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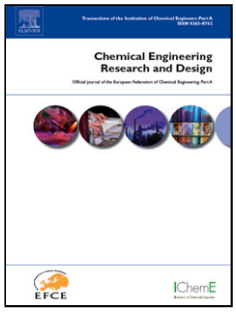

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Process analysis and optimization of single stage flexible fibre biofilm reactor treating milk processing industrial wastewater using response surface methodology (RSM)

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Abstract

Milk processing industries consume significant volumes of sustainable water during processing and operating steps to meet hygienic requirements. Therefore, such industries are recognized as a potential source for producing large volume of contaminated water which generally contains biodegradable matters, and are subsequently suitable to be biologically treated. Based on this, many researchers have been moving towards milk processing wastewater (MPW) treatment via biological approaches. In present study, the performance of a single stage flexible fibre biofilm reactor (SS-FFBR) in mitigation of organic loading of the MPW was assessed. The performance of SS-FFBR in terms of the biological treatment was statistically modelled and analysed in terms of HRT and COD_{in} by employing response surface methodology (RSM). From the results, it is clear increase in HRT and decrease in the COD_{in} led to incrementing the COD removal percentage. In addition, a significant mutual interaction occurs between the variables and other responses. The optimum removal was reported at an HRT of 8 h and COD_{in} of 3922 mg/L corresponded to high OLR of 11.67 kg COD/m^3.d. The experimental results were in good agreement with model prediction.

Keywords: SS-FFBR**,** milk processing wastewater, RSM

1. Introduction

The milk processing industry as one of food processing sectors consumes a large amount of freshwater to ensure the quality control and hygiene standards, thereby, large volumes of wastewater generate (Buabeng-Baidoo et al., 2017). In addition, milk processing wastewater (MPW) is recognized as a potential source of water pollution owing to high level of nutrients and organic matters (Su et al., 2012). Therefore, there is an urgent need to treat such wastewaters before discharge, otherwise deterioration of the water supplies as well as

environment destruction would be a big concern in the near future. As the MPW is extremely biodegradable, therefore, it is amendable to biological treatment (Gil-Pulido et al., 2018). In this regard, various wastewater treatment options, either biological or physico-chemical methods, are applied to remediate the dairy wastewater (Asadi et al., 2017; Bickers and Bhamidimarri, 1998; Gavala et al., 1999; Omil et al., 2003; Rahimi et al., 2016; Rezaee et al., 2015; Tawfik et al., 2008; Zinadini et al., 2015).

Most recently, biological attached growth systems have gained more attention and are proven to be efficient alternative for treatment of carbonaceous and nitrogenous pollutants (Zhang, L. et al., 2015; Zinatizadeh and Ghaytooli, 2015). In the study of Dias et al., the physical properties effect of two types of carrier as a medium was investigated on duration of starting up a moving attached growth system and also COD and ammonia removal (Dias et al., 2018). Zhu et al. reported removal efficiency of 94.9 and 97.7 % in terms of ammonia nitrogen and phosphorus, respectively, with the aid of anabaena flosaquae biofilm growth using the polyvinyl chloride (PVC) media (Zhu et al., 2018). A two-stage attached growth system was developed by research team of Luo et al., to enhance the removal of organic matters and nutrients (Luo et al., 2014). In another work, the nitrogen removal was promoted by integrated reactions of anaerobic ammonium oxidation (anammox) and denitrifying anaerobic methane oxidation (DAMO) in a membrane biofilm reactor (MBfR) (Xie et al., 2018). The development of an active thin layer of microorganisms is identified as biofilm, which plays an important role in the success of any fixed film process. The long residence time of the sludge and the high density of the microorganisms attached to the support media can lead to the process being much more resistant to various shock loads (Gray, 2004). Zinatizadeh and his co-workers applied a moving bed biofilm reactor (MBBR) in order to co-removal of organic compounds and nutrients from a real urban wastewater (Zinatizadeh and Ghaytooli, 2015). The research group of Zhang et al. employed an integrated fixed-biofilm activated

sludge reactors as pilot and full scale to provide nitritation-anammox process (Zhang, L. et al., 2015). In another study, a single-stage anaerobic fluidized ceramic membrane bioreactor (AFCMBR) has been utilized for low-strength wastewater treatment (Aslam et al., 2017). Talwar and his colleague studied the performance of rotating biological contractor (RBC) as a subsequent treatment after photocatalysis to raise degradation of a real pharmaceutical wastewater (Talwar et al., 2018). Zhang et al. surveyed the improved nitrogen removal from rural wastewater using a combined system i.e. a vertical flow trickling filter (VFTF) and a horizontal flow multi-soil-layering (HFMSL) bioreactor (Zhang, Y. et al., 2015). In spite of many studies done using such attached growth systems, biofilm reactors can be more complicated in operation and may require high capital and operating costs. Additionally, the main drawback of attached growth processes is that a high bulk dissolved oxygen (DO) concentration is needed to drive the diffusion of oxygen into the biofilm (Vanhooren et al., 2002).

Response surface methodology (RSM) is known as a collection of mathematical and statistical approaches beneficial to model and analyse problems where a response of interest is influenced by several variables, and the aim is to optimize the response (Montgomery, 2005). Also, the RSM is recognized as a significant application in process designing and optimizing as well as developing design present. This methodology makes a research strategy systematic and efficient to survey the interaction effects of numerous parameters with the help of statistical procedures (Hong et al., 2009). Eventually, the overall effects of the parameters on the method can be represented by RSM (Baş and BoyacI, 2007). Additionally, the RSM provides more information from a few numbers of experiments. Conventional techniques are extremely laborious and time consuming; in addition, such methods do not guarantee the determination of true optimum conditions and are unable to detect synergistic interactions between two or more operating variables (Wu et al., 2009).

In recent years, the application of the RSM has been used effectively by some researchers to optimize and study interactive effects of independent variables in various fields (Asfaram et al., 2015; Schenone et al., 2015; Shaykhi and Zinatizadeh, 2014; Witek-Krowiak et al., 2014, Pirsaheb et al., 2009). As stated by Montgomery (2005), in most problems of RSM, the relationship between the response and the independent variables is unidentified and may be complicated in most cases. Hence, the first stage in the RSM is to appropriately approximate a functional relationship between responses, *y,* and the independent variables. In overall, this methodology utilizes a polynomial with low-order in some areas of the independent variables. The approximating function is a first-order model provided that the response is modelled well by a linear function of the independent variables. Otherwise, a polynomial with higher order must be employed to estimate the response, that is analysed to locate the optimum, i.e. the set of independent variables, such that the partial derivatives of the model response with respect to the individual independent variables is equal to zero. Mainly the majority of RSM problems use one or both of these approximating polynomials (Khuri and Cornell, 1996; Mason et al., 2003; Montgomery, 2005). The ultimate aim of RSM is to determine the optimum operating conditions for the system, or to find out the region that complies with the operating requirements. There are generally three various types of experimental design methods including Doehlert, Box–Behnken and central composite design (CCD) which are extensively used in different studies (Shaykhi and Zinatizadeh, 2014). For example, Box–Behnken, which provides more information from interaction among the minimum factors, is employed in several environmental studies (Qiu et al., 2016; Wei et al., 2017; Yang et al., 2016; Zhao et al., 2015). Additionally, CCD known as one of the most common methods is used to model the RSM, and also identification and optimization of the independent variables (Shaykhi and Zinatizadeh, 2014).

Up until now, there have been few studies regarding the establishment of flexible fibre biofilm reactors treating MPW (Abdulgader et al., 2009; Yu, Q et al., 2003; Yu, QJ et al., 2003). The aim of current research is to investigate the performance and optimization of a single stage flexible fibre biofilm reactor (SS-FFBR) treating MPW using RSM. This approach can overcome all the problems associated with traditional biofilm systems. The flexible fibres provide a very high surface area for microorganisms to grow on (around 2200 m^2/m^3). Sludge bulking in the conventional biofilm reactor can be eliminated through the intensive motion of the fibre caused by air mixing. The flexible fibre biofilm reactors have the potential as an appropriate substitute system for the conventional processes in the treatment of dairy wastewaters, because of their high effluent quality and efficient performance, robustness, moderate cost, ease of construction, low maintenance and simple operation. The system performance was modelled and analysed using RSM under different influent COD concentrations and HRT where 8 interrelated parameters were assessed as response. Finally, an optimal region of the main parameters was determined using the RSM.

2. Materials and Methods

2.1. Wastewater preparation

The wastewater used in this research was taken from the final collection wastewater basin at National Foods Milk Ltd, Brisbane, Queensland, Australia. The factory uses approximately 400,000 litres of water each day over a six-day operation. The water is predominantly for cleaning and plant operation. An extensive cleaning begins with flushing out the milk residual from tanks and pipes; general working environment, external surfaces of tanks, lines and machinery. All ends up in the wastewater along with cleaning chemicals. Raw wastewater samples were continuously collected in 20 L plastic containers from the factory wastewater effluent stream. After the samples were collected, they were directly transported to the laboratory at Griffith University for initial characterization and sustained in a

refrigerator at 0-4 ºC before being used for further investigation. Typical characteristics of raw milk processing wastewater are given in Table 1.

Table 1. Specifications of raw milk processing wastewater.

COD	4000-14250
BOD	3000-8910
pH	11.70
Total Solids	5790-6380
Total Suspended Solid	1420-3540
Volatile Suspended Solid	1350-3480
Total Nitrogen (as N)	N/A
Total phosphate (as P)	37
Oil and Grease	N/A

N/A=Not Analysed

2.2. Flexible fibre

The configuration of the flexible fibre used in this study is illustrated in Figure 1. The flexible fibre media chosen as the packing material of this study was manufactured from rayon fibre with a specific gravity of 1.02, and a unit weight of 3 kg/m^3. The fibres were bundled and circularly attached to a rope for support at intervals of about 80 mm. The length of the fibre when straightened was about 75 mm and the diameter of the fibre was 0.07 mm. The rope was attached to a support frame and fixed at the centreline of each compartment. The length of the rope used in this experiment was 450 mm. The fibre packing had a specific contact surface area of around 2164 m^2/m^3 of tank volume and a void fraction of more than 99 % (Table 2). This provided a large surface area for bacteria growth and attachment. The arrangement of packing media in the reactor is depicted in Figure 2.

Table 2. Flexible fibre media characteristics (Huang and Hung, 1987).

Parameters	Value
Material	Rayon fibre
Specific weight	1.02
Unit weight (kg/m^3)	3
Void ratio	0.99
Aldehyde content	25%
Anti-pulling strength (g/fibre)	6.8-7.1
Maximum elongation ratio	12%

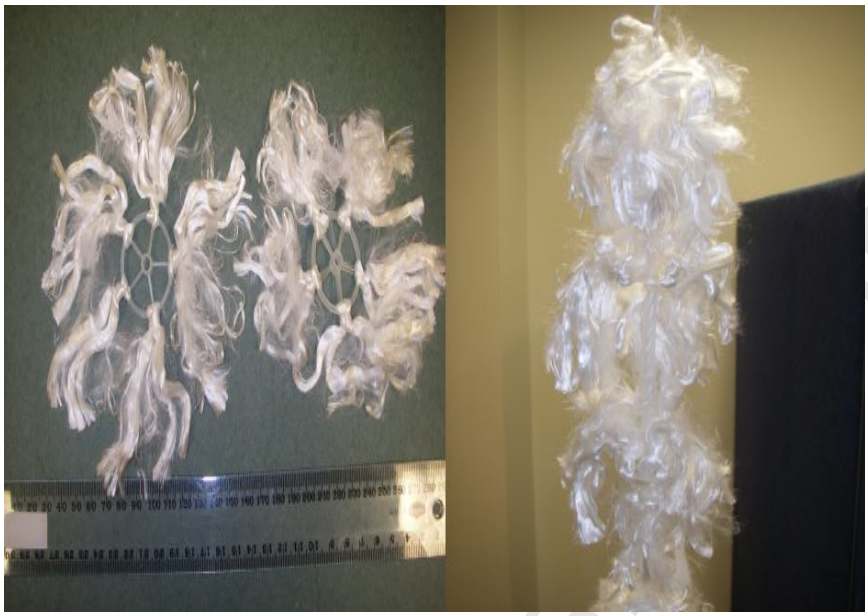


Figure 1. Photo of flexible fibre bundles.

2.3. Bioreactor configuration and start up

In this part of the study, the experiments were conducted in a laboratory-scale single stage flexible fibre biofilm reactor (SS-FFBR). The schematic diagram of the SS-FFBR is illustrated in Figure 3. The square shape SS-FFBR was fabricated using acrylic plastic transparent sheet with a thickness of 6 mm. A transparent malarial was used to build the reactor in order to observe wastewater throughout the experiment. Such material was thick enough to withstand the wastewater working volume pressure on the tank wall. The reactor had an effective working 8 L volume of with a height of 650 mm and width of 125 mm. The aeration was employed using two air diffusers placed at the bottom. The influent port was located at the top of the reactor with a diameter of 20 mm, while the effluent sample port was in the bottom, also with 20 mm diameter. The reactor was provided with a support frame where the flexible fibre was attached. The reactor was supplied with 7 flexible fibre bundles and attached to 450 mm rope length. A peristaltic pump (Cole Parmer, Masterflex, Model

7523-60) with 16 inches diameter Tygon tubing was used for feeding the wastewater into the reactor. The effluent port was connected to a settling tank with a volume of 12.5 L and HRT used in the settling tank were 12.2 h, 18.9 h, and 23 h. The SS-FFBR was initially inoculated with a mixed seed sludge culture in order to start the biofilm development. The reactor was operated in a SBR mode to acclimatize the microorganisms in the reactor. The COD_{in} concentration was gradually increased to the targeted COD concentration. After the acclimatization period, the reactor was run for a couple of days as a SBR mode until the biofilm growth was established, and after that the continuous operation of the reactor was initiated at 12 h HRT and an 11 mL/min feed flow rate. The AFR was not kept constant and varied upon the DO level in the reactor to maintain the DO value at higher than 2 mg/L. The influent pH values were adjusted to the range of 6.5-7.5, and the influent COD concentration were diluted to the desired level by using tap water. The normal investigation of the performance of the SS-FFBR was initiated. During the start-up period, the COD removal, DO, turbidity, and volatile suspended solids (VSS) were also monitored daily. In the first week, the reactor was fed step-wise with an influent COD concentration varying from 500 to 800 mg/L, at a HRT of 12 h.

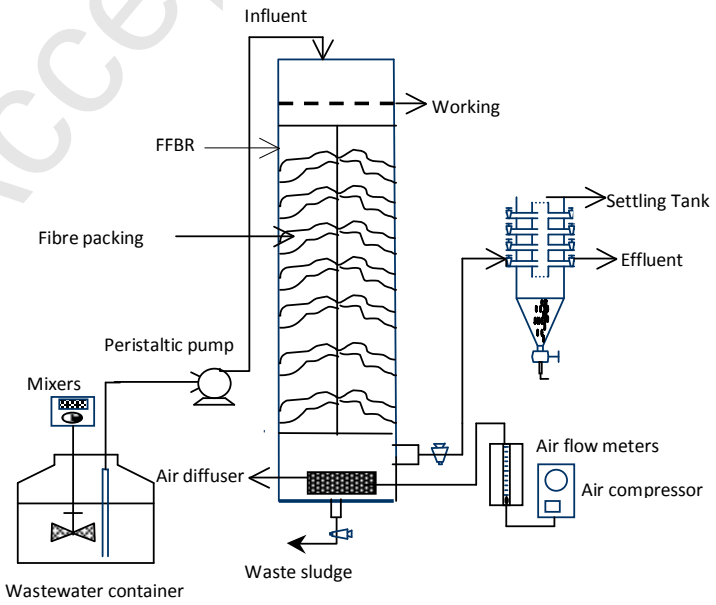


Figure 2. Schematic diagram of SS-FFBR experimental set-up.

2.4. Bioreactor operation

This part of the experiments was conducted to evaluate the effectiveness and performance of a SS-FFBR in the treatment of milk processing wastewater. A 500 mL of a mixture of activated sludge was poured into the reactor. The sludge mixture had a VSS concentration of 4189 mg/L and VSS/SS ratio of 0.79. Initially, the reactor was batch-fed daily with diluted milk processing wastewater, starting with a COD concentration of 500 mg/L and the concentration increased gradually to an average concentration of 2417 mg COD/L. This step took about 7 days till the biofilm was developed on the fibre. Continuous experiments were started by feeding the reactor with an initial influent COD concentration of 2400 mg/L corresponding to an OLR of 3.6 g/L.d and a HRT of 16 h. Different dilutions of raw milk processing wastewater were prepared using tap water. The pH of the feed was kept constant at 7 using diluted HCL and NaOH solutions. In this set of experiments, the steady state condition was not reached till after 20 days due to interruption occurring in the reactor. To investigate the effectiveness of the single stage reactor and the effect of OLR on the system performance, the HRT and wastewater flow rate were retained constant at 16 h, and 12.96 L/d; the influent COD concentration was changed between 800 mg/L and 4000 mg/L. In addition, the reactor was operated at HRTs of 12, and 8h and the flow rate of wastewater was kept controlled at 15.84 L/d, 24.48 L/d, respectively. The wastewater was fed with COD concentrations of 800 mg/L, 2400 mg/L, and 4000 mg/L. The biomass concentration in the reactor was evaluated in every experimental run. Throughout the experiments, the influent pH was monitored and kept at 7. The air flow rate of all experiments was adjusted based on keeping the level of dissolved oxygen concentration > 2 mg/L in the reactor. The temperature of the reactor was maintained at room temperature.

Suitable inoculums were prepared by acclimatizing the seed with wastewater. Activated sludge samples were used for acclimatization. The activated sludge sample was collected from Logan City Council Wastewater Treatment Plant in Loganholme and Brisbane City Council Wastewater Treatment Plant at Oxley Creek. In both locations, the raw sewage is mainly domestic in origin. The collected sludge from both plants was tested in the laboratory. The total suspended solid was more than 5000 mg/L and SV30 was 180 ml/L. The sludge volume index (SVI) value was accounted to be 77 g/L.

2.5. Experimental design

The interactive effects of HRT of the bioreactor and influent COD concentration on the responses were described using the one variable at a time technique, 9 continuous experiments on SS-FFBR were conducted at HRTs of 8, 12 and 16 h, chosen based on literature (Yu, Q et al., 2003; Yu, QJ et al., 2003), each with the influent COD concentration was 800, 2400, 4000 mg/L. For this experimental design, both HRT and influent COD concentration were considered as independent variables. The parameters that were considered as dependent output responses are TCOD removal efficiency, TCOD removal rate, SRT, turbidity, specific substrate utilization rate (U), and VSS/TSS ratio.

After experiments were completed, the coefficients of the polynomial model were obtained using equation below (Khuri and Cornell, 1996; Zinatizadeh et al., 2009).

$$Y = \beta_0 + \beta_i X_i + \beta_j X_j + \beta_{ii} X_i^{\,2} + \beta_{jj} X_j^{\,2} + \beta_{ij} X_i X_j + \ldots \qquad (1)$$

where i and j are the linear and quadratic coefficients, respectively, and β is the regression coefficient. The experimental data were analysed and processed for Eq. 1 using Design Expert software (Stat-Ease, Version 6.0.6, Inc., Minneapolis, MN) including analysis of variance (ANOVA), which is suitable for experimental design. The coefficient of determination R^2, adjusted R^2 and adequate precision were employed to evaluate the fit

quality of the polynomial model equation. Model terms are chosen or rejected according to the probability of error (P) value with 95% confidence level. The polynomial equation fitted was expressed as 3D surface plots to visualize the relationship between the responses and the experimental level of each factor utilized in the design. The optimum region is also identified based on the main parameters in the overlay plot. The analysis of the data was accomplished by using the general factorial design of response surface methodology (RSM). The experimental conditions and results are displayed in Table 3. The results were analysed using analysis of variance (ANOVA), carried out by Design Expert Software (version 6.0, State-Ease, Inc., Minneapolis, MN) (Table 4).

Table 3. Experimental conditions and results investigated in this study.

Run	Variable		Response							
	Factor1 A:HRT (h)	Factor2 B:COD_{in} (mg/L)	TCOD removal (%)	TCOD removal rate (g/L.d)	TSS removal, (%)	Specific substrate utilization rate (U), (Q(S0-S)/X.V), g CODrem/g VSS.L.d	SRT (d)	VSS/TSS ratio of biomass (Suspended + Attached) Average	Effluent pH	Effluent Turb, NTU
1	8	2480	90.3	6.72	90.2	0.987	38	0.91	7.73	36.25
2	12	2477	96.6	4.78	93.0	0.772	86	0.92	7.99	6.01
3	12	4010	91	7.31	92.2	0.975	43.7	0.93	7.5	41.6
4	8	836	95.0	2.38	91.4	0.41	76.8	0.93	7.58	5.85
5	16	3750	94.5	5.32	93.6	0.94	104.8	0.91	8.02	28.4
6	12	830	94.9	1.57	93.1	0.218	196.4	0.956	7.5	2.73
7	16	2266	95	3.20	93.5	0.582	91.8	0.94	7.9	18.25
8	16	763.3	94	1.07	96.7	0.213	78.5	0.929	7.65	4.64
9	8	3921.8	89.9	10.5	89.7	1.42	14.9	0.95	7.80	44.6

Table 4. ANOVA results for the equations of the Design Expert 6.0.6 for studied responses.

RESPONSE	TRANSFORMATION	MODIFIED EQUATIONS WITH SIGNIFICANT TERMS	PROBABILITY	R^2	ADJ. R^2	ADEQ. PRECISION	SD	CV	PRESS	PROBABILITY FOR LACK OF FIT
COD removal	-	93.85 +1.48A -1.42B +1.42AB	0.009	0.7073	0.6098	8.478	1.23	1.31	38.86	0.0269
Specific substrate Utilization rate (U)	-	0.75-0.18A+0.42B+0.08 A^2 -0.11 B^2 -0.071AB	<0.0001	0.9843	0.9732	32.421	0.054	7.34	0.15	0.0269
VSS/TSS ratio	-	0.93+2.833E-3B-5.88E-3B^2-5.5E-3AB	<0.0006	0.8410	0.7880	12.370	2.4E-3	0.26	2.17E-4	0.0628
Effluent Turbidity	Base 10 log	0.87+ 0.48B + 0.35A^2	<0.0001	0.8482	0.8179	15.294	0.18	17.43	0.58	0.0295
SRT	Base 10 log	1.95 + 0.23A - 0.21B - 0.17A^2 +0.2AB	<0.0001	0.9416	0.9123	20.40	0.081	4.34	0.35	-

A: first variable, HRT (h), B: second variable, influent COD (mg/L), R^2: determination coefficient, Adj. R^2: adjusted R^2, Adeq. Precision: Adequate precision, SD: standard deviation, CV: coefficient of variation, PRESS: predicted residual error sum of square

2.6. Analytical methods

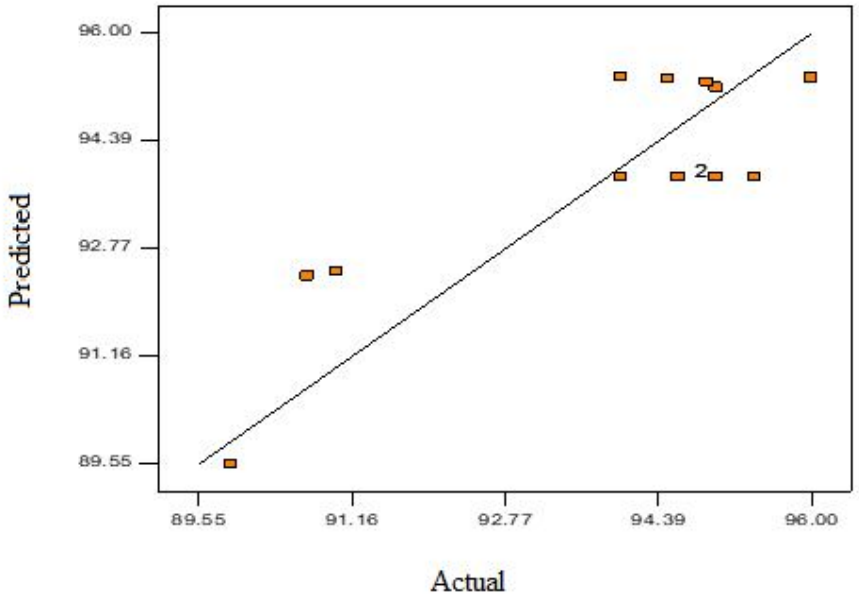
Influent and effluent samples were analysed in each experiment. The parameters analysed were pH, DO, COD, BOD, TSS, VSS, Turbidity and biomass concentration. The analytical testing procedures were in accordance with the standard APHA methods (APHA, 1995). In addition, mixed liquor biomass concentration, and wasted sludge, as well as attached biomass were determined. The pH was measured using a pH meter model 90-FL supplied by TPS Pty Ltd (Australia). Dissolved oxygen (DO) measurement in the aeration tank was monitored daily with a DO probe (YSI Model: 5010). The digital DO meter model YSI/5000 was provided from the YSI Company, USA. The DO concentration was maintained between 3-6 mg/L for all experiments conducted by adjusting the airflow rate. The air flow rates were controlled by an air flow meter model Porter F665-AV1, Parker Hannifin Coporation, USA, and the range of the flow rates operated in these experiments was varied between 0.250-15 L/min. Turbidity was determined by using a turbidity meter model 2100 A, supplied by (HACH Company, USA), based on the nephelometric method, and the results of the measurements were reported as nephelometric turbidity units (NTU).

3. Results and discussion

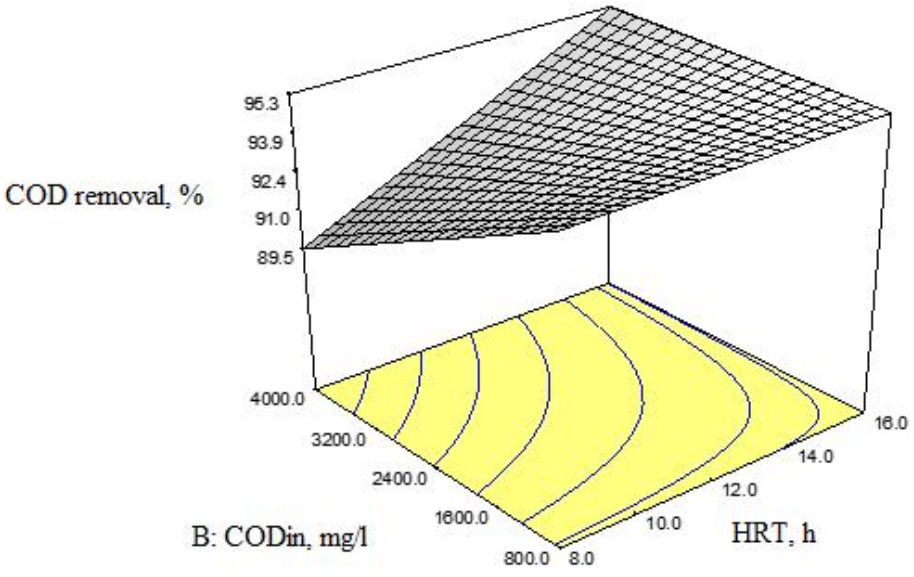
3.1. Effects of influent COD concentration and HRT on COD Removal and COD Removal Efficiency

The response surface of COD removal efficiency was described by two reduced quadratic models within the range of the factors (Table 4). Where, A and B are referred to as HRT and COD_{in}, respectively. Diagnostic plots such as the anticipated versus actual values aid to judge the model validity. The correlation between the experimental and simulated data of the COD removal efficiency presented in Figure 3a with a value of R^2=0.70, that indicates a relatively low agreement between the real data and those acquired from the models. Furthermore,

adequate precision (AP) value obtained was more than 4 (Table 4) for COD removal efficiency, which confirms the predicted model can be used to navigate the design space defied by the central composite design (CCD). For graphical interpretation of the interaction, the use of the three-dimensional plots of the regression model is highly recommended. Figure 3b presents the concurrent effects of the two independent variables on the COD removal efficiency. As shown in the response surface plot, the response increases by increasing the HRT at higher COD_{in}, whereas the response remained constant by increasing HRT at the lowest COD_{in}, indicating that minimum HRT was sufficient. The interaction verified that the HRT and COD_{in} played a significant role in the COD removal efficiency during the process. The lowest COD removal efficiency of 89.9% was gained by the SS-FFBR at the highest COD_{in} concentration (corresponding to 11.7 kg COD/m^3.d OLR) at lowest HRT of 8 h. This result is comparable with those results (COD removal efficiency 96.3 %) obtained by Rodgers et al. (2006) using a horizontal flow biofilm reactor to treat synthetic dairy wastewater at a lower OLR of 2.3 gCOD/m^2.d and longer 36 h HRT. In another study, Najafpour et al. (Najafpour et al., 2006) treated food cannery wastewater using a three stage RBC, and achieved only 93.7% of COD removal efficiency at long HRT of 40 h. An 82.11% COD removal efficiency was obtained at maximum OLR of 10.2 kg COD/m^3.d when dairy wastewater was treated using an aerobic fluidized bed biofilm reactor (FBBR) (Resmi and Gopalakrishna, 2004). The good performance of SS-FFBR might be in relation to the higher amount of biomass accumulated in the reactor owing to large surface area provided by the flexible fibre, and that may increase the substrate consumption in the bioreactor. The research group of Mansouri et al., achieved COD removal of 93.5 % at retention time of 6.5 h and COD_{in} of 1000 mg/l (Mansouri and Zinatizadeh, 2017). In another study, the COD removal of 99.5 % was obtained in a lab-scale sequencing batch biofilm reactor (SBBR) at 18 h and 5000 mg/l as COD_{in} (Zinatizadeh et al., 2011).



(a)

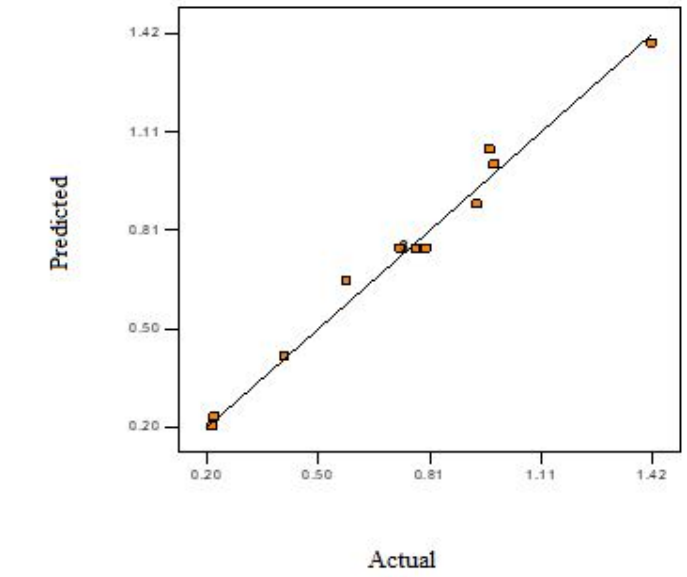


(b)

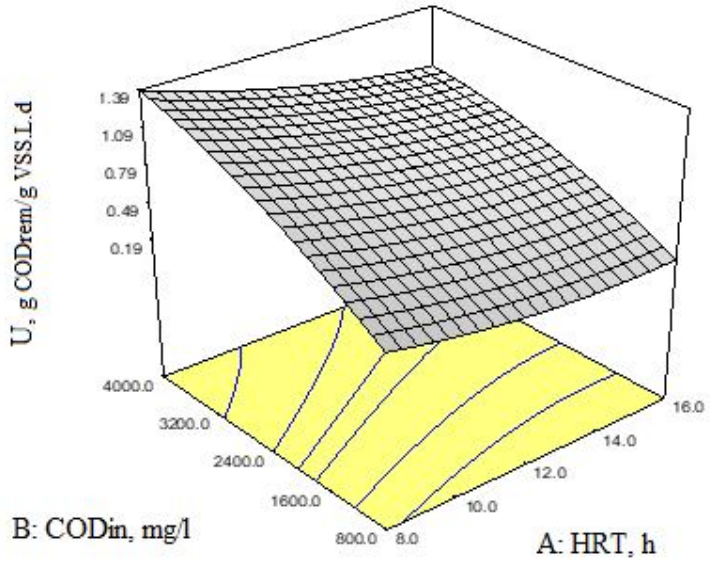
Figure 3. (a) Actual versus predicted values of COD removal, (b) Response surface plot for COD removal.

3.2. Effect of Influent COD Concentration and HRT on Specific Substrate Utilization Rate (U)

The regression equation of specific substrate utilization rate as an empirical model in terms of coded factors is represented in Table 3. Where, U is specific substrate utilization rate, A is HRT and B is COD_{in}. A high correlation was observed between actual and predicted values of specific substrate utilization rate (U) with R^2=0.984 as shown in Figure 4a. This confirms the adequacy of the equations of model to anticipate values for U. The observed points on this plot demonstrate that the actual values are distributed fairly close to the straight line and show a significant correlation. Figure 4b depicts the variation of U as a function of the two variable factors, HRT and influent COD concentration. The Figure shows that a significant mutual interaction occurs between the variables on specific substrate utilization rate as a response. As a general trend, the specific substrate utilization rate (U) increased with increase in COD_{in} and decrease in HRT due to the increase in OLR and biological activity of the microbial population. The maximum value of U was modelled to be 1.39 $gCOD_{rem}$/gVSS.L.d, whereas the actual value was 1.42 $gCOD_{rem}$/gVSS.L.d. However, the modelled minimum value of U was 0.19 $gCOD_{rem}$/gVSS.L.d while the actual value was 0.213 $gCOD_{rem}$/gVSS.L.d. This condition was obtained at 16 h HRT and low COD concentration. Hamoda (1989) found a similar observation of increase in U with the increase of substrate concentration. It can be observed that as a higher COD is applied to a reactor, the higher specific substrate utilization rate is obtained. This indicates that the influent COD concentration applied to the reactor has not achieved its limiting value and shows the ability of the process to treat a higher range of COD concentration. Sonwani et al., in their work obtained U_{max} of 0.769 g/L.d in an HRT of 5 d using a moving bed bioreactor (MBBR) treating the naphthalene contained wastewater (Sonwani et al., 2019).



(a)



(b)

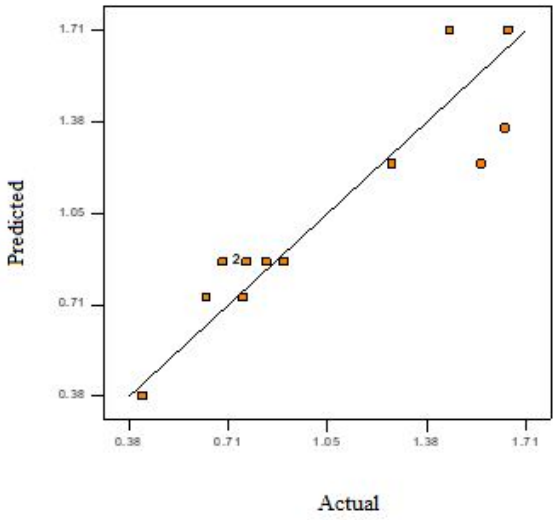
Figure 4. (a) Actual versus predicted values of specific substrate utilization rate, (b) Response surface plot for specific substrate utilization rate.

3.3. Effects of HRT and CODin on effluent turbidity

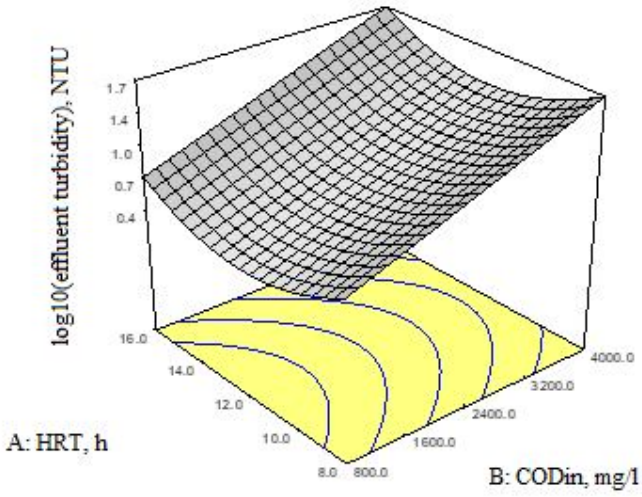
For modelling the interactive effects of the variables viz. HRT and COD_{in}, on the effluent turbidity as a process response, from Table 4, the quadratic model is obtained as the empirical model (built with codified factors). Figure 5a demonstrates a high agreement (R^2=0.8482)

between the experimental and modelled data, indicating that 15.18% of the total variation is not explained by the model. The observed points on this plot indicate that the actual value is distributed away from the straight line. The regression model was performed after transforming the actual raw data to a function of log base 10, as error is a function of the magnitude of the responses. Figure 5b presents the simultaneous effects of the variables on the effluent turbidity. It is obvious from the figure that the effluent turbidity has a proportional increase when the influent COD_{in} increased and HRT decreased. This might be attributed to less integrity in the microbial biofilm formed. For example, at the highest COD_{in} of 4000 mg/L, the effluent turbidity declined from 1.649 to 1.45 as the HRT increased from 8 h to 16 h. This was expected due to the increase in the biomass growth rate and the washed out sludge, especially with increase in the influent flow rate. It can be noted from the figure that the maximum value of the effluent turbidity was 1.649 at the higher OLR, corresponded to HRT 8h and COD 3921.8 mg/L, whereas the lowest effluent turbidity quantity was 0.43 which was measured at an HRT of 12 h and COD_{in} of 830 mg/L. At the lower HRT value, a high effluent turbidity was obtained, and this was also affected by the amount of the COD applied to the system. The decrease of the effluent turbidity at 12 h HRT and COD_{in} of 800 and 2400 mg/L was noticeably lower (0.43 and 0.77) than the values obtained at HRT 8 h and 16 h. This observation is somewhat in agreement with the findings by Najafpour et al. (Najafpour et al., 2006) for a RBC reactor. Such a low effluent turbidity may be due to a lower range of OLR than the previous run that operated in a similar range of OLR. It suggests that due to low influent TSS contents, which microorganisms can easily degrade, low effluent turbidity is resulted. The low level of turbidity expresses the reduced amount of sloughed biomass and the sludge had good settling property in the settling tank. At a low HRT of 8 h, the reactor effluent turbidity gradually increased from 0.76 to 1.649, which may be due to the increase in the TSS concentration of the influent stream and increase in the OLR that may

have an effect on the attached biomass. Despite some variation of the effluent turbidity, particularly at the HRT of 12 h, it can be suggested that HRT is a significant and effective variable amongst the operated variables.



(a)



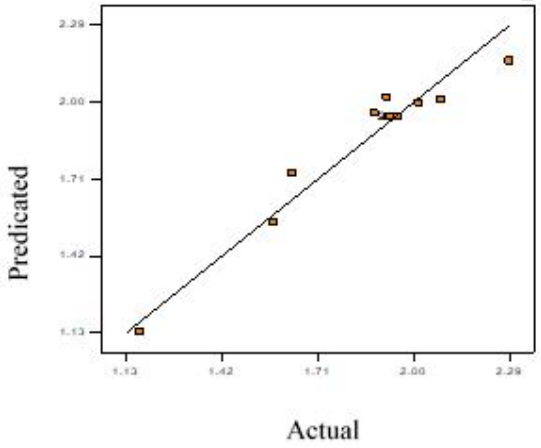
(b)

Figure 5. (a) Actual versus predicted values of turbidity, (b) Response surface plot for turbidity.

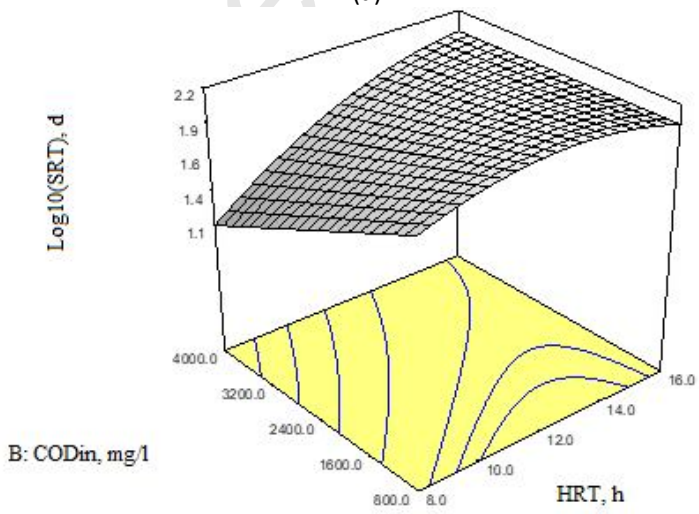
3.3. Effect of HRT and COD_{in} on SRT

SRT, introduced as a significant parameter to control process, was evaluated by measuring VSS inside the reactor and in the effluent at different COD_{in} concentrations. The high SRT values demonstrate the effective role of packing media on the process stability due to carrying a high amount of biomass, which allows the microorganisms to resist and survive with changes in OLR. The SRT influences the growth of various microorganism's kinds as well as the extent to which different reactions will take place. In order to achieve a desirable SRT in the reactor, the rate of sludge wastage must be controlled at a desired value. Generally, undesired accumulation of biomass results in different destructive effects depending on the type of application. For example, some challenges observed in gas bio-filtration; clogging, channeling, extreme head loss, and reduction in performance; are related to excess sludge accumulation (Yang et al., 2010). For modelling the interactive effects of the variables (COD_{in} and HRT) on the process control responses, SRT, from Table 3 the quadratic model (built with codified factors) was obtained. Figure 6a shows good agreement (R^2=0.941) between the experimental and predicted data of SRT. Figure 6b depicts the effect of the two operating variables on SRT. The main effect of HRT (A) and COD_{in} (B) were significant model terms. It should be stated that the ratio of the maximum to minimum SRT was about 13. Therefore, a logarithmic function with base 10 was applied to the SRT model. As can be seen from the 3D figure, the effect of increase in HRT on increase in SRT was more at the higher values of the COD_{in}, implying more effect of HRT relative to COD_{in} at higher values of COD_{in}. In order to achieve a high and long SRT, whilst maintaining a short level of HRT, it is necessary to ensure that the biomass in the reactor does not wash out in the effluent. In the SS-FFBR, the provided packing media helps to achieve a high SRT as biofilm attachment on the packing media. In this study, the minimum and the maximum value of SRT

were evaluated based on a logarithmic function with base 10 applied in order to fit the data. This curve shows that at HRT of 8 h and COD concentration of 3921 mg/L the reactor achieved a lowest SRT value of 1.17 day. However, the highest SRT (2.29 day) achieved lower COD concentration of 830 at 12 h HRT. A general decrease on the SRT at low HRT was expected and that was due to the increase in the feed flow rate. Zinatizadeh et al. (2006) observed a similar trend when treated Palm Oil Mill Effluent (POME) wastewater using an UASB reactor.



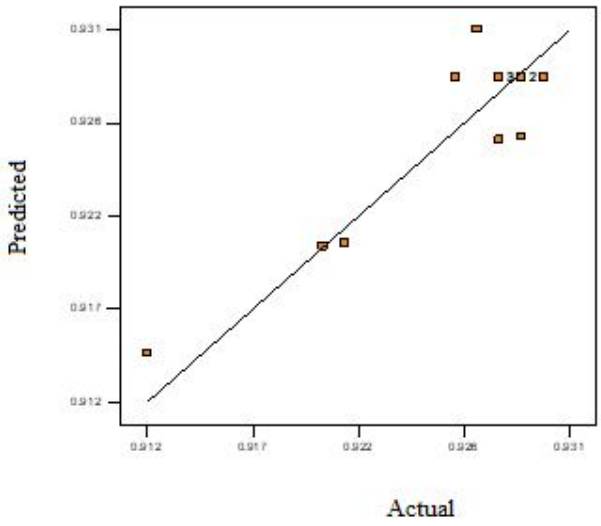
(a)



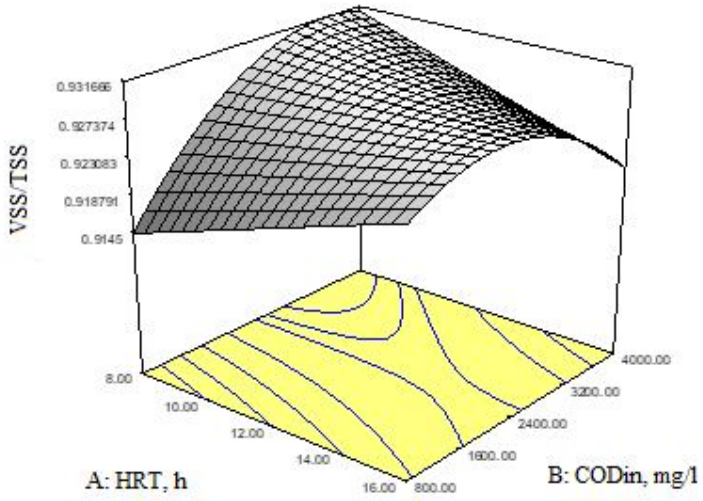
(b)

Figure 6. (a) Actual versus predicted values of $\log_{10}$ (SRT), (b) Response Surface plot for SRT.

The ratio of VSS to TSS concentration (VSS/TSS) in a bioreactor indicates the organic component in the solids content. Figure 7a exhibits the anticipated versus the actual values for VSS/TSS ratio with a correlation coefficient of R^2=0.841. Figure 7b demonstrates 3D plots of the model for VSS/TSS ratio as COD_{in} and HRT. The figure indicates that the ratio of VSS/TSS sharply dropped as the COD_{in} concentration decreased to 800 mg/L and with increase the in HRT. The typical value of the VSS/TSS ratio should be in the range 0.6-0.8, whereas in this study the ratio was too high and was in the range of 0.91-0.956. This value was beyond the typical value recommended, and this higher value of VSS/TSS ratio indicates the high fraction of organic matter in influent total suspended solids. The COD_{in} was the most effective variable. Hu and Liu, (2002) obtained a similar ratio (0.93) when they treated a synthetic wastewater.



(a)



(b)

Figure 7. (a) Predicted versus actual values for VSS/TS, (b) Response surface plots for VSS/TSS ratio.

3.5. Process optimization analysis

With multiple responses, the optimum condition in which all parameters concurrently meet the proposed removal criteria could be inspected visually by superimposing the contours graphs of the response surface in an overlay plot. Graphical optimization denotes the area of desirable response values in the factor space. Figure 8 displays the graphical optimization, which shows the area of feasible response values (shaded area) in the factors space. The optimum condition was identified by taking into account COD removal, SRT and substrate utilization rate (U). The shaded area in Figure 8 indicates the optimum conditions for reactor performance. The region that satisfies the constraints is in yellow color, whereas the region which does not meet the criteria is in gray color. And the blue lines and red dots show the boundary conditions and different runs, respectively. The optimum removal was obtained at a HRT of 8 h and COD_{in} 3922 mg/L, corresponded to high OLR of 11.67 kg COD/m^3.d. The results implied the raw milk processing wastewater could be well treated at a high OLR and

Low HRT as indicated above. The results also showed a high treatment capacity of the SS-FFBR.

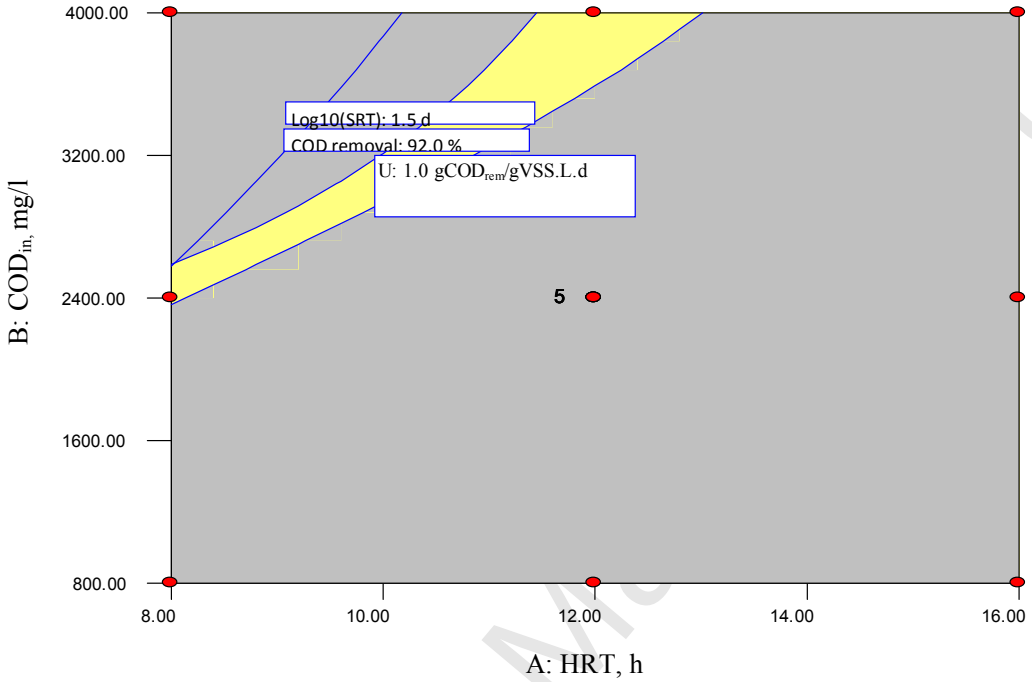


Figure 8. Overlay plot for optimal region.

(Yellow district satisfies the constraints; Red dots are design points; Blue lines are boundary conditions.)

4. Conclusions

Lab scale flexible fibre biofilm reactor, SS-FFBR, was successfully developed, fabricated and operated for effective milk processing wastewater treatment. Response surface methodology (RSM) was successfully applied to model and analyse the process as well as determination of the optimum operating conditions. The interactive effects of two studied variables, HRT and COD_{in}, on the process performance were extensively evaluated. The removal efficiency of the COD was incremented with increasing the HRT and decreasing the COD_{in}. A significant mutual interaction observed between the variables and other responses. By applying RSM, the optimum removal was obtained at a HRT of 8 h and COD_{in} 3922 mg/L, corresponded to high OLR of 11.67 kg COD/m^3. d. The experimental findings indicated good agreement with model prediction.

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Highlights

- SS-FFBR was an effective design in treatment of milk processing wastewater.
- The biological treatment process was successfully modelled and optimized using RSM.
- The experimental findings were in good agreement with model prediction.