PERFORMANCE ASPECTS OF *PARACOCCUS PANTOTROPHUS* TREATING URABAN SOLID WASTE LEACHATE

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Abstract

In this study, applicability of a mixotrophic bacterium *Paracoccus pantotrophus* in medium containing urban solid waste leachate was investigated. The patterns of growth were studied in 1 l shake flasks in batch mode. Experiments were carried out in two phases. In the first phase two media concentrations with variation in carbon source were used for different experimental runs, in which combination of glucose and acetate was selected as the optimum medium. In the second phase effect of leachate loading on microbial growth was investigated in six batch reactors, containing optimum mineral medium mixed with leachate. Addition of leachate was done on the basis of Chemical Oxygen Demand (COD) contribution, which ranged from 10% to 50%. After 149 h incubation, 79.6 to 89.4% COD and 20 to 37.5% $NH4^+$– nitrogen was found to be removed. Although, higher concentration resulted in a decrease in microbial growth and COD removal capacity, *P. pantotrophus* exhibited its capabilities of utilizing substrate from leachate. The system's performance indicated that a single stage aerobic system can be developed using *P. pantotrophus* for simultaneous removal of carbon and nitrogen from dilute leachate.

Key words– batch reactor, solid waste leachate, microbial growth, *Paracoccus pantotrophus*, shake flask

Introduction

Urban solid waste leachate has both high concentration of carbon and nitrogen content, besides having other organic and metallic pollutants. One of the major restraints in most of the leachate treatment technologies is that, COD and biochemical oxygen demand (BOD) are treated, and then nitrification is performed in a separate sophisticated system; the additional costs of which can be very high. *Paracoccus pantotrophus* (previously known as *Thiosphaera pantotropha*), a heterotrophic nitrifier and aerobic denitrifier, is reported to simultaneously remove carbon and nitrogen from the wastewater. This paper presents some studies on applicability of *P. pantotrophus* for urban solid waste leachate diluted with synthetic wastewater.

Methods

Two synthetic mineral medium concentrations were used for acclimatization studies. The first medium containing 1000 mg/l COD and 110 mg/l $NH4^+$-N, had two variations in carbon source. The first (medium 1a) had a combination of sodium acetate and glucose while the second (medium 1b) had only sodium acetate as the carbon source. The second concentration containing 500 mg/l COD and 55 mg/l $NH4^+$-N (medium 2), had only sodium acetate as the carbon source.

For first phase of batch experiments, the synthetic mineral medium (500 ml) were placed in 1 l shaking flasks. 5% (v/v) pre-culture of *P. pantotrophus* was inoculated in the shake flasks and incubated at 37°C, at 150 rpm for 267 h. Samples (2 ml) were taken periodically for optical density (OD_{430}nm), pH measurement and chemical analysis. To evaluate the bacterial acclimatization in leachate, medium 1a was used as the basal medium for all the second phase batch experiments. Different concentrations of leachate ranging from 10% to 50% (on COD basis) were added in medium 1a, thereby keeping the initial COD in all the reactors constant. Leachate mixed basal mineral mediums (500 ml) were placed in 1 l shaking flasks, and the pre-cultures of *P. pantotrophus* were inoculated at 5% (v/v) each and incubated at 37°C, at 150 rpm for 300 h.

Results and discussion

To evaluate optimum composition of synthetic mineral medium, three batch culture reactors were operated. The media used in this study were mineral medium 1(a), 1(b) and medium 2. Under similar growth conditions *P. pantotrophus* grew better in medium 1(a) and 1(b) than in medium 2 (Figure 1). It reached a maximum growth (OD_{430} 1.14) in medium 1 (a) after 58 h, and the time taken to reach the peak was lesser for 1(a). Also, better sustenance of microbial growth in medium 1(a) as indicated by higher OD_{430} signifies better growth of *P. pantotrophus* in this medium. Similarly, an increase in the maximum specific growth rate was observed when a culture was exposed to mixtures of carbon sources in comparison to growth with either of these substrates as single carbon sources (Harte and Webb, 1967; Wood and Kelly, 1977; Brinkmann and Babel, 1992). Thus medium 1(a) being the optimum medium was selected as basal medium for further batch studies.

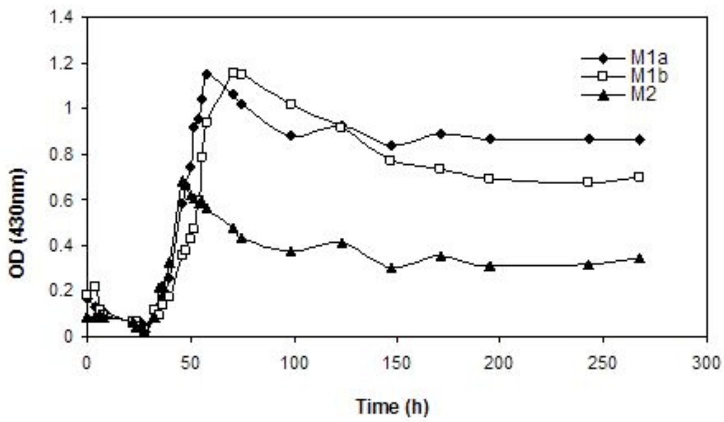

Figure 1 Growth curves for P. pantotrophus grown in synthetic mineral medium

To study the effect of leachate concentration on the growth of *P. pantotrophus*, six shake flask batch reactors were studied. Table 1 presents the effect of leachate loading on the removal of nitrogen and COD. In all the reactors initial COD was maintained at 1000 mg/l, but initial nitrogen was not controlled and it kept increasing with increase in leachate load. It was observed that this increase in ammonium nitrogen load resulted in a corresponding increase in ammonium nitrogen removal. Reactor A ($NH4^{+}$-N load, 112 mg/l) which contained only basal medium, showed a lowest 20% removal, whereas, reactor F (containing 50 % leachate) with highest ammonium load (179.2 mg/l) showed a maximum removal of 37.5%. A straight line correlation was observed for $NH4^{+}$-N load vs. removal. A similar result was obtained by Gupta and Gupta (2001), while treating high strength domestic wastewater by *P. pantotrophus* (then *Thiosphaera pantotropha*), in a rotating biological contactor.

In general, ammonia nitrogen can be removed only by assimilation into biomass or by nitrification. Here, a substantial removal in alkalinity (Table 1) indicates its consumption during heterotrophic nitrification, resulting in ammonium removal. At the same time removal of TKN (Table 1) suggests a simultaneous aerobic denitrification taking place, as it indicates reduction in total nitrogen. However, a decrease in TKN removal with increase in leachate concentration also suggests that microbes are not able to utilize organics from increasing concentration of leachate. A decrease in both, COD removal and average peak OD_{430} (Table 1) with increase in leachate concentration also supports this possibility.

Table 1 Effect of leachate loading on the removal of nitrogen and COD

Reactor	Leachate (%)	NH4+-N Initial (mg/l)	NH4+-N Removal (mg/l)	Alkalinity (as CaCO3) Initial (mg/l)	Alkalinity (as CaCO3) Final (mg/l)	TKN (mg/l) Initial	TKN (mg/l) Removal	Peak OD430 (average)	CODa Remaining
A	0	112.0	19.4	310	180	115.36	17.36	1.19	106
B	10	123.2	29.4	320	210	131.04	52.64	1.05	115
C	20	134.4	35.84	360	270	148.96	59.36	0.97	121
D	30	151.2	52.24	394	290	159.04	30.24	0.96	153
E	40	173.6	61.6	455	320	192.64	33.04	0.84	166
F	50	179.2	67.2	498	377	213.00	19.8	0.74	204

Conclusions

The batch test results indicate that *P. pantotrophus* could utilize substrate from a leachate mixed synthetic mineral medium. After 149 h incubation, 79.6 to 89.4% COD and 20 to 37.5% NH_4^+– nitrogen was found to be removed. A linear decrease in microbial growth and COD removal capacity with increase in leachate concentration in the culture medium was attributed to the presence of toxic inhibitory substances present in leachate. Although, higher concentration resulted in a decrease in microbial growth and COD removal capacity, the system's performance indicated that a single stage aerobic system can be developed using *P. pantotrophus* for simultaneous removal of carbon and nitrogen from dilute leachate.

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