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ScienceDirect
Journal of Hazardous Materials 149 (2007) 60–66
Journal of Hazardous Materials
www.elsevier.com/locate/jhazmat

Characteristics of phenol biodegradation in saline solutions by monocultures of *Pseudomonas aeruginosa* and *Pseudomonas pseudomallei*

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Received 26 June 2006; received in revised form 14 March 2007; accepted 14 March 2007
Available online 19 March 2007

Abstract

Phenol is a highly toxic and carcinogenic compound and its biodegradation is very important to meet the environmental regulations. Two bacterial strains capable of utilizing phenol as a sole source of carbon were isolated from the wastewater of a pharmaceutical industry. On the basis of morphological and biochemical characteristics these strains were identified as *Pseudomonas aeruginosa* and *Pseudomonas pseudomallei*. Both of these strains were very efficient for phenol degradation. *P. pseudomallei* degraded phenol at a maximum concentration of 1500 mg L^{-1} within seven days with a specific growth rate of 0.013 h^{-1} and phenol degradation rate of 13.85 mg L^{-1} h^{-1}. Maximum initial concentration of phenol utilized by *P. aeruginosa* was 2600 mg L^{-1} with 0.016 h^{-1} specific growth rate and 26.16 mg L^{-1} h^{-1} phenol degradation rate. Moreover, the effect of various salts i.e., NaCl, KCl, Na_2SO_4 and K_2SO_4 on the growth of these strains and phenol degradation rate (at 1000 mg L^{-1}) was studied. In the presence of these salts, *P. aeruginosa* showed up to 1.53 and 1.34 times faster phenol degradation rate and specific growth rate, respectively as compared to *P. pseudomallei.* In addition, *P. aeruginosa* exhibited higher chemical oxygen demand (COD) and biochemical oxygen demand (BOD) reduction rates as compared to the strain *P. pseudomallei*.
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Keywords: Biodegradation; Pharmaceutical industry; Phenol; Salts; Wastewater

1. Introduction

The growth of industry has brought with it the problem of pollution and hazardous wastewater. The challenges for the industries are to solve these critical problems. Phenolic compounds are hazardous pollutants that enter the environment through wastewater discharges from a variety of industries like leather, textiles, phenol-formaldehyde resin, oil refinery, coking plant, pharmaceutical, coal conversion, etc. and also through the decomposition of attached algae and phytoplanktons [1–3].

Phenol has adverse effect on aquatic life, plants and is toxic to bacterial growth, however there are some microorganisms that can tolerate and use it as a carbon and energy source [4]. Phenol biodegradation studies have been performed with different bacterial species [5,6] including halophilic, moderately halophilic species of *Halomonas* sp. [7], and adapted cells of *Pseudomonas* sp. [8]. With a phenol-degrading thermophile *Bacillus thermoleovorans,* very high specific growth rate (2.8 h^{-1}) was measured at 15 mg L^{-1} phenol concentration, while at higher phenol concentrations (100–500 mg L^{-1}) growth rates were less as compared to those at low phenol concentration [9]. Straube et al. [10] reported that at higher phenol concentration ($\geq$250 mg L^{-1}), substrate inhibition was observed by increase in lag phase and decrease in growth rates. *Candida parapsilopsis* among 22 species isolated from phenol containing wastewater was found to be capable of growth in a medium with 1000 mg L^{-1} phenol [11].

Many aerobic phenol-degrading microorganisms have been isolated and the pathways for the aerobic degradation of phenol are now definitely well established. The first step consist of oxygenation of phenol by phenol hydroxylase enzymes to form catechol, followed by ring cleavage adjacent to or in-between the

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doi:10.1016/j.jhazmat.2007.03.046

two hydroxyl groups of catechol. Phenol hydroxylases ranging from simple flavoprotein monooxygenases to multicomponent hydroxylases, as well as the genes coding for these enzymes, have been described for few aerobic phenol-degrading microorganisms [4].

In addition to the phenol, industrial wastewater contains various salts that can inhibit the growth of microorganism used for phenol biodegradation. Although salts can be removed by physical processes such as reverse osmosis, ion exchange or electrodialysis, these methods are expensive. Another option to decrease the salt concentration for better biodegradability of organic matter is dilution of wastewater. However, this proves to be an inefficient use of fresh water resources and also creates a problem of significant increase in wastewater volume. Hence, there is considerable interest in the use of bacteria for phenolic wastewater treatment. To cope with relatively extreme conditions of industrial wastewater, more important are the bacteria that can grow in extreme conditions like pH, temperature and at high salt concentration [12].

Biodegradation of phenol in the presence of high concentration of salts, particularly sodium chloride has been reported [12–16]. Lower rate of phenol degradation was observed [13] at NaCl concentration of about 3% by using same media and culture. More than 99.5% phenol degradation was observed at 15% NaCl using halophilic bacteria in biofilm and sequencing batch reactors [14,15]. Halophilic bacteria have been used to remove phenol at concentrations of up to 320 mg L^{-1} in a medium containing 10% NaCl [16]. *Halomonas* sp. have been reported to degrade 100 mg L^{-1} phenol in model industrial waste containing NaCl (varying between 1 and 14% (w/v)) and showed optimum phenol utilization and hence growth at about 5% (w/v) NaCl [7]. Although the effect of NaCl on biodegradability of phenol has been studied well, the presence of other salts/anions/cations mostly present in industrial effluents that can affect biodegradation of phenol have been ignored. As industrial wastewater contains many salts at high concentrations, the strains capable of degrading phenol with high specific growth rate in the presence of high concentrations of different salts are particularly desirable.

The major aim of this work was to isolate and characterize microbial strains that besides their capability to degrade phenol in the presence of high salt concentrations have the ability to reduce chemical oxygen demand (COD) and biochemical oxygen demand (BOD) load as well. We have isolated two strains from the wastewater of a pharmaceutical industry and utilized these for biodegradation of phenol in the presence of various salts. The amount of phenol and the salts used in biodegradation studies were comparable to their magnitude in the industrial wastewater.

2. Materials and methods

2.1. Chemicals

All the chemicals used in these studies were of analytical grade and purchased from Sigma, Merck and BDH and were used as received. All the solutions/media were made in double distilled water. Bacteriological media were purchased from Difco and Oxoid.

2.2. Characterization of wastewater

Five representative composite wastewater samples were collected, from main outlet of a pharmaceutical industry in Kasur, Pakistan with 7 days intervals over a period of 35 days. Each time the wastewater was collected hourly for a whole working day and mixed to get a composite sample. The wastewater samples were shifted to the lab in iceboxes. Phenol concentration, COD, BOD, cations (Na^+ and K^+) and anions (Cl^- and ${SO_4}^{2-}$) in wastewater samples were determined according to standard methods [17]. The results thus obtained were employed to formulate media to be used for studying the effect of salts on phenol biodegradation (Section 2.6).

2.3. Enrichment, isolation and selection of bacterial strains

Bacterial strains used in these studies were isolated from a pharmaceutical industrial sludge and were identified based on the morphological, physiological, biochemical and carbon source utilization tests [18]. All microbial isolations were performed in mineral salts (MS) medium containing (g L^{-1}): K_2HPO_4, 4.3; KH_2PO_4, 1.7; $(NH_4)_2SO_4$, 2.69; $MgSO_4$, 0.2 and $CaCl_2$, 0.03. Phenol (analytical grade) was used as a sole source of carbon and energy. Autoclaved phenol solution (100 g L^{-1}) was added directly to the autoclaved MS medium (pH 7.0) to obtain desired phenol concentrations. Bacterial strains were isolated by enrichment culture techniques using 5 g of wet sludge as inoculum in 100 ml MS medium containing 100 mg L^{-1} phenol. The flasks were incubated at 37 °C with shaking at 120 rpm. Samples were withdrawn after 24 h and appropriate dilutions were plated onto MS−agar plates containing 50 mg L^{-1} phenol. Isolated colonies were re-streaked and the individual colonies were then transferred on nutrient agar plates to check purity of single colony. Liquid medium (MS) was used for further studies. The purity of the strains was further checked by microscopic examination. Eight morphologically different strains were isolated which could survive and grow in the presence of 200 mg L^{-1} phenol. These strains were further exposed successively to higher concentrations (up to 1500 mg L^{-1}) of phenol. Only two strains capable of vigorous growth in MS medium containing phenol (1500 mg L^{-1}) at 37 °C were obtained and purified. These two strains were named as NIBGE MB and NIBGE 3B before identification. Strains NIBGE MB and NIBGE 3B were exposed successively to higher concentrations of phenol (up to 3000 mg L^{-1}) in MS medium. NIBGE 3B could not grow above 1500 mg L^{-1} phenol, but NIBGE MB could utilize an initial phenol concentration of 2600 mg L^{-1}. These two bacterial strains were selected for phenol degradation studies.

2.4. Preparation of inoculum

Seed cultures of the selected bacterial strains were grown in 200 ml nutrient broth medium (0.8%) in 1 L conical flasks

equipped with side arms to facilitate turbidity measurements and incubated for 18 h on an orbital shaker (120 rpm) at 37 °C. Cells were recovered by centrifugation (15,000 × *g*, 15 min), and resuspended in autoclaved normal saline (0.85% NaCl in distilled water) and the optical density (OD) at 600 nm (Spectro UV–vis RS Spectrophotometer) was adjusted to 0.7 with normal saline. This inoculum was used at 2% (v/v) to study biodegradation of phenol unless otherwise mentioned.

2.5. Biodegradation of phenol

Phenol degradation and bacterial growth was observed by growing the selected bacterial strains in MS medium containing phenol to a maximum concentration of 1500 $mg\,L^{-1}$ (for strain 3B) and 2600 $mg\,L^{-1}$ (for strain MB) for 7 days at 37 °C in triplicates. For each experiment freshly prepared 2% (v/v) inoculum of OD 0.7 was used. Samples were taken at 24 h intervals and analyzed for bacterial growth and phenol concentration. Phenol degradation rate (Q_S, $mg\,L^{-1}\,h^{-1}$) and cell yield coefficient ($Y_{X/S}$, mg cell mg^{-1} phenol) during the time course study of the biodegradation processes were determined by using the method reported by Pirt [19]. Q_S was determined from the maximum slope in plot of phenol concentration ($mg\,L^{-1}$) versus time of incubation. Cell yield coefficient ($Y_{X/S,}$) was calculated as the dry cell mass per mass of phenol utilized. Specific growth rate per hour ($\mu = [dx/dt]/X$) was determined as suggested in Baileys and Ollis [20].

2.6. Experimental design to study the effect of salts on phenol biodegradation

Effect of salts on the biodegradation of phenol was determined by adding NaCl, KCl, Na_2SO_4, and K_2SO_4 separately to the growth medium to obtain approximately the same concentrations of anions or cations as found in pharmaceutical industry wastewater in triplicates. These studied were carried out in MS medium containing 1000 $mg\,L^{-1}$ phenol. The amounts ($g\,L^{-1}$) of salts added separately into the growth medium (MS) were: NaCl, 2.54; KCl, 3.23; Na_2SO_4, 6.125 and K_2SO_4, 3.77. Concentrations of different cations and anions present in pharmaceutical industry waste water and those used during phenol biodegradation studies are mentioned in Table 1.

2.7. COD and BOD reduction versus phenol degradation

Chemical oxygen demand and biochemical oxygen demand reduction following phenol biodegradation and bacterial growth was studied in 200 ml MS medium (in 500 ml flasks) containing 1000 $mg\,L^{-1}$ phenol as sole source of carbon and energy (no extra salts were added to the growth medium for COD and BOD reduction studies). Adapted cultures of *P. aeruginosa* and *P. pseudomallei* in their log phase of growth were used as inoculum (2%, v/v) and the cultures were incubated in an orbital shaker at 120 rpm and 37 °C. Samples were taken at 24 h intervals, centrifuged at 15,000 rpm and 4 °C for 10 min. Cell-free supernatant was used to determine phenol concentration, COD and BOD.

2.8. Analytical methods

The bacterial growth was measured as optical density at 600 nm (OD_{600}) using a UV–vis spectrophotometer and correlated to biomass concentration (dry cell mass $mg\,L^{-1}$). The optical density–biomass correlation was linear up to the cell concentration range of 300 $mg\,L^{-1}$ dry cells with an $OD_{600} = 1.00$. Phenol concentration, COD and BOD were determined using standard methods [17].

3. Results and discussion

3.1. Wastewater characteristics

The wastewater collected from the pharmaceutical industry showed a large variation in composition in terms of concentra-

Table 1
Characteristics of the pharmaceutical industrial wastewater, MS media and quantities of salts used to study effect of various cations and anions on phenol biodegradation by strains of *P. aeruginosa* and *P. pseudomallei*

Parameter	Characteristics of wastewater[a]	Characteristic of MS media[b]	Quantity of salt added to MS medium
pH	6.6 ± 2.07	7.0	–
Phenol ($mg\,L^{-1}$)	1053 ± 162	1000	–
COD ($mg\,L^{-1}$)	5097 ± 698	2032	–
BOD ($mg\,L^{-1}$)	1032 ± 216	773	–
Na^+ ($mg\,L^{-1}$)	1176 ± 182	998	NaCl, 2.5 $g\,L^{-1}$
		1183	Na_2SO_4, 6.125 $g\,L^{-1}$
K^+ ($mg\,L^{-1}$)	1612 ± 143	1692	KCl, 3.23 $g\,L^{-1}$
		1690	K_2SO_4, 3.37 $g\,L^{-1}$
Cl^- ($mg\,L^{-1}$)	1572 ± 214	1540	KCl, 3.23 $g\,L^{-1}$
		1570	NaCl, 2.5 $g\,L^{-1}$
SO_4^{2-} ($mg\,L^{-1}$)	2280 ± 30	2080	K_2SO_4, 3.37 $g\,L^{-1}$
		4140	Na_2SO_4, 6.125 $g\,L^{-1}$

[a] Each value is a mean of five composite samples of wastewater collected at different times and ± indicates standard deviation among them.
[b] COD and BOD of media (MS media + phenol + salts) are due to added phenol. Concentrations of ions mentioned in this column were obtained by adding certain salts at concentrations mentioned in the following column.

tions of phenol, anions/cations and pH. This might be due to the different production units operating in the pharmaceutical industry at different sampling times. Table 1 summarizes such variations indicating the mean value and the standard deviation. There is high chemical oxygen demand to biochemical oxygen demand ratios (COD/BOD) i.e. 4.93 indicating low biodegradability of phenolic and other compounds present in wastewater of pharmaceutical industry. High COD value (5097 mg L^{-1}) as compared to phenol concentration (1053 mg L^{-1}) showed that in addition to phenol other organic matter was also present in the wastewater (an aqueous solution containing 1000 mg L^{-1} phenol has COD value of approximately 2000 mg L^{-1}). The pH values were around 6.6 ± 2.07. The concentration of cations i.e. Na^+ and K^+ in the wastewater were 1176 ± 182 mg L^{-1} and 1612 ± 143 mg L^{-1}, respectively. Two anions Cl^- and $SO_4{}^{2-}$ were found at concentrations of 1572 ± 214 mg L^{-1} and 2280 ± 30 mg L^{-1}, respectively. These observations (concentrations of cations and anions) were exploited to formulate media (by determining the right amount of salt to be added to MS medium) to study phenol biodegradation in the presence of salts.

3.2. Isolation and characterization of bacterial strains

Eight morphologically different bacterial strains capable of biodegrading phenol were isolated from activated sludge of a pharmaceutical industry using enrichment culture technique. Two strains capable of tolerating high phenol concentrations with high degrading efficiency and relatively rapid growth in minimal medium i.e. NIBGE-3B and NIBGE-MB were selected for phenol degradation studies. These strains were identified based on morphological, physiological and biochemical tests to be *P. pseudomallei* (NIBGE 3B) and *P. aeruginosa* (NIBGE MB). Maximum initial concentration of phenol utilized by *P. pseudomallei* was 1500 mg L^{-1}, however *P. aeruginosa* could utilize up to 2600 mg L^{-1}phenol concentration.

3.3. Biodegradation of phenol

The time course for phenol (1500 mg L^{-1}) degradation by *P. pseudomallei* is presented in Fig. 1. This strain was able to completely utilize phenol within 7 days with 0.013 h^{-1} and 13.85 mg L^{-1} h^{-1} specific growth rate and substrate consumption rate, respectively (Fig. 1). Whereas *P. aeruginosa* could completely degrade 2600 mg L^{-1} phenol within 7 days (data not shown) with 0.016 and 26.16 mg L^{-1} h^{-1} specific growth rate and substrate consumption rate, respectively. *P. pseudomallei* could not tolerate this high concentration of phenol in the medium. Both the strains showed 24 h lag phase for growth/phenol biodegradation. However, at lower phenol concentrations (<1000 mg L^{-1}) no or little lag phase was observed for growth and phenol biodegradation by both *P. aeruginosa* and *P. pseudomallei* (data not shown). Straube et al. [10] reported that at higher phenol concentration (≥250 mg L^{-1}), substrate inhibition was observed by increase in lag phase. Also, the lower the initial concentration of phenol in the growth medium, the less time it took to be degraded completely by both *P. pseudomallei* and *P. aeruginosa* (data not shown). Moreover, when adapted cultures of *P. pseudomallei* or *P. aeruginosa* were used as inoculum, there was no lag phase and the system reached to the exponential growth phase quickly. The previous reports on biodegradation of phenol in batch processes were to a maximum concentration of 1750 mg L^{-1} [1]. To our knowledge, capacity of phenol degradation by *P. aeruginosa* isolated in this study is higher than that reported by other workers (Table 2). This might

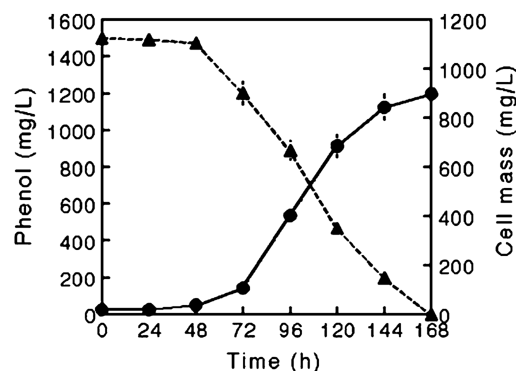

Fig. 1. Cell mass formation (●) and phenol utilization (▲) during growth of *P. pseudomallei* in shake flask cultures grown in MS medium containing phenol (1500 mg L^{-1}) as sole source of carbon and energy. Error bars represent standard deviation among three replicates.

Table 2
Comparisons of bacterial strains isolated in this study to previously characterized phenol degrading microorganisms based on the capacity to degrade initial phenol concentrations in the medium

Sr. no.	Reference	Bacterial strain	System	Phenol (mg L^{-1})	Other conditions (°C/pH)
1	[21]	Mixed culture I	Batch	900	28/6.6
		Mixed culture II (filamentous organisms)	Batch	1000	28/6.6
2	[22]	*P. putida*	Batch/continuous	700	30/6.2–6.7
3	[23]	*P. putida*	Continuous	500	30/60
		T. cutaneum	Continuous	900	28/6.8
4	[24]	*P. putida*	Continuous	700	30/6.2–6.8
5	[25]	Mix culture (*Acinetobacter* sp. and *Pseudomanas* sp.)	Batch	500	30/6.2–6.7
6	[1]	*B. brevis*	Batch	1750	34/8.01
7	[26]	*P. putida*	Batch	50	30/7.0
8	This study	*P. aeruginosa*	Batch	2600	37/7.0
		P. pseudomallei	Batch	1500	37/7.0

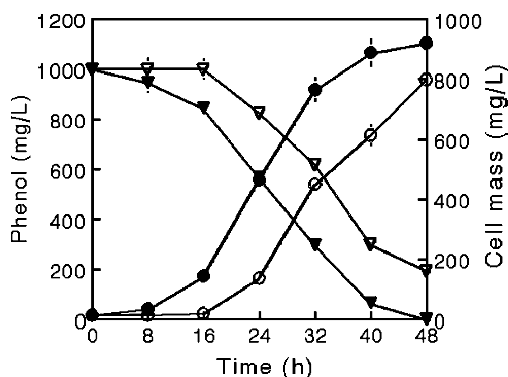

Fig. 2. Cell mass formation by *P. pseudomallei* (○); *P. aeruginosa* (●) and phenol degradation by *P. pseudomallei* (▽) and *P. aeruginosa* (▼) in the presence of 2.5 g L^{-1} NaCl in shake flask cultures. Phenol (1000 mg L^{-1}) and NaCl were added to the MS medium and inoculated with 2% (v/v) un-adapted cultures and incubated at 37 °C and 120 rpm. Error bars indicate standard deviation among three replicates.

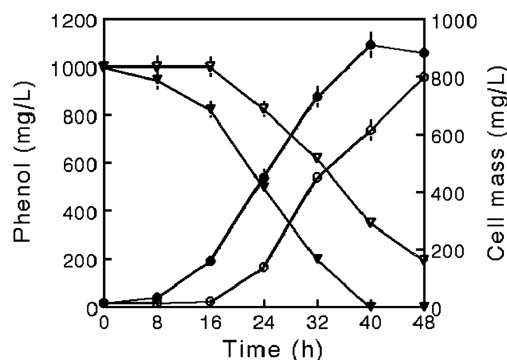

Fig. 3. Cell mass formation by *P. pseudomallei* (○); *P. aeruginosa* (●) and phenol degradation by *P. pseudomallei* (▽) and *P. aeruginosa* (▼) in the presence of 6.125 g L^{-1} Na_2SO_4 in shake flask cultures. Phenol (1000 mg L^{-1}) and Na_2SO_4 were added to the MS medium and inoculated with 2% (v/v) un-adapted cultures and incubated at 37 °C and 120 rpm. Error bars indicate standard deviation among three replicates.

be due the adaptation of microbes at high phenol concentration in the sludge of a pharmaceutical industry where untreated wastewater has been added for many years.

3.4. Effect of salts on phenol biodegradation

As industrial wastewater contains many anions and cations at high concentrations, lab scale studies for biodegradation of phenol were also carried out in the presence of salts containing these ions. Various salts were added to the MS medium to obtain approximately the same concentration of ions (Na^+, K^+, Cl^- and $SO_4{}^{2-}$) as found in the wastewater samples collected from pharmaceutical industry (Table 1). The salts were: NaCl, 2.5; Na_2SO_4, 6.125; KCl, 3.23 and K_2SO_4, 3.37 g L^{-1}. Concentrations of Na^+, K^+, Cl^- and $SO_4{}^{2-}$ obtained by the addition of salts are mentioned in Table 1. Phenol concentration (1000 mg L^{-1}) for this experiment was kept very close to that in the industrial wastewater (1053 ± 162 mg L^{-1}). *P. aeruginosa* completely degraded 1000 mg L^{1-} phenol within 48 h with 8 h lag phase in the presence of 2.5 g L^{-1} NaCl (Fig. 2). These results were comparable to the controls where no salt was added (data not shown). Figs. 2 and 3 showed that while growing in the presence of NaCl and Na_2SO_4. *P. aeruginosa* had higher efficiency for phenol degradation, showed rapid growth and shorter lag period than *P. pseudomallei*. Similar observations were recorded with added KCl and K_2SO_4 (data not shown). In the presence of all salts (mentioned above) *P. aeruginosa* had higher μ (specific growth rate), Q_S and $Y_{X/S,}$ as compared to *P. pseudomallei* (Table 3). The *P. aeruginosa* exhibited up to 1.34- and 1.53-times more μ, and Q_S, respectively than *P. pseudomallei*.

In this study low concentrations of salts were used as compared to those used by other workers to observe their effect on biodegradation of phenol [13,15,16]. Microbial survival and growth in the presence of all selected salts may be due to the fact that in this study low concentration of salts were used. Also, these bacterial strains might have been surviving at these salt concentrations in sludge of pharmaceutical industry wastewater for long time. It has been reported that adaptation enhances/modifies the enzyme activities possessed by the organism [4]. More generally, results indicate that *P. aeruginosa* may prove useful for treatment of phenolic effluents containing high concentrations of salts.

3.5. COD and BOD reduction by P. aeruginosa and P. pseudomallei

While growing in the presence of 1000 mg L^{-1} phenol, *P. aeruginosa* reduced COD and BOD of the culture medium with

Table 3
Phenol degradation rate (Q_S), specific growth rate (μ) and cell yield coefficient ($Y_{X/S,}$) for phenol (1000 mg L^{-1}) utilization by *P. pseudomallei* and *P. aeruginosa* in the presence of various salts

Salt	Microorganism	Q_S[a] (mg L^{-1} h^{-1})	μ[a] (h^{-1})	$Y_{X/S}$[a] (mg cells mg^{-1} phenol)
Sodium chloride (2.54 g L^{-1})	*P. pseudomallei*	29.04	0.041	0.82
	P. aeruginosa	34.83	0.049	0.93
Sodium sulphate (6.125 g L^{-1})	*P. pseudomallei*	26.92	0.040	0.82
	P. aeruginosa	37.22	0.049	0.88
Potassium chloride (3.23 g L^{-1})	*P. pseudomallei*	26.30	0.055	0.69
	P. aeruginosa	34.91	0.064	0.80
Potassium sulphate (3.77 g L^{-1})	*P. pseudomallei*	23.95	0.059	0.68
	P. aeruginosa	36.77	0.071	0.75

[a] Each value is a mean of three replicates.

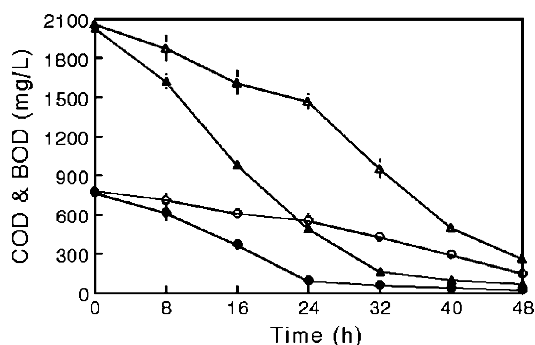

Fig. 4. BOD removal by *P. pseudomallei* (◯) and *P. aeruginosa* (●), and COD reduction by *P. pseudomallei* (△) and *P. aeruginosa* (▲) in shake flask cultures. Phenol (1000 mg L^{-1}) was added to the MS medium and inoculated with 5% (v/v) adapted cultures and incubated at 37 °C and 120 rpm. Error bars indicate standard deviation among three replicates.

removal rate of 71 and 27 mg L^{-1} h^{-1}, respectively (Fig. 4). *P. aeruginosa* took 40 h to reduce COD and BOD values from an initial of 2032 and 773 mg L^{-1} down to 69 and 26 mg L^{-1}, respectively. As presented in Fig. 4, *P. pseudomallei* was less efficient for COD and BOD reduction as COD and BOD decreased from 2032 and 773 mg L^{-1} to 269 and 153 mg L^{-1}, respectively within 48 h of growth. As adapted cultures of *P. aeruginosa* and *P. pseudomallei* were used as inoculum, there was no lag phase for phenol degradation and hence COD/BOD reduction. Although phenol (1000 mg L^{-1}) was completely removed, COD and BOD remained to some extent which might be due the release of toxic/soluble metabolites in the growth media that can not be biodegraded by monocultures of *P. aeruginosa* and *P. pseudomallei* but degradable at lower concentrations (due to dilution in BOD bottles) by the mixed cultures used for BOD determination. These results are better as compared to those previously reported by Babu et al. [8] whereby 2000 mg L^{-1} COD (due to 1000 mg L^{-1} phenol) was reduced to about 100 mg L^{-1} by adapted cells in 65 h.

4. Conclusion

Two strains capable of degrading phenol at high initial concentrations were isolated from the wastewater sludge of a pharmaceutical industry. These strains were identified as *P. aeruginosa* and *P. pseudomallei*. *P. aeruginosa* degraded phenol to a maximum initial concentration of 2600 mg L^{-1} as compared to *P. pseudomallei* which could degrade phenol to a maximum initial concentration of 1500 mg L^{-1}. Time taken to completely degrade phenol by both the strain (at their respective concentrations) was ≅150 h. *P. aeruginosa* proved to be a better strain in terms of degrading higher concentration of phenol and phenol degradation rates. At higher initial phenol concentrations, there was a lag phase whereas at lower initial phenol concentrations, the lag phase was absent or very short. The presence of salts had no or little affect of phenol biodegradation by both the strains. *P. aeruginosa* was more efficient as compared to *P. pseudomallei* for phenol degradation, and COD/BOD reduction as compared to *P. pseudomallei*. The strain *P. aeruginosa* could be used as a potential candidate for bioremediation of phenolic wastewater because of its high phenol degrading efficiency. The results also indicate that *P. aeruginosa* can be very efficiently used for treating phenolic wastewater containing high concentrations of cations/anions such as Na^+, K^+, Cl^- and SO_4^{2-}. These studies could be helpful for the design of batch and/or continuous bioreactors for the treatment of high strength phenolic wastewater.

Acknowledgement

We are thankful to the Army Welfare Pharmaceutical Industry, Kasur, Pakistan for the funding of this project and their assistance for the collection of samples.

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