Asian Journal of Chemistry Vol. 17, No. 2 (2005), 1171–1184

Growth Model for Biodegradation of VOCs in Batch and VOCs Removal from Contaminated Air in Biofilter Using *Pseudomonas putida* and Isolated Strain NTPM1

GHASEM D. NAJAFPOUR* and YAP YOK MIAN
School of Chemical Engineering, Engineering Campus, Universiti Sains Malaysia
Sri Ampangan, Nibong Tebal, 14300 Penang, Malaysia
E-mail: najafpour8@yahoo.com chghasem@eng.usm.my
Tel: +604-593 7788 Ext 5404; Fax: +604-594 1013

Biodegradation of VOCs in polluted air was investigated in biofilter. Biofilter was a kind of continuous process loaded with active cells. *Pseudomonas putida* F1 and isolated strain *NTPM1* were immobilized in packed bed biofilter. Contaminated air, contained acetone was biologically oxidized by passing through the biofilter. The filter medium with an active biofilm was acting as effective adsorbing surface. Extensive studies were conducted to determine removal efficiency of the biofilter. Acetone-air flow rate with acetone concentrations of 8 to 16 g/L, was set at 100, 200 and 300 cm^3/min equivalent to empty bed residence time of 2, 3 and 6 min, respectively. At air flow rate of 100 cm^3/min, maximum acetone removal efficiency of 90% was obtained. In batch experiment, low concentration of acetone in the range of 150–550 mg/L and phenol, also 150–950 mg/L of hexane, benzene and toluene with an increment of 100 mg/L were used. *P. putida* F1 and isolated strain *NTPM1* were compared. *P. putida* F1 showed higher removal efficiency in shorter period of time in comparison to isolated strain *NTPM1*. The batch results demonstrate the treatability and efficient removal of acetone, phenol, hexane, benzene and toluene in the biofilter at high concentration of 16 g/L. A growth model, based on microbial biodegradation of VOCs in batch process was developed.

Key Words: Volatile organic compounds, Hazardous air pollutants, *Pseudomonas putida*, Biofilter, Growth Model.

INTRODUCTION

Volatile organic compounds (VOCs) may cause cancer or other serious health effects or adverse environmental and ecological effects. These pollutants are emitted to air in significant quantities. Emission sources are mobile (vehicles) or stationary (factories, refineries, power plants). Based on US-EPA Priority Pollutant List, the 1990 Clean Air Act Amendment, there are 189 potential chemicals in the Priority List of Hazardous Substances, required to be controlled and reduced by 90%[1].

Air pollution control using biofiltration technology is getting popular among the industries. As industries are encouraged to invest for treatment of waste gases and contaminated air for VOCs removal, biofiltration may get extra attention such as cost effectiveness, a competitive alternative to the existing conventional technologies such as incineration and activated carbon adsorption[2–4]. Biofiltration is an air pollution control technology that holds active microorganisms to degrade organic pollutants in biological processes[5–7]. Microorganisms are grown as active biofilm on filter media which is holding moisture and nutrients. Contaminated air passes through the filter media and then the contaminants are transferred from gas phase to biofilm. The penetrated VOCs are degraded by microorganisms[2, 8].

The moisture content, temperature, pH, pressure, nutrients and empty bed retention time are the major factors affecting the performance of biofilter. It has been believed that 75% of problems associated with biofilter are caused by poor humidification. The optimal moisture content of the biofilm established on packing materials is in the range of 40–60 %. By losing moisture of the fixed film in biofilter, the resulted dried filter media may cause decline in biofilter performance[9].

The major concerns in industrial application of biofilter would be: how well a filter material works, determine the effectiveness of the biofilters in removal of VOCs and odour from a polluted air stream. Biological degradation is taking place in aqueous phase of the filter material. Inadequate nutrient and moisture level in the biolayer may cause microorganism failure in removal of VOCs and odour from the air stream[10].

Biofilters are suitable for treating large volumes of gas containing low concentration of pollutant[11]. It is a technology that is well suited to treat low concentration emissions where combustion or adsorption technologies are not appropriate. Conventional techniques for removal of VOCs from contaminated air such as incineration or adsorption by activated carbon are costly and may generate secondary waste streams. Nowadays industries are mostly looking forward to biological technology, which provides a more economical solution and an environmentally benign process[12].

Hence, biological degradation and purification processes are now widely used in various industries. It is commercially proven in Europe. Since the early 1990s, biofiltration has been used successfully to control pollution in a number of air contaminants such as noxious odours, volatile organic compounds stemming from a wide range of industrial and public sector sources[2, 5, 8].

Biological waste air treatment processes offer a zero waste generation, no nitrogen oxides and also do not generate any secondary waste stream. Pollutants are generally converted to carbon dioxide with the growing microorganism. Good operational stability and non-dangerous processes encourage the development of biological air treatment.

Mono-aromatic hydrocarbons such as benzene, toluene and phenol are the best choices for biodegradation studies. These toxic chemicals are used as solvent in fuels, production of plastics, synthetic fibres and pesticides[13, 14]. Biofiltration theory involves the use of mathematical models. The key component of the process is active biofilm. The active microbe layer exists within the matrix of the

filter media. Living microorganisms in the column degraded organic compounds to intermediate metabolites. Finally carbon dioxide and water are harmless biological products and may be discharged without further treatment. These processes need to control living organisms and make sure that the system obtains enough energy for reproduction of cell bodies[6].

Biofiltration theory involves the use of a model system. Microbial degradation kinetics plays a vital role in design and sizing of biofilter. Monod rate equation is commonly used to describe the growth law for a number of bacteria, which is similar to Michaelis-Menten kinetics[2, 15-17]. To characterize the growth curves, logistic equation was used, which has additional benefit in representing the entire growth curve[16]. Growth curve includes the lag phase, the exponential growth and the stationary phase. Theoretically, cell growth rate is expressed as follows:

$$\frac{dX}{dt} = \mu X \tag{1}$$

where X is the cell dry weight concentration (g/L) and is the specific growth rate (h^{-1}). The specific growth rate for the logistic model is:

$$\mu = \mu_m \left(1 - \frac{X}{X_{max}} \right) \tag{2}$$

where μ_m is the maximum specific growth rate (h^{-1}) and X_{max} is the maximum cell dry weight concentration (g/L). By substitution of eqn. (2) into eqn. (1) and performing integration, the following equation for the cell concentration was obtained:

$$X = \frac{X_0 \exp(\mu_m t)}{1 - \left(\frac{X_0}{X_{max}} \right) (1 - \exp(\mu_m t))} \tag{3}$$

The above equation was used to predict the cell growth in batch experiments. In this research, inoculation volumes were kept constant for batch experiments, where the logistic model was a fair approximation of the growth curve. Polymath computer software was used to define logistic growth kinetic parameters. The biodegradation of acetone and phenol, hexane, benzene and toluene at various concentrations have been determined using *P. putida* F1 and isolated strain *NTPM1*. Various air flow rates of inlet pollutants were introduced into a continuous biofilter to investigate the removal efficiency. The purpose of this research was to demonstrate the biodegradation of organic toxic compounds in batch and the application of biofilter, an extremely fast process for removal of contaminants (in gas phase) from the air stream.

EXPERIMENTAL

Pseudomonas putida F1, ATCC 700007 was obtained from the American Type Culture Collection, USA. A strain of microorganisms was isolated from paper mills wastewater (Nibong Tebal Paper Mill Sdn. Bhd, Nibong Tebal, Malaysia) named *NTPM1*. The microorganisms were grown aerobically at 25°C and pH 6.7

in nutrient broth. The initial media for seed culture consist of yeast extract, peptone and dextrose with concentration of 1, 3 and 5 g/L, respectively. The medium used for batch and continuous experiments consists of: yeast extract 0.2 g, $MgSO_4$ 0.1 g/L, $CaCl_2$ 0.08 g/L, ferric citrate 0.08 g/L, K_2HPO_4 0.4 g/L, KH_2PO_4 0.016 g/L, EDTA 0.05 g/L and $Fe_2(SO_4)_3 \cdot H_2O$ 0.6 mg/L, $ZnSO_4 \cdot 7H_2O$ 0.2 mg/L, $CuSO_4 \cdot 5H_2O$ 0.2 mg/L, $MnSO_4 \cdot 5H_2O$ 0.2 mg/L, $CoSO_4 \cdot H_2O$ 0.2 mg/L. *Pseudomonas putida* F1 and *NTPM1* were adapted in a medium with 250 mg/L of acetone, phenol, hexane, benzene and toluene before any experiments were carried out. Culture screening was conducted and the purity of the isolated culture was often controlled by culturing organisms on petri-dish and gram stain.

The continuous experimental set-up is shown in Fig. 1. The biofilter was constructed from acrylic plastic transparent tube with internal diameter of 46 mm and 950 mm height (L/D = 20). The biofilter column was filled with packing material. The packing material was 7 mm glass Raschig rings, coated with 2% agar. The height of packing was 945 mm. The column empty bed volume after the establishment of biofilm was about 600 mL.

Biofiltration experiments

Batch biodegradation experiments were carried out in 250 mL conical flask containing 100 mL of medium and single chemical pollutant concentration range 150 to 550 mg/L, increment of 100 mg/L. Flasks were placed on an orbital shaker (B. Braun, Germany) at 100 rpm and 25°C. Sample was taken periodically to determine pollutant concentration and biomass concentration. The experimental set-up for continuous biofilter (Fig. 1) consists of feed and nutrient tank, nutrient pump, air blower, air flow meter and fixed film biofilter with inlet and outlet sample ports. The organic compounds (VOCs) were added in to substrate tank

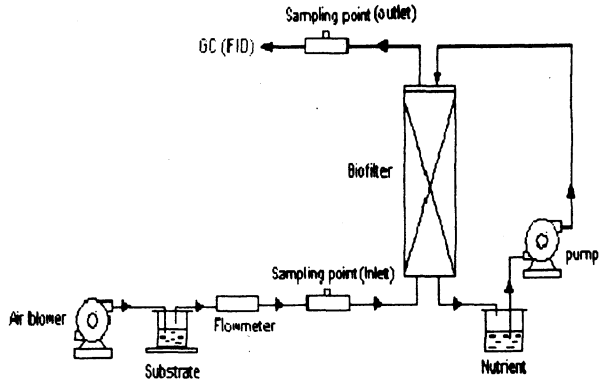

Fig. 1. Experimental set-up for continuous biofilter process flow diagram

with define concentration. An air stream was bubbled through the organic solution. The air was used as carrier gas for organic contaminants. The inlet and outlet concentrations of organic compounds were determined by gas chromatograph. The contaminated air was passed through the filter bed. The continuous biodegradation in biofilter was conducted with contaminated air at 3 different flow rates: 100, 200 and 300 cm^3/min, with empty bed residence time (EBRT) of 6, 3 and 2 min, respectively. Acetone concentration in the substrate tank resulted 8 and 16 g/L, with mass fraction of 1 and 2 wt % of contaminants in the air stream, respectively. The nutrients were supplied down-flow, at flow rate of 5.5 mL/min, to maintain biofilm moisture. The nutrients were mainly yeast extract and peptone with concentration of 1 and 3 g/L, respectively. The trace metals and minerals as stated in batch media were used. The trace metals and minerals acted as growth stimulants. During the continuous experiment, gas samples were taken from the inlet and outlet sampling ports and analyzed by gas chromatograph.

Analytical methods

Biomass concentrations were measured with cell optical density at 600 nm using a spectrophotometer (Cecil model 1000). The biomass concentration was calculated from a correlation between optical density and dry cell weight. Analysis of liquid sample from substrate tank was conducted to determine acetone concentration. The sample was filtered using polyether sulfone membrane, 25 mm diameter, 0.25 μm pore size. The filtered sample was analyzed by gas chromatograph (HP 5890 series II). The GC was equipped with flame ionization detector (FID). The GC column was packed with 3% OV-101, 80/100 Chromosorb. The temperatures of injector, oven and detector were 220, 200 and 250°C, respectively. Nitrogen (35 mL/min) was used as carrier gas. For continuous biofiltration, gas samples were collected at the inlet and outlet of biofilter using gas-tight syringe (Hamilton CO., Reno, Nevada).

RESULTS AND DISCUSSION

The microorganisms used in this research work were *P. putida* F1 obtained from ATCC and isolated strain from pulp and paper mill wastewater, *NTPM1*. Three species of dominant microorganisms were isolated from the wastewater sample. In primary screening, a potential isolated strain was selected for further studies. The isolation and screening of the organisms was based on utilization of aliphatic and aromatic compounds and to obtain energy for growth. The isolated strains were inoculated into medium containing 250 mg/L acetone as sole carbon source. After 24 h of incubation at 32°C, the medium was very turbid and cell growth was observed. The isolated organism after sequential screening was grown on acetone and compared with *P. putida* F1. The prepared gram-negative stained slides for microscopic views are shown in Figs. 2 and 3 for *P. putida* F1 and isolated strain *NTPM1*, respectively. The isolated strain was able to grow on phenol, hexane, benzene and toluene, also utilizing mono-aromatic compounds as the sole carbon source. The inoculums of isolated strain *NTPM1* were

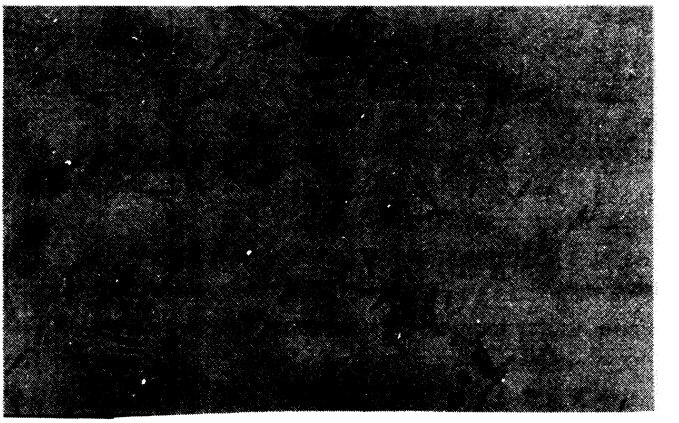

Fig. 2. Isolated strain of bacteria, *NTPM1*

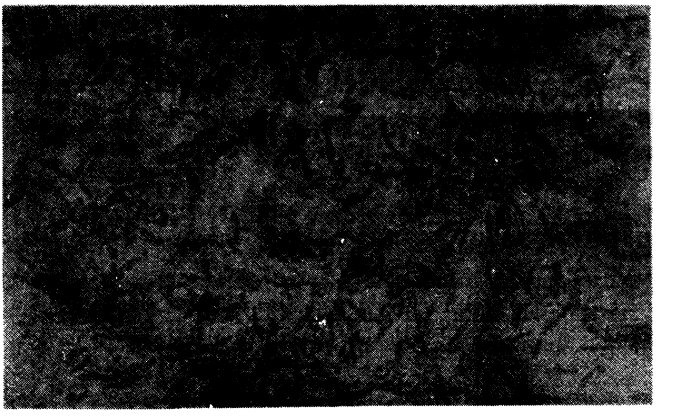

Fig. 3. *Pseudomonas putida* F1, ATCC 700007

introduced into four flasks of media each containing 250 mg/L of hexane, phenol, benzene and toluene as carbon sources. This experiment was conducted to examine the growth-ability of the isolated strain, *NTPM1* on various carbon sources. The results showed that the isolated strain *NTPM1* was able to grow well in media containing mono-aromatics for 24 h incubation at 32·C. The isolated strain was a gram-negative, non-spore, motile rod-shaped bacterium. The morphological colony formed on petri dish was rounded hard, semi-translucent, regular, entire, low convex, smooth and shiny.

Biodegradation of acetone in batch experiments was carried out with *P. putida* F1 and isolated strain *NTPM1* in the media, with a concentration range of 150–550 mg/L; an increment of 100 mg/L. Figs. 4 and 5 present the rapid decrease of

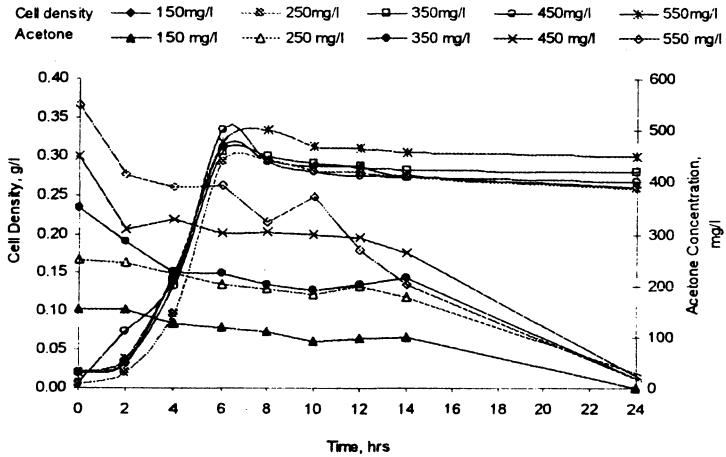

Fig. 4. Reduction of acetone and production of biomass using *P. putida* F1

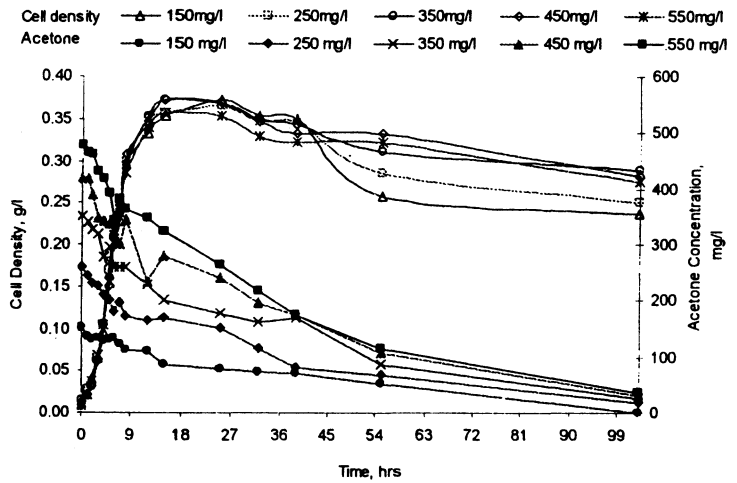

Fig. 5. Reduction of acetone and production of biomass using *NTPM1*

acetone and increase in cell concentration of *P. putida* F1 and isolated strain *NTPM1*, respectively. The increasing trend of cell density for an incubation period of 24 h was about the same, except that *P. putida* F1 showed faster growththan the isolated strain *NTPM1*. The maximum cell density was about 0.33 and 0.35 at 8 and 18 h for *P. putida* F1 and isolated strain *NTPM1*, respectively. The batch system was agitated (100 rpm) to enhance oxygen transfer rate into the media; thus higher cell growth was obtained. It was well reported in literature that *P. putida* F1 has the potential to oxidize single ring aromatics[4, 9, 12, 14, 15]. Therefore the isolated strain *NTPM1* was also experimented in batch culture with aromatic carbon sources. At first, the cell growth was carried out with 250 mg/L of organic compounds and then the adapted culture was used for biodegradation of phenol, hexane, benzene and toluene. The concentrate range of studies for phenol was

from 150 to 550 mg/L with an increment of 100 mg/L. The concentration range for other saturated aromatic such as hexane and unsaturated aromatics such as benzene and toluene was 150–950 mg/L with 100 mg/L increment. Fig. 6 shows

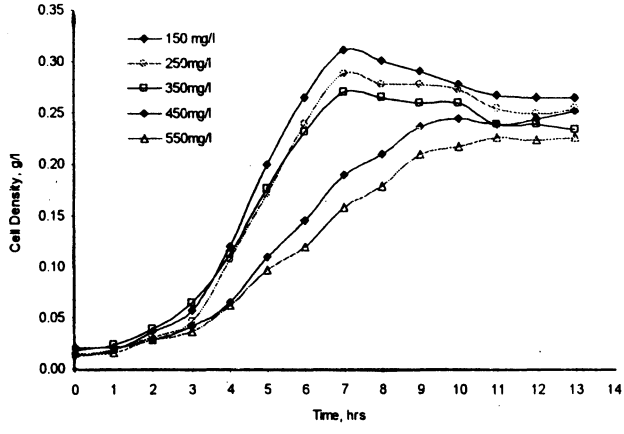

Fig. 6. Cell concentration of *NTPM1* grown on phenol

the biodegradation of phenol with the isolated strain, *NTPM1*. The growth of gram-negative *NTPM1* was retarded with additional phenols. Once the phenol concentration was increased from 150 to 450 mg/L, the cell density dropped from 0.3 to 0.2 g/L in 8 h.

Higher concentrations of organic sources: hexane, benzene and toluene, were utilized and the cell densities are shown in Figs. 7, 8 and 9, respectively. There was a 2 h lag phase for most of the aromatic compounds. The cell density trends in Figs. 7, 8 and 9 were about the same; even at high concentration, 950 mg/L

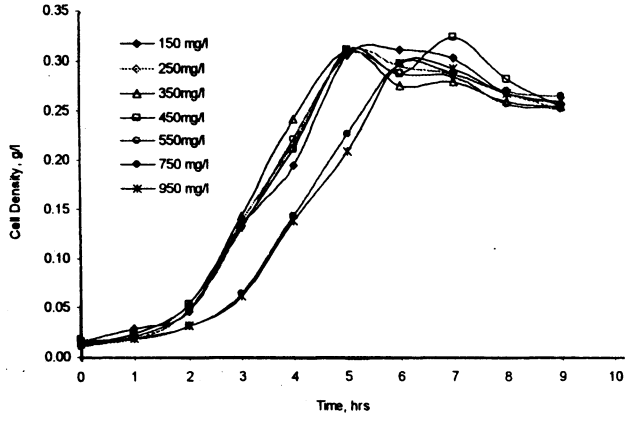

Fig. 7. Cell concentration of NTPM1 grown on hexane

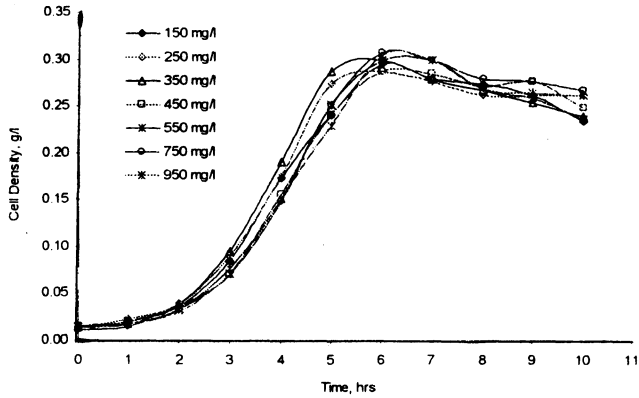

Fig. 8. Cell concentration of *NTPM1* grown on benzene

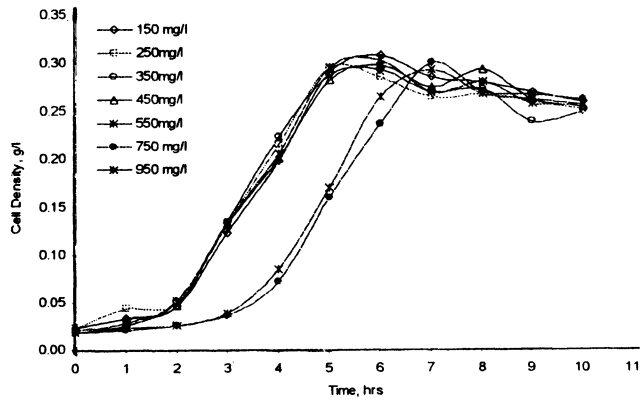

Fig. 9. Cell concentration of *NTPM1* grown on toluene

of aromatics, the growth curve followed the same trend. The exponential phase for phenol, hexane, benzene and toluene was observed in all experiments at approximately 4 h.

.Continuous operations of biofilter loaded with *P. putida* F1 and isolated strain *NTPM1* were conducted with polluted air flow rates of 100, 200 and 300 cm^3/min. Fig. 10 presents acetone removal efficiency of the biofilter for continuous mode of operation in a duration of 290 h. Acetone was used as pollutant and air was the carrier gas. The filter efficiency was sensitive to air flow rate and acetone concentration. Nutrient solution was periodically sprinkled every 2–4 h to keep the required moisture of the biofilm. Flow rate for the nutrient solution was 5.5 mL/min. The pollutant degradation performance on the biofilter is expressed in terms of the pollutant removal efficiency. The removal efficiencies widely varied

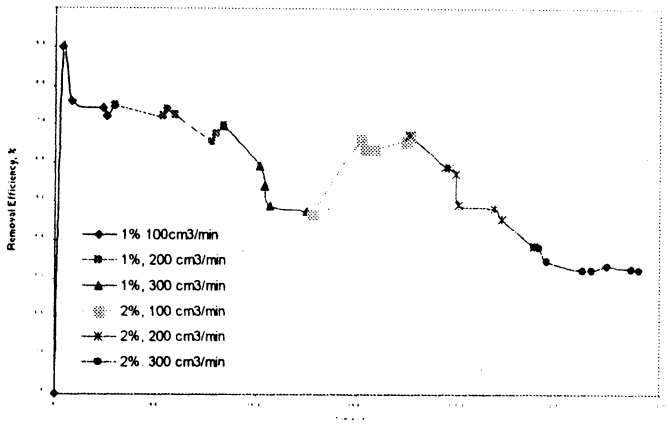

Fig. 10. Acetone removal efficiency in continuous biofilter

from 30 to 76% and were apparently linked to inlet gas flow rate or inlet acetone concentration. From the beginning of this experiment, 4 h after start up, the first part of Fig. 10, the inlet acetone concentration was 8 g/L (1 wt %); the removal efficiency was 90%. For the first 30 h after start up, the removal efficiency was maintained above 75% with air contained acetone flow rate of 100 cm^3/min. The removal efficiency was steady and maintained constant at 75%. After 54 h of operating, the air flow rate was doubled (200 cm^3/min). The removal efficiency fluctuated and was maintained in the range of 67–74%, for air flow rate of 200 cm^3/min. After 102 h of operating, the flow rate was tripled (300 cm^3/min). Low efficiency about 33% of acetone removal was observed. The removal efficiency dropped to about 47–59%. In the next stage of operation, after 152 h, the acetone concentration at the inlet air increased to 16 g/L (2 wt %). The removal efficiency with air flow rate of 100 cm^3/min was maintained in the range of 64–68%. The second flow rate, 200 m^3/min was experimented after 194 h of operation. The removal efficiency dropped to 60% at steady state condition. At high air flow rate, the system showed poor removal efficiency. Lowest removal efficiency was observed after 290 h, which was 32% when the flow rate was tripled, 300 cm^3/min of polluted air. The organic loading was directly proportional to gas flow rate, or inversely proportional to corresponding empty-bed-gas-residence-times (EBRT) in the biofilter.

Fig. 11 shows the impact of EBRT on the average removal efficiency. It can be noted that the removal efficiency increased with the additional increment of EBRT. Longer residence time resulted in higher removal efficiency. For long EBRT (6 min) corresponding to air flow rate of 100 cm^3/min, 80% efficiency was achieved. Compared to shorter EBRT (3 min) corresponding to 200 cm^3/min air flow rate, the removal efficiency dropped to 73%. The longer residence time gave longer contact time between the microorganism and acetone. For shorter EBRT (2 min) corresponding to higher air flow rate (300 cm^3/min), the removal efficiency dropped to 51%. Even though higher flow rate gave higher concentra-

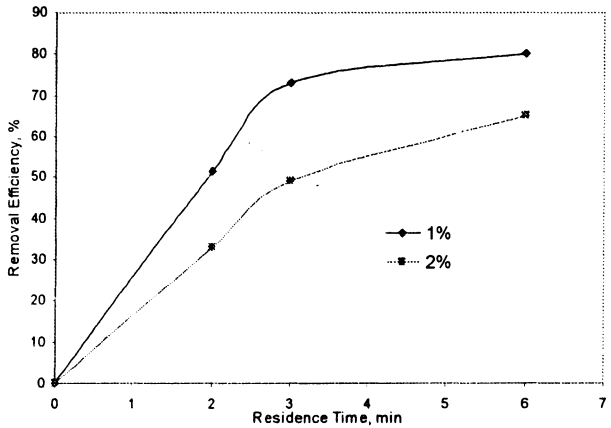

Fig. 11. Acetone removal efficiency with respect to residence time in continuous biofilter

tion of acetone, but the contact time between the microorganism and acetone was shorter. The air flow rate of 300 cm^3/min resulted in 2 min of EBRT in the column, but 100 cm^3/min allowed 6 min of EBRT in the column. A three times longer retention time for acetone remaining in the column gave longer contact time and thus removal efficiency was increased by 60%. For shorter EBRT, the microorganism did not have sufficient time to perform a good biodegradation rate on the available acetone. Similar phenomena happened on acetone with higher concentration, which was 16 g/L (2 wt %). For longer EBRT (6 min), 65% of removal efficiency was achieved. On the other hand, shorter EBRT gave poor removal efficiency, which was only 33%.

The linear model, Lineweaver Burk Plot for *P. putida* F1 and *NTPM1* grown on acetone, is shown in Fig. 12. The lines are parallel with the same slopes and

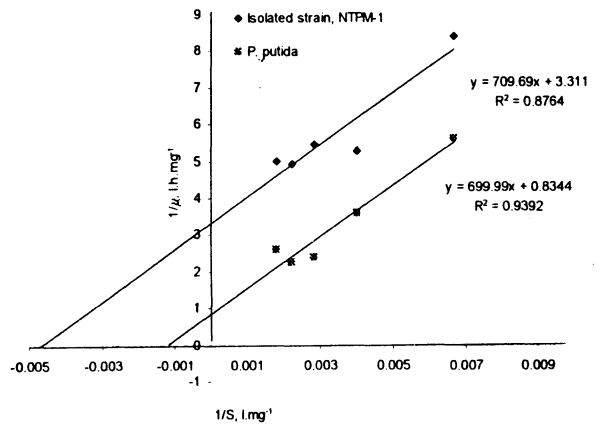

Fig. 12. Lineweaver Burk Plot for *P. putida* F1 and *NTPM1* grown on acetone

represent the growth kinetic parameters. The maximum specific growth rates for *P. putida* F1 and *NTPM1* were 1.2 and 0.3 h^{-1}, respectively. All experimental results showed that the growth of *P. putida* F1 was faster than *NTPM1*.

Figs. 13 and 14 show the experimental data for growth curves and the logistic models were well fitted for *P. putida* F1 and *NTPM1*, respectively. The solid line

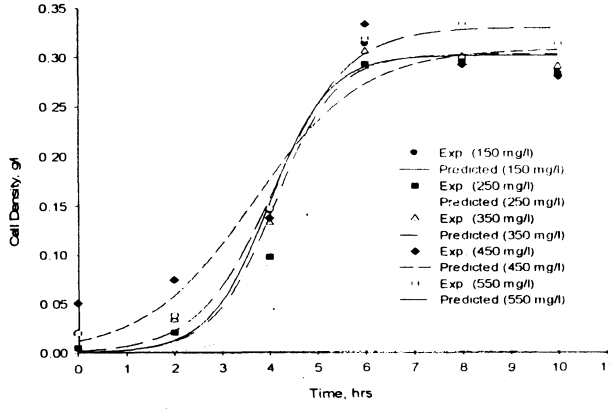

Fig. 13. Cell growth model for *P. putida* F1, on acetone

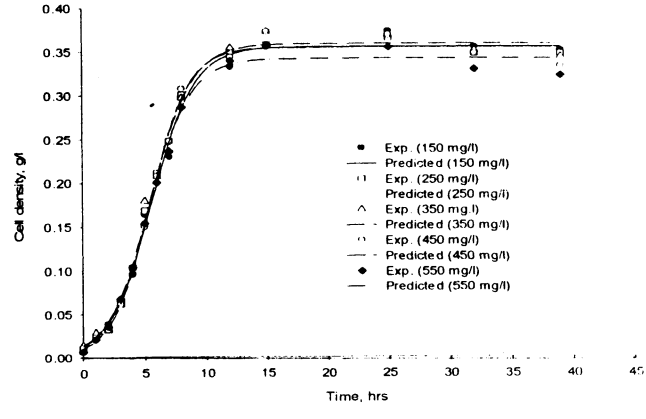

Fig. 14. Cell growth model for *NTPM1*, on acetone

(the model) exactly followed the actual data. Tables 1 and 2 summarize the kinetic parameters defined in eqn. (3). The coefficients were calculated with Polymath software. The R^2 values for all acetone concentrations (150–550 mg/L) were from 0.90 to 0.99. That indicates that good agreement of the model with the experimental data exists. A 2 h lag phase was observed in the projected model and experimental data.

TABLE-1
KINETIC PARAMETERS DEFINED IN LOGISTIC MODEL USING POLYMATH AND R^2 VALUE FOR *PSEUDOMONAS PUTIDA* F1

Acetone concentration	Variable	Initial guess	Value (Model)	95% Confidence	R^2	X_0/X_m
150 mg/L	X_0	0.1	0.0005	0.005	0.982	1.68×10^{-3}
	μ_m	1.0	1.585	2.488		
	X_m	0.6	0.30	0.045		
250 mg/L	X_0	0.1	6.5×10^{-6}	0.0001	0.995	2.23×10^{-5}
	μ_m	1.0	2.506	5.55		
	X_m	0.6	0.29	0.028		
350 mg/L	X_0	0.1	0.0005	0.004	0.993	1.65×10^{-3}
	μ_m	1.0	1.55	2.133		
	X_m	0.6	0.3	0.05		
450 mg/L	X_0	0.1	0.012	0.00001	0.90	0.0380
	μ_m	1.0	0.88	0.0001		
	X_m	0.6	0.3	0.0003		
550 mg/L	X_0	0.1	0.002	0.000004	0.99	5.76×10^{-3}
	μ_m	1.0	1.26	0.00005		
	X_m	0.6	0.33	0.00001		

TABLE-2
KINETIC PARAMETERS DEFINED IN LOGISTIC MODEL USING POLYMATH AND R^2 VALUE FOR *NTPM1*

Acetone concentration	Variable	Initial guess	Value (Model)	95% Confidence	R^2	X_0/X_m
150 mg/L	X_0	0.1	0.014	0.00575	0.995	0.039
	μ_m	1.0	0.575	0.0794		
	X_m	0.6	0.355	0.011		
250 mg/L	X_0	0.1	0.0121	0.00003	0.998	0.034
	μ_m	1.0	0.6218	0.0005		
	X_m	0.6	0.3540	0.0002		
350 mg/L	X_0	0.1	0.014	0.00002	0.993	0.039
	μ_m	1.0	0.586	0.0003		
	X_m	0.6	0.359	0.0001		
450 mg/L	X_0	0.1	0.01	0.00001	0.995	0.030
	μ_m	1.0	0.64	0.0003		
	X_m	0.6	0.35	0.0001		
550 mg/L	X_0	0.1	0.013	0.000005	0.995	0.037
	μ_m	1.0	0.60	0.00008		
	X_m	0.6	0.34	0.00003		

Conclusion

The purpose of batch experiment was to demonstrate the biodegradation of organic pollutants using microorganisms. In batch biodegradation, the cells were able to consume aliphatic and mono-aromatic toxic chemical compounds. *P. putida* F1 (ATCC strain) was able to consume most of the toxic chemicals faster than the isolated strain *NTPM1*. It can be concluded that *P. putida* may yield slightly higher removal efficiency in biodegradation process, compared to isolate strain *NTPM1* in the same environment. High removal efficiency (80%) was achieved in the biofilter with 1 wt % acetone and an air flow rate of 100 cm^3/min. It was proved that the filter holding the moisture with medium sparkling at constant flow rate as the microorganism was easily attached on the agar surface. The durabiiity of the biofilter was continuously tested for 290 h.

ACKNOWLEDGEMENTS

The present research was made possible through an IRPA grant No. 03-02-05-9016, sponsored by Ministry of Science, Technology and Innovations (MOSTI), Malaysia and Universiti Sains Malaysia (USM). The authors wish to thank Research Creativity and Management Office (RCMO), Universiti Sains Malaysia and MOSTI for their financial support.

REFERENCES

1. M. McGinness, A Novel New Approach to VOC and HAP Emission Control, Process of the Industrial Energy Technology, Houston, Texas, USA (2000).
2. S. Peishi, Y. Xianwan, H. RuoHua, H. Bing and Y. Ping, *J. Cleaner Prod.*, **12**, 95 (2002).
3. J.H. Sz-Chwun, L. Chi-Mei, L. Huei-Chuan and P. Huey Fang, *J. Biotechnol.*, **105**, 83 (2003).
4. K. Okamoto, M. Izawa and H. Yanese, *J. Biosc. Bioeng.*, **95**, 633 (2003).
5. M.C. Delhomenie, L.L. Bibeau, N. Bredin, S. Roy, S. Broussau, R. Brzezinski, L.J. Kugelmass and M. Heitz, *Adv. Environ. Res.*, **6**, 239 (2002).
6. G. Spigno, C. Pagella, M.D. Fumi, R. Molteni and D.M. De Faveri, *Chem. Eng. Sci.*, **58**, 739 (2003).
7. H.H.J. Cox and M.A. Deshusses, *Chem. Eng. J.*, **878**, 101 (2002).
8. P. Oyarzun, F. Arancibia, C. Canales and G.E. Aroca, *Process Biochem.*, **39**, 165 (2003).
9. Y. Sun, X. Quan, J. Chen, F. Yang, D. Xue, Y. Liu and H. Yang, *Process Biochem.*, **38**, 109 (2002).
10. S. Ji-Hyeon, R. Javier and K.A. Kinney, *Water Res.*, **37**, 4497 (2003).
11. L. Yonghui, Q. Xie, S. Yumei, C. Jingwen, X. Daming and C.J. Shik, *J. Haz. Mat.*, **95B**, 199 (2002).
12. M. Zilli, E. Palazzi, L. Sene, A. Converti and M. Del Borgh, *Process Biochem.*, **37**, 423 (2001).
13. L. Malhautier, C.C. Gracian, R. Jean-Claude, F. Jean-Louis and C. Pierre, *Le Chemosphere*, **50**, 145 (2002).
14. Jong-O Kim, *Process Biochem.*, **39**, 1 (2003).
15. T. Abu-Hamed, E. Bayraktar, T. Mehmetoglu and U. Mechmetoglu, *Process Biochem.*, **39**, 27 (2003).
16. K. Harun, E. Inci, G. Ufuk, Y. Meral and T. Lemi, *Inter. J. Hydrogen Energy*, **28**, 381 (2003).
17. W. Shyh-Jye, T. Hsiu-Mu and W. Wen-Chuan, *Environ. Prog.*, **16**, 187 (1997).

(*Received:* 4 August 2004; *Accepted:* 19 January 2005) AJC-4064