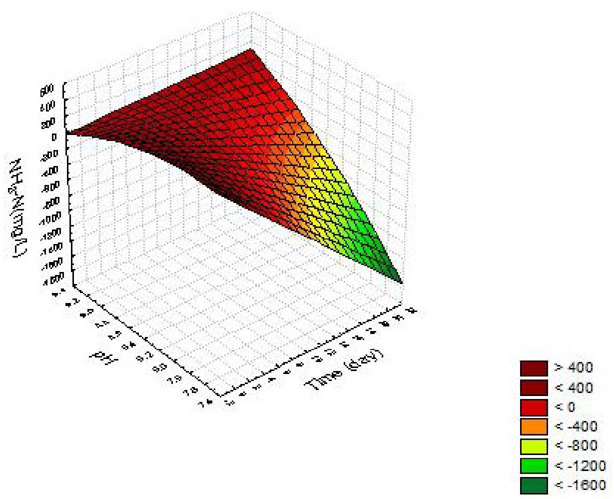

Fig. 4 Surface response of NH_3-N concentrations

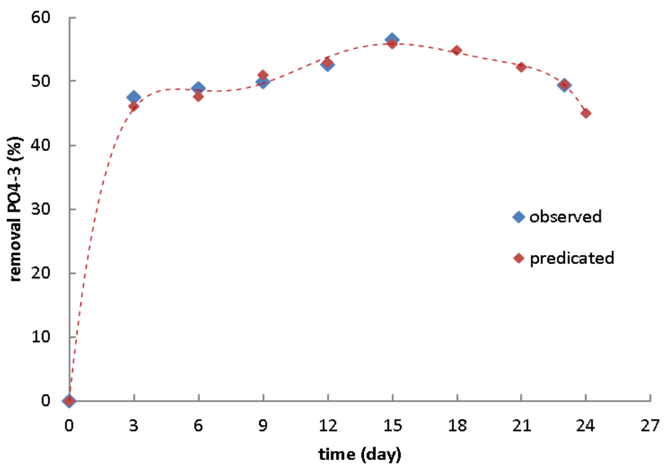

Fig. 5 Effect on retention time on % removal of PO_4^{-3}

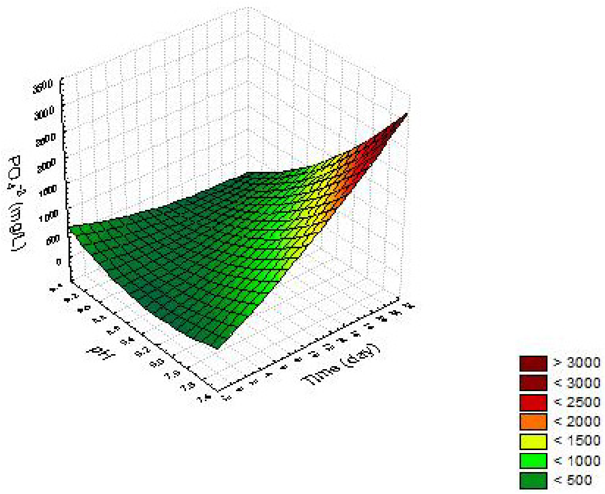

Fig. 6 Surface response of PO_4^{-3} concentrations

The optimum conditions for % removal of PO_4^{-3} are:

Time = 15 days, and pH = 9.2 gives maximum removal of 56.5%. Fig. 5 shows the optimum retention time, and Fig. 6 shows the effect of time and pH on % removal of PO_4^{-3} as a response surface.

For COD, TSS, and Turbidity, the optimum conditions are:

Time = 15 days, and pH = 9.2 gives minimum values of 1650 mg/L, 50 mg/L, and 21 NTU for COD, TSS, and Turbidity respectively.

Figs. 7-9, show the optimum retention time values, while, Figs. 10-12 show the effect of operating parameters on the COD, TSS, and turbidity as a response surface.

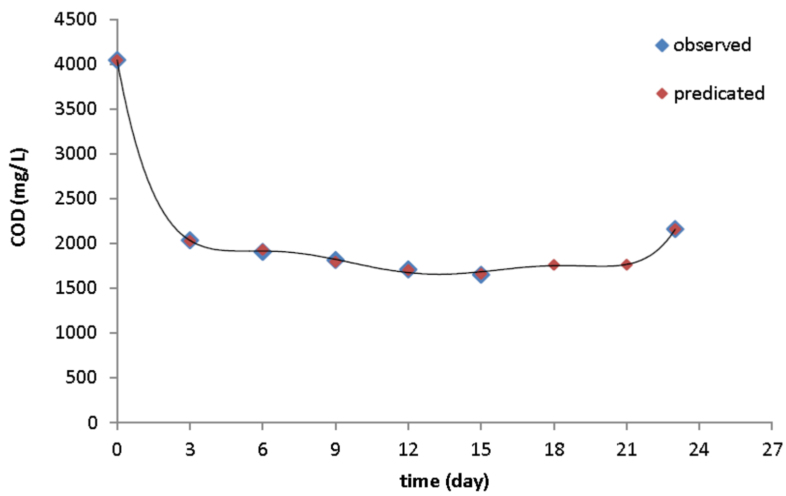

Fig. 7 Effect of retention time on COD concentrations

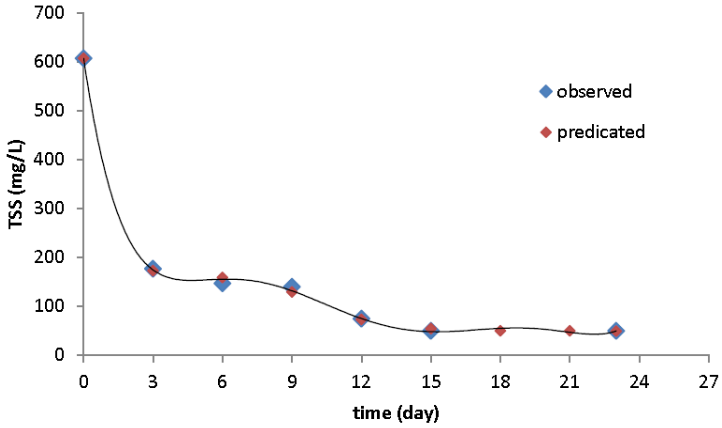

Fig. 8 Effect of retention time on TSS values

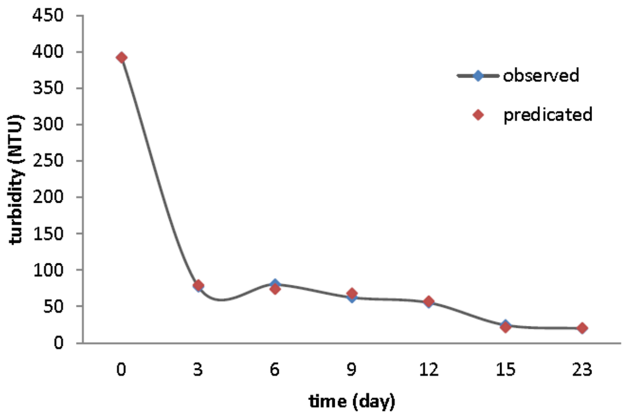

Fig. 9 Effect of retention time on turbidity values

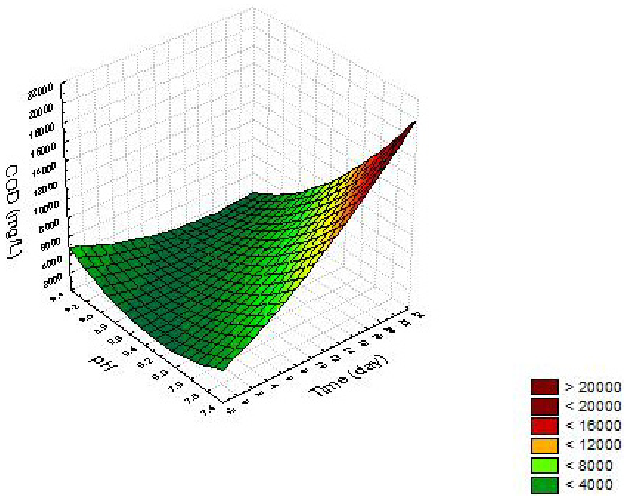

Fig. 10 Surface response of COD

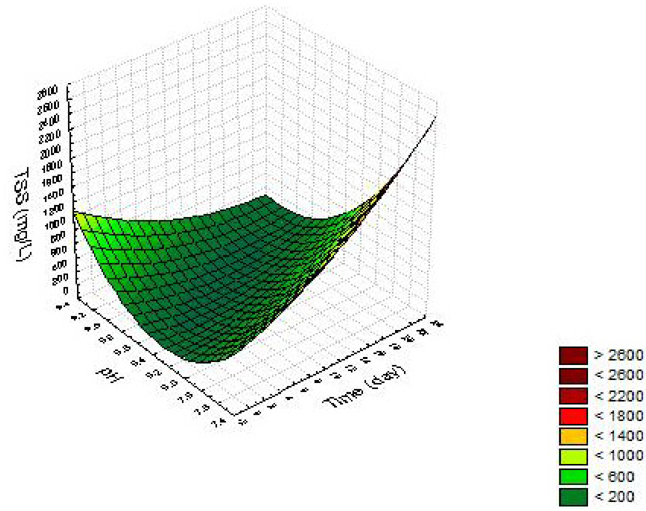

Fig. 11 Surface response of TSS

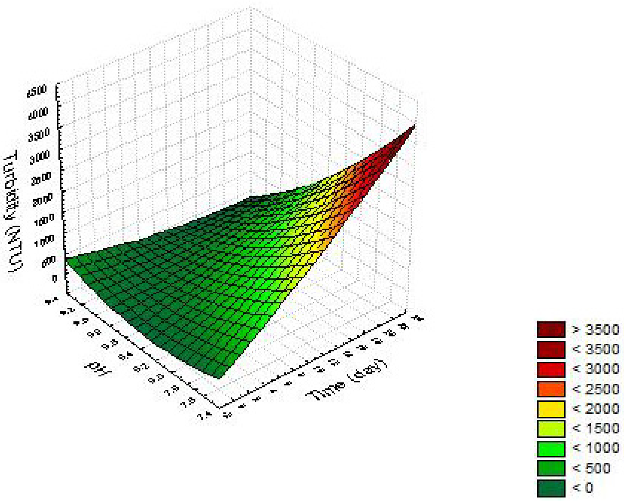

Fig. 12 Surface response of turbidity

The retention time day 15 represents the optimum retention time for PO_4^{-3}, COD, TSS, and turbidity. It is the same reason for NH_3-N removal, dealing with equilibrium and mass conservation of $PO_4^{-3}$and COD. As for TSS and turbidity, it depends on the amount of solid particles in the solution.

Since uptake is the main mechanism of nutrient removal by microalgae, the microalgal population growth rate directly affects the nutrient removal rate. Meantime, nitrogen and phosphorus could be simultaneously utilized and removed efficiently only if the N/P ratio in wastewater is in a proper range. The growth kinetics of microalgae also influences the lipid accumulation in microalgal cells: the microalgal population growth rate affects lipid accumulation rate, and the nutrient condition determines the lipid productivity and lipid content per microalgal biomass. Normally microalgae contain three kinds of organic material: protein, carbohydrate and natural lipid. Beneath environmental pressure the stopping of microalgal cell division is observed and the synthesis of CO_2 is transformed to lipid as storage of energy, so the lipid content per microalgal biomass is raised.

Fig. 13 shows the effect of pH of the POME solution on NH_3-N, PO_4^{-3}, TSS concentrations, and turbidity. From the figure we can clearly recognize that pH value of 9.2 has lower values of TN, TP, TSS, and turbidity, means it was the optimum value, proved by experiments and model simulation. The concentration of nutrients can be affected by pH. The changes in pH can affect the solubility of ammonium and orthophosphates present in the substrate medium. Not only did the microalgae helped reduction of NH_3-N and PO_4^{-3} by assimilating into their bodies but the high pH condition also encouraged ammonia stripping by converting ammonium ions into ammonia gas and orthophosphorus precipitation with calcium in the form of calcium phosphate, that further reduce the concentrations in the medium. Thus, better removal of nutrients was observed as a result of higher pH.

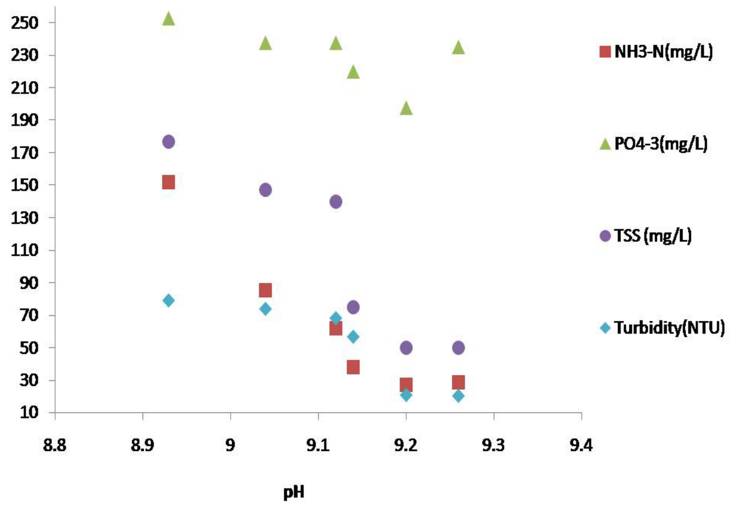

Fig. 13 Effect of pH on NH_3-N, PO_4^{-3}, TSS concentrations and turbidity values

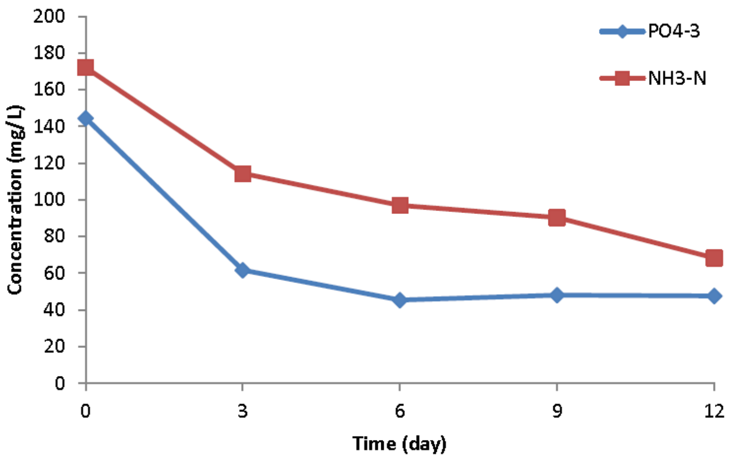

Fig. 14 NH_3-N and PO_4^{-3} concentrations with time during UF membrane separation

6.3 Effect of time of UF membrane separation

Figs. 14-16, show the ultrafiltration membrane separation of POME wastewater, we can see clearly that the concentration of NH_3-N, PO_4^{-3}, COD, TSS, and turbidity were decreased sharply at first three days of membrane filtration process, after that, the decreasing in concentration become slight until day 6 which indicates lower values of PO_4^{-3}, COD, and TSS. After day 6, the concentration of these parameters has little increased due to membrane fouling which affect the membrane separation efficiency. For turbidity, and NH_3-N, the lowest values are indicated at day 9, and day 12, respectively, this is due to high initial concentrations of total nitrogen present already in POME wastewater samples required more time in order to be removed, in addition to the selectivity of the membrane itself. As for turbidity, it increased slightly after day 9 because of expected membrane pore blocking with solid particles as recognized in Fig. 13.

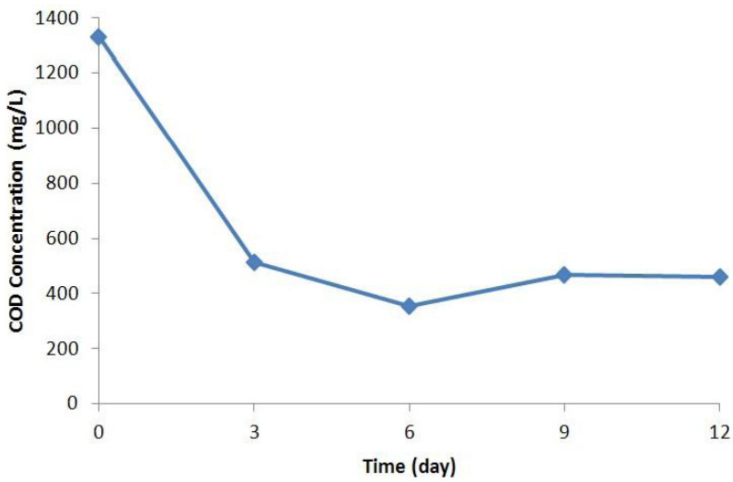

Fig. 15 COD concentrations during UF membrane separation

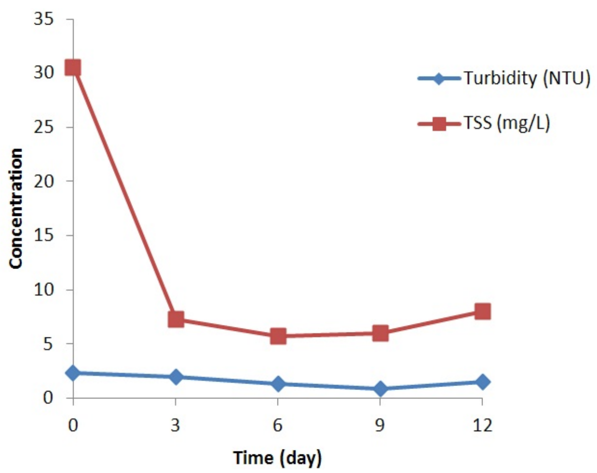

Fig. 16 Turbidity and TSS values during UF membrane separation

7. Conclusions

From this experimental work and optimization technique, we can conclude that: biotreatment using microalgae is considered as a promising method for POME wastewater treatment in addition to biofuel production, distinguished as totally green process without any chemicals used. From optimization done on operating variables of microalgae treatment, it was shown that retention time is a very important parameter needs to be estimated carefully in order to keep the treatment process useful and cost effective. According to optimization process using WinQSB technique, the optimum conditions for % removal of NH_3-N was 18 days retention time, and pH value equals to 9.22, while for % removal of PO_4^{-3}, COD, TSS, and turbidity, the optimum conditions were: retention time equals to 15 days, and pH value equals to 9.2. Also, the ultrafiltration membrane process is useful to the complete purification of POME samples as verified by experiments and testing methods, forms an integrated process for POME treatment and purification.

Acknowledgments

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