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A Study on Degradation of Phenol by *Pseudomonas species*

Sivasankari.P*, Prakash.B, Jeyaprakash.S.T, Thiyagarajan.D, Palanivel.P

Department of Biotechnology, PGP college of Arts and Science, Namakkal, India

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Sree Narayana Guru College, K.G. Chavadi, Coimbatore, India-641105.

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***Corresponding author**
Sivasankari.P
Department of Biotechnology
PGP College of Arts and Science,
Namakkal, India
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Abstract

The industrialization and extensive use of pesticides in agriculture and the pollution of environment with man-made organic compound has become a major problem. Phenolic compounds are abundant in the biosphere as components of the complex polymer lignin, humicacids and as environmental pollutants resulting from various industrial activities. Phenol pollution is mainly associated with pulp industries, coal mines and refineries. In industries the chemical process of degradation of various hazardous materials has been replaced by bioprocess. In this study *Pseudomonas pseudoalcaligens and Pseudomonas putida* obtained from IMTECH. Mineral medium was prepared and the microorganisms were inoculated. The two cultures were taken for conical flask and bioreactor method. The samples were collected and analyzed for phenol estimation method, COD, UV-visible spectrometry. The higher amount of phenol degradation was observed in *Pseudomonas putida* when compared to *Pseudomonas pseudoalcaligens*. Phenol degradation results indicate bioreactor method is highly effective when compared to conical flask method.

Keywords: Degradation, *Pseudomonas pseudoalcaligens, Pseudomonas putida*

1. Introduction

Bioremediation is a pollution control technology that uses biological systems to catalyze the degradation a transformation of various toxic chemicals to less harmful forms. [1]. Phenol is a man made as well as naturally occurring aromatic compound and an important intermediate protect in the biodegradation of natural & industrial aromatic compounds.[2]. Aqueous phenolic effluents or relatively common industrial waste being produced in several industries and operation such as tannery, yas & coke oven industries.[4].

2. Materials and Methods

Nutrient broth media

Peptone-5g
Sodium chloride-5g
Yeast extract-3g
Beef extracte-3g
Distilled water-1000ml

Organisms.

Pseudomonas pseudoalcaligens (IMTECH 2651)
Pseudomonas pudida (IMTECH 1192)

Inoculum preparation

Primary inoculum:

The bacterial cultures obtained from IMTECH were incubate in a 100 ml nutrient broth. The culture were incubated at 37^0 C for 24 hrs.

Secondary inoculum

1ml of primary inoculum was taken and incubated in a 200ml of nutrient broth. The culture was then incubated at 37^0 C for 24 hrs.

Assessment of Phenol Tolerance by *Pseudomans sp.*

The bacterial culture was taken in nutrient agar plates with varying concentration of phenol.yhe concentration ranges from(0.2ml,0.4ml,0.6ml,0.8ml,1.0ml) per liter. The inoculate agar plates were observed for growth after incubation period. The plate of highest concentration showing growth was taken as the tolerance limit.[4]

Estimation of Phenol–Bromination method

The brominating winkles solution is prepared by dissolving 15g of potassium bromide and 2.8g of potassium in per litre.5ml of sample was taken from the medium the medium and centrifuged at 2000 rpm for 20 minutes. The supernatant was collected and taken into iodination flask and made up to 50ml.To this 50 ml winkles solution and 5ml of Con HClwas added and kept in shaker for 30 minutes. After 30 minutes 20 ml of potassium iodine was added and titrated against 0.1 N sodium thiosulphate by staras an indicator.The end point was the disappearance of blue colour.[4]

Degradation study

Conical flask method :

500µl of phenol was added to 1 litre of the mineral medium. To that 10 ml of two different *Pseudomonas* cultures were added to two conical flask containing mineral medium with phenol and kept in the orbital shaker for 5 days. The sample was collected at 0 and every 24 hours of interval for 5 days and the sample were centrifuged at 2000 rpm for 20 minutes.[6,7]

Bioreactor method:

The mineral medium was sterilized in autoclave at 121 c for 15 minutes. 100 ml of this broth was dispensed into the reactor and was sterilized insitu at 1 atm for 15 minutes and to that 10 ml of *Pseudomonas sp* were inoculated.the sample was collected and subjected to COD and phenol estimation test.[8,9].

3. Results and Discussion

Assessment of phenol tolerance:

The *Pseudomonas sp* were streaked on nutrient agar plate. The plate of highest concentration showing growth at 0.6 ml concentration was found.

Estimation of phenol degradation:

Pseudomonas pseudoalcaligens was observed to degrade 23.52% in conical flask and 29.41%in bioreactor at the end of 120hrs.

Table 1

Time in hours	Sample	
	Blank	*Pseudomonas pseudoalcaligens*
0	48.1	46.4
24	48.3	46.7
48	48.3	46.8
72	48.2	46.8
96	48.3	47.0
120	48.2	46.9

Pseudomonas putida was observed to degrade 41.17% in conical flask and 47.05%in bioreactor at the end of 120hrs.

Table 2

Time in hours	Sample	
	Blank	***Pseudomonas putida***
0	48.1	46.4
24	48.4	46.8
48	48.3	46.8
72	48.3	46.9
96	48.3	47.0
120	48.3	47.3

Chemical Oxygen Demand:

Pseudomonas pseudoalcaligens was observed to degrade conical flask 36.36% and bioreactor 40.90% of the phenol present in the solution at the end of 120 hours.

Pseudomonas putida was observed to degrade conical flask 63.63% and bioreactor 68.12% of the phenol present in the solution at the end of 120 hours.

UV Visible spectrum analysis:

Appearance of a distinct peak in the wave length 286nm and the OD value 0.257 for the sample *Pseudomonas pseudoalcaligens* after 24 hours of incubation the intensity of the characteristics peak at 0.256 decrease indicating the reduction in the phenol content. *Pseudomonas putida* after 24 hours of incubation the intensity of the characteristics peak at 0.256 decrease indicating the reduction in the phenol content.

4. References

1. A.R.Kulanthaivel, V.Venkadeshwaran, R.Veerasamy, Basic principles of practical chemistry, pp: 290-292.
2. G.Gonzalez, G.Herrera, T.Garacia, M.Pena, Biodegration of industrial wastewater in a fluidized bed reactor with immobilized cells of *Pseudomonas putida* .Bioresource Technology, **2001,** 80: 137- 142.
3. P.S.Santhanam. Studies on the phenolic constitutents of Dhawa leaves: Part2:Affinity studies. Leather science, **1996**, Vol. 19. pp: 303-306.
4. Seun Mi Park *el al* ,Hoo Hwi park, Woon Ki Lim, Hae Ja Shin. A new variant activator involved in the degration of phenolic compounds from a strain of *Pseudomonas putida*. Journal of Biotechnology, **2003**, 103, pp: 227-236.
5. V.Sridevi and M.V.V.Chandana Lakshmi. Batch Kinetic studies in phenol degradation. Indian Journal of Environmental Protection, **2007**, Vol.27 ,IJEP27
6. Zee Ronen and Aharon Abeliovich. Anaerobic-Aerobic process for Microbial degradation of Tetrabromobisphenol. Applied and Environmental microbiology. **2000**, Vol-66, No-6, pp: 2372-2377.
7. B.Madhan, J.Raghava Rao, T.Ramasami. Comparision of the toxicity threshold of water pollutants to bacteria, algae and protozoa in the cell multiplication inhibition test. **1980**, pp: 231-241
8. J. Galindez–maye, J.Ramon-Gallegos, N.Ruiz-ordaz. Biodegration by yeast *Candida albicans* in a fluidized bed reactor. Journal of Biochemical engineering.Vol:38, pp: 147-157
9. T.Godjevargova and G.Burdelova. Biodegration of phenol by immobilized *Trichosporon cutaneum.* Biochemical Engineering Journal. **2006**, Vol-41, pp: 2342-2346.