



## Investigate the simultaneous effect of pH, temperature, and hydraulic retention time in moving bed biofilm reactor: optimization and modeling using response surface methodology

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### ABSTRACT

In recent years, the application of moving bed biofilm reactor (MBBR) in the biological treatment of municipal and industrial wastewater has been developed. Herein, following the construction of the MBBR reactor, the effect of three important parameters of temperature, hydraulic retention time (HRT), and pH simultaneously on the MBBR efficiency was investigated. The effluent wastewater of the anaerobic unit of Kanyar Dairy Company located in Quchan – Iran was treated as the model wastewater. Response surface methodology was applied to design the experiments of system operating conditions. The analysis of variance was applied to determine the most important parameters affecting the performance of the MBBR reactor in order to increase the chemical oxygen demand (COD) removal percentage. The amount of COD was measured as the process response. Based on the results and the analysis of variance, the response of the experiments was in accordance with the predicted model. The *p*-value in all the three parameters of temperature, HRT, and pH was less than 0.05, therefore making them important while having a significant effect on the performance of the reactor. Moreover, only the interaction of HRT and pH affects the performance of the reactor and should be controlled. Under the optimal conditions (pH = 7, temperature = 27°C, and HRT = 11 h), COD removal efficiency reached 93%. According to the results, an optimized MBBR is a good option for dairy wastewater treatment.

**Keywords:** Analysis of variance; Chemical oxygen demand removal efficiency; Dairy wastewater; Moving bed biofilm reactor; Optimization

### 1. Introduction

The wastewater of the dairy industry contains a high level of soluble and insoluble fats, carbohydrates, and proteins [1], thereby resulting in a high level of chemical oxygen demand (COD). The quality of dairy industry wastewater is quite different and dependent on the types of products. Thus, constructing wastewater treatment plants in these factories is inevitable and necessary [2].

Biological treatment is the most effective method to remove the biodegradable organic pollutants from industrial wastewaters using anaerobic and aerobic techniques. Studies have revealed the pollutants removal process in this method with a more economic way [3,4].

Today, different solutions have been proposed to optimize the performance of wastewater treatment methods; for instance, it has been stated that biofilm the nitrification–anaerobic ammonium oxidation (ANAMMOX) process to

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be present and having degradation possibilities, for biogas effluent treatment, using the oxidation–reduction potential method to improve the performance of the nitrogen-rich wastewaters treatment process [5] or use ANAMMOX-denitrification biomass in a microbial fuel cell to enhance electricity generation and nitrogen removal efficiency [6].

Aerobic treatment is the last stage of COD and biochemical oxygen demand removal, in which the effluent from anaerobic units enters [7]. The most important aerobic units are the active sludge, rotating biological contactors (RBC), membrane bioreactor (MBR), sequencing batch reactors (SBR), integrated fixed-film activated sludge (IFAS), and moving bed biofilm reactor (MBBR) [8–10].

The process of Kaldnes moving bed, known as the bio-reactors with moving bed, is developed by Kaldnes Company with the collaboration of SINTEF Research Center in 1987, patented in the European Innovation Center [11,12]. Over time, modifications were made to these reactors. Over the last recent years, research has been done on moving bed and attached growth modification in order to increase the efficiency of MBBR and minimize its disadvantages [13–15].

MBBRs have high efficiency in removing organic compounds [16,17]. The benefits of this system include being compact, using all the space of the reactor, the possibility of enough growth of biomass, the impossibility of system blockage, cost-effectiveness; additionally, it requires little investment close to the activated sludge process, costs less than other aerobic methods, and there is no need to return the sludge in this system [18–20].

The MBBR process is an ideal priority for simultaneous nitrification and denitrification attributing to the longer sludge age and aerobic/anoxic microenvironment along with biofilm [21]. The heterotrophic denitrification, as one of the main reactions in BNR, can also be used together in the A/O/A system with Fenton reaction as the biological stage [22].

In the past years, the usage of biological treatment systems as the moving bed bio-reactors has been reported for the treatment of municipal and industrial wastewaters, for instance, the production of paper and cardboard pulp, dairy products, slaughterhouses, and chemical industry [23–25].

The optimization of the biodegradation process could increase the efficiency of contaminant removal because it can be affected by environmental conditions. There are various methods to optimize these systems, one of the most important ones would be the design of the experiment [26–29].

The main problem of the old optimization methods, such as “single factor” is a large number of experiments and the failure to study the interactions between variables, thus, optimization using multivariate statistical methods [30] is more desirable. Among several methods, response surface methodology (RSM) is the most suitable one to study the interactions between parameters and determine the optimal condition [31]. The most common RSM methods are central composite design (CCD) and Box–Behnken [32].

In this paper, we used an MBBR in Kanyar Dairy Company (Quchan – Iran) to treat the effluent wastewater from an anaerobic filter to investigate and optimize the pH, temperature, and hydraulic retention time (HRT) of MBBR. Due to the high variety of products in Kanyar Dairy, this

effluent had unique properties, such as high pH changes and high COD.

The pH, temperature, and HRT are the most important operating conditions in biological wastewater treatment. Careful control of operating conditions increases the efficiency of removing organic pollutants from the effluent and reduces treatment costs.

So far, a lot of research has been done on offering various methods to reduce the cost of wastewater treatment [33,34]. In the current work, the MBBR was used to reduce the cost of wastewater treatment which, as mentioned before, has a lower start-up and operating cost than other aerobic methods. In addition, optimizing the operating conditions of working with this reactor will raise the efficiency of this reactor without increasing the cost of wastewater treatment. Moreover, if these three variables are optimized, the amount of COD reduction will rise significantly. To date, in none of the studies on the optimization of operational variables in the MBBR, the simultaneous effects of the main three, pH, temperature, and HRT, have not been investigated [35–38].

Herein, for the first time, the effect of operational variables (HRT, pH, and temperature) on Kanyar Dairy wastewater treatment using MBBR was investigated simultaneously, being modeled and optimized by RSM and central composite design. Due to the type of effluent and the optimization of three important operational variables that significantly reduce COD, these data are unique.

## 2. Materials and methods

### 2.1. Experimental set-up

In this study, a MBBR was utilized with an effective volume of 13.275 L and an external dimension of the length of 36 cm, a width of 15 cm, and a height of 33 cm. It was made up of plexiglass with a wall thickness of 4 mm. Moreover, 40% of the reactor was filled with plastic flexible corrugated conduit pipe hose media with a specific surface area of 500 m<sup>2</sup>/m<sup>3</sup>, a height of 2.5 cm, a diameter of 1.5 cm, and a density of 0.96 g/cm<sup>3</sup>.

The required air to provide the dissolved oxygen and circulate the biofilm carriers inside the bio-reactor was supplied by an air compressor, HEALIA model, made in China and three air dispensers installed at the bottom of the bio-reactor. To keep the carriers in the bio-reactors, a sieve (with an opening of 5 mm) was placed at the outlet of the bio-reactor. The volume of the air by a flow valve is controlled to provide suitable dissolved oxygen. This reactor is equipped with a primary feed tank to control the temperature and pH. As previously mentioned, flexible corrugated conduit pipe hose was used as media in this reactor due to the reduction of treatment costs, in addition, the large size of this media has caused more growth of microorganisms in this medium. A feed tank for precise control of the temperature and pH was also installed before MBBR; thus, the operating conditions could be easily controlled before the effluent enters the MBBR, which can be said to be the distinguishing feature of this system [35,39,40].

The flow rate was controlled through the use of a peristaltic pump DLSMA made in Italy. The temperature

sensor was installed inside the feed tank to detect and control wastewater temperature. Fig. 1 presents the schematic of the feed tank, MBBR, and the settling tank.

## 2.2. Material

The materials used in this investigation are as follows:

Raw wastewater (the effluent from the anaerobic filter reactor (AFR)) was obtained from Kanyar Dairy Company located in Quchan – Iran. Sulfuric acid 98% and ammonium bicarbonate were purchased from Merck Company. Sodium hydroxide 98.5% was obtained from Arax Shimi (Iran).

## 2.3. Wastewater characteristics

The raw wastewater was collected from the outlet of the AFR of Kanyar Dairy Company located in Quchan – Iran. Characteristics of the wastewater are listed in Table 1.

## 2.4. Analytical methods

An MBBR pilot was installed after the AFR at Kanyar Dairy Company, Quchan – Iran. The MBBR was then seeded with the waste sludge from an active sludge of the aerobic unit of the plant's treatment, with which 50% of the reactor volume was filled. Subsequently, the remaining space was filled with the effluent wastewater from an anaerobic filter reactor. The effluent was diluted with water to obtain the desired amount of COD. At first, the reactor was operating discontinuously with effluent wastewater from an anaerobic filter reactor with COD = 200 mg/L with the COD:N:P ratio of 100:50:15 to adapt and grow microorganisms for a month. Ammonium bicarbonate was used as a nitrogen source at this stage. Thus, the system is operated continuously with the organic loading rate (OLR) of 1 kg COD/m<sup>3</sup> d (in many research, the OLR unit is expressed as kg COD/m<sup>3</sup> d, but multiplying the number of OLR by the number of a

specific surface area of media can also express the value of OLR as 2 g COD/m<sup>2</sup> media d). Effluent wastewater from an anaerobic filter reactor with the characteristics mentioned is represented in Table 1. The amount of air in the reactor was measured between 4–5 mg/L in the MBBR during the experiment via the Winkler method [41]. The wastewater enters the bio-reactor from the feed tank continuously due to gravity. The present study aimed to investigate the effect of the temperature, HRT, and pH on the MBBR performance and define the optimum conditions of operation. The temperature varied between of 24°C, 28°C, and 32°C using a heater element installed in the feed tank equipped with a temperature sensor. To ensure a constant temperature in the reactor, the inlet temperature and flow rate were measured per hour. The pH was measured via a pH meter during the examination and was adjusted using acid and alkali in a feed tank in the three pH levels of 6, 7, and 8. The HRT was adjusted in the range of 5, 10, and 15 h. It varied due to controlling the inlet and outlet flow rate to the MBBR in the three ranges of 0.11, 0.05, and 0.036 l/min. Each level of independent parameters in the reactor was adjusted while the reactor operated in the new conditions until it reached a steady-state (its characteristics were examined three times each day to ensure the conditions remain stable in those conditions). Furthermore, the effluent was taken from the outlet valve

Table 1  
Characteristics of Kanyar dairy wastewater

Test	Result
Temperature, °C	25
pH	7
COD, mg/L	997
TSS, mg/L	165
TDS, mg/L	237
Turbidity, NTU	274

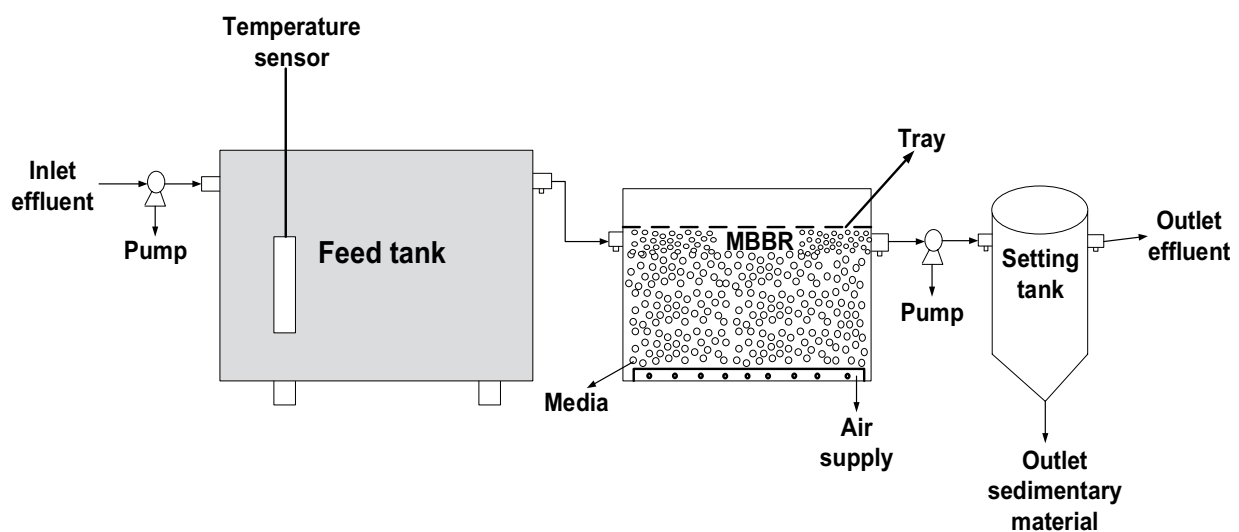


Fig. 1. The schematic of the feed tank, MBBR, and settling tank setup for the Kanyar dairy wastewater treatment.

three times at regular intervals and the average values of the samples were used for interpretation and analysis [36].

The COD of the wastewater (before and after biotreatment) was measured spectrophotometrically (spectrophotometer AAS 240 – German). The pH was measured with the pH meter TES1381 made in Taiwan. A German-made AL125 reactor was used to heat the COD vial.

2.5. Statistical analysis of data

To design the experiments and to analyze the data statistically, Design-Expert Version 11 software was employed. To design the experiments via Design-Expert software, the response surface model was utilized with the central composite method which was quadratic. The temperature, HRT, and pH were considered as the effective factors in the performance efficiency of the MBBR for their optimization. In dairy wastewater treatment modeling and optimization, COD removal efficiency was considered as a response to indicate the performance of the MBBR reactor.

3. Results and discussion

3.1. COD removal from dairy wastewater modeling

The second-order model was used to create a relationship between the variables and their interactions on the response (COD removal efficiency) of the treatment process. Eq. (1) was applied to establish a relationship between the variables and their interactions on the response with the regression analysis. The response of the treatment process is the percentage of COD removal.

$$R = 85.77 + 17.4A + 4.4B + 9C - 0.32A \times B - 2.63A \times C - 1.88B \times C - 7.61A^2 - 11.61B^2 - 24.61C^2 \tag{1}$$

Following the analysis of variance (ANOVA) and the removal of parameters with no effect on the response, Eq. (1) is changed to Eq. (2):

$$R = 85.77 + 17.4A + 4.4B + 9C - 2.63A \times C - 7.61A^2 - 11.61B^2 - 24.61C^2 \tag{2}$$

In Eqs. (1) and (2) *R* represents the percentage of COD removal (%), *A*, *B*, and *C* indicate HRT (h), temperature (°C), and pH, respectively.

A positive sign of factor coefficients will state a direct relationship between the variable and the response while a negative sign will hurt the response. Thus, increasing the variables, *A*, *B*, and *C* will increase the system response whereas the interaction between *A* and *C* will have the opposite effect on the system response.

Fig. 2 represents the result of the analysis of variance, displaying the interaction between two variables (*A* and *B*). According to Fig. 2, increasing HRT and decreasing the pH leads to a rise in the percentage of COD removal in the reactor. In general, any simultaneous increase or decrease of pH and HRT has a direct effect on the performance of the MBBR. The pH is closely related to alkalinity. Thus, the higher the simple alkalinity, the higher the concentration of hydroxides and carbonates; accordingly, the higher the pH, the more alkaline the solution is. Moreover, research has shown that higher pH disrupts the operation of the MBBR reactor [42,43]. The alkalinity of the effluent entering the MBBR was 840 mg/L, which was increased in the effluent and its amount reached 1,200 mg/L, which is due to the formation of HCO<sup>3-</sup> [3–5].

3.2. ANOVA analysis of the model

The percentage of COD removal efficiency as a response was analyzed via Design-Expert software. Designed

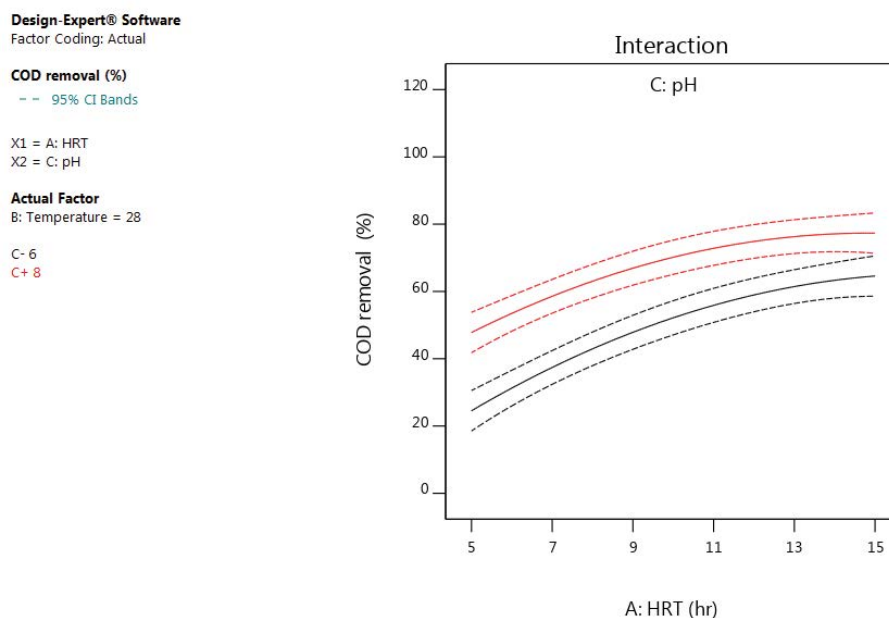


Fig. 2. Effect of HRT and pH interaction on the COD removal at temperature of 28°C.

experiments and their responses are represented in Table 2. ANOVA analysis of variables as linear, quadratic, and the interaction between them is summarized in Table 3. The  $p$ -value indicates the function of the achieved experimental results to study a statistical hypothesis. A  $p$ -value of less than 0.05 (typically 0.05%) is statistically significant. A value of  $P$  above 0.05 ( $>0.05$ ) is not statistically significant and indicates strong evidence for the null hypothesis [40]. If the  $p$ -value of each parameter was lower than the threshold value of 0.05, that parameter was considered significant and effective at that level [44,45]. As indicated in Table 3, all three independent factors, temperature, HRT, pH (linearly and squarely), and the interaction between pH and HRT have a significant impact on the test process ( $P < 0.05$ ) while the interactions of HRT, temperature, and pH were not significant. The amount of the  $p$ -value of the model demonstrates that the regression model is significant ( $P < 0.0001$ ) and depicted that the quadratic model was sufficient for the necessary correlation between the response and the variables. Large  $F$ -value and small  $p$ -value generally imply that this model has a significant coefficient [43]. The lack of fit rate illustrates the lack of fit rate of the data achieved with replications of the center point. Because the  $p$ -value of the lack of fit was less than 0.05 and considered insignificant the model has the necessary fit.

The value of adjusted  $R^2$  reveals the proper overlap of laboratory data predicted utilizing the CCD model. In this model, the adjusted  $R^2$  was 0.9854, which was reduced to 0.9832 following the simplification of the model. The predicted  $R^2$  of 0.9623 is in reasonable agreement with the adjusted one.

In similar studies, few researchers reported that the coefficient of determination ( $R^2$ ) was more than 0.8

indicating that the regression model was well consistent with the data [46].

Fig. 3 illustrates the normal distribution of the main variables and all their interactions. The normal probability diagram is employed to check the intrinsic deviation from the normal state. The deviation of each parameter of the straight line represents a deviation from the normal state. A residual normal distribution graph was applied to determine the model's accuracy. As shown in Fig. 3, most of the data are around the centerline, thereby being normal and acceptable [47].

### 3.3. Investigation of contour diagrams of regression models

Contour graphs are useful tools to investigate the effects of independent variables on the response. Through the use of these graphs, the effect of independent variables on the response could be determined at different points.

Fig. 4 presents the contour diagram of temperature and HRT on the COD removal efficiency. As can be seen in Fig. 4, the points closer to the bold orange color represent a higher COD removal efficiency. According to Fig. 4, the highest COD removal efficiency belongs to temperatures between 24°C–30°C and high HRT (higher than 9 h). This may be attributed to the fact that at temperatures between 24°C and 32°C, the growth rate of microorganisms was enough, and at less than 9 h HRT, the contact time between the microorganisms and biomass was not enough. Consequently, Fig. 4 indicates no signs of the two variables, temperature and HRT, affecting each other.

Based on Fig. 5, in the green part of the counter diagram, the COD removal efficiency is low, and in the orange area, the COD removal efficiency is high. The highest COD removal efficiency belongs to high HRTs and pH

Table 2

List of designed experiments and the experimental and theoretical response for dairy effluent wastewater treatment using MBBR

Std	Run	Factor 1	Factor 2	Factor 3	Response
		A: pH	B: Temperature, °C	C: HRT, h	COD removal %
16	1	7	35	22	83.5
5	2	6	30	32	37
9	3	6	35	22	60
15	4	7	35	22	82
2	5	8	30	12	34
14	6	7	35	32	85
10	7	8	35	22	75
11	8	7	30	22	54
17	9	7	35	22	80
1	10	6	30	12	10
7	11	6	40	32	42
4	12	8	40	12	45
3	13	6	40	12	30
8	14	8	40	32	65
6	15	8	30	32	60
13	16	7	35	12	59
12	17	7	40	22	66

Table 3  
ANOVA analysis of the dairy wastewater treatment modeling using MBBR by RSM and CCD

Source	Sum of squares	df	Mean square	F-value	p-value
Model	9,777.24	9	1,086.36	121.16	<0.0001
A-HRT	3,027.60	1	3,027.60	337.67	<0.0001
B-Temperature	193.60	1	193.60	21.59	0.0024
C-pH	810.00	1	810.00	90.34	<0.0001
AB	1.13	1	1.13	0.1255	0.7336
AC	55.13	1	55.13	6.15	0.0422
BC	28.13	1	28.13	3.14	0.1198
A <sup>2</sup>	154.98	1	154.98	17.29	0.0043
B <sup>2</sup>	360.87	1	360.87	40.25	0.0004
C <sup>2</sup>	1,622.11	1	1,622.11	180.92	<0.0001
Residual	62.76	7	8.97		
Lack of fit	62.76	5	12.55		0.0920
Pure error	0.0000	2	0.0000		
Cor. total	9,840.00	16			

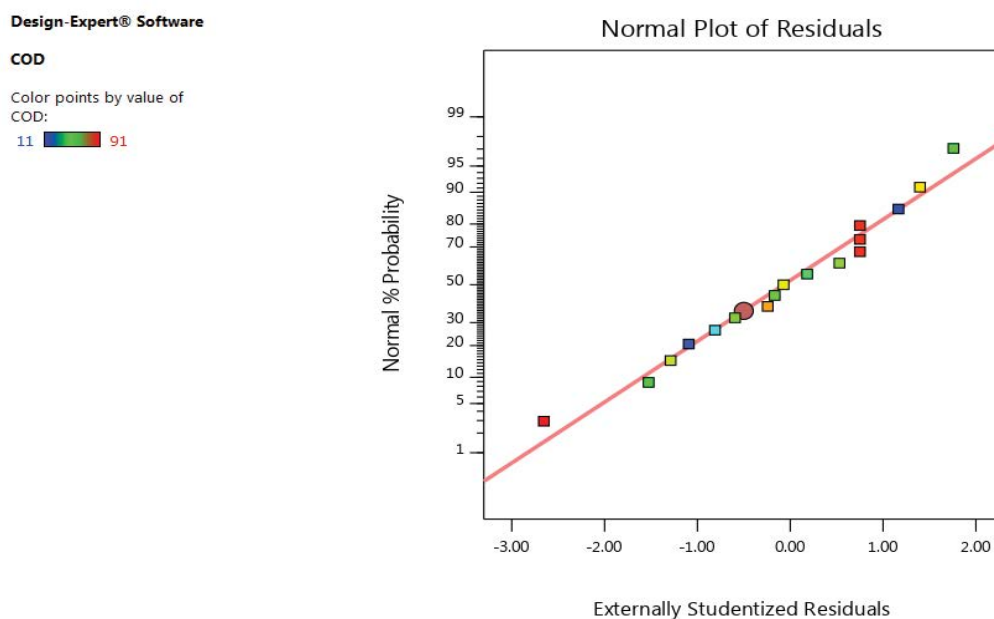


Fig. 3. Normal scatter chart of the Kanyar dairy wastewater treatment modeling by CCD.

between 6.5 and 7.5. The HRT and pH have a significant effect on each other and the interaction between them results in the opposite effect on the system response.

Following the increase in pH up to 6.5, the effluent produces sludge, and the rate of sludge production rises too. However, with increasing the pH value up to 7.5, the sludge production becomes so high that the effluent is out of its normal state and the so-called effluent becomes toxic. In the orange part of Fig. 6 in which pH is below 6.5, the removal percentage is not increased by raising the temperature. Moreover, at temperatures above 33°C, increasing pH does not affect the removal efficiency of COD. This observation can be related to the non-interaction between these two factors (The *p*-value of more than 0.05 confirms this result).

### 3.4. Effect of HRT

The efficiency of the wastewater treatment process with the activated sludge was studied in three HRTs, including 5, 10, 15, and 20 h. As depicted in Fig. 7, in a pH value of 7 and a temperature of 28°C, the COD removal efficiency in the HRT of 5, 10, and 15 and 20 h were 59%, 85%, 91%, and 93%, respectively; and the amount of total suspended solids (TSS) in the HRT of 5, 10, and 15 and 20 h were 90, 60, 54 and 52 mg/L, respectively. These results can be explained by the fact that the biofilm was not enough in the HRT of 5 h. In the HRT of 10 h with a reduction in the dissolved oxygen and the ratio of food to microorganisms, the system reached a steady-state condition, and the

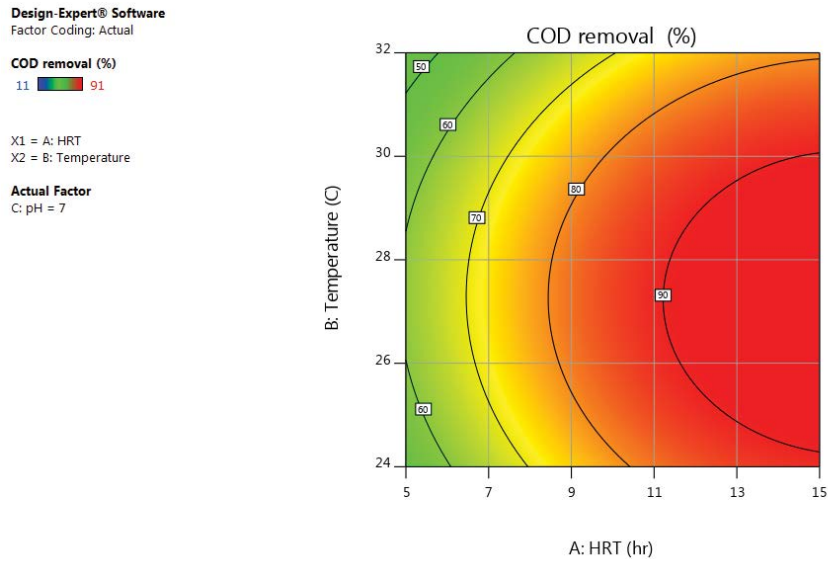


Fig. 4. Contour diagram of simultaneous effect of temperature and HRT of COD removal efficiency.

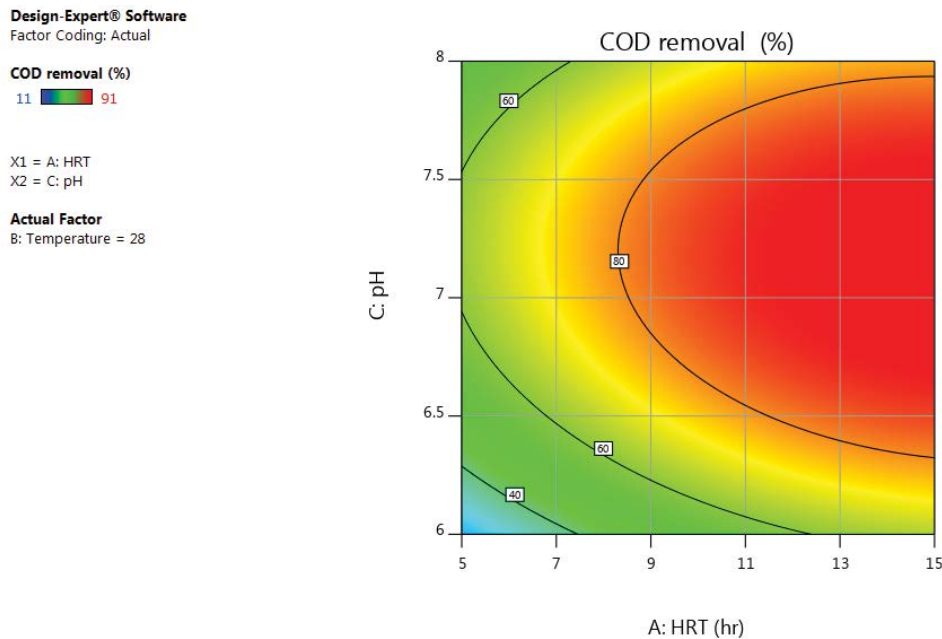


Fig. 5. Contour diagram of simultaneous effect of pH and HRT on COD removal efficiency.

biofilm becomes thicker, which led to a significant increase in efficiency. In the HRT of 15 h, the thickness of the biofilm increased more, thereby increasing the efficiency of COD removal from dairy wastewater more [48,49].

3.5. Effect of temperature

Fig. 8 reveals the efficiency of the effluent treatment process with the activated sludge at four temperature levels of 24°C, 28°C, 32°C, and 36°C. Following the temperature change of each stage, in which the reactor operated for a week and its properties were tested in three stages every day to reach a steady-state in those new conditions and the

effluent properties were then measured. As demonstrated in Fig. 8, at a pH of 7 and HRT of 10 h, the total dissolved solids (TDS) at temperatures of 24°C, 28°C, 32°C, and 36°C were 170, 82, 130, and 230 mg/L, respectively, and the turbidities were 110, 70, 80, and 120 NTU, respectively. The optimum temperature in these conditions was 28°C, meaning that with increasing the temperature, the percentage of COD removal also increased to reach the desired temperature, which also varied depending on the type of microbe in each sludge. The process gradually continued until it reached a point where the treatment process stopped. The effect of temperature on this process is very large and has a great effect on the settling and clotting properties [50,51].



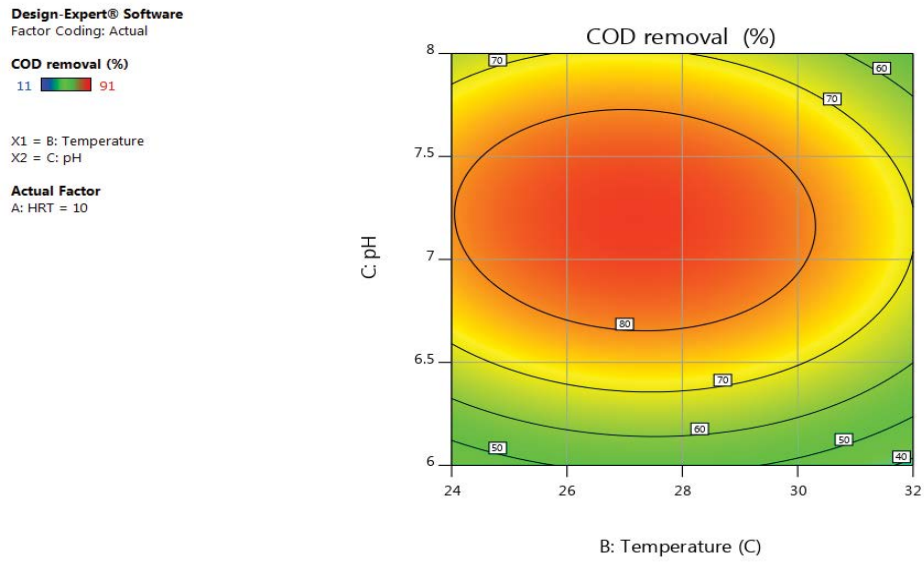


Fig. 6. Contour diagram of simultaneous effect of pH and temperature on the of COD removal efficiency.

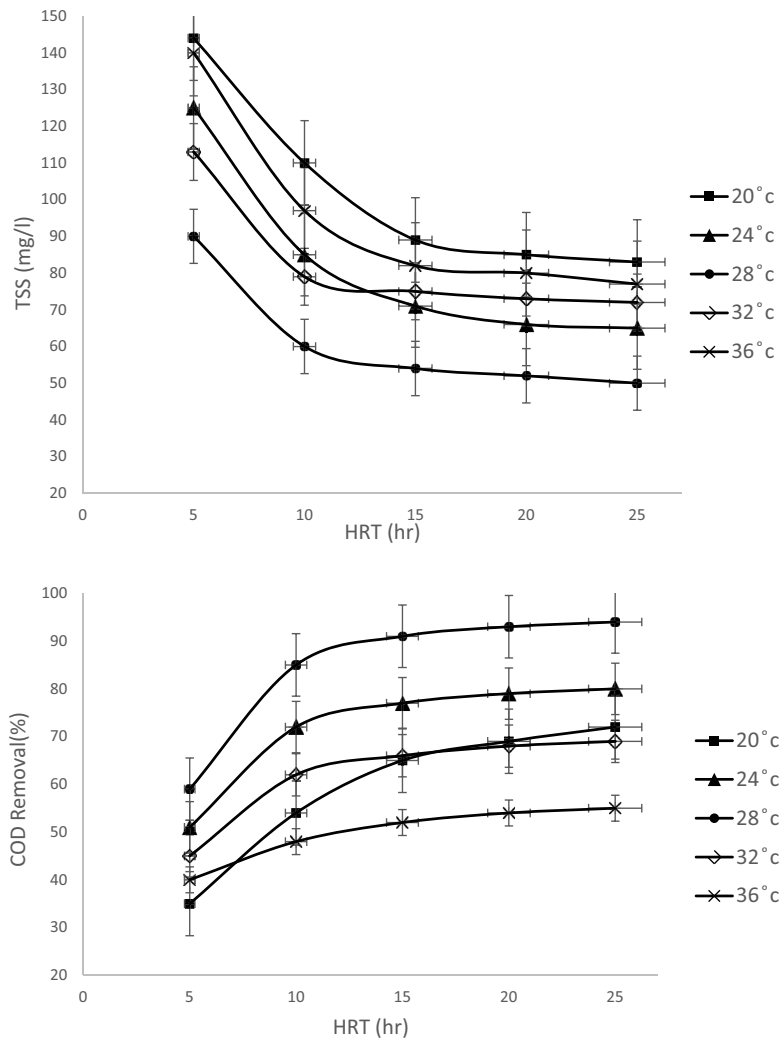


Fig. 7. Effect of HRT and temperature at pH = 7 on COD removal efficiency and TSS removal.



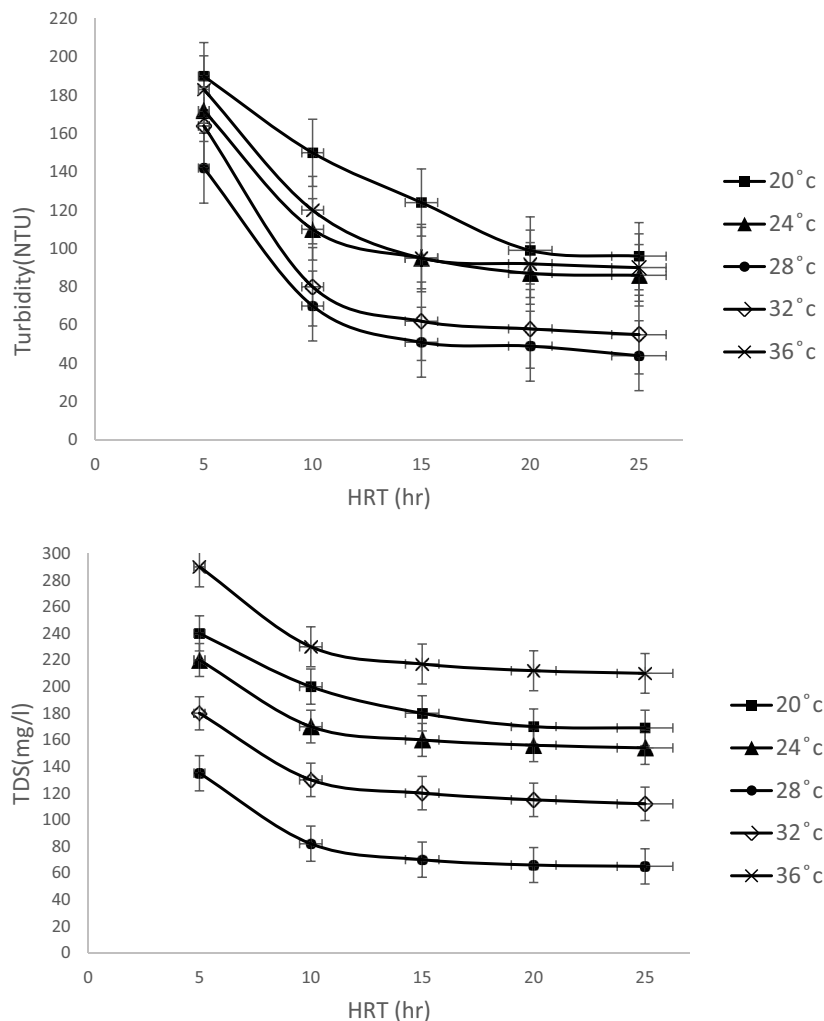


Fig. 8. Effect of HRT and temperature at pH = 7 on TDS removal efficiency and turbidity removal.

Table 4  
Optimal conditions of the dairy wastewater treatment modeling by RSM and CCD

Response	COD removal
Predicted mean response	91.0982
Predicted median response	91.0982
Std. Dev.	2.99435
<i>n</i>	1
SE pred.	3.25306
95% PI low	83.406
Data mean	92.5
95% PI high	98.7905

### 3.6. Effect of pH

The neutral pH for the microorganism activation is reported as the optimal pH value in most studies. Therefore, we considered the best pH to be 7 according to Fig. 7 and other studies. For more reassurance of the accuracy of results,

these experiments were repeated with pH values of 6, 7, and 8 (Fig. 9) while the temperature and HRT were constant at 28°C and 10 h. The COD removal efficiency, TSS, TDS, and turbidity in pH values of 6, 7, and 8 were 53%, 93%, and 68%; 114, 52, and 90 mg/L; 136, 66, and 112 mg/L; and 194, 49, and 100 NTU, respectively. This can be attributed to the fact that the pH affects microorganism metabolism and performance. It can be concluded that increasing the pH value in the specified range contributes to the efficiency increasing while with changing pH from 7 to 8, the percentage of organic matter removal decreases. Similar results were achieved with pH = 7.3 [52].

### 3.7. Optimization

The obtained optimal amounts for each of the independent factors were considered to achieve maximum COD removal efficiency from dairy wastewater using MBBR. At best condition (pH = 7, temperature = 27°C, and HRT = 11 h COD) removal efficiency was obtained 93%. According to Table 4, to confirm the accuracy of the optimal point, the COD removal experiment was repeated three

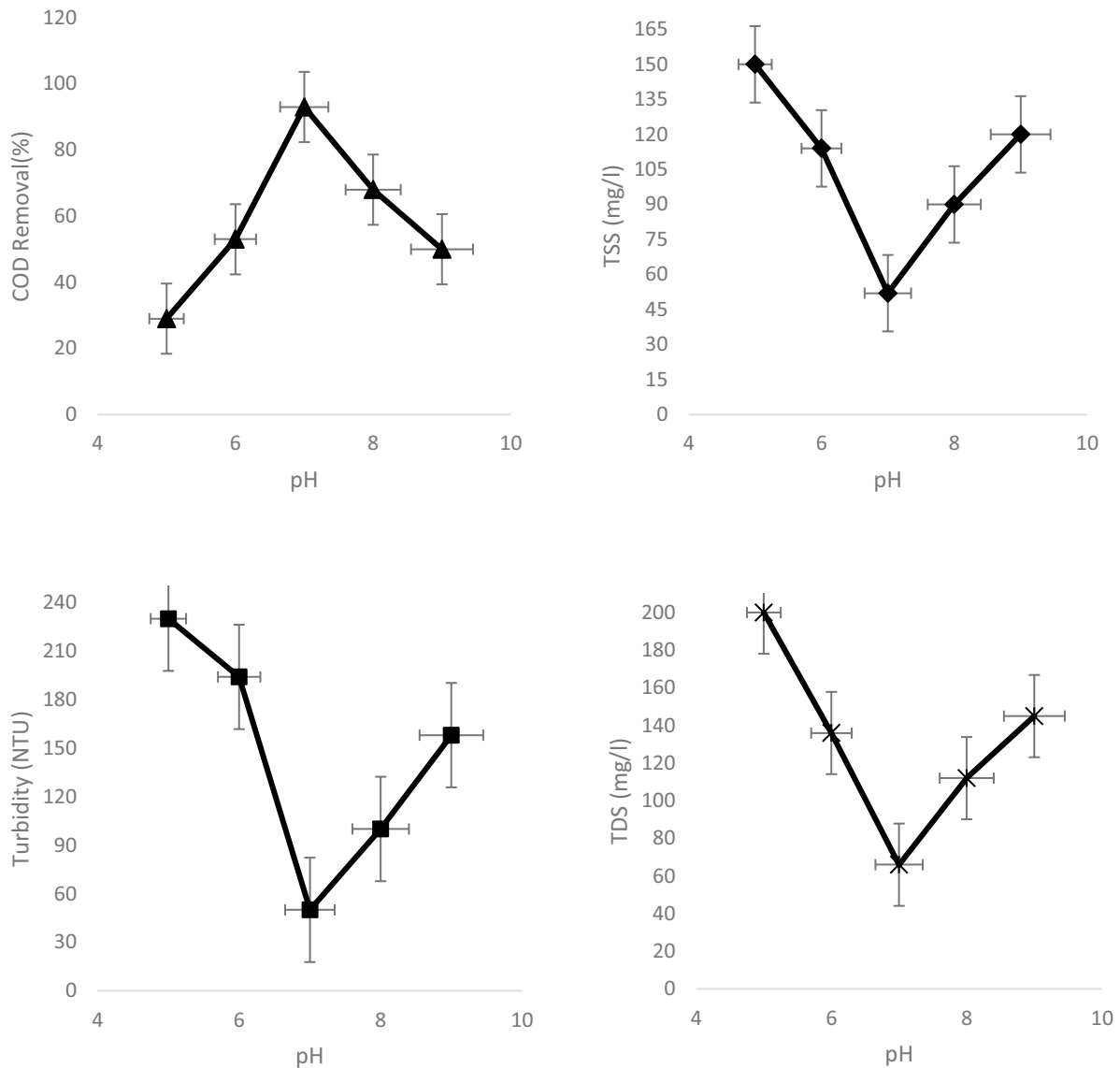


Fig. 9. Effect of pH on COD removal efficiency at temperature of 28°C and HRT of 10 h.

times and the percentage of COD removal in each step was calculated. The obtained experimental results at the optimal operating condition indicated an excellent agreement with the predicted response by the models.

#### 4. Conclusion

The present study represented the possibility of MBBR for effluent wastewater treatment from the anaerobic filter of Kanyar Dairy Company in Quchan – Iran. We aimed to determine the optimal operating conditions of MBBR in the treatment of dairy effluent and to investigate the simultaneous effect of temperature, HRT, and pH on the performance of the MBBR. The setup of MBBR was prepared for three months and was seeded with waste sludge from the activated sludge unit of Kanyar Dairy Company.

The results revealed that with increasing temperature, the percentage of COD removal increases in order to reach the desired temperature. Furthermore, lowering the temperature reduces the growth synthesis and the speed of biological reactions and thus decreases the removal efficiency.

The percentage of COD removal increases with increasing HRT, but does not exceed a certain limit and high HRT and overburden concentrations raise the stability of the system. Based on the results, the reactor performance in acidic and alkaline environments decreased and the best performance of both reactors was when pH was 7.

According to the statistical analysis of the results and the *p*-value of the independent variables, it was found that all three parameters, temperature, HRT, and pH, are effective and cannot be ignored.

Moreover, it was found the *p*-value is less than 0.05 only in the interaction between the two parameters of pH and HRT. Furthermore, only the interaction of these two parameters

with each other reduces the efficiency of COD removal, which is of great importance in the MBBR aerobic reactor.

The value of adjusted  $R^2$  indicates the proper overlap of laboratory data, predicted via the CCD model. In this model, adjusted  $R^2$  was 0.9854, which was reduced to 0.9832 after simplifying the model. The predicted  $R^2$  of 0.9623 is in line with the adjusted one.

The highest COD removal efficiency belonged to the temperature of 24 to 30, pH of 6.5 to 7.5, and an HRT of more than 7 h. Prior to the optimization, the removal percentage was 85%, however, after this process pH = 7, temperature = 27°C, and HRT = 11 h were obtained as the optimal condition points with a 93% of COD removal efficiency. Based on other research conducted under optimal conditions in other important aerobic biological treatment methods, such as active sludge, SBR, MBR, and RBC, the COD removal percentage is reported to be 65%, 80%, 94%, and 96%, respectively [53,54].

In brief, on account of the low price, simplicity, and high efficiency of COD removal compared to other aerobic methods, the MBBR reactor exhibited a good performance

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### References

- [1] C. Liang, N. de Jonge, P.N. Carvalho, J.L. Nielsen, K. Bester, Biodegradation kinetics of organic micropollutants and microbial community dynamics in a moving bed biofilm reactor, *Chem. Eng. J.*, 415 (2021) 128963, doi: 10.1016/j.cej.2021.128963.
- [2] E. Zkeri, A. Iliopoulou, A. Katsara, A. Korda, M. Aloupi, G. Gatidou, M.S. Fountoulakis, A.S. Stasinakis, Comparing the use of a two-stage MBBR system with a methanogenic MBBR coupled with a microalgae reactor for medium-strength dairy wastewater treatment, *Bioresour. Technol.*, 323 (2021) 124629, doi: 10.1016/j.biortech.2020.124629.
- [3] E. Rikmann, I. Zekker, T. Tenno, A. Saluste, Inoculum-free start-up of biofilm- and sludge-based deammonification systems in pilot scale, *Int. J. Environ. Sci. Technol.*, 15 (2018) 133–148.
- [4] I. Zekker, O. Artemchuk, E. Rikmann, K. Ohimai, G. Dhar Bhowmick, M. Madhao Ghangrekar, J. Burlakovs, T. Tenno, Start-up of ANAMMOX SBR from non-specific *inoculum* and process acceleration methods by hydrazine, *Water*, 13 (2021) 350, doi: 10.3390/w13030350.
- [5] I. Zekker, A. Kivirüüt, E. Rikmann, A. Mandel, M. Jaagura, T. Tenno, O. Artemchuk, S. dC Rubin, T. Tenno, Enhanced efficiency of nitrating-ANAMMOX sequencing batch reactor achieved at low decrease rates of oxidation–reduction potential, *J. Environ. Eng. Sci.*, 36 (2019) 0225, doi: 10.1089/ees.2018.0225.
- [6] I. Zekker, G.D. Bhowmick, H. Priks, D. Nath, E. Rikmann, M. Jaagura, T. Tenno, K. Tamm, M.M. Ghangrekar, ANAMMOX-denitrification biomass in microbial fuel cell to enhance the electricity generation and nitrogen removal efficiency, *Biodegradation*, 31 (2020) 249–264.
- [7] M. Ahmadi, M. Ahmadmoazzam, R. Saeedi, M. Abtahi, S. Ghafari, S. Jorfi, Biological treatment of a saline and recalcitrant petrochemical wastewater by using a newly isolated halo-tolerant bacterial consortium in MBBR, *Desal. Water Treat.*, 167 (2019) 84–95.
- [8] S.A. Osmani, A. Rajpal, A.A. Kazmi, Upgradation of conventional MBBR into Aerobic/Anoxic/Aerobic configuration: a case study of carbon and nitrogen removal based sewage treatment plant, *J. Water Process Eng.*, 40 (2021) 101921, doi: 10.1016/j.jwpe.2021.101921.
- [9] A. Tsitouras, N. Al-Ghussain, R. Delatolla, Two moving bed biofilm reactors in series for carbon, nitrogen, and phosphorus removal from high organic wastewaters, *J. Water Process Eng.*, 41 (2021) 102088, doi: 10.1016/j.jwpe.2021.102088.
- [10] M. Kozak, K. Cirik, S. Başak, Treatment of textile wastewater using combined anaerobic moving bed biofilm reactor and powdered activated carbon-aerobic membrane reactor, *J. Environ. Chem. Eng.*, 87 (2021) 105596, doi: 10.1016/j.jece.2021.105596.
- [11] M. Zhang, T. Song, C. Zhu, Y. Fan, A. Soares, X. Gu, J. Wu, Roles of nitrate recycling ratio in the A<sup>2</sup>O - MBBR denitrifying phosphorus removal system for high-efficient wastewater treatment: Performance comparison, nutrient mechanism and potential evaluation, *J. Environ. Manage.*, 270 (2020) 110887, doi: 10.1016/j.jenvman.2020.110887.
- [12] H. Yuan, Y. Li, K. Wang, Effect of influent ammonia nitrogen concentration on microbial community in MBBR reactor, *Water Sci. Technol.*, 83 (2021) 162–172.
- [13] M. SeyedSalehi, J. Jaafari, C. Hélix-Nielsen, G. Hodaifa, M. Manshouri, S. Ghadimi, H. Hafizi, H. Barzanouni, Evaluation of moving-bed biofilm sequencing batch reactor (MBSBR) in operating A<sup>2</sup>O process with emphasis on biological removal of nutrients existing in wastewater, *Int. J. Environ. Sci. Technol.*, 15 (2018) 199–206.
- [14] J. Jaafari, A.B. Javid, H. Barzanouni, A. Younesi, N.A.A. Farahani, M. Mousazadeh, P. Soleimani, Performance of modified one-stage Phoredox reactor with hydraulic up-flow in biological removal of phosphorus from municipal wastewater, *Desal. Water Treat.*, 171 (2019) 216–222.
- [15] D. Naghipour, E. Rouhbakhsh, J. Jaafari, Application of the biological reactor with fixed media (IFAS) for removal of organic matter and nutrients in small communities, *Int. J. Environ. Anal. Chem.*, 98 (2020) 1–11.
- [16] A. Shitu, G. Liu, Y. Zhang, Z. Ye, J. Zhao, S. Zhu, D. Liu, Enhancement of mariculture wastewater treatment using moving bed biofilm reactors filled with modified biocarriers: characterisation, process performance and microbial community evaluation, *J. Environ. Manage.*, 291 (2021) 112724, doi: 10.1016/j.jenvman.2021.112724.
- [17] A. Duyar, V. Ciftcioglu, K. Cirik, G. Civelekoglu, S. Urus, Treatment of landfill leachate using single-stage anoxic moving bed biofilm reactor and aerobic membrane reactor, *Sci. Total Environ.*, 776 (2021) 145919, doi: 10.1016/j.scitotenv.2021.145919.
- [18] M.A.S. Hafiz, C. Baumann, J.K. Sher, H. Schönberger, F. Weber, Performance evaluation of anaerobic moving bed bioreactor (An-MBBR) for pretreatment of desizing wastewater, *Desal. Water Treat.*, 181 (2020) 123–130.
- [19] A.F.M. Udaiyappan, H.A. Hasan, M.S. Takriff, S.R.S. Abdullah, N.H.M. Yasin, B. Ji, Cultivation and application of *Scenedesmus* sp. strain UKM9 in palm oil mill effluent treatment for enhanced nutrient removal, *J. Cleaner Prod.*, 294 (2021) 126295, doi: 10.1016/j.jclepro.2021.126295.
- [20] E. Edefell, P. Falàs, S. Kharel, M. Hagman, M. Christensson, M. Cimbritz, K. Bester, MBBRs as post-treatment to ozonation: degradation of transformation products and ozone-resistant micropollutants, *Sci. Total Environ.*, 754 (2021) 142103, doi: 10.1016/j.scitotenv.2020.142103.
- [21] A. Mandel, I. Zekker, M. Jaagura, T. Tenno, Enhancement of anoxic phosphorus uptake of denitrifying phosphorus removal process by biomass adaption, *Int. J. Environ. Sci. Technol.*, 16 (2019) 5965–5978.
- [22] K. Klein, E. Kattel, A. Goi, A. Kivi, N. Dulova, A. Saluste, I. Zekker, M. Trapido, T. Tenno, Combined treatment of pyrogenic wastewater from oil shale retorting, *Oil Shale*, 34 (2017) 82–96.
- [23] R. Shankar, S. Kumar, A.K. Prasad, P. Khare, A.K. Varma, V.K. Yadav, Biological wastewater treatment plants (WWTPs) for industrial wastewater, *Microbiol. Ecol. Wastewater Treat. Plants*, 342 (2021) 193–216.

- [24] D. Hosseinlou, Determination of design loading rates for simultaneous anaerobic oxidation/partial nitrification-denitrification process and application in treating dairy industry effluent, *J. Environ. Chem. Eng.*, 9 (2021) 105176, doi: 10.1016/j.jece.2021.105176.
- [25] Y. Achour, L. Bahsis, E.-H. Ablouh, H. Yazid, M.R. Laamari, M. El Haddad, Insight into adsorption mechanism of Congo red dye onto *Bombax Buonopozense* bark activated-carbon using central composite design and DFT studies, *Surf. Interfaces*, 23 (2021) 100977, doi: 10.1016/j.surfin.2021.100977.
- [26] T.M. Laid, K. Abdelhamid, L.S. Eddine, B. Abderrhmane, Optimizing the biosynthesis parameters of iron oxide nanoparticles using central composite design, *J. Biomol. Struct.*, 1229 (2021) 129497, doi: 10.1016/j.molstruc.2020.129497.
- [27] F. Jiang, M. Zhang, X. Yang, X. Zeng, Z. Liang, D. Li, Research on optimization design of dual derrick for offshore platform based on RSM, *IOP Conf. Ser.: Earth Environ. Sci.*, 440 (2020) 052049.
- [28] N. Rezanian, M.H. Zonoozi, M. Saadatpour, Coagulation-flocculation of turbid water using graphene oxide: simulation through response surface methodology and process characterization, *Environ. Sci. Pollut. Res.*, 28 (2021) 14812–14827.
- [29] Y. Li, C. Liu, L. Zhang, B. Sun, A partition optimization design method for a regional integrated energy system based on a clustering algorithm, *Energy*, 219 (2021) 119562, doi: 10.1016/j.energy.2020.119562.
- [30] A.B. Jasni, H. Kamyab, S. Chelliapan, N. Arumugam, S. Krishnan, M.F.M. Din, Treatment of wastewater using response surface methodology: a brief review, *Chem. Eng. Trans.*, 78 (2020) 535–540.
- [31] P. Xiao, J. Zhou, X. Luo, B. Kang, L. Guo, G. Yuan, L. Zhang, T. Zhao, Enhanced nitrogen removal from high-strength ammonium wastewater by improving heterotrophic nitrification-aerobic denitrification process: insight into the influence of dissolved oxygen in the outer layer of the biofilm, *J. Cleaner Prod.*, 297 (2021) 126658, doi: 10.1016/j.jclepro.2021.126658.
- [32] I.D. Boateng, X.-M. Yang, Process optimization of intermediate-wave infrared drying: screening by Plackett–Burman; comparison of Box–Behnken and central composite design and evaluation: a case study, *Ind. Crops Prod.*, 162 (2021) 113287, doi: 10.1016/j.indcrop.2021.113287.
- [33] I. Zekker, M. Raudkivi, O. Artemchuk, E. Rikmann, H. Priks, M. Jaagura, T. Tenno, Mainstream-sidestream wastewater switching promotes ANAMMOX nitrogen removal rate in organic-rich, low-temperature streams, *Environ. Technol.*, 34 (2020) 1–10.
- [34] E. Rikmann, I. Zekker, M. Tomingas, P. Vabamäe, K. Kroon, A. Saluste, T. Tenno, A. Menert, L. Loorits, S.S. dC Rubin, Comparison of sulfate-reducing and conventional ANAMMOX upflow anaerobic sludge blanket reactors, *J. Biosci. Bioeng.*, 118 (2014) 426–433.
- [35] A. di Biase, M.S. Kowalski, T.R. Devlin, J.A. Oleszkiewicz, Moving bed biofilm reactor technology in municipal wastewater treatment: a review, *J. Environ. Manage.*, 247 (2019) 849–866.
- [36] R.K. Sonwani, G. Swain, B.S. Giri, R.S. Singh, B.N. Rai, A novel comparative study of modified carriers in moving bed biofilm reactor for the treatment of wastewater: process optimization and kinetic study, *Bioresour. Technol.*, 281 (2019) 335–342.
- [37] A.D. Santos, R.C. Martins, R.M. Quinta-Ferreira, L.M. Castro, Moving bed biofilm reactor (MBBR) for dairy wastewater treatment, *Energy Rep.*, 6 (2020) 340–344.
- [38] K. Tang, P. Rosborg, E.S. Rasmussen, A. Hambly, M. Madsen, N.M. Jensen, A.A. Hansen, C. Sund, H.G. Andersen, E. Torresi, C. Kragelund, H.R. Andersen, Impact of intermittent feeding on polishing of micropollutants by moving bed biofilm reactors (MBBR), *J. Hazard. Mater.*, 403 (2021) 123536, doi: 10.1016/j.jhazmat.2020.123536.
- [39] X. Chen, Q. Zhang, Y. Zhu, T. Zhao, Response of wastewater treatment performance, microbial composition and functional genes to different C/N ratios and carrier types in MBBR inoculated with heterotrophic nitrification-aerobic denitrification bacteria, *Bioresour. Technol.*, 42 (2021) 125339, doi: 10.1016/j.biortech.2021.125339.
- [40] M. Xu, W. Zhou, X. Chen, Y. Zhou, B. He, S. Tan, Analysis of the biodegradation performance and biofouling in a halophilic MBBR-MBR to improve the treatment of disinfected saline wastewater, *Chemosphere*, 269 (2021) 128716, doi: 10.1016/j.chemosphere.2020.128716.
- [41] X. Cao, N. Ren, G. Tian, Y. Fan, Q. Duan, A three-dimensional prediction method of dissolved oxygen in pond culture based on Attention-GRU-GBRT, *Comput. Electron. Agric.*, 181 (2021) 105955, doi: 10.1016/j.compag.2020.105955.
- [42] T. Tenno, K. Uiga, A. Mashirin, I. Zekker, E. Rikmann, Modeling closed equilibrium systems of H<sub>2</sub>O–dissolved CO<sub>2</sub>–solid CaCO<sub>3</sub>, *J. Phys. Chem.*, 121 (2017) 3094–3100.
- [43] T. Tenno, E. Rikmann, K. Uiga, I. Zekker, A. Mashirin, T. Tenno, A novel proton transfer model of the closed equilibrium system H<sub>2</sub>O–CO<sub>2</sub>–CaCO<sub>3</sub>–NH<sub>4</sub><sup>+</sup>, *Proc. Est. Acad. Sci.*, 67 (2018) 260–270.
- [44] M. Zhang, J. Gao, Y. Fan, Q. Liu, C. Zhu, L. Ge, C. He, J. Wu, Comparisons of nitrite accumulation, microbial behavior and nitrification kinetic in continuous stirred tank (ST) and plug flow (PF) moving bed biofilm reactors, *Chemosphere*, 278 (2021) 130410, doi: 10.1016/j.chemosphere.2021.130410.
- [45] A.C. Affam, W.C. Chung, P.L. Lau, O.A. Johnson, K.C. Seong, L. Baloo, B.W.L. Peng, F. Xinru, Design Expert Application for Optimization of Ag/AgBr/TiO<sub>2</sub> Visible Light Photocatalyst Preparation, B.S. Mohammed, N. Shafiq, M. Rahman, S. Kutty, H. Mohamad, A.L. Balogun, Eds., Proceedings of the International Conference on Civil, Offshore and Environmental Engineering, ICCOEE 2021: ICCOEE2020, Vol. 132, Springer, Singapore, 2021, pp. 93–102.
- [46] S.J.S. Chelladurai, K. Murugan, A.P. Ray, M. Upadhyaya, V. Narasimharaj, S. Gnanasekaran, Optimization of process parameters using response surface methodology: a review, *Mater. Today Proc.*, 37 (2021) 1301–1304.
- [47] J. Jaafari, H. Barzanouni, S. Mazloomi, N.A.A. Farahani, K. Sharafi, P. Soleimani, G.A. Haghghat, Effective adsorptive removal of reactive dyes by magnetic chitosan nanoparticles: kinetic, isothermal studies and response surface methodology, *Int. J. Biol. Macromol.*, 164 (2020) 344–355.
- [48] S.I. Gadow, Y.-Y. Li, Development of an integrated anaerobic/aerobic bioreactor for biodegradation of recalcitrant azo dye and bioenergy recovery: HRT effects and functional resilience, *Bioresour. Technol. Rep.*, 9 (2020) 100388, doi: 10.1016/j.biteb.2020.100388.
- [49] A.B. Fanta, A.M. Nair, S. Sægrov, S.W. Østerhus, Phosphorus removal from industrial discharge impacted municipal wastewater using sequencing batch moving bed biofilm reactor, *J. Water Process Eng.*, 41 (2021) 102034, doi: 10.1016/j.jwpe.2021.102034.
- [50] A. Dargahi, R. Shokoohi, G. Asgari, A. Ansari, D. Nematollahi, M.R. Samarghandi, Moving-bed biofilm reactor combined with three-dimensional electrochemical pretreatment (MBBR–3DE) for 2,4-D herbicide treatment: application for real wastewater, improvement of biodegradability, *RSC Adv.*, 11 (2021) 9608–9620.
- [51] L. Daija, A. Selberg, E. Rikmann, I. Zekker, T. Tenno, T. Tenno, The influence of lower temperature, influent fluctuations and long retention time on the performance of an upflow mode laboratory-scale septic tank, *Desal. Water Treat.*, 57 (2016) 18679–18687.
- [52] A. Micek, K. Józwiakowski, M. Marzec, A. Listosz, T. Grabowski, Efficiency and technological reliability of contaminant removal in household WWTPs with activated sludge, *Appl. Sci.*, 11 (2021) 1889, doi: 10.3390/app11041889.
- [53] G. Xu, Z. Zhang, F. Gao, Effect of COD/N ratios and DO concentrations on the NOB suppression in a multi-cycle SBR, *J. Environ. Chem. Eng.*, 9 (2021) 105735, doi: 10.1016/j.jece.2021.105735.
- [54] A.M. Abdelkader, Comparative study between membrane bioreactor MBR and rotating biological contactors RBC for greywater treatment, *Int. J. Environ. Sci. Dev.*, 12 (2021) 107–111.