International Scholars Journals

African Journal of Water Conservation and Sustainability ISSN 2375-0936 Vol. 13 (1), pp. 001-010, January, 2025. Available online at www.internationalscholarsjournals.org © International Scholars Journals

Full Length Research Paper

Biodegradation of Phenol by Mixed Microbial Culture in Batch Reactor: Kinetic Analysis and Performance Assessment

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Accepted 22 November, 2024

Mixed microbial culture collected from effluent treatment plant of a coke oven industry has been studied for its phenol biodegrading potential under aerobic condition in a batch reactor. The result showed that, after acclimatization, the culture could biodegrade up to 700 mg/l of phenol. The results showed that specific growth rate of microorganisms and specific substrate degradation rate increased up to 300 mg/l of initial phenol concentration and then started decreasing. The biodegradation kinetics is fitted to different substrate inhibition models by MATLAB 7.1©. Among all models, Haldane model was best fitted (Root Mean Square Error = 0.0067) for phenol degradation. The different biodegradation constants (K_s, K_i, S_m, μ_{max}, $Y_{X/S}$, k_d) estimated using these models showed good potential of the mixed microbial culture in phenol biodegradation.

Key words: Mixed culture, phenol biodegradation, kinetics, inhibition model.

INTRODUCTION

Phenolic compounds are commonly found in various industrial waste streams emanating from resin manufacturing units, coal gasification plants, petroleum refineries, coke oven industries etc (Juang and Tsai, 2006; Yan et al., 2006). Phenolics are considered in the top of the priority pollutant list given by Environmental Protection Agency (EPA, USA) (Yan et al., 2006). These compounds have high stability, high toxicity and are carcinogenic in nature, along with odor problem even at very low concentration. Phenol containing wastewater needs careful handling before they are discharged to the receiving water bodies.

The treatment of phenolic compounds from wastewater is widely practiced, although ongoing economical cost studies have shown, physical-chemical studies are costly and caused additional production of toxic chemical sludge. Biodegradation of such recalcitrant compounds from wastewater emerged as a challenging job to the researchers worldwide for obtaining an innocuous practice. Thus the biological phenol removal would be a useable alternative because it will produce no toxic end products and also use low cost technology.

Aerobic degradation of phenol has been studied extensively. For example *Pseudomonas putida* has been widely used for biodegradation of phenols (Agarry et al., 2008a) . Study on microbial degradation of phenol have shown that phenol can be aerobically degraded by wide variety of fungi and bacteria cultures such as *Candida tropicalis* (Chang et al., 1998; Ruiz-ordaz et al., 1998; Ruiz-ordaz et al., 2001), *Acinetobacter calcoaceticus* (Paller et al., 1995), *Alcaligenes eutrophus* (Hughes et al., 1984; Leonard and Lindley, 1998), *P. putida* (Hill and Robinson, 1975; Kotturi et al., 1991; Nikakhtari and Hill, 2006) and *Burkholderia cepacia* G4 (Folsom et al., 1990 ; Solomon et al., 1994).

Since the specific single bacteria seldom available in nature and also difficult to maintain in the field, it is urged that biodegradation study of phenol and phenolic

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compounds would be carried out in presence of mixed population of phenol degrading bacteria. But a very few works have been done on phenol biodegradation by indigenous mixed culture bacteria, that is, not on any specific single bacteria. In 1997, Kumaran and Paruchuri have estimated some biokinetic constants of an activated sludge based phenolic wastewater treatment systems (60 - 500 mg/l of phenol) and compared those kinetic constants for pure culture in synthetic wastewater containing phenolic compounds. Later, biokinetic parameters estimation of phenolic compounds on activated sludge (thOD 0.78 -2.58 mg /mg) was carried by respirometric method by Orupold et al. (2001). They have analyzed substrate dependent oxygen uptake data by Michaelis-Menten growth kinetic model. A further investigation was also carried out with indigenous mixed microbial culture by Tziotzios et al. (2005) in packed bed and suspended growth reactors separately. They found that, olive pulp bacteria enriched culture used as inoculums show better efficiency in packed bed reactor than suspended growth reactor corresponding to phenol degradation rate, though packed reactor was found to be more shock resistant to higher phenol concentration. Agarry et al. (2008b) have studied inhibition kinetics of phenol biodegradation by binary mixed culture of *Pseudomonas aeruginosa* and *Pseudomonas fluorescence* from steady state and washout data, where they found the maximum reaction rate to be 0.322 mg/mg/h (upto 100 mg/l of phenol). Agarry et al. (2009) have also studied the substrate inhibition kinetics of phenol by pure *P. fluorescence* in continuous bioreactor. They have observed the maximum phenol degradation rate at 100 mg/l concentration to be 0.246 mg/mg/h under washout cultivation by this organism. From their experiments it is evident that phenol biodegradation by mixed culture is more efficient than pure culture. Saravaran et al. (2008) have studied substrate inhibition kinetics of phenol by indigenous mixed culture in a batch reactor and found that substrate degradation rate is maximum at 400 mg/l of phenol with a degradation rate of 15.7 mg/L/h. They have fitted the kinetics data in both Haldane and Han-Levenspiel model, where Han-Levenspiel model was found to be better fitted for their experiment. Bajaj et al. (2009) have studied phenol degradation by mixed culture for 2.5 - 7.0 mmol/L of phenol in batch system. They have used Haldane model to estimate the kinetic parameters and calculated the yield factor to be in the range of 0.10 - 0.16 showing that phenol-containing wastewater can be treated by mixed culture used as biocatalyst.

A variety of substrate utilization and inhibition models have been used to describe the dynamics of microbial growth on phenol. The substrate inhibition models along with their mathematical forms have been described below. The earliest model on microbial growth kinetics, the Monod model (1949), relates the growth rate of microorganism to the concentration of a single growth controlling substrate represented by the following equation:

$$\mu = \frac{\mu_{max} S}{Ks + S} \quad (1)$$

Where is specific growth rate of mixed microbial culture (hr^{-1}) = $X^{-1}\frac{dX}{dT}$, S is limiting substrate concentration (mg/l), $_{max}$ is maximum specific growth rate of the culture (h^{-1}), K_S is half saturation constant (mg/l).

Different working groups (Kumar et al., 2005; Nuhoglu and Yalcin, 2005; Okpokwasili and Nweke, 2005) have proposed several mathematical models to express the culture growth and substrate utilization. Microbial growth can also be modeled by simple Monod equation (Kovar and Egli, 1998). However this equation became unpopular for growth in presence of some inhibitory substance. In such situation Haldane model are normally used to represent the growth in both lower and higher concentration of inhibitory substance. Haldane model has the form (Wang and Loh, 1999) as:

$$\mu = \frac{\mu_{max} S}{Ks + S + \frac{S^2}{Ki}} \quad (2)$$

Where *K*i is the substrate inhibition constant (mg/l).

Due to its significance, this model was widely adopted by most of the researchers. Aiba et al. (1968) proposed a model to express microbial growth rate as given by Equation (3):

$$\mu = \frac{\mu_{max} S \exp(-S/Ki)}{Ks + S} \quad (3)$$

Yano and Koga (1969) proposed a model, based on a theoretical study on the dynamic behavior of single vessel continuous fermentation subject to the growth inhibition at high concentration of rate limiting substrate, e.g. the acetic acid fermentation from ethanol, the gluconic acid fermentation from glucose, etc. The model form is given in the following equation:

$$\mu = \frac{\mu_{max}}{Ks + S + \frac{S^2}{K_1} + \frac{S^3}{K_2}} \quad (4)$$

Where K_1, K_2 are the positive constants. Similarly,

Edward (Webb, 1970) proposed the modified form of Haldane model as given by equation (5):

$$\mu = \mu_{\max} \frac{S(1+S/K)}{S + Ks + (S^2/Ksi)} \quad (5)$$

Where Ksi is the substrate inhibition constant in mg/l.

But it was found that this model did not show any significant improvisation to Haldane model (Mulchandani and Luong, 1989). Edward (Teisser, 1970) proposed another model to predict substrate inhibition at higher substrate concentration as given by the equation (6):

$$\mu = \mu_{\max} [\exp(-S/Ki) - \exp(-S/Ks)] \quad (6)$$

The model proposed by Luong (1987) as in Equation (7) appeared to be useful for representing the kinetics of substrate inhibition. Though this model is of generalized Monod type, but accounts for substrate stimulation at its both, low and high concentrations. The model has the capability to predict the values of S_m, the maximum threshold substrate concentration, above which the growth is completely inhibited (Luong 1987).

$$\mu = \frac{\mu_{\max} S}{S + Ks} \left[\frac{1 - S^n}{Sm}\right] \quad (7)$$

Where n is an empirical constant.

Han and Levenspiel (1988) proposed a model to express substrate degradation rate (Equation 8). This model involves a delay function, which has an exponential form and incorporates the critical product or substrate concentration corresponding to the inflection point on the growth (Han and Levenspiel, 1988; Okpokwasili and Nweke, 2005).

$$q = \frac{q_{\max} S [1 - S/Sm]^n}{S + Ks - [1 - S/Sm]^m} \quad (8)$$

Where q is the specific substrate degradation rate (h^{-1}), q_{max} the maximum specific substrate degradation rate (h^{-1}), Sm is the critical inhibitor concentration (mg/l) above which the reactions stops, and m and n are the empirical constants.

The corresponding form of this equation for microorganism is:

$$\mu = \frac{\mu_{\max} S [1 - S/Sm]^n}{S + Ks - [1 - S/Sm]^m} \quad (9)$$

However, a mixed microbial community is needed for complete biodegradation of phenol and phenolic compounds present in the wastewater from industries. Real wastewater treatment processes have to deal with mixture of phenolic compounds and maintenance of mixed microbial culture is easier. In view of above, the present study was carried out to investigate the biodegradation study of phenol in wastewater by mixed microbial consortium. The objective of the investigation was to study the kinetic coefficients and the rate of biodegradation of phenol by an indigenous mixed microbial system at different initial phenol concentration. It was also aimed to fit different substrate inhibition models for the phenol biodegradation and to compare the goodness of fit for those models to test for the best- fit one representing the present work. Novelty in this work is the simultaneous estimation of biodegradation kinetic coefficients by substrate inhibition models along with effective yield and decay coefficients of a mixed microbial culture taken from a real life activated sludge comprising of several microorganisms of coke oven industry wastewater treatment plant.

MATERIALS AND METHOD

Microorganisms and culture acclimatization condition

The mixed microbial sludge was collected from an effluent treatment plant of a coke oven industry situated in Durgapur, West Bengal, India. The existing effluent treatment plant is operated on the principle of suspended growth biological reactor facilitated with extended aeration system. The sludge was collected from the recirculation line as well as from the top of the sludge drying bed. The indigenous mixed microbial culture was first acclimatized to phenol, so that, microbes can produce phenol degrading enzymes in the laboratory condition. Therefore, acclimatization of the mixed culture was carried out to make the microbial cell compatible to take up phenol as the sole carbon and energy source during its biodegradation. For acclimatization of sludge with phenol, first the culture was grown at very low concentration of phenol (5 mg/l) in 250 ml conical flask containing 100 ml of Mineral Salt (MS) media with 100 mg/l of glucose and 100 mg/l of beef extract, under continuous stirring (110 rpm). The composition of MS media is given as (mg/l): $(NH_4)_2SO_4$ 230, $CaCl_2$ 7.5, $FeCl_3$ 1.0, $MnSO_4.H_2O$ 100, $MgSO_4.7H_2O$ 100, K_2HPO_4 500, KH_2PO_4 250 (pH 7.0 ± 0.2). Then glucose and beef extract concentration were gradually decreased by 20 mg/l in every batch and supplemented by increased concentration of phenol. Batch process was adopted for acclimatization of sludge. After three month of acclimatization period, the sludge was changed to mineral salt medium (MS media) with phenol as sole carbon and energy source upto a concentration of 700 mg/l.

Analytical procedure

Phenol concentration was analytically estimated by High Performance Liquid Chromatography (HPLC) (Shimadzu) equipped with Ultraviolet-Visible (UV-VIS) detector and C18 column. The mobile phase used was acetonitrile and water mixture (60:40). The flow rate of the eluent was set to 1 ml/min and the detection wavelength was 275 nm. The biomass growth in the sample was

monitored by measuring its absorbance at 600 nm wavelength using UV-VIS Spectrophotometer (Shimadzu). Then biomass concentration was calculated from a standard graph plotted as dry cell mass of microbial culture vs. optical density measured at 600 nm (Saravanan et al., 2008).

Experimental set up

All phenol biodegradation experiments using the mixed microbial culture were carried out in a 3 L capacity bioreactor with provisions for air supply for necessary aeration. In every batch the total volume of wastewater was taken as 1 L. A mini air compressor was used for the purpose of air supply. Compressed air was fed in the reactor at a rate of 5L/min by filtering the air by air-filter. Samples were withdrawn at predetermined time interval, after which, the biomass concentration and the residual phenol concentrations were analyzed. The phenol concentration was analyzed in the supernatant obtained after centrifugation of the sample for 5 min.

Batch biodegradation study

The biodegradation study was performed in the laboratory (Heritage Institute of Technology, Kolkata) to biodegrade inhibitory phenolic substances in wastewater coming out from phenol handling industries. All biodegradation experiments using the mixed culture were performed in batch bioreactor containing MS media with phenol as sole carbon and energy source at concentration range of 100 - 700 mg/l. 60ml inoculum was added to bioreactor for each set of experiment. This was accomplished by transferring directly (under aseptic conditions), freshly phenol-acclimatized culture to MS media with phenol at different concentrations (100 – 700 mg/l) at step up concentration interval of 100 mg/l. For each initial concentration of phenol, experiments were carried out in triplicate under identical condition and the average values are reported. All the experiments were done until the concentration of residual phenol in reactor was found to reach at equilibrium concentration at the specific time. For each batch of reaction, specific growth rate of the culture have been calculated and fitted in several substrate inhibition models: Monod model (1949), Haldane model (1968), Aiba model (1968), Yano and Koga model (1969), Edward model (1970), Luong model (1987) and Han-Levenspiel model (1988).

Study of decay kinetics of culture for phenol degradation

The typical growth curve in microbial assays usually shows a declining trend in the cell population after a complete consumption of substrate. But in suspended growth system where mixed microbial culture is used for biodegradation, the growth model can be represented in the following way:

$$\frac{dX}{d\theta} = Y_{x/s}\frac{dS}{d\theta} - k_d X$$

$$\frac{1}{X}\frac{dX}{d\theta} = Y_{x/s}\frac{dS}{d\theta} - k_d$$

$$\frac{1}{X}\frac{(X - Xo)}{\theta} = Y_{x/s}\frac{(So - S)}{\theta} - k_d \qquad (10)$$

Where $\frac{1}{X}\frac{(X-Xo)}{\theta}$ is the specific cell growth rate and $\frac{(So-S)}{X\theta}$ is specific substrate utilization rate (U day^{-1}), $Y_{x/s}$ is microbial yield coefficient (mg/mg), k_d is the death coefficient of microbial mass in day^{-1}.

The biodegradation data of each of the initial phenol concentration were plotted as $\frac{1}{X}\frac{(X-Xo)}{\theta}$ vs. $\frac{(So-S)}{X\theta}$ and slope of plot gave value of yield coefficient ($Y_{x/s}$) and negative intercept gave endogenous death coefficient (k_d) for 100 mg/l-700 mg/l of phenol concentration.

Modeling the kinetics of phenol biodegradation

At various initial phenol concentrations, the specific growth rates ( ) have been calculated and data were tested in different deterministic models as per Equations 1 - 9. The modeled equation were solved by nonlinear regression method using MATLAB© 7.1 and directly applied on the experimental data of specific growth rates at different initial phenol concentration. However, specific substrate de-gradation rate terms, q and q_{max} in the original equation of the Han-Levenspiel model have been replaced by μ and μ_{max}, to represent specific growth rate and maximum specific growth rate of the culture, respectively.

RESULTS AND DISCUSSION

Effect of initial phenol concentration on its own biodegradation

Figure 1 shows the time course profile of the phenol biodegradation by the mixed culture. It can be seen that the mixed culture is able to degrade completely upto 700 mg/l phenol in almost 36 h. It is evident from Figure 1 that the time taken by the mixed culture to degrade phenol is depended on its initial concentration. It was found that biodegradation rate increases with the increase in phenol concentration upto 300 mg/l, but then starts decreasing. A maximum rate $\frac{dS}{d\theta}$ of 24.5 mg/L/h is obtained at initial phenol concentration of 300 mg/l. At concentration 400 mg/l of phenol, degradation rate has been evaluated as 20 mg/L/h. Rate is less than 20 mg/L/h for 100 mg/l and 700 mg/l of phenol concentration.

Effect of phenol concentration on the growth of the culture

Phenol concentration has been shown to have an inhibitory effect at higher concentration (Kumar et al, 2005; Gabriela et al. 2006; Stoilova et al., 2006). Microbial growth was observed at phenol concentration upto 700 mg/l. The growth profile of the culture at different initial phenol concentration is shown in Figure 2. It was observed that phenol concentration below 300 mg/l shows almost no inhibitory effect, as lag phase of growth was very short. For phenol concentrations higher than300 mg/l, lag phase took longer times, which is also observed in Figure 1 as increasing trend of time taken to degrade phenol of higher initial concentration. At concentrations above 300 mg/l of phenol, a distinct substrate inhibition was found. This is quite similar to the result obtained by

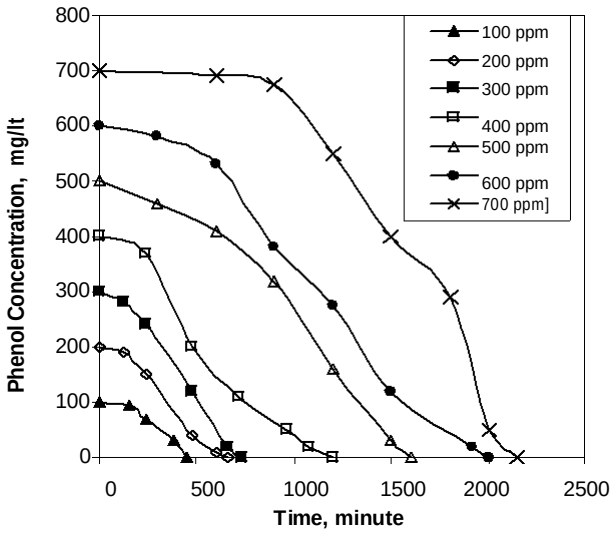

Figure 1. Phenol biodegradation profile with time.

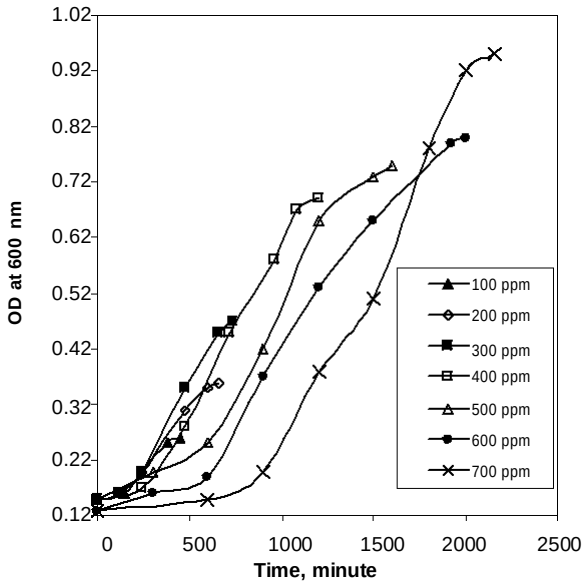

Figure 2. Microbial growth profile with time.

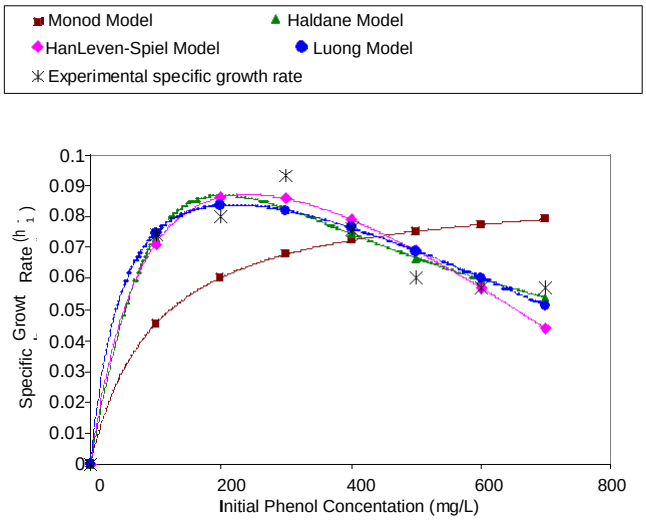

Figure 4. Predicted specific growth rate of the culture due to Monod, Haldane, Han-Levenspiel and Luong model.

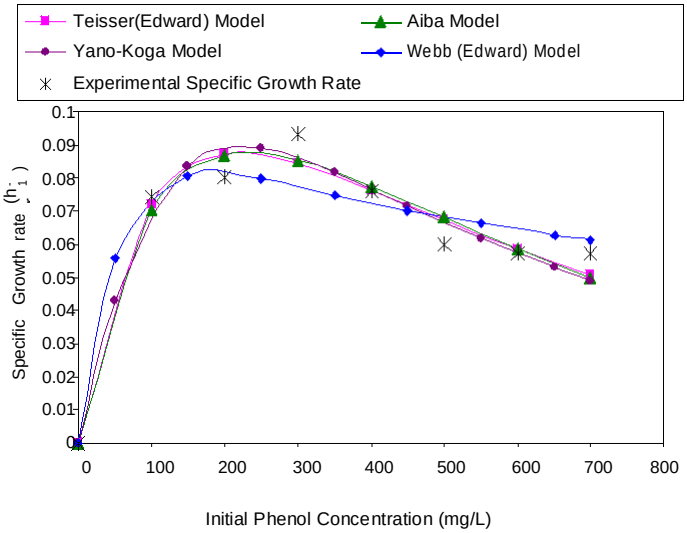

Figure 5. Experimental and predicted specific growth rate of the culture due to Teisser (Edward), Webb (Edward), Yano-Koga and Aiba model.

Bajaj et al. (2009) where they have found the mixed microbial culture is showing substrate inhibition after 4 mM (376.44 mg/l) of phenol. Saravanan et al. (2008) have also reported to have substrate inhibition above 300 mg/l of phenol as seen by the decrease in the specific growth rate. Figure 3 shows that, in the present study, until 300 mg/l of phenol concentration, the specific growth rate of the culture increased (highest μ =0.093 h^{-1}). For concentration higher than 300 mg/l, specific growth rate decreases and became almost constant at 600 mg/l (μ = 0.057 h^{-1}) and 700 mg/l (μ = 0.057 h^{-1}) of phenol.

Exploration of best-fit kinetic model for phenol biodegradation

Family of plots in Figure 1 reveals the pattern of phenol removal throughout the respective biodegradation period is similar to each other irrespective of initial phenol concentration. Figure 4 and 5 showed the comparative plots of experimental specific growth rates and the model predicted ones as given by equation 1 - 9 and solved by MATLAB© 7.1. Among several models used to fit the present experimental data for specific growth rates versus different initial phenol concentration, not only Haldane

Table 1. Summary of growth kinetics parameter values obtained from different models during biodegradation of phenol by mixed microbial culture used in present work.

Model	max (hr^{-1}).	Ks (mg/l)	Ki	Ksi	K	S_m	n	m	RMSE
Monod	0.09	100.6	-	-	-	-	-	-	0.0208
Haldane	0.3057	257.5	162.6	-	-	-	-	-	0.0067
Hanlevenspiel	0.2901	252.1	-	-	-	720	1	1	0.0218
Luong	0.1291	59.39	-	-	-	1148	0.9	-	0.0072
Edward	0.1675	95.05	-	200	1000	-	-	-	0.0090
Aiba	0.2579	200.3	502	-	-	-	-	-	0.0078
Teisser	0.1386	95.04	699.5	-	-	-	-	-	0.0075
Yano-Koga	0.2981	286.2	-	-	K_1 = 261.7 K_2 = 499.8	-	-	-	0.0079

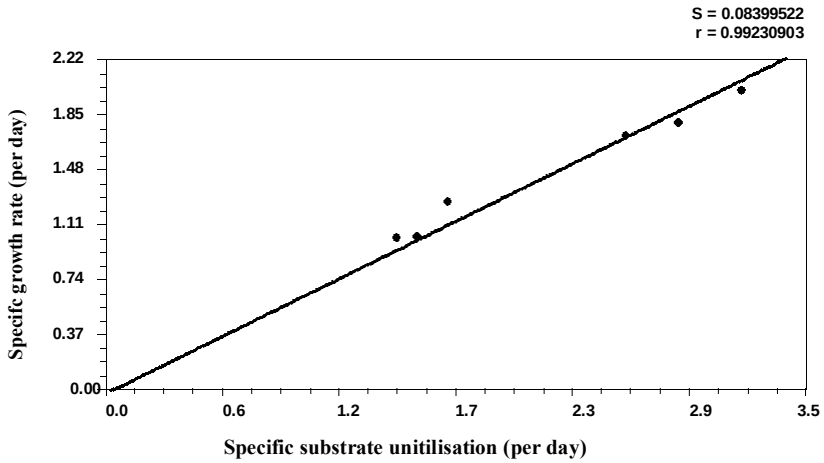

Figure 6. Plot for determining yield and death coefficient of culture for phenol biodegradation.

model but also Luong model showed fit well reasonably as determined by the root mean square error (RMSE) calculated between experimental and the model predicted specific growth rate values. This could be attributed based on the models themselves, which are considered more refined from the standpoint of development of these models.

The biokinetic constants of growth of the culture obtained from these models along with root mean square error between experimental and predicted rate values are shown in Table 1. The Table also reports the value of K_S and μ_{max} as per Monod model by nonlinear regression method. Haldane and Han-Levenspiel model predicts marginal differences in both K_S and μ_{max} values, but differs a lot in RMSE calculated between experimental and model predicted specific growth rates. This may be due to the fact that Haldane model takes care of the value of inhibition constant Ki, that is an important parameter in understanding the kinetics of the microorganism in the system. Luong and Han- Levenspiel models also predicted the critical substrate concentration (S_m) value, at which specific growth rate fall to zero (1148 and 720 mg/l, respectively). But Han- Levenspiel predicted S_m value (720 mg/l), agrees well with the experimental result. Experimental data shows that the specific growth rate at 700 mg/l of initial phenol concentration is 0.057 h^{-1} and after that, it becomes very low. Though RMSE value for Han- Levenspiel model is high enough than Luong model, but that may be because of the good fitting of Luong model at lower initial phenol concentration. The difference in the models predicted kinetic constant values for the present experiment perhaps due to the fact that the two models were originally developed for systems containing a different microorganism and substrate. Table 2 shows the comparison of biokinetic constants as obtained by different investigators as well as in the present study.

Evaluation of microbial yield and death coefficient during phenol degradation

A plot of specific growth rate vs. specific substrate consumption rate (Figure 6) according to Equation (10)

Table 2. Summary of growth kinetics obtained in various studies for the treatment of phenolic wastes.

S/ No.	Authors	Bacterial strain	System	Concentration range (mg/l)	Monod model		Haldane model		
					μ_{max} (hr^{-1})	Km (mg/l)	μ_{max} (hr^{-1})	Ks (mg/l)	Ki (mg/l)
1	Powlowsky and Howell	Mixed culture I Mixed culture II (Filamentous organism)	Batch Batch	0-900 0-1000	- -	- -	0.260 0.223	25.4 5.86	173.0 934.5
2	Livingstone and Chase	NCIB8250 *Actino bacter* sp +NCIB10535 (*Pseudomonus* sp.) +NCIB1015(*Pseudomonus* sp)	Batch	0-500	-	-	0.418	2 .9	370
3	Huchinson and Robinson	*P. putida* F1 ATCC 17484	Batch	<200	-	-	0.388	1.06	903
4	Hill and Robinson	*P. putida* (ATCC 17484, Stainer 110) *A. calcoacetius* (phenol only) *P. fluoroscens* 2218 (phenol only)	Batch/ Continuous	0-700	-	-	0.534	0.015	470
5	Kumaran and Paruchuri	Pooled culture (Phenols) *P. fluoroscens,* *P. putida, P. cepacia, A. calcoacetius,* *C. tropicalis*	Batch	60-500	0.465	3 0.96	0.542	36.2	145.0
6	Okaygun et al.	*Pseudomonus* sp. and *Klebsiella* sp.	Batch	100-170	-	-	0.325	8.2	170.0
7	Arutchelvan, V	*Bacillus brevis*	Batch	750 - 1750	-	-	0.026 - 0.078	2.2 - 29.3	868 - 2434.7
8	Buitron G	Mixed culture	Batch	40	-	-	0.258	3.9	121.7
9	Marrot B	Mixed culture	Continuous	2500	-	-	0.438	29.5	72.4
10	Saravanan et al.	Mixed culture	Batch	100 - 800	0.37	144.68	0.3085	44.92	525.00
11	Bajaj et al.	Mixed culture	Batch	23.5 - 659	-	-	0.3095	74.65	648.13
12	Present study	Mixed culture	Batch	100 - 700	0.0995	35.76	0.150	51.8	404.04

was used to evaluate the yield and death coefficient for the phenol biodegrading micro-organisms. The yield coefficient in the present study (0.66 mg/mg) is found to be close to those reported in literature as $Y_{X/S}$ = 0.6 ± 0.12 mg/mg (Livingston and Chase, 1989) . Bajaj et al. (2009) have reported the yield factor as 0.1 - 0.17 (A580 units or OD at 580 nm/mmol of phenol) for mixed microbial culture. The calculated decay coefficient for the present study was 0.00103 h^{-1} similar to values published by Kumaran and Paruchuri (1997), who obtained decay coefficient to be 0.005 hr^{-1} for phenol degradation by mixed culture. Kumar et al. (2005) have calculated a decay coefficient value of 0.0056 h^{-1} of *P. putida* MTCC 1194 for phenol degradation in batch culture. Arutchelvan et al. (2006) have reported decay coefficient to be 0.003 - 0.12 h^{-1} and yield coefficient of 0.293 - 0.571 for phenol removal by *Bacilus bravis*. This yield coefficient is slightly less and decay coefficient is slightly more than the

value obtained in the present experiment. The kinetic data as estimated by different scientists deriving from either pure or mixed culture including the present study is listed in Table 2. The wide variation of kinetic coefficients values perhaps is due to the fact that most of the investigators have carried out the research either under pure culture system in different initial test condition. The present study deals with mixed microbial culture containing several species, whose death rate is not same, for which the overall death coefficient marginally deviated from the available literature data.

Conclusion

Kinetics of phenol degradation was studied under aerobic condition in batch reactor using an indigenous mixed microbial culture, isolated from an effluent treatment section of a coke oven plant. The culture could grow and biodegrade phenol upto 700 mg/l. However, phenol exhibited inhibition to both specific growth rate and substrate degradation rate above 300 mg/l of initial phenol concentration. Specific growth rates of the culture under different initial phenol concentration from 100mg/l to 700 mg/l have been calculated. By fitting specific growth rates on suitable substrate inhibition models, biokinetics constant that are necessary to understand the kinetics of biodegradation process were evaluated by MATLAB 7.1© software. Root Mean Square Error values between the experimental specific growth rates and the model predicted ones have been calculated for different substrate inhibition models. It is observed that the best model that fit the present study is Haldane model having lowest RMSE value of 0.0067 and predicting reasonable kinetic coefficient values. Therefore, the mixed culture used in the present work is a potential culture that can be used for phenol biodegradation under aerobic condition in real life wastewater treatment.

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List of symbols:

S	mg/l	Substrate concentration
So	mg/l	Initial substrate concentration
X	mg/l	Biomass concentration
Xo	mgl	Initial biomass concentration
	hr^{-1}	Specific growth rate
$_{max}$	hr^{-1}	Maximum specific growth rate
K_1, K_2	mg/l	Positive constants
Ks	mg/l	Half saturation constant
Ki	mg/l	Substrate inhibition constant
n, m	-	Empirical constant
Yx/s	mg/mg	Microbial yield coefficient
K_d	hr^{-1}	Microbial decay coefficient
	Day	Batch time