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Phosphorus Removal from Dairy Wastewater in Batch Systems under Simultaneous Aerobic/Anaerobic Conditions: Application of Response Surface Methodology

M. Amini*

Department of Environmental Science, Faculty of Natural Resources, University of Jiroft, Jiroft, Iran

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ABSTRACT

The objective of this paper is to demonstrate phosphorus removal from a dairy wastewater in a batch system with simultaneous aerobic and anaerobic process. The system consists of a granular sequencing batch reactor (SBR) working under alternating aerobic/anaerobic conditions. In order to analyze the process, four significant variables viz. MLSS, COD/N ratio, aeration time and cycling time and four dependent parameters as the process responses were studied. The experiments were designed using a central composite design (CCD) and the data were processed under response surface methodology (RSM). The optimal removal percentage of PO_4^{3-} obtained 98.34% at SVI, MLSS, and MLVSS of 44.03 ml/g, 5645.97 mg/l and 4435.99 mg/l, respectively. High-magnification SEM showed the morphological diversity of the microorganisms that inhabited the SCNP (Simultaneous carbon, nitrogen and phosphorous removal) in batch conditions, which included spherical, rod and long filamentous-shape bacteria microorganisms. This study showed a successful high phosphorus removal from dairy wastewater with microbial granules cultivated under the alternating aerobic–anaerobic conditions in batch regime.

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1. INTRODUCTION

The nitrogen (N) and phosphorus (P) contents of discharged wastewaters are the main causes for eutrophication of water bodies. It is therefore necessary to remove these substances from wastewaters in order to reduce the harm they cause to the environment. In most continental waters, phosphorus is the limiting nutrient for biological growth; therefore, its removal from wastewaters is considered[1]. Although chemical precipitation was originally the usual procedure for phosphorus removal from wastewaters, the advantages provided by the biological phosphorus removal have promoted its implementation in many wastewater treatments. Enhanced biological phosphorus removal (EBPR) is a wastewater treatment process aimed at achieving low phosphorus concentration effluents [2]. Microorganisms responsible for EBPR are characterized by two different metabolisms. In an anaerobic condition, accumulation of readily biodegradable carbon substrates and hydrolysis of intracellular polyphosphate occur, which is released in the bulk liquid in the form of orthophosphate. While in an aerobic condition, phosphate accumulation happens in the form of intracellular polyphosphate from readily biodegradable carbon substrates as energy source. It has been recommended that the anaerobic conditions be spatially compartmentalized for the efficient removal of phosphorus [3]. Dairy wastewater is treated using biological treatment methods because reagent costs are high and the soluble COD removal is poor in physical–chemical treatment processes. Anaerobic-aerobic treatment processes are commonly used together for dairy wastewater treatment, in order to achieve the effluent discharge limits for industry wastewaters [4]. Granular systems can couple with other treatment units to complement benefits from both processes [5, 6]. Biogranules were formed by self-immobilization of microorganisms without the presence of any medium for attachment. This work is the first study to evaluate the ability of batch experiments to investigate

*Corresponding Author's Email: amini.malihe@gmail.com (M. Amini)

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simultaneous aerobic and anaerobic conditions in one unit together for remove P components from dairy wastewaters [7, 8]. The response surface methodology (RSM) has an important application in the process modeling and design [9, 10]. This methodology can be usefully applied in the development of suitable water treatment technology [11]. In order to analyze the process, four significant independent variables, viz. mixed liquor suspended solids (MLSS) (mg/l), COD/N ratio, aeration time (min/h) and cycling time (h) and four dependent parameters as process responses (PO_4^{3-} removal percentage, SVI, MLSS and MLVSS) were studied. Heretofore, no research has been done to compare above four factors together to verify the extent of their influence on wastewater treatment process. Thus, in this study, the interactions among the variables as well as their direct impacts on the process are discussed.

2. MATERIALS AND METHODS

2. 1. Wastewater Composition and Seed Sludge Growth

The composition of the Synthetic wastewater was as follows: (g/l): sucrose 1, NH_4Cl 0.95, KH_2PO_4 0.08, $MgSO_4$ 0.2, $FeSO_4$ 0.01, $CaCl_2$ 0.2, $NaHCO_3$ 0.073, $CaCO_3$ 0.005 [12]. It was used for sludge growth before start experiments. All chemicals used for medium were analytical grade [7]. Dairy wastewater was collected from an industrial dairy (Shir Kameh) and stored at 2 to 6°C in refrigerator. Shir Kameh industry is located in Noor city,North of Iran. The characteristics of the raw dairy wastewater fed to the experiments are shown in Table 1. The activated sludge was obtained by acclimating mixed liquor suspended solids collected from the Municipal Wastewater Treatment Plant at Chamestan, Noor, North of Iran.

2. 2. Experimental Design

P–PO_4^{3-} removal percentage from dairy industry wastewater in batch system was designed via response surface methodology (RSM) [11]. The statistical method of factorial design of experiments (DOE) eliminates systematic errors with an estimate of the experimental error and minimizes number of experiments. The central composite design (CCD) is the most frequently used under RSM design. RSM was used to assess the relationship between responses and independent variables, as well as to optimize the relevant conditions of variables in order to predict the best value of responses. The optimum values of the selected variables were obtained by solving the regression equation at desired values of the process responses as the optimization criteria [13]. Each independent factor was coded at five levels: $-\alpha$, -1, 0, +1 and $+\alpha$. The range and level of the variable in coded units from RSM studies are given in Table 2. The design consisted of 2^k factorial points augmented by 2^k axial points and a center point, where k is the number of variables. Therefore, 30 experiments ($2^k+2k+6=30$) were conducted with 25 experiments organized in afactorial design (including 16 factorial points, 8 axial points and 1 center point) and the remaining 5 involving the replication of the central point to get a good estimate of the experimental error [9, 13]. To carry out experiments in batch conditions, 4 dependent parameters were measuredas response. The regression equation after the analysis of variances (ANOVA) gave the level of PO_4^{3-} removal percentage, SVI, MLSS and MLVSS as a function of MLSS (X_1), COD/N (X_2) , Aeration time (X_3) and Cycling time (X_4) [9]. Table 3 shows the results of CCD experiments for studying the effect of four independent variables along with the predicted mean and observed responses.

2. 3. Experimental Approach

The total working volume was kept at one liter in each assay. The required amount of sludge in each assay was added as a concentrated solution, as indicated in design [14]. In this stage, dairy wastewater provided enough sucrose (concentration of total sucrose was about 1400 mg/l) and NO_3^- and PO_4^{3-} sources as little as possible.Different amounts of nutrients were added into erlenmeyers at the beginning of the study. Biomass concentration was varied between 1500 and 5500 mg/l in varied runs. Biomass concentration (MLSS mg/l) was determined at the beginning and the end of the experiment.

TABLE 1. Characteristics of Shir kameh industry wastewater

Component	Content, mg/l
pH	10.26
Oil & grease	1042.2
Total solids (SS)	2348
Total suspended solids (TSS)	454
Volatile suspended solids (VSS)	239
Total COD (TCOD)	2176
Soluble COD (SCOD)	1408
Total BOD (TBOD)	1800
N-TKN	2.6
N-NO_3^-	26.307
P-PO_4^{3-}	8.259

TABLE 2. Experimental ranges and levels of the independent variables

Independent variables	Range and Level				
	-α	-1	0	+1	+α
MLSS, mg/l	1500	2500	3500	4500	5500
COD/N	10	15	20	25	30
Areation time, min/h	35	40	45	50	55
Cycling time, h	10	20	30	40	50

Air was introduced through a diffuser at erlenmeyers using an air pump. The upflow shear force was adjusted by changing air flow rate [15]. The cycling time for aeration in batch experiments was 1h and in this cycle aerobic phase was from 35 to 55 min/h and anaerobic phase 5 to 25 min/h. The experiments were performed at room temperature. After end of each run among 10, 20, 30, 40 and 50 h of the aerobic/anoxic process, the supernatant was taken from each flask and filtered through a wathman filternumber 42. Samples were withdrawn and centrifuged at 15000 rpm for 10 min to remove suspended biomass from the liquid phase (Hermle, 236 HK, Germany). After centrifuging the top of supernatant was used for analysis.

2. 4. Analysis Item and Methods Wastewater samples in batch runs were analyzed for PO_4^{3-}, SVI, MLSS and MLVSS using standard methods in American Public Health Association [16, 17]. The phosphorus concentration was determined spectrophotometrically using the absorbance value at 400 nm with a UV/Vis spectrophotometer (Perkin Elmer, Lambda 25, USA) according to standard methods [18]. The results were used to analyze the process using analysis of variance (ANOVA) which was automatically performed by Design Expert Software.

3.RESULTS AND DISCUSSIONS

3. 1. Microscopic Observation Scanning electron microscopy (SEM) was used to visualize the surface, inner and microbial morphology of granules in the batch conditions. For exactly monitoring of the granule structure and characteristics, SEM analysis was performed several times. Figure 1 demonstrates in detail the morphology of granules formed in this process. Results of SEM showed that granules at the steady state conditions had a smooth outer surface with a compact and regular appearance. As shown in thisfigure, the granules were mainly composed of bacilli bacteria and slightly composed of cocci bacteria. These granules had a compact structure, in the shape of rod, round and long rope-like on the surface and inner of sludge's granules (Figure 1a). Filamentous bacteria on the surface of granules also the rod and round shapes were connected together by a slime matrix. Therefore, it seems that vigorous shaking makes the granules weaker as could be observed. Only a limited number of bacteria near the granule surface were directly exposed to the nutrients components in the wastewater, most of the bacteria were located in the interior and thus protected [12].

TABLE 3. Full factorial central composite design of orthogonal and real values with observed responses for removal of PO_4^{3-} %, SVI, MLSS, and MLVSS.

Run order	PO43-, %		SVI, ml/g		MLSS, mg/l		MLVSS, mg/l	
	Actual value	Predicted value	Actual value	Predicted value	Actual value	Predicted value	Actual value	Predicted value
1	90.49	90.38	44.03	43.92	4638.72	4587.75	3715.69	3706.87
2	88.43	87.58	48.47	48.94	3328.9	3273.43	3084.14	3084.33
3	91.19	90.16	49.21	49.3	5646.97	5701.07	4435.99	4448.94
4	90.91	92.47	48.67	48.58	2756.5	2744.09	1965.63	1950.04
5	86.39	87.58	49.11	48.94	3231	3273.43	3069.49	3084.33
6	87.84	87.58	48.82	48.94	3297.44	3273.43	3087.57	3084.33
7	95.67	94.98	49.6	49.74	3310.76	3318.88	2076.65	2079.7
8	86.34	85.45	46.65	46.56	4547.59	4533.89	3700.1	3694.81
9	88.72	87.58	48.99	48.94	3283	3273.43	3097.32	3084.33
10	94.62	94.78	49.57	49.62	3527.62	3534.79	2952.2	2949.74
11	88.95	89.65	49.93	50.1	4403.79	4360.87	3359.89	3364.79
12	87.66	87.58	49.07	48.94	3232.22	3273.43	3096.32	3084.33
13	89.19	89.96	49.56	49.52	2878.46	2898.35	2394.8	2404.31
14	89.91	89.01	51.28	51.19	3550.16	3545.21	2851.86	2845.39
15	73.55	74.78	47.31	47.32	3961	4028.79	3396.88	3404.68
16	84.71	85.09	49.53	49.53	3335	3369.52	2908	2907.81
17	98.34	99.7	47.03	47.01	3843.3	3907.63	2794.35	2800.11
18	92.39	91.57	51.62	51.69	4270.78	4248.14	3455.59	3447.18
19	82.18	82.34	51.51	51.46	5350.93	5294.93	4021.89	4017.27
20	94.41	94.58	49.88	49.82	4827.43	4816.59	3451.68	3442.17
21	93.2	92.34	49.03	48.77	2722.09	2729.5	2058.12	2064.35
22	86.44	87.58	49.13	48.94	3268	3273.43	3071.15	3084.33
23	90.9	90	46.27	46.29	2968.79	2950.98	2410.68	2422.64
24	89.39	89.56	51.47	51.35	4083.14	4084.9	3671.15	3681
25	84.55	84.25	50.89	50.82	3818.63	3835.66	2867.13	2880.85
26	88.05	87.36	47.46	47.67	2813.09	2763.46	2484.02	2472.61
27	85.29	85.2	46.6	46.79	2783.83	2786.93	2367.19	2374.44
28	81.83	82.51	50.02	49.87	4083.14	4107.14	3596.86	3589.11
29	88.75	89.39	50.45	50.58	4872.05	4870.83	4045.87	4043.85
30	80.62	79.93	49.13	49.18	3235	3208.9	2644.01	2633.57

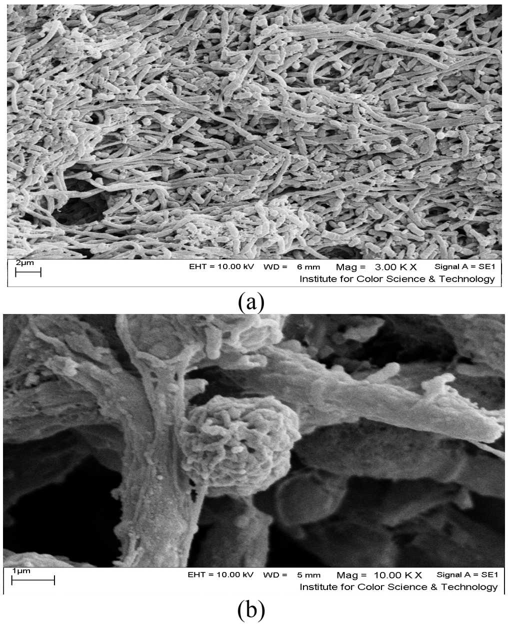

Figure 1. The SEM images from bacterial cells on the surface and inner of granules (2 μm) (a) and cavities in the outer surface of aerobic granules (1 μm) (b).

Large cavities (Figure 1b) could be observed in the outer surface of aerobic granules, which couldbe used to enhance the transport of oxygen, carbon source and nutrients (N, P) into the inner cores of the granules [19]. The difference from these experiments and other researches in appearance of cavity or smooth outer surface of cultivated granules in the reactor was explained by the characteristics of the seed sludge and wastewater. However, further investigations are needed in the formation mechanism of the granule in thesesystems [20].

3. 2. Statistical Analysis RSM approach and analysis of variance ANOVA has conducted for second-order response surface model and the results for all responses are summarized in Table 4. According to Equation (1) the significance of each coefficient was determined by the F-values and values of probability >F [21]. The larger the magnitude of the F-values and the smaller p-values, the more significant is the corresponding coefficients.Values of "prob > F" less than 0.05 also indicate high significant regression at 95% confidence level [22]. In this case, the first-order main effects, square effects and interaction effects of factors were significant model terms. However, the model F-value was from between 50.82 to 4708.45 for PO_4^{3-} removal percentage and MLVSS models, respectively and values of prob > F (<0.0001) for all responses model indicated that the model terms are significant.The insignificant value of lack of fit (more than 0.05) indicated that the quadratic model was valid forresponse. The examination of the fit summary output revealed that the quadratic model is statistically significant for the response and therefore, it was used for further analysis [23]. The goodness of fit of the model was checked by the determination of the coefficient (R^2) [24]. The empirical relationship between responses (Y) and the four variables (factors) in coded values obtained by the application of RSM are given in Table 5. Values of Prob > F were less than 0.05 indicate that the model terms are significant as presented in this table. The square of correlation coefficient for each response was computed as the R-square (R^2) that is a measure of the amount of variation around the mean explained by the model. In this case, the value of the determination of coefficient obtained was about 1 to 0.97 for different responses, which revealed that this regression is statistically significant and only 0-4% of the total variations are not explained by the model. The value of the predicted determination of the coefficient (Pred. R^2 = 0.89-1) is in reasonable agreement with the value of adjusted determination of the coefficient (Adj. R^2 = 0.95-1). At the same time, a relatively lower value of the coefficient of variance (CV = 0.41-1.27% for all responses indicates good precision and reliability of the experiments which were carried out.

3. 3. Process Analysis

3. 3. 1. Phosphorus Removal The batch experimental tests were conducted with various four factors (MLSS (A), COD/N (B), aeration time (C) and cycling time (D)) on phosphorus removalpercentage. All batch runs in this part of study were fedwith dairy wastewaters containing phosphorus concentrations of 30, 50, 70, 90 and 110 mg/l.These experimental designs were according to DOE software. The highest phosphorus removal percentage was observed in 17 run (98.34%) with MLSS 3500 mg/l, COD/N ratio 20, aeration time 55 min/h and cycling time 30 h while minimum value for phosphorus removal percentage was obtained in 15 run (73.55%) with MLSS 3500 mg/l, COD/N ratio 20, aeration time 45 min/h and cycling time 10 h. Also lowest value for COD removal percentage was observed in run 15 therefore this run do not have good conditions for COD and phosphorus removal percentage. At the end of runs phosphorus removal percentages were about 73.55to98.34% and therefore phosphorous was completely consumed.From the ANOVA results (see Table 5). Values of "Prob > F" less than 0.0500 indicate model terms are significant. In this case A, B, C, D, AB, CD, A^2, B^2, C^2, D^2 are significant model terms. Insignificant model terms, which have limited influence, such as AD and BC were excluded from the study to improve the model. Only two model terms in this case are insignificant and model for phosphorus removal percentage is desirable. The regression equations are the empirical model in terms of coded and actual factors for phosphorous removal. The results show the combined effects of MLSS and COD/N ratio were effective on removal percentage of phosphorous. According to this part; maximum removal percentage of phosphorous (91.03%) was obtained in lowest value of MLSS (2500 mg/l). COD/N ratio in highest value (25) is desirable for dairy wastewater treatment. Also other part shows the combined effects of MLSS and cycling time were influenced on removal percentage of phosphorous. Maximum phosphorous removal percentage of factor (89.03%) was shown in the lowest MLSS and also in the middle of cycling time (2500 mg/l and 35 h). These are confirming the results of experiments. The combined effects of COD/N ratio and aeration time on removal percentage of phosphorous showed that the maximum phosphorous removal percentage (92.77%) was shown in the highest values of COD/N ratio and aeration time (25 and 50 min/h). Generally in anoxic condition maximum phosphorous removal percentages were occurred. In this part; incontrary to COD and TKN removal percentages maximum value for aeration time (50 min/h) is desirable and it is good that phosphorous removal percentage is desirable in aerobic and anaerobic conditions together. Also the combined effects of aeration time and cycling time on removal percentage of phosphorous showed that 91.3% of phosphorous removal percentage was obtained in maximum and middle values of aeration time and cycling time (50 min/h and 35 h). Aeration time in maximum value is good for wastewater treatment. About COD/N ratio maximum value in all experiments is desirable for highest removal of nutrients from dairy wastewaters.

An increase in aeration time causes a decrease in the anaerobic time when the PAOs accumulate poly hydro butyrate (PHB) from the produced volatile fatty acids (VFAs). In this process, glucose as the individual source of VFAs requires sufficient time for acidification. Another reason for decrease in phosphorous removal at high aeration time was due to presence of nitrate, which inhibits the fermentation processes producing VFAs in the anaerobic zone. The anaerobic/anoxic operation provided the suitable condition for the enrichment of phosphate accumulating organisms [25]. Therefore, to obtain high removal percentages of phosphorus, anaerobic conditions are required for the uptake and storage of phosphorus to be easily on biodegradable organic matter (structure of sludge in batch conditions of wastewater treatments) [25]. SBR under alternating anaerobic/anoxic condition was used to achieve biological removal of phosphorus from wastewater samples. Phosphorus removal efficiency of 94% indicated that SBR was very desirable to receive high phosphorus removal percentage [26]. In addition to, the biological removal of phosphorus from nutrient-rich wastewater using granular sludge has been investigated by Yilmaz et al. [27]. The removal efficiency of soluble phosphorus was 89%. However, the high suspended solids in the effluent limited the overall removal efficiency to 74% for TP. The good nutrient removal was achieved through the process known as simultaneous nitrification, denitrification, and phosphorus removal [27]. As the nitrification gradually improved, more carbon was needed to denitrify the increasing amount of NO_x produced. Therefore, the extra acetate was gradually increased from 330 to 750 mgCOD/l. It shows similar to present study that COD/N ratio in higher value is better for maximum removal percentage of phosphorus from wastewaters.

3. 3. 2. Sludge Volume Index (SVI) SVI was constant to be about 44.03 to 51.62 ml/g during all the experiments and minimum value (best condition) of this response was obtained in the first run (MLSS 4500 mg/l, COD/N ratio 25, aeration time 50 min/h and cycling time 20 h) and maximum value was 51.62 ml/g in run 18 that it was not suitable for wastewater sludge. Although difference of minimum and maximum values for SVI in these batch experiments are insignificant. In general, results demonstrate it is better that SVI will be obtained in minimum values with stable conditions. Dairy wastewaters were treated in successive cycles each lasting a few hours according to runs from design expert software [28]. At the end of every cycle, the biomass was settled before the effluent is withdrawn. Granules with excellent settling properties are essential for the effective functioning of biological systems treating wastewater because of sludge bulking is a common problem encountered in suspended growth biological nutrient removal processes [29]. Microbial granules can form under alternating aerobic–anaerobic conditions. The sludge volume index (SVI) of aerobic granules can be lower than 51.62 ml/g in these experiments, which is much lower than that of conventional bioflocs. In fact, alternating aerobic–anaerobic conditions were found to promote the growth of good settling bioflocs and contribute to increases of denitrifying fraction in the bioflocs. This implies that from an engineering perspective, the settleability of sludge can be improved significantly through the formation of aerobic granules so that it can be settled in a more compact clarifier [28]. In these experiments; from the analysis carried out (see Table 5), a quadratic model was selected to describe the variation of the response. The model terms, A, C, D, AB, AC, AD, BC, BD, CD, B^2, C^2, D^2 are significant factors.

Insignificant model terms were found to be B, implying that only COD/N ratio have smaller effect on the response. The regression equation obtained in term of coded factor for SVI is presented in Table 5.

According to results in all runs, SVI was low and there are not significant different between results. Generally in these experiments SVI was around 47.16 to 48.67 ml/g. Therefore SVI almost became stabilized at 48 ml/g. These are best conditions for sludge in wastewater treatment. In this part, inverse with other sections for SVI; MLSS is desirable in lowest value and it is similar to before responses.

According to results, generally COD/N ratio in high value (25) are optimum for best results and also it is excellent for nutrient removal from wastewaters. SVI in this section is 48.67 ml/g.MLSS and aeration time together are affected on SVI in wastewaters. Both these factors are desirable in high value (4500 mg/l and 50 min/h). Therefore, MLSS does not have very significant effect on SVI incompare to other factors. These results are confirmed that for obtain minimum value of SVI; high amount of aeration time is essential. Therefore, a reverse impact of the aeration time on SVI was observed as the variable increased. Increasing in aeration time caused an increase in the response due to high sludge production as well as the favored condition for denitrification resulted from high DO consumption rate.

Therefore, improve in SVI values was greatly influenced by the operating DO concentration (aeration time). In addition, continuous operation of batch systems under high aeration time, the nitrifiers and fast-growing heterotrophic bacteria gradually enriched. Consequently, the attached-growth sludge in batch system became thicker, and the dissolved oxygen transferred into the inner of the sludge was hindered. Depth of the anoxic layer of the biofilm depends on the oxygen supply rate and depletion rate by microbes. Hence, nitrification takes place at the carrier interface, which is an aerobic layer, and denitrification occurs in the deeper layer of the biofilm, where anaerobic bacteria are present. As a result, the SVI via SND increased [30].

According to C, E and F sections; 20 h of cycling time is desirable for do the treatment process. It is optimum for economy respect to removal nutrient in low time in the best conditions. Also, COD/N ratio in D and E sections is reported in high value at minimum value of SVI. SVI in all sections is (48.67, 47.8, 48.06, 47.8, 48.06 and 47.16 ml/g) that all of these values are very important and desirable for wastewater treatment plant to prevent sludge to be condensed.

Activity of nitrifiers in high value of aeration time and also COD/N ratio was confirm in study of Xia et al. [31]. In SBRs strategy was involved in stepwise increase in ammonium (NH_4^+-N) concentration in the influent. Results showed that the activity of nitrifying bacteria significantly increased with gradually increasing NH_4^+-N. SVI was below 25 ml/g, the mean settling velocity was in excellent value. The NH_4^+-N removal efficiency averaged above 99% and total nitrogen (TN) removal was greatly enhanced and could reach to 68% [31]. Also Dulekgurgen et al.[32] in SBR system obtained a sludge volume index less than 40 ml/g, with securing a removal efficiency of 95% for carbon, 99.6% for phosphate, and 71% for nitrogen. When SVI started to decrease, the structure of the activated sludge flocs started to change from fine particles to small granules. Therefore, minimum value for SVI was showed in high value of COD/N ratio and aeration time that is used in granulation [32].

3. 3. 3. MLSS/ MLVSS Mixed Liquor Volatile Suspended Solids (MLVSS) is a part of Mixed Liquor Suspended Solids (MLSS). In fact, the MLVSS concentration displayed a similar trend to that of MLSS, but the MLVSS: MLSS ratio decreased with respect to time [33] and samples after MLSS measurement were used to burn and calculation of MLVSS. Therefore, MLSS/MLVSS results in experiments for four factors are near together. Results show the comparative effects of four factors (MLSS (A), COD/N (B), aeration time (C) and cycling time (D)) on MLSS/MLVSS in the batch runs. In run 3, the maximum values of the MLSS/MLVSS concentration were obtained to be 5646.97 and 4435.99 mg/l, respectively. Also the minimum values of the response were found to be 2722.09 and 2058.12 mg/l (run 21) and 2756.5 and 1965.63 mg/l (run 4). From the ANOVA results shown in Table 5, the model terms including A, B, C, D, AB, AC, AD, BC, BD, CD, A^2, B^2, C^2, D^2 for MLSS and A, B, C, D, AB, AC, AD, BD, CD, A^2, B^2, C^2, D^2 for MLVSS are significant model terms. Only BC interaction is insignificant model term for MLVSS response therefore was excluded from the study to improve the model. The empirical model in terms of coded factors for MLSS/MLVSS is stated in Table 5.

MLSS and COD/N ratio were found as the most influential factors on MLSS/MLVSS in the design space studied. These factors in A, B, C, D and E sections were demonstrated that in 4500 mg/l of MLSS and in the total range of COD/N ratio; MLSS/MLVSS have maximum values. Results showed a simultaneous increase in the variables MLSS and COD/N ratio improved the values of sludge concentrations in batch systems. Therefore, MLSS in highest value is importance factor for the best MLSS/MLVSS in wastewater treatment and these responses were increased with increasing in granule concentration. It is not essential to separate waste sludge from the system in this period. Effect of aeration time and cycling time on MLSS/MLVSS are desirable in lowest values.

TABLE 4. Analysis of variance (ANOVA) for the response surface quadratic model

Response	Source	Sum of Squares	df	Mean Square	F Value	p-value, Prob > F
PO_4^{3-} removal percentage, %	Model	689.23	12	57.44	50.82	< 0.0001
	Residual	19.21	17	1.13		
	Lack of Fit	14.41	12	1.2	1.25	0.4286
	Pure Error	4.8	5	0.96		
	Total	708.44	29			
SVI, ml/g	Model	86.46	13	6.65	166.54	< 0.0001
	Residual	0.64	16	0.04		
	Lack of Fit	0.32	11	0.03	0.46	0.8681
	Pure Error	0.32	5	0.06		
	Total	87.1	29			
MLSS, mg/l	Model	18253558.51	14	1303825.61	581.94	< 0.0001
	Residual	33607.32	15	2240.49		
	Lack of Fit	26334.28	10	2633.43	1.81	0.2659
	Pure Error	7273.04	5	1454.61		
	Total	18290000	29			
MLVSS, mg/l	Model	11006379.75	14	786169.98	4708.45	< 0.0001
	Residual	2504.55	15	166.97		
	Lack of Fit	1787.51	10	178.75	1.25	0.4272
	Pure Error	717.04	5	143.41		
	Total	11010000	29			

TABLE 5. ANOVA results for response parameters

response	R^2	Adjusted R^2	Predicted R^2	Adequate Precision	SD	Mean	CV	PRESS
PO_4^{3-}, %	0.97	0.95	0.89	35.61	1.06	88.36	1.2	75.28
SVI, ml/g	0.99	0.99	0.98	56.89	0.2	49.01	0.41	2.17
MLSS, mg/l	1	1	0.99	88.78	47.33	3728.98	1.27	162158.65
MLVSS, mg/l	1	1	1	273.49	12.92	3071.07	0.42	11328.59

X_1, MLSS, mg/l; X_2, COD/N/P; X_3, Aeration time, min/h; X_4, Cycling time, h; SD, standard deviation; DF, degree of freedom; CV, coefficient of variation; *PRESS*, predicted residual error sum of squares.

MLSS/MLVSS as a function of these two factors were investigated while the other variables were kept constant and value of MLSS/MLVSS were 3951.80 and 3403.27 mg/l in 40 min/h aeration time and 20 h cycling time. Also nutrient removal and MLSS/MLVSS of each run was mainly achieved to high value in the aerobic part because increase of aeration time to caused nitrification of autotrophic bacteria and assimilation of carbonaceous bacteria [13]. In addition, formation and performance of granular sludge in SBR treating an abattoir wastewater was studied by Cassidy and Belia [33]. Influent concentrations averaged 1520 mg/l MLVSS, 7685 mg/l COD, 1057 mg/l TKN and 217 mg/l TP. The granules had a density of 62 gVSS/l and a sludge volume index (SVI) of 22 ml/g. Removal of COD and TP were over 98%, and removal of TKN and MLVSS were over 97%. Nitrification and denitrification occurred simultaneously during reaction. From an operational standpoint, excessive loss of biomass can upset the balance of the COD, N, and P removing capacity [33]. Removal of COD, N, and P were over 95% during startup, indicating that the strategy was effective. Therefore, MLSS/MLVSS in high value was confirm for the best treatment of nutrients in wastewaters. The conventional SBRs treating wastewaters with flocculating sludge can be used for desirable nutrients removal in high value of MLSS/MLVSS.

4.CONCLUSION

In this study, phosphorus removal occurred inside granules in aerobic/anaerobic phases in batch systems. RSM experimental design was employed to obtain the operational conditions for the maximum removal rates of this substance. Variables for wastewater treatment process were: MLSS, COD/N ratio, aeration time and cycling time. These variables were studies in laboratory and optimum value of each variable for obtaining the maximum value of responses were determined. Results showed that the optimum conditions were found to be 98.34% PO_4^{3-} removal and amount of 44.03 ml/g SVI, 5646.97 mg/l MLSS and 4435.99 mg/l MLVSS in batch runs, respectively. Analysis using Design Expert revealed the obtained approximating models for PO_4^{3-}, SVI, MLSS and MLVSS to be satisfactorily fitted, with R^2 of 0.97, 0.99, 1 and 1, respectively. This study has shown that the use of combined anaerobic/aerobic conditions for phosphorus removal from dairy wastewater could be an effective and economical method in batch system.

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Phosphorus Removal from Dairy Wastewater in Batch Systems under Simultaneous Aerobic/Anaerobic Conditions: Application of Response Surface Methodology

M. Amini

Department of Environmental Science, Faculty of Natural Resources, University of Jiroft, Jiroft, Iran

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چکیده

موضوع این مقاله بررسی میزان حذف فسفر از یک فاضلاب لبنی درسیستم ناپیوسته تحت شرایط همزمان هوازی / بی هوازی است. سیستم شامل رآکتور ناپیوسته (SBR) و تحت شرایط متغیر هوازی و بی هوازی کار می کند. به منظور آنالیز فرآیند؛ چهار متغییر مستقل MLSS ، نسبت COD/N، زمان هوادهی و زمان دوره تصفیه و چهار متغییر وابسته به عنوان نتیجه تاثیر متغییرهای مستقل بررسی شدند. آزمایشات با استفاده از طرح نقاط مرکزی طراحی و داده ها با استفاده از روش پاسخ سطحی آنالیز شدند. درصد حذف بهینه فسفر ٪۹۸.۳٤ به ترتیب در SVI ، MLSS وMLVSS: ٤٤.۰۳ml/g ، ٥٦٤٥.۹۷mg/l و ٤٤۳٥.۹۹ mg/l به دست آمدند. بزرگنمایی بالای میکروسکوپ الکترونی تنوع در اشکال میکروارگانیسم های حذف کننده همزمان کربن، نیتروژن و فسفر را در شرایط ناپیوسته نشان داد که شامل میکروارگانیسم های باکتریایی گرد، میله ای و شبکه ای بودند. نتایج این تحقیق درصد بالای حذف فسفر را از فاضلاب لبنی با استفاده از گرانول های میکروبی تحت شرایط متغییر هوازی و بی هوازی در سیستم ناپیوسته نشان داد.

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