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Short Communication

Identifying the influential priority of the factors governing PHB production by activated sludge with integration of uniform design and grey relational analysis

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ABSTRACT

Uniform design coupled with grey relational analysis approach was used in this study to evaluate the influence of four main factors governing PHB production by activated sludge in a sequencing batch reactor (SBR). Influent chemical oxygen demand (COD), nitrogen and phosphate concentrations and initial pH were considered for process evaluation. First, a mixed-level uniform design methodology was employed to arrange the experiments. Then, a grey relational analysis method was used to investigate the influential priorities of these four factors on PHB yield, production rate and the overall performance. The results indicate that influent COD concentration was the most important factor governing PHB yield, while pH showed stronger impacts on the PHB production rate and the overall PHB production performance. This approach provides an effective way to identify the influential priority of operating factors on PHB production process, and it can also be appropriately extended to other complex biological wastewater treatment processes.

1. Introduction

Polyhydroxybutyrate (PHB) is a common bacterial storage polymer in pure cultures and activated sludge [1–6]. It offers many advantages over conventional petrochemical-based plastics such as complete biodegradability, high biocompatibility and abundant source availability [5,7–9]. However, the high cost of manufacturing PHB from pure cultures significantly weakens its market competitiveness [7,10]. Compared with pure-culture approach, producing PHB from activated sludge with organic wastes is attracting increasing interests from researchers and engineers due to its high economical viability [5,11–14]. However, this process is rather complex and can be influenced by many operating factors, namely the influent organic load, nutrient concentrations, pH, feeding pattern and so on [7,10,15,16]. Therefore, to maximize PHB production, it is necessary to optimize these operating factors. In this regard, uniform design methodology in combination with grey relational analysis may provide a useful tool.

Uniform design methodology is widely recognized as one of the best statistical design techniques [17], and has been extensively applied for influencing factor analysis due to its high-efficiency, robustness and flexibility [18,19]. It is capable of producing samples with high representation in the studied experimental domain and accommodating the largest possible number of levels for each factor [18], thus effective evaluation of multiple parameters can be achieved at limited number of experiments [20]. On the other hand, grey relational analysis is well known for its capability of finding out complicated interrelationships among multiple factors and variables at minimal data required [21–23]. In grey relational analysis, the influential degree of a compared series on the reference series is represented by the relative distance between them in an imaging grey space, thus prior assumption about the distribution type is not needed [24].

Considering the limitations and advantages of these two statistical methodologies and the complexity of the activated sludge PHB-producing process, it is, therefore, expected that an integration of the two methods may open a useful avenue for such factor analysis and evaluation. Therefore, this study aims to develop a novel approach for quantitative evaluation of the influences of operating factors on PHB production process. Uniform design methodology is first employed to design the experiments, and then grey relational analysis was used for factor priority evaluation.

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2. Material and methods

2.1. Sludge and wastewater

A laboratory-scale sequencing batch reactor (SBR), with a working volume of 2 L, was utilized to enrich PHB-producing activated sludge under periodic conditions. The reactor was operated in successive cycles of 4 h each. Each cycle consisted of 3 min of influent feeding, 222 min of aeration, 10 min of settling, and 5 min of effluent withdrawal. The hydraulic retention time was 8 h and the temperature was maintained at 20 °C. Air was introduced through an air diffusion by an air pump at the bottom of the reactor.

The composition of the synthetic wastewater is listed in Table 1, and trace element was supplied as described by Fang et al. [25]. The pH of the mixed liquors was kept constantly by feeding 4 M NaOH or 2 M HCl solutions via respective peristaltic pumps.

2.2. Experimental arrangement with uniform design

The experiments were arranged by uniform design [17]. The influent chemical oxygen demand (COD), nitrogen and phosphate concentrations and initial pH were selected as independent inputs variables. Mixed levels uniform design was adopted due to the different level numbers of the experimental factors [19], where two 12-levels factors (influent COD and nitrogen concentrations) and two 6-levels factors (influent phosphate concentration and initial pH) were included. Thus, the $U_{12}(12^2 \times 6^2)$ uniform design table was used (Table 2). The PHB yield, PHB production rate and the overall PHB production performance were considered as output variables.

2.3. Grey relational analysis

Grey relational analysis approach was used to evaluate the influence of the above four factors on PHB production. Based on the experimental results of uniform design, the compared series and the reference series were selected in the first place. To evaluate the influential degrees, the four operating factors were taken as the compared series ($Y_j = \{Y_j(k) | j = 1, 2, \ldots r;\ k = 1, 2, \ldots n\}$, where Y_j represents the compared series), whereas the output variables of PHB yield, PHB production rate and overall PHB production performance were used as the reference series ($Y_0 = \{Y_0(k) | k = 1, 2, \ldots n\}$, where Y_0 represents the reference series).

After that, it is necessary to normalize the original data in the range between zero and one prior to grey relation analysis. Therefore, the input four operating factors and the output variables were normalized to have the same order for the reduction of error. The following three equations were used for the normalization [23,26]:

(A) If the expectancy is larger-the-better, then it can be calculated using the following equation:

$$y_j(k) = \frac{Y_j^{(0)}(k) - \min Y_j^{(0)}(k)}{\max Y_j^{(0)}(k) - \min Y_j^{(0)}(k)} \quad (1)$$

(B) If the expectancy is smaller-the-better, then it can be expressed as:

$$y_j(k) = \frac{\max Y_j^{(0)}(k) - Y_j^{(0)}(k)}{\max Y_j^{(0)}(k) - \min Y_j^{(0)}(k)} \quad (2)$$

(C) If the expectancy is nominal-the-better, then it can be expressed as:

$$y_j(k) = 1 - \frac{|Y_j^{(0)}(k) - a|}{\max\{\max[Y_j^{(0)}(k)] - a, a - \min[Y_j^{(0)}(k)]\}} \quad (3)$$

where y_j is the grey relation value, $\min[Y_j^{(0)}(k)]$ is the minimum value of $Y_j^{(0)}$, $\max[Y_j^{(0)}(k)]$ is the maximum value of $Y_j^{(0)}$ and a is the best value of $Y_j^{(0)}(k)$.

Based on the normalized data, the grey relational coefficients were calculated to establish the relationship between the compared series and the reference series. The grey relational coefficients $\gamma(y_0(k), y_j(k))$ can be computed by Deng [27]:

$$\gamma(y_0(k), y_j(k)) = \frac{\min_j \min_k |y_0(k) - y_j(k)| + \xi \max_j \max_k |y_0(k) - y_j(k)|}{|y_0(k) - y_j(k)| + \xi \max_j \max_k |y_0(k) - y_j(k)|} = \frac{\Delta_{\min} + \xi \cdot \Delta_{\max}}{\Delta_{0j}(k) + \xi \cdot \Delta_{\max}} \quad (4)$$

where $y_0(k)$ is the series of experimental results, $y_j(k)$ is series of the influenced factors, $\Delta_{0j}(k) = |y_j(k) - y_0(k)|$, also, $\Delta_{\max}$ is defined as the element of maximum value in the matrix and $\Delta_{\min}$ is the element of minimum value in the matrix, ξ $(0 < \xi \leqslant 1)$ is a distinguishing coefficient used to adjust the range of the comparison environment and to control the level of differences of the relational coefficients. Here, ξ value of 0.5 was taken for calculation because it is generally adopted in most studies [23].

Finally, the grey relational grade was determined as the average value of all grey relational coefficients, which represents the influential priority of a factor on the entire system. The grey relational grade was calculated by the following equation:

$$\gamma(y_0, y_j) = \frac{1}{n} \sum_{k=1}^{n} \gamma_{0j}(k) \quad (5)$$

In this way, the influential priorities of the four factors on PHB yield, PHB production rate and overall PHB production performance could be identified.

Table 1
Composition of the synthetic wastewater.

Component	Concentration (mg/L)
Acetate	1000
NH_4Cl	25
KH_2PO_4	5
$MgSO_4$	10
Trace element solution	1 (mL/L)

Table 2
Uniform design matrix defining COD, nitrogen, phosphate concentrations in the influent and initial pH ($U_{12}(12^2 \times 6^2)$).

Run	Coded values[a]				Real values			
	X_1	X_2	X_3	X_4	x_1	x_2	x_3	x_4
1	1	8	5	6	200	45	38	8.5
2	2	3	4	5	600	20	30	8.0
3	3	11	2	4	1000	60	14	7.5
4	4	6	1	3	1400	35	6	7.0
5	5	1	6	2	1800	10	46	6.5
6	6	9	4	1	2200	50	30	6.0
7	7	4	3	6	2600	25	22	8.5
8	8	12	1	5	3000	65	6	8.0
9	9	7	6	4	3400	40	46	7.5
10	10	2	5	3	3800	15	38	7.0
11	11	10	3	2	4200	55	22	6.5
12	12	5	2	1	4600	30	14	6.0

[a] X_1 = COD (mg/L); X_2 = NH_4^+ (mg/L); X_3 = PO_4^{3-} (mg/L); X_4 = initial pH.