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ORIGINAL ARTICLE

A new approach for development of kinetics of wastewater treatment in aerobic biofilm reactor

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Abstract Biofilm process is widely used for the treatment of a variety of wastewater especially containing slowly biodegradable substances. It provides resistance against toxic environment and is capable of retaining biomass under continuous operation. Development of kinetics is very much pertinent for rational design of a biofilm process for the treatment of wastewater with or without inhibitory substances. A simple approach for development of such kinetics for an aerobic biofilm reactor has been presented using a novel biofilm model. The said biofilm model is formulated from the correlations between substrate concentrations in the influent/effluent and at biofilm liquid interface along with substrate flux and biofilm thickness complying Monod's growth kinetics. The methodology for determining the kinetic coefficients for substrate removal and biomass growth has been demonstrated stepwise along with graphical representations. Kinetic coefficients like K, k, Y, b_t, b_s, and b_d are determined either from the intercepts of X- and Y-axis or from the slope of the graphical plots.

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Introduction

Biofilms are natural, heterogeneous, multiphase material consisting primarily of microbial cells in slouble extracellular polymeric substances (EPS), and interstitial water (Hinson et al. 1996) attached on a solid surface which are used to combat high organic strengths in wastewater. Due to some, inherent advantages like low energy consumption, easy maintenance, better physical and chemical stability, and excellent biomass retention capacity, importance of biofilm is increasing in biological treatment of wastewater. A unique feature of biofilm process lies in the operation of continuous flow mode with minimal biomass loss compared to the traditional suspended growth bioreactor. Biofilm has an additional advantage having no contamination at its inner depth under inhibitory condition. There is a limited number of methods available for determining the kinetic coefficients required for the solution of the model of an aerobic biofilm bioreactor. In the process design of an aerobic biofilm bioreactor, the kinetic coefficients act as an essential tool for predicting the substrate utilization and biomass growth under a specific condition. The biomass yield coefficient (Y) indicates the portion of the biomass growth from the utilization of substrates. Specific biomass growth rate (μ) represents the biomass growth rate per unit biomass per day. Maximum value of specific biomass growth rate (μ_{max}) is attained when substrates are not in limiting condition. Half-velocity constant (K) is the substrate concentration corresponding to half of maximum specific biomass growth rate (μ_{max}). The maximum specific substrate utilization rate (k) represents the substrate

utilization rate per day per unit biomass corresponding to μ_{max}. Biomass decay coefficient (b_d) is the rate of biomass loss due to endogenous decay and is taken into consideration for determining the net biomass growth rate.

In the biofilm process, the rate of total biomass loss (b_t) consists of the rates of biomass shear loss (b_s) and biomass decay (b_d).Y, k, K, b_t, b_s, and b_d are the kinetic coefficients required for the solution of a specific biofilm model. Substrate mass balance is accomplished with the simultaneous occurrence of both diffusion and utilization of substrates. The molecular diffusion coefficient of the substrate in water (D) and in the biofilm (D_f) represent internal mass transport resistance in the bulk liquid and in the biofilm, respectively. The thickness of the effective diffusion layer of the bulk liquid (L) represents the external mass transport resistance. The biomass concentration (X_f) in the biofilm can be determined from the total biofilm thickness (L_f). Biofilm thickness primarily depends on the substrate concentration in the bulk liquid and the liquid hydraulics. The biomass per unit surface area, i.e., (X_fL_f) is constant indicating the steady state condition of biofilm. Various research studies have been reported on modeling of biofilm reactor (Williamson and McCarty 1976; Rittmann and McCarty 1980; Suidan and Wang 1985; Strand 1986; Kim and Suidan 1989; Golla and Thomas 1990; Heath et al. 1990; Saez and Rittman 1991; Lee 1997; Rauch et al. 1999; Tsuno et al. 2002; Pritchett and Dockery 2001; Perez et al. 2005; Shaoying et al. 2005; Hsien and Lin 2005; Mudliar et al. 2008; Jiang et al. 2009; Qi and Morgenroth 2005; Rao et al. 2010; Liao et al. 2012; Eldyasti et al. 2012; Gullicks et al. 2011), but no simplified method was developed for determining the kinetic coefficients. Indeed, the kinetic coefficients are needful for solving the biofilm model and hence for the process design of the biofilm reactor. A kinetic study was performed in an aerobic biofilm bioreactor for evaluation of efficiency of COD removal from organic wastewater (Dey and Mukherjee 2010). The half-velocity constant (K) was determined from the experimental results using Monod's kinetics. The inhibition kinetics was used in another research work for predicting the response of biofilms to toxic compounds in wastewater (Gheewala and Annachhatre 2003). Although the relationship between the effectiveness factor of biofilm and the half-velocity constant (K) was shown, no guideline was stipulated for arriving the values of K. Hsien and Lin (2005) has conducted one batch test for determining the kinetic coefficients for simulation study in a biofilm reactor.

Therefore, development of kinetics for an aerobic biofilm reactor for the treatment of wastewater finds its relevance and having an importance in this research work. The present method employs a series of biofilm model equations complying Monod's growth kinetics. The graphical solutions for evaluating the kinetics coefficient like K, k, Y, b_t, b_s, and b_d for both the substrate removal and biomass growth have been presented sequentially.

Materials and methods

The schematic diagram of Biofilm model considering purely attached biomass is shown in Fig. 1.

The concept of biofilm model is based upon three basic assumptions, i.e., (1) the nature of the kinetics for substrate utilization in suspended growth and attached growth are in similar nature, (2) external substrate transport from the bulk liquid to the biofilm through biofilm Liquid interface follows Fick's first law of molecular diffusion and (3) the substrate transport within the biofilm follows the Fick's second law of molecular diffusion. Now, the equation for external mass transport according to Fick's first law

$$J = \frac{D}{L} \times (S_0 - S_s) \tag{1}$$

where, J is the substrate flux into the biofilm (mg/cm^2/day), D is the Molecular diffusion coefficient in liquid (cm^2/day), L is the thickness of effective diffusion layer (cm), S_0, S_s is the substrate concentrations in the bulk liquid and at the biofilm liquid interface, respectively (mg/cm^3). Now, from the substrate mass balance within the biofilm is.

$$S_0 - S_w - aJ\theta = 0 \tag{2}$$

where, a is the specific surface area of supporting media (cm^{-1}), θ is the Hydraulic Retention time (h), S_w is the effluent substrate concentration (mg/cm^3), therefore, from Eq. 2 we get,

$$J = \frac{S_0 - S_w}{a\theta}$$

Again from the mass balance equation for substrate in the biofilm considering Monod's' kinetics we get,

$$\frac{d^2 S_f}{dz^2} = \frac{kX_f S_f}{D_f (K + S_f)} \tag{3}$$

where, S_f is the substrate concentration at a point in the biofilm (mg/cm^3), k is the maximum specific rate of

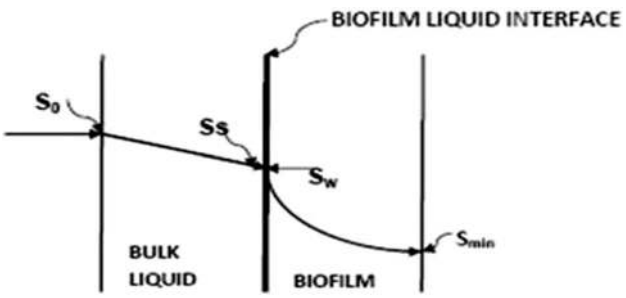

Fig. 1 Schematic diagram of biofilm model

substrate use (per day), X_f is the active biomass density within the biofilm (mg/cm^3), D_f is the molecular diffusion coefficient of the substrate in the biofilm (cm^2/day), K is the half-velocity coefficient (mg/cm^3).

Referring Fig. 1, the boundary conditions for solving above second-order differential Eq. (3) may be taken as below:

At the attachment surface (i.e., at $z = 0$) there will be no flux, i.e., $\frac{dS_f}{dz} = 0$.

At the biofilm/liquid interface, i.e., at $z = L_e$,

$$J = D_f * \frac{dS_f}{dz} = D * \frac{(S_0 - S_s)}{L}$$

Now, the solution of the above second-order differential Eq. (3) on substrate utilization considering both the boundary condition within the biofilm is shown below:

$$J = \sqrt{2kX_f D_f[(S_s - S_w) + K \ln[(K+S_w)/(K+S_s)]}$$

$$J^2 = 2kX_f D_f[(S_s - S_w) + K \ln[(K+S_w)/(K+S_s)] \quad (4)$$

Analytical procedure for determining the kinetic coefficients

To determine the kinetic coefficient from, the Biomass growth curve, Eqs. (1), (2) and (4) can be systematically used in the following Steps.

Step 1

$$\mu = \frac{(X_2 - X_1)}{(X_1) * \theta} \quad (5)$$

μ is the specific growth rate of biomass for biofilm bioreactor (h^{-1}), X_2 is the final Biomass density for attached phase at the end of batch period (mg/cm^3), X_1 is the initial Biomass density for attached phase at the end of batch period (mg/cm^3), θ is the batch period under consideration (h).

Now, biomass density can be determined by washing the biofilm attached surface with 25 ml 0.1 (N) NaOH after heating the solution at 80 °C (Lazarova et al. 1994; Cox and Deshusses 1999) and subsequently by measuring the protein concentration with Lowry's method as below:

Steps for determining the protein concentration by Lowry's method:

1. Preparation of following reagents are as follows:
   (a) 5 % Na_2CO_3 solution.
   (b) $CuSO_4$ solution (0.5 mg $CuSO_4$, $5H_20$ + 100 ml of 1 % sodium potassium tartrate solution)
   (c) Alkali copper reagent(50 ml of reagent a + 2 ml of reagent b)
   (d) Folin reagent solution (folin reagent:distilled water = 1:1
   (e) 1 (N) NaOH solution
   (f) Standard BSA solution of concentration 200 mg/l.
2. 0.5 ml of the cell suspension (BSA solution) + 0.5 ml 1(N) NaOH solution were kept in boiling water bath for 5 min.
3. The content was then cooled in cold water. A 5 ml of freshly prepared reagent (c) was added to the content and allowed to stand for 10 min.
4. Now 0.5 ml folin reagent solution was added and the whole mixture was allowed to stand for 30 min for color development.
5. The blank was prepared taking 0.5 ml distilled wastewater instead of bacterial suspension and was treated in the same way as above.
6. The absorbance value was measured at 750 nm using visible spectrophotometer against distilled water blank.

BSA standards ranged between 0 and 200 µg/ml were processed in the same manner as samples for developing the calibration curve.

Step 2

The 'μ' values can be plotted in Y-axis with respect to 'S_w' values in X-axis, which should have an ascending asymptotic nature as shown in Fig. 2. The half-velocity constant K can be obtained corresponding to half of the maximum specific growth rate, i.e., μ_m, as indicated in the same figure.

Step 3

Using Eqs. 1, 2 and 4,

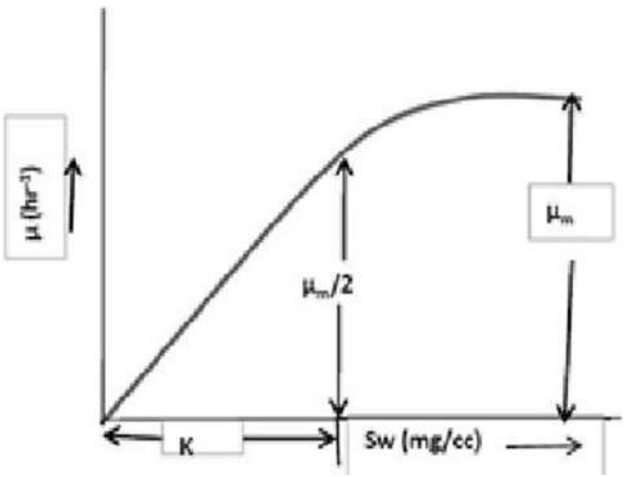

Fig. 2 Biomass growth curve for determination of half-velocity constant K

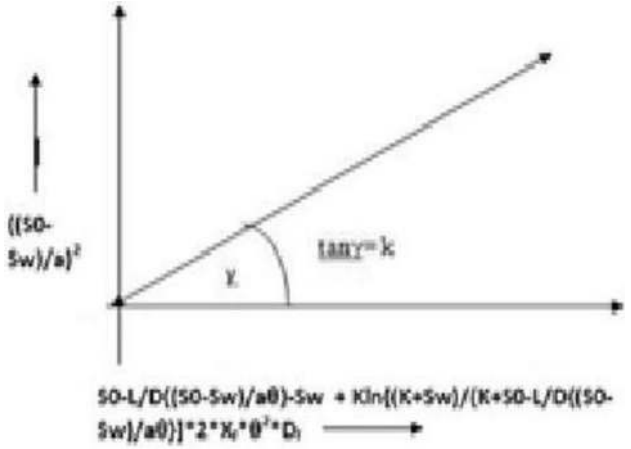

Fig. 3 Biofilm growth for determination of k and Y

$$\left(\frac{S_0-S_W}{a}\right)^2 = 2kX_fD_f\theta^2\left[\left\{S_0-\frac{L}{D}\frac{S_0-S_W}{a\theta})-Sw\right\}+ks\ln\left\{\frac{k+S_W}{k+s_o-\frac{L}{D}\left(\frac{S_0-s_w}{a\theta}\right)}\right\}\right].$$

Now, $\left(\frac{S_0-S_W}{a}\right)^2$ values can be plotted with respect to $2X_fD_f\theta^2\left[\left\{S_0-\frac{L}{D}\left(\frac{S_0-S_W}{a\theta}\right)-S_W\right\}+K\ln\left\{\frac{k+S_W}{k+s_0-\frac{L}{D}\left(\frac{S_0-s_w}{a\theta}\right)}\right\}\right]$ and a best-fit line can be drawn as shown in Fig. 3 below. The slope of a best-fit line passing through the origin will result in the value of k.

Step 4

Now the total biofilm thickness L_f can be estimated as $L_f = \frac{JY}{X_f b_t} \Rightarrow L_f = \left(\frac{S_0-S_W}{X_f a\theta bt}\right)Y$where, J is the substrate flux, X_f is the Biofilm density, b_t is the overall specific biofilm loss rate in (h^{-1}), $\mu_m = kY$ or $Y = \mu_m/k$. Therefore, the value of $L_fX_f\theta$ can be plotted with respect to $\left(\frac{S_0-S_W}{a}\right)Y$ to find out b_t.

Step 5

To determine the shear loss coefficient (b_s), the sloughed off biomass from the attached surface may be equated to the final suspended biomass after batch period in the reactor. $Y{\cdot}a{\cdot}\theta{\cdot}J{\cdot}\frac{b_s}{b_t} = X$, the portion of biomass which is sloughed off from the attached biofilm and take part into the suspended one. or $\frac{Ya\theta J}{b_t} = \frac{X}{b_s}$ or, $\frac{Y(S_0-S_w)}{b_t} = \frac{X}{bs}$, where θ is the HRT (Hydraulic retention time), X is the suspended biomass contributed from the attached biofilm in the reactor. Now, $Y{\cdot}\left(\frac{S_O-S_W}{b_t}\right)$ can be plotted with respect to X as shown in Fig. 4 below. The slope of the best-fit line will denote the value of $1/b_s$.

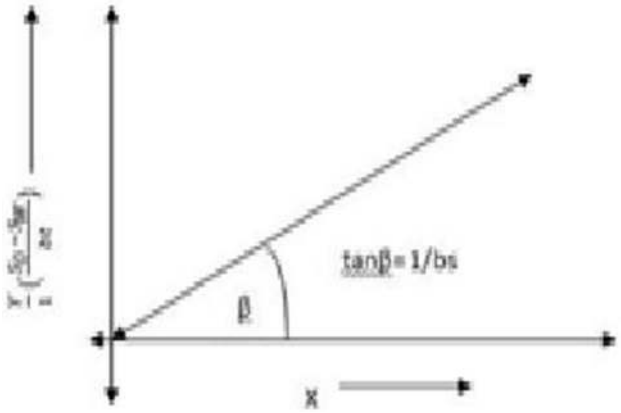

Fig. 4 Determination of b_s (Biomass shear loss)

Table 1 Semi Batch study results of Biofilm reactor

Batch slot	Batch period in hours	Attached biomass concentration (mg/cc)	Effluent COD (mg/cc)	μ (h^{-1})
1st day	0	0.278	0.112	
	3	0.370	0.92	0.11
	6	0.453	0.82	0.105
	9	0.532	0.79	0.1015
	12	0.607	0.77	0.0986
2nd day	0	0.470	0.100	
	3	0.657	0.78	0.133
	6	0.691	0.70	0.078
	9	0.768	0.60	0.070
	12	0.843	0.54	0.066
3rd day	0	0.408	0.130	
	3	0.672	0.100	0.216
	6	0.840	0.80	0.177
	9	0.888	0.75	0.131
	12	0.948	0.70	0.11

Results

The semi-batch study results are tabulated in the Table 1 below.

Illustrative example

In the laboratory, semi-batch studies on municipal wastewater in an aerobic biofilm bioreactor in three consecutive days were performed at an interval of 3 h. 'μ' is calculated from the expression $\mu = \frac{(X_{attached2}-X_{attached1})}{(X_{attached1})\theta}$ as shown in Table 1. Consequently, 'μ' *Vs.* 'S_w' graph is plotted as shown in Fig. 5 to find out μ_{max} and K.

From Fig. 5, μ_{max} and K are obtained as $\mu_{max} = 0.11$ - $h^{-1} = 2.64$ day^{-1} and $K = 60$ mg/l $= 0.06$ mg/cc. Now,

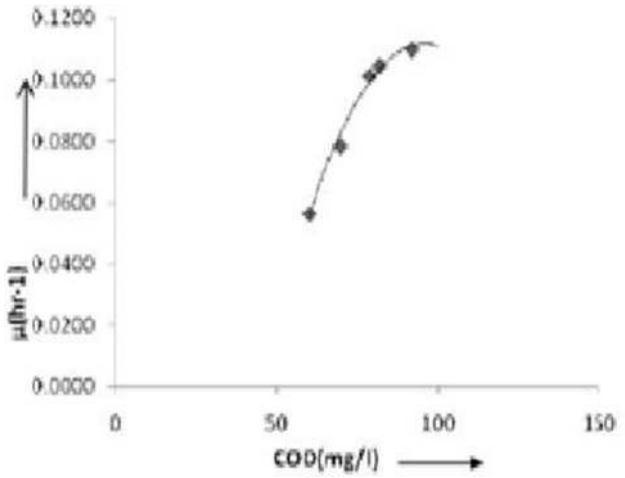

Fig. 5 Biomass growth curve for determination of half-velocity constant K

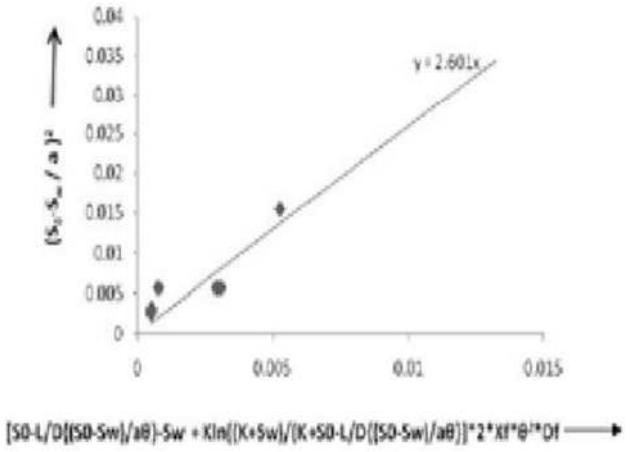

Fig. 6 Biofilm growth for determination of k and Y

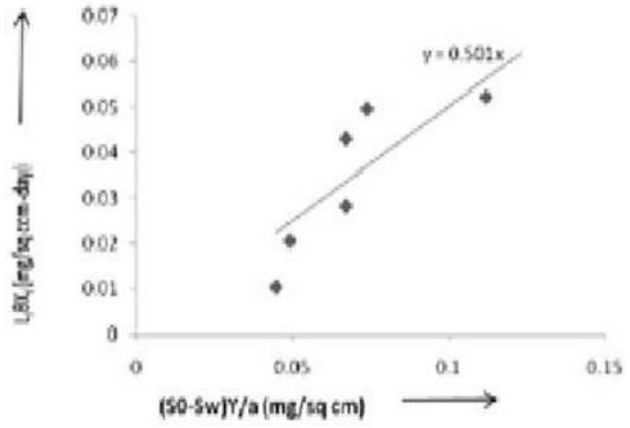

Fig. 7 Biomass balance for attached growth for determination of b_t

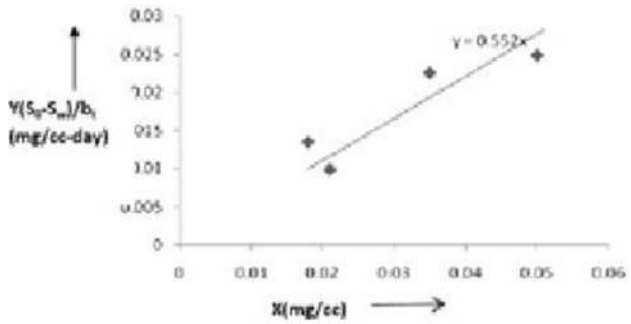

Fig. 8 Determination of b_s (Biomass shear loss)

the values of $(S_0 - S_w/a)^2$ are plotted against $2*X_f*D_f*\theta^2*[S_0 - L/D\{(S_0 - S_w)/a\theta\} - S_w + K\ln\{(K + S_w)/(K + S_0 - L/D\{(S_0 - S_w)/a\theta\}]$ as shown in Fig. 6 to determine the value of k and Y.

From the Fig. 6, it is observed that $k = 2.6$ day^{-1} Therefore, $Y = \mu_{max}/k = 2.64/2.6 = 0.99$. Now, $L_f\theta X_f$ are plotted against $(S_0 - S_w)Y/a$ as shown in Fig. 7 to determine the value of b_t.

It is observed from Fig. 7 that $1/b_t = 0.501$ day, hence total biomass loss rate, $b_t = 1.996$ day^{-1}. Now, $Y(S_0 - S_w)/b_t$ are plotted against X as shown in Fig. 8 to determine the value of b_s.

From Fig. 8, $1/b_s = 0.552$ day, hence biomass shear loss, $b_s = 1.81$ day^{-1}. Hence biomass decay $b_d = 0.186$ day^{-1}.

The simulation study was conducted with the proposed model varying the organic loadings and hydraulic retention times and the performance of the biofilm reactor was envisaged as shown in Table 2.

The kinetic coefficients and physical data in this regard are as follows.

$k = 8$ day^{-1}, $Y = 0.5$, $K = 0.01$ mg/cm^3, $b_t = 0.1$ day^{-1}, $D = 0.8$ cm^2/day, $Df = 0.64$ cm^2/day, $L = 0.01$ cm.

Discussion

The specific growth curve of biomass as shown in Fig. 2 essentially depicts the Monod's kinetics, which is valid for non-inhibitory environment. In case of inhibitory environment, the specific growth curve will show a maximum value (i.e., μ_m) and then it will move downward. As the specific growth curve significantly influences the values of μ_m and K, it should be plotted with a set of data as large as possible. The maximum specific substrate utilization rate (k) can be calculated from the slope of Fig. 3, provided the biomass density of the biofilm reactor is known. On the other hand, the 'b_t' value evaluated from the slope of Fig. 9

Table 2 Data of simulation study for evaluating performance of biofilm reactor considering proposed model

Trial no	S_0 (mg/cc)	θ (h)	X_f (mg/cc)	S_s (mg/cc)	S_w (mg/cc)	J (mg/cm^2/day)
01	0.1	2.4	40	0.089	0.017	0.83
02	0.1	0.8	40	0.078	0.041	1.77
03	0.3	2.4	40	0.269	0.058	2.42
04	0.1	4.8	40	0.094	0.009	0.46
05	0.05	2.4	40	0.044	0.008	0.42
06	0.001	2.4	40	0.0009	0.00034	0.007

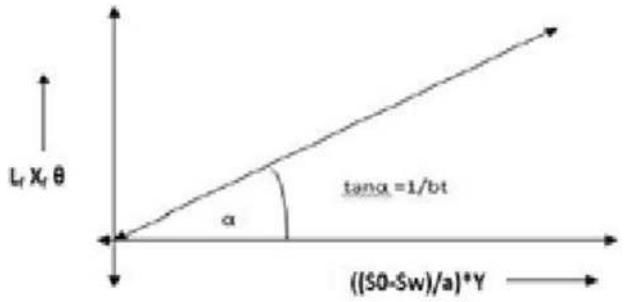

Fig. 9 Biomass balance for attached growth for determination of b_t

represents the rate of total biomass loss in the biofilm reactor. The rate of shear loss (b_s) is evaluated by plotting $Y \cdot (\frac{S_0 - S_W}{b_t})$ against X as shown in Fig. 5. In these cases also the slope of the 'best-fit line' considerably influences the values of b_s and b_d and therefore Figs. 5, 9 should be drawn with a large set of data. The use of the present method in determining the kinetic coefficient for the treatment of municipal wastewater in a biofilm reactor clearly demonstrates its efficacy and compatibility. All such kinetic data also show good removal rate of organic substrate in the biofilm reactor as indicated in Table 2. The output results of Table 2 were also compared with Pseudo-analytical solution by Rittman and McCarty (1980) with the same set of kinetic coefficients and found okay. The values of kinetic coefficients obtained in batch study are more or less in similar nature of that of suspended growth process in earlier research work (Ref Mardani et al. 2011)

Conclusion

So far, no simplified method for determining the kinetic coefficients of the biofilm bioreactor was developed in any earlier research. An attempt was made to present a simple method with straight-line equations and graphical representation for determining the kinetic coefficients of an aerobic biofilm reactor. The proposed development for kinetic coefficients of an aerobic biofilm reactor thus finds its relevance for process design of an aerobic biofilm bioreactor. The proposed method is an accurate, fast, and simplified one to find out the kinetic coefficients of an aerobic biofilm bioreactor. It is possible to simply calculate the kinetic coefficients from the graphs either from the intercepts of both x- and y-axis or from the slope of the graph applying mass balance equations both for substrates and biomass. All necessary kinetic coefficients like K, k, Y, b_s, b_t, and b_d can be determined by the proposed method required for the solution of the modeling of an aerobic biofilm bioreactor. In reality the application of biofilm bioreactor model suffers due to lack of accuracy of kinetic models and uncertainty in the kinetic parameters. Successful modeling of the biofilm reactor therefore requires accurate determination of biokinetic parameters.

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