Research Journal of Chemical Sciences _____ ISSN 2231-606X
Vol. **2(6),** 60-65, June **(2012)** *Res.J.Chem.Sci.*

Biodegradation of Waste gas Containing Benzene by using Corn-Cob based Biofilter

Rahul*[1], Mathur Anil Kumar[2] and Balomajumder Chandrajit[1]
[1]Chemical Engineering Department, Indian Institute of Technology, Roorkee-247667, INDIA
[2]Uttar Pradesh, Pollution Control Board, Agra-282007, INDIA

Available online at: www.isca.in
(Received 6th April 2012, revised 13th April 2012, accepted 16th April 2012)

Abstract

In this present study, a bacterial strain is capable of utilizing benzene as a sole carbon source was isolated from biofilter. Based on the biochemical test the microbes was identified as Bacillus sphaericus. Performance of a biofilter packed with corn-cob packing media was studied for biolfiltration. The removal of benzene was evaluated for various concentration range from 0.0893 to 0.1006 and at empty bed residence time (EBRT) varying from 3.06 to 1.15 s. The experiment was conducted for a period of 68 days in five different phases. When the benzene loading was less than 1.9033 $gm^{-3}h^{-1}$, nearly 99.95% removal could be achieved. A maximum elimination capacity of 2.377 $gm^{-3}h^{-1}$ was obtained at a loading of 2.5109 $gm^{-3}h^{-1}$ with an empty bed residence time (EBRT) of 138 s in phase II.

Keywords: Biofiltration, benzene, bacillus sphaericus, elimination capacity.

Introduction

Due to high releases of wide variety of pollutants in environment results in an increasing number of environmental related problems. These xenobiotic compounds are usually removed slowly and tend to accumulate in the environment, their accumulation can cause severe environmental problems. With increasing public concern about deteriorating environment air quality, stringent regulations are being enforced to control air pollutants[1]. Volatile organic compounds (VOCs) belong to a special category of air pollutants that can adversely affect our health. From an environmental point of view, mainly benzene, toluene, ethyl benzene, *o*-xylene (BTEX) and pyridine are important industrial solvents that are frequently encountered in industrial operations and contaminated sites. From the priority list of hazardous substances comprehensive environmental response, compensation and liability Act (CERCLA) (USEPA, 2005), benzene is ranked sixth and total BTEXs are ranked seventy eight from 275 substances identified to pose the most significant potential threat to human health[2]. Benzene is also classified as hazardous substance in the EPA list of priority pollutants (USEPA 1996). Because of its confirmed carcinogenic properties, the standard set by USEPA 2002 is 5µg L^{-1} for benzene in drinking water[3,4].

Industrial operation is an important source of VOCs and there are number of removal technologies available to treat polluted stream, among them biological treatment method is an option which uses the natural ability of microorganisms to consume the pollutants[5]. It is cheaper, cost-effective and very efficient removal without any secondary air pollutants. This technique has been applied successfully to control a number of air pollutants such as odours, VOCs and hazardous substances[6-7]. The biological treatment is more often used for low concentration VOCs treatment (<3000 mgm^{-3}) and has advantage of simple configuration, low capital and operating costs and minimum secondary pollutant production[8-9]. Biofilteration or "bio-oxidation" is an emerging energy efficient technology for control of VOCs[10]. It is used to eliminate contaminants from air using microorganisms, which are immobilized on a surface of solid support media. The aim of present study is to evaluate the performance of a biofilter packed with corn-cob as packing material and inoculated with a mixed culture in treating benzene vapour stream under variable loading conditions. Performance was assessed by determining benzene removal efficiencies, elimination capacities and microbial concentration by varying the process parameters and operating conditions.

Material and Methods

Microorganisms: A bacterial strain namely *Bacillus sphaericus* was used in this study. *B. sphaericus* was isolated from the biofilter unit which was used for the treatment of benzene. The details of the method for isolation and identification are presented in previous paper[11]. The isolated bacterial strain was identified from MTCC and IMTECH, Chandigarh, India, as *Bacilus sphaericus* and were assigned a number MTCC-8103. The bacterial strain was further confirmed by partial 16S rDNA sequence analysis[12].

Acclimatization of Culture Media: Cultivation of microorganism were performed by *B. sphaericus* in 500 mL flask containing 100 mL of the nutrient solution with benzene as the sole carbon source. The nutrient solution used had the following composition per liter of water: K_2HPO_4 (0.91 g),

$Na_2HPO_4.2H_2O$ (2.39 g), $FeSO_4.7H_2O$ (0.2 g), $MgSO_4.7H_2O$ (2.0 g), $MnSO_4.7H_2O$ (0.88 mg), $Na_2MoO_4.2H_2O$ (1 mg), $CaCl_2$ (3 mg), $ZnSO_4.7H_2O$ (0.04 mg), and $CoCl_2.6H_2O$ (0.04 mg). The cultures were acclimatized to benzene by exposing the culture in a series of shake flasks. The startup of acclimatization was obtained by inoculating 100 mL of basal salt medium (BSM) with *B. sphaericus* from nutrient agar slants under sterile conditions in presence of 10 mgL^{-1} of benzene. After 48 h of incubation at 30 ^oC, 5 mL of this culture was added to the fresh BSM as inoculum in presence of 10 mgL^{-1} of benzene. 48 h later, a third fresh BSM was also inoculated with 5 mL of the last culture to insure that the both bacteria were already adapted to benzene.

Experimental set-up: In this experiment a laboratory scale biofilter was set-up as shown in figure-1. The biofilter was made of Persplex (acrylic glass) tube with an internal diameter of 14 cm and packing bed height of 60 cm. The packing material was supported by the acrylic sieve plate. The nutrient fed was collected at the bottom of the biofilter. The biofilter was filled with a packing material which supported thin biological film. The packing material consisted of corn-cob with equivalent-volume diameter of 2.5 cm and porosity of 66.1%. The activated sludge obtained from the secondary clarifier of a wastewater treatment plant was used as the microbial seed in the biofilter. The suspended microorganisms were allowed to settle for 12 h and the supernatant water was then discarded to obtain concentrated sludge. The packing material was inoculated with benzene acclimated mixed microorganisms. The bed material plays a important role in the performance and steady operation of biofilter; therefore, it is desirable to develop novel low-cost and high-efficiency filter media[13]. Furthermore, environmental factors such as temperature and water content, and microbial community of biofilter need to be monitored, as they also have effects on the efficiency of biofilter[14]. The biofilter was placed within a temperature control box to regulate the temperature of benzene gas stream. At first, compressed air was passed through a filter to remove particulate matter. After purification, the air stream was distributed to humidifier and benzene bottle. Then humidified air and benzene vapor were mixed in a mixing chamber and the mixture was passed through the packed bed. The corn-cob compositions are presented in table-1. The full size corn-cob were procured from a local agricultural site, dried in an oven and cut into equal size cylindrical pieces. Almost equal sized pieces, having size 1.5 cm length and 2.0 cm diameter were prepared and they were washed twice with distilled water and then with Millipore water and finally dried in an oven at 105^oC for 5 h. After drying, the pieces were sterilized at 15 psi for 20 min. All experiments were conducted in a temperature controlled chamber at 30 ± 2 ^oC.

Analytical Method: Benzene gas concentrations were analyzed by using a MICHRO 9100 gas chromatograph equipped using a capillary column type HP 5 and a flame ionization detector (FID) connected with a computing integrator. The injector, oven and detector temperature were maintained at 210^oC, 60^oC and 230^oC, respectively. The fuel gas and carrier gas was hydrogen with a flow rate of 5 $mLmin^{-1}$. The air samples were collected from the various sampling ports in an airtight syringe. 50 µL of air samples were injected into the FID gas chromatograph for analyze. Air samples with known benzene were used for the calibration according to the standard procedure[15]. Gas samples were collected at a regular time intervals from the inlet, outlet as well as from the sampling ports provided at the biofilter by using an airtight syringe and analyzed for residual benzene.

Performance evaluation of biofilter: The parameters to measure performance of the biofilter are removal efficiency (%) and elimination capacity (gm^{-3} h^{-1}). They have been estimated by the following equations:

Inlet loading rate, $$LR = \frac{QC_i}{V_b} \quad (1)$$

Elimination capacity, $$EC = \frac{Q(C_i - C_o)}{V_b} \quad (2)$$

Removal efficiency, $$RE(\%) = \left(1 - \frac{C_o}{C_i}\right) \times 100 \quad (3)$$

where C_i and C_o is the inlet and outlet benzene concentration. V_b is the volume of bed. Performance of biofilter was investigated with respect to the loading rate (LR) of benzene in $gm^{-3}h^{-1}$. To do that, loading rate was expressed in terms of the combination of two parameters: benzene concentration (gm^{-3}) and flow rate ($Lmin^{-1}$). These two parameters were actually varied in the reactor to study the performance of the reactor to maintain the pre calculated loading rate[6].

Results and Discussion

Performance evaluation of biofilter under variable loading conditions: To evaluate the biofilter performance with fluctuating loading conditions, step changes in benzene concentration or in empty bed residence time (EBRT) were conducted for 68 days (phase I - V) in the bioreactor. The relative proportions of the benzene for each loading condition are shown in table-2. Phase I, II, III, IV and V lasted for 18, 10, 12, 14 and 14 days, respectively. Average loading rate applied to biofilter was gradually increased from 7.58 to 20.25 $gm^{-3}h^{-1}$. corresponding EBRT also decreased from 3.06 to 1.15 min. In each phase the inlet concentration effects on the total performance of the reactor were investigated. The different gas flow rates ranged from 3 to 8 lpm and various concentrations with the maximum amount of 0.1006 gm^{-3} were applied to the bed. To ensure accuracy and repeatability of results, various conditions were tested for several times and in different time intervals. Overall performance of the bed in the form of inlet, outlet concentration and removal efficiency for a period of more than two months is presented in figure-2. The system was started with flow rate of 3 lpm corresponding to an EBRT of 3.06 min with an inlet benzene concentration of 0.0941 gm^{-3}. Gradual increase in removal efficiency was observed (figure-2)

to about 99.8% after about 18 days. The results are consistent with the reported acclimation periods from several weeks to several months[16]. Inoculation of the biofilter media with adapted microbial aggregates greatly reduces the acclimation time of biofilter[17-19] to as low as 2 days[20]. Steady-state conditions were observed after day 16 of the operation in phase I.

Performance evaluation of Biofilter under Removal Efficiency: To study of the performance of the reactor in the term of removal efficiency are presented in the figure-2. The reactor had been operated into five phases and total time of operation was 68 days. The each run was operated till pseudo steady state is achieved. Pseudo steady state was presumed when the changes in the benzene removal efficiency were within 5% for three successive days. The removal efficiencies in each run gradually increases, reached stable, and then rapidly decreases after a sudden change in EBRT or influent concentration. As in figure-2, after 1 day of operation in phase I, overall efficiency in biofilter was 69.6% and by day 6 it was more than 81.4%. Performance was continuing to improve but at slower rate, reaching 91.5% on day 10 and more than 99% on day 18. On day 19, the start of phase II, loading rate has been increased by nearly 1.5 times from average 1.90 g m^{-3} h^{-1} to 2.51 g m^{-3} h^{-1}. In this phase flow rate was constant at 4 lpm, but concentration was varied from 0.0901 to 0.0982 g m^{-3}. Due to sudden changed in loading rate to the reactor, the removal efficiency was declined at 80.3%. The percentage of benzene removed in the biofilter increased with time, but at less rapid rate. It reached to an average removal of more than 96% by day 23. Phase II was lasted for almost 10 days. On day 29, at the beginning of period III, the flow rate entering the biofilter was increased by near to a factor of 1.5 times of initial phase. The residence time was 1.84 min while the target influent benzene average concentration was 0.09603 gm^{-3}. Thus, the organic loading rate to the biofilter was increased to 3.132 g m^{-3} h^{-1}. As shown in figure-2, one day following the increase in loading rate, only 77% of the influent benzene was removed. Same response similar to phase II had been observed once again. There were initial high sudden decrease in removal efficiency from 99% to less than 77% and then there was the recovery in the removal efficiency up to 78%. In the fourth phase, gas flow rate was fixed at 6 lpm. Only the concentration of benzene was changed as 0.0968 gm^{-3}. Due to change in the concentration of benzene the loading rate was also changed from 3.132 g m^{-3} h^{-1} to 3.789 g m^{-3} h^{-1}. During this phase it was observed that the removal efficiency was also changed in initial days. The removal efficiency of benzene again achieved from 82 to 99% . On day 55, the start of phase V, the flow rate was further increased at 8 lpm. To achieve the loading rate of 5.0386 g m^{-3} h^{-1}, inlet concentration was increased up to 0.0965 gm^{-3}. This was done to understand performance of the reactor. Here residence time was low but the concentration was high. In this phase the removal efficiency was reached high as 75% as compare to the phase three. It is clear from figure that if the residence time is high, the degradation is less.

Performance evaluation of Biofilter under Elimination Capacity: The biofilter performance was also evaluated in terms of the elimination capacity (EC) of benzene for the various loading rate which is, defined as the amount of benzene for the various loading rate, degraded per unit of reactor volume and time. The EC, which reflects the capacity of the biofilter to remove the pollutants, is plotted in figure-3 as a function of inlet benzene load. The elimination capacities of benzene increased with the increase in influent benzene concentration. In figure-3, symbols represent the experimental data of benzene while a dotted line indicates the 100% removal. Significant variation of the EC in various phases was observed due to the change in influent concentration and removal rate. From the this figure it is clear that when the influent benzene loadings were less than 2.510 g m^{-3} h^{-1}, nearly 100% removal could be achieved in phase II. The maximum elimination capacity of the biofilter was 4.492 g m^{-3} h^{-1} at inlet benzene load of 5.038 g m^{-3} h^{-1} in phase V. It was observed that the maximum elimination capacities of benzene removal in biofilters inoculated with *Pseudomonas* sp. NCIMB 9688 and packed with raw and sieved sugarcane bagasse and with peat were 3.2, 6.4 and 26 g m^{-3} h^{-1} at 6.1, 12 and 31 g m^{-3} h^{-1} loading rates, resulting in 52, 53 and 84% removals, respectively[21]. In previous study, the removal of benzene vapor from gaseous streams in two identically sized lab-scale biofiltration columns were used and observed a maximum elimination capacities, obtained at an inlet load of 6.12 g m^{-3} h^{-1} as 3.50 and 3.80 g m^{-3} h^{-1} with raw and ground sugarcane bagasse, respectively[22]. The biodegradation of ethanol in bioreactor inoculated with *Candida utilis (C. utilis)* and packed with sugar cane bagasse also found in other prvious study[23]. At a higher aeration rate (ethanol load of 153.8 g m^{-3} h^{-1}), the biofilter displayed an average removal efficiency of 70% at an elimination capacity of 107.7 g m^{-3} h^{-1}.

Conclusion

The performance of a lab scale biofilter and effects of various operating parameters were studied. After inoculation, microbial acclimation needed approximately 30 days and simultaneously biodegradation of benzene was observed. Steady state was achieved within 18 days of operation. The results demonstrated the use of corn-cob as a support media in the biofilter for the treatment of air streams contaminated with benzene compounds vapors is reliable, efficient and easy to operate and maintain. When the influent benzene loadings were less than 1.9033 g m^{-3} h^{-1}, nearly 99.95 % removal could be achieved. The corn-cob packing material is appeared to be efficient packing materials for low to medium loading rate. The maximum elimination capacity of the biofilter was 2.377 g m^{-3} h^{-1} at inlet benzene load of 2.5109 g m^{-3} h^{-1} at EBRT of 138 s in phase II. Moreover, the pure strain *B. sphaericus*, isolated in this study, has higher potential for degradation of benzene.

References

1. Mohseni M. and Grant A.D., Bio filtration of mixtures of hydrophilic and hydrophobic volatile organic compounds, *Chem. Eng. Sci.*, **55**,1545–1558 **(2000)**

2. Comprehensive Environment Response, Compensation and Liability Act (CERCLA), USEPA **(2005)**

3. Shim H., Shin E. and Yang, S.T., A continuous fibrous-bed bioreactor for BTEX biodegradation by a co-culture of *Pseudomonas putida* and *Pseudomonas fluorescens, Advan. Environ. Res.,* **7**, 203-216 **(2002)**

4. ASTDR, Priority list of hazardous substances, Agency of toxic substances and disease registry **(1997)**

5. Thakur Prabhat Kumar, Rahul, Mathur Anil Kumar and Balomajumder Chandrajit, Biofiltration of Volatile Organic Compounds (VOCs) – An Overview, *Res. J. Chem. Sci.,* **1(8)**, 83-92 **(2011)**

6. Mathur A. K., Majumder C. B. and S Chatterjee, Combined removal of BTEX in air stream by using mixture of sugar cane bagasse, compost and GAC as biofilter media, *J. Hazard. Mater.,* **148**, 64–74 **(2007)**

7. Mathur A. K., Sundaramurthy J. and Balomajumder C., Kinetics of the removal of mono-chlorobenzene vapour from waste gases using a trickle bed air biofilter, *J. Hazard. Mater.,* **137 (3)**, 1560–1568 **(2006)**

8. Khan F. I. and Ghoshal A. K., Removal of volatile organic compounds from polluted air, *J. Loss Prev. Process Ind.,* **13**, 527–545 **(2000)**

9. Burgess J.E., Parsons S.A. and Stuetz R.M., Developments in odour control and waste gas treatment biotechnology: a review, *Biotechnol. Adv.,* **19**, 35–63 **(2001)**

10. Bohn H., Consider biofilter for decontaminating gases, *Chem. Eng. Prog.,* **88(4)**, 34-40 **(1992)**

11. Rahul, Mathur Anil Kumar and Balomajumder Chandrajit, Biodegradation of Waste Gas containing Mixture of BTEX by *B. Sphaericus*, *Res. J. Chem. Sci.*, **1(5)**, 52-60 **(2011)**

12. Kristiansen Anja, Pedersen Kristina Hadulla, Nielsen Per Halkjer, Nielsen Lars Peter, Nielsen Jeppe Lund and Schramm Andreas, Bacterial community structure of a full-scale biofilter treating pig house exhaust air, *Systematic and Applied Microbiol,* **34**, 344-352 **(2011)**

13. Babbitt C.W., Pacheco A. and Lindner A.S., Methanol removal efficiency and bacterial diversity of an activated carbon biofilter, *Bioresour Technol,* **100**, 6207–6216 **(2009)**

14. Moon L.E., Lee S.Y., Lee S.H., Ryu H.W. and Cho K.S., Earthworm cast as a promising filter bed material and its methanotrophic contribution to methane removal, *J. Hazard. Mater.,* **176**, 131–138 **(2010)**

15. Lodge J.P., Methoda of Air Sampling and Analysis, Lewis Publishing Inc, New York **(1989)**

16. Juneson C., Ward O.P. and Singh A., Microbial treatment of a styrene-contaminated air stream in a biofilter with high elimination capacities, *J. Ind. Microbiol Biotechnol*, **26**1, 96-202 **(2001)**

17. Shareefdeen Z and Baltzic B.C., Biofiltration of toluene vapor under steady-state and transient conditions: theory and experimental results, *Chem Eng Sci,* **49,** 4347-4360 **(1994)**

18. Acuna M.E., Perez F., Auria R. and Revah S., Microbiological and kinetic aspects of a biofilter for the removal of toluene from waste gases, *Biotechnol Bioeng,* **63**, 175-184 **(1999)**

19. Jorio H., Payre G. and Heitz M., Mathematic modeling of gas-phase biofilter performance, *J. Chem Technol Biotechnol*, **78**, 834-846 **(2003)**

20. Arnold M., Reittu A., VonWright A., Martikainen P. J. and Suihko M. L., Bacterial degradation of styrene in waste gases using a peat filter, *Appl Microbiol Biotechnol*, **48**, 738-744 **(1997)**

21. Zilli M., Daffonchio D., Di Felice1 R., Giordani M. and Converti A., Treatment of benzene-contaminated airstreams in laboratory-scale biofilters packed with raw and sieved sugarcane bagasse and with peat, *Biodegradation*, **15**, 87-96 **(2004)**

22. Sene L., Converti A., Felipe M. G. A. and Zilli M., Sugar Cane Bagasse as Alternative Packing Material for Biofiltration of Benzene Polluted Gaseous Stream -a Preliminary Study, *Biores Techn,* **83**, 153-157 **(2002)**

23. Christen P., Domenech F., Michelena G., Auria R., and Revah S., Biofiltration of volatile ethanol using sugar cane bagasse inoculated with *candida utilis*, *J. Hazard. Mater.*, **B89**, 253-265 **(2002)**

Table-1
Characteristics of the corn-cobs for biofilter

Parameters	**Corn-Cob**
Physical compositions	
Particle size (length /diameter) (cm)	1.5/2.0
pH	6.11
Moisture content (%) (at field capacity)	56.3
Dry filled weight (g)	187
Chemical compositions (%)	
C	43.2
H	7.097
N	0.0
S	0.0

Table-2
Operating conditions of benzene for each phase of experiments in corn-cob based biofilter

Phase	**Operating period (days)**	**Flow rate (L min^{-1})**	**Benzene concentration (g m^{-3}) range**	**Average loading (g m^{-3} h^{-1})**	**EBRT (min)**
I	0-18	3	0.0941-0.0992	1.9033	3.06
II	19-28	4	0.0901-0.0982	2.5109	2.3
III	29-40	5	0.0893-0.1006	3.1316	1.84
IV	41-54	6	0.0932-0.0988	3.7891	1.53
V	55-68	8	0.0919-0.0987	5.0386	1.15

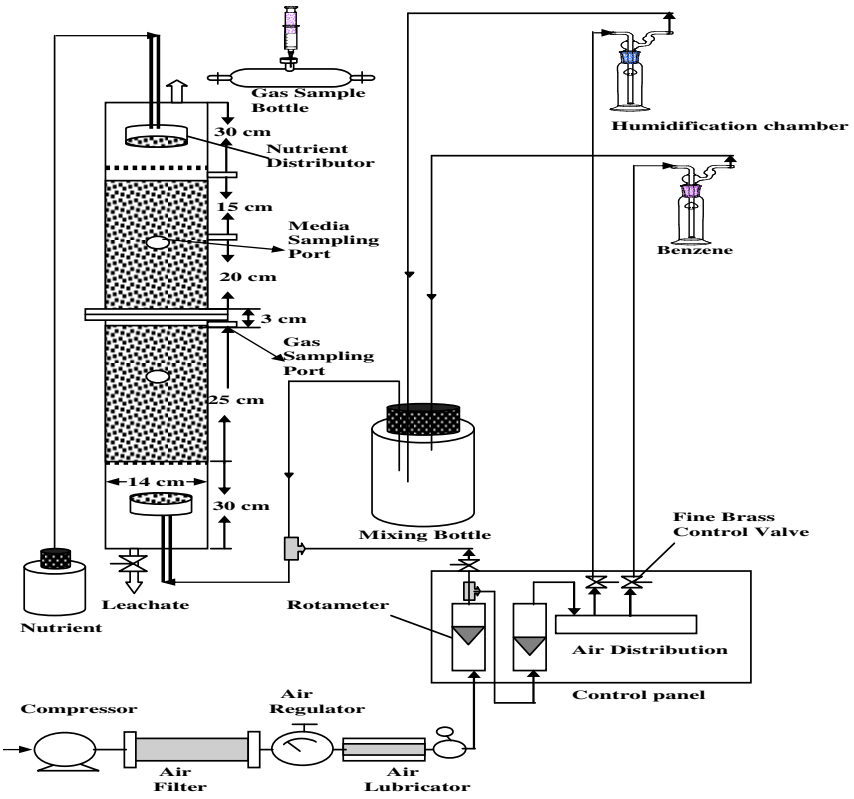

Figure-1
Schematic of biofilter unit treating benzene

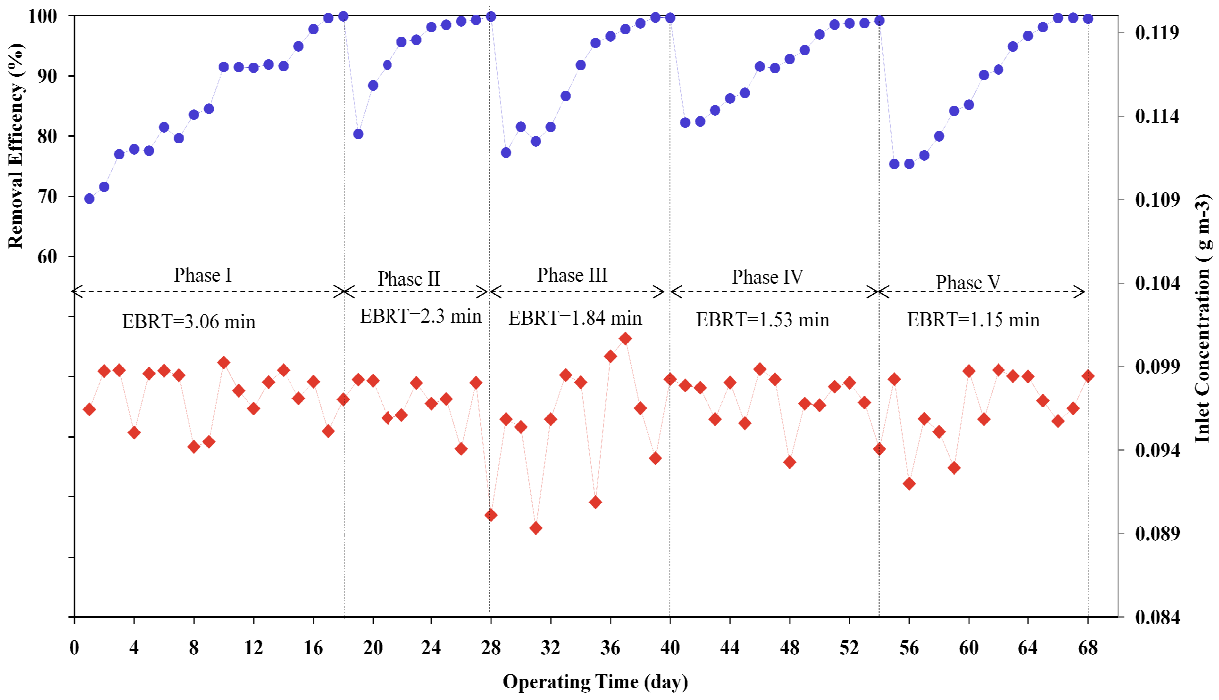

Figure-2
Overall performance of corn-cob based biofilter in removal of benzene with in time

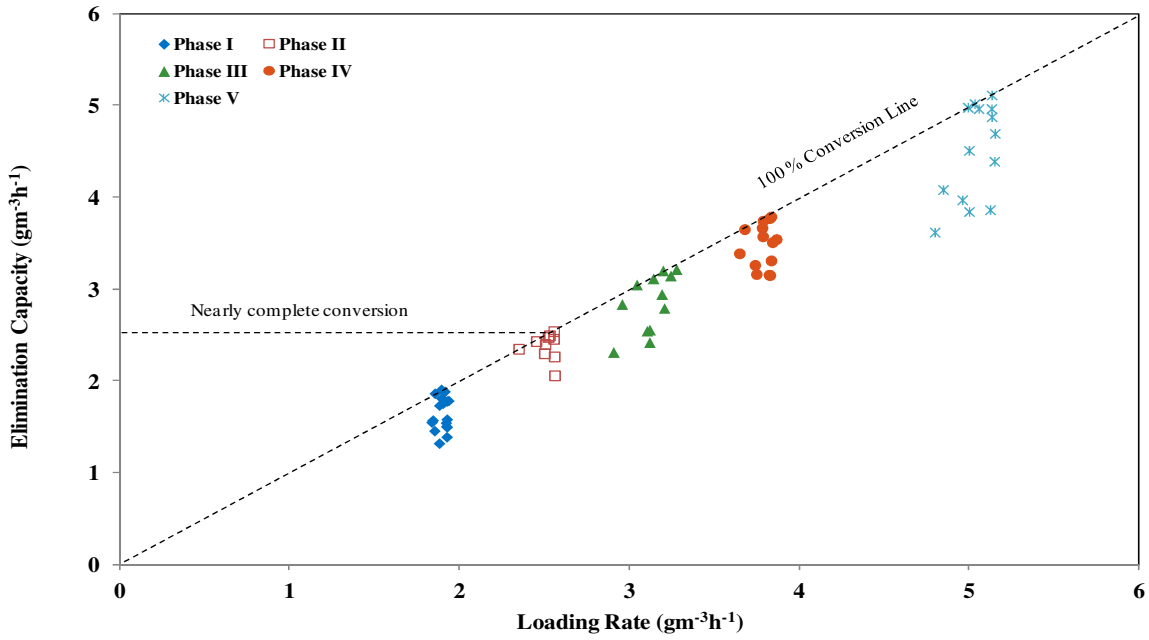

Figure-3
Overall elimination capacity in the biofilter as a function of inlet load of benzene