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Treatment of arsenic contaminated water in a laboratory scale up-flow bio-column reactor

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Abstract

The present paper describes the observations on the treatment of arsenic contaminated synthetic industrial effluent in a bio-column reactor. *Ralstonia eutropha MTCC 2487* has been immobilized on the granular activated carbon (GAC) bed in the column reactor. The synthetic water sample containing As(T) (As(III):As(V) = 1:1), Fe, Mn, Cu and Zn at the initial concentrations of 25, 10, 2, 5, 10 ppm, respectively, was used. Concentrations of all the elements have been found to be reduced below their permissible limits in the treated water. The significant effect of empty bed contact time (EBCT) and bed height on the arsenic removal was observed in the initial stage. However, after some time of operation (approximately 3–4 days) no such effect was observed. Removal of As(III) and As(V) was almost similar after ∼2 days of operation. However, at the initial stage As(V) removal was slightly more than that of As(III). In absence of washing, after ∼4–5 days of operation, the bio-column reactor was observed to act as a GAC column reactor based on physico-chemical adsorption. Like arsenic, the percent removals of Fe, Mn, Cu and Zn also attained minimum after ∼1 day and increased significantly to the optimum value within 3–4 days of operation. Dissolved oxygen (DO) has been found to decrease along with the increasing bed height from the bottom. The pH of the solution in the reactor has increased slightly and oxidation–reduction potential (ORP) has decreased with the time of operation.

Keywords: Arsenic; Bio-reactor; Adsorption; EBCT; *Ralstonia eutropha MTCC 2487*

1. Introduction

Arsenic contaminated ground water enters into the human body and causes various types of cancers. Along with natural sources arsenic can also enter into the water stream through industrial effluents like metal-processing industries, semiconductor industries, etc., and acid mine drainage. Important role of mine drainage on the arsenic poisoning has recently been observed in Koudikasa village of Rajnandgaon district in Chhattisgarh, India [1]. Arsenic concentration in acid mine drainage is normally very high. The normal range of the concentration of As(T) in the acid mine drainage, in Carnoules Creek, France has recently been reported as 0–250 ppm [2]. The As(III) concentration in this acid drainage has also been reported to be around 60–90% of the As(T). Concentration of arsenic, copper, zinc and iron in the water of Borah and Maids creeks and drainage from Conrad mine, Australia, has recently been reported as high as 8.6, 5, 11 and 9.7 ppm, respectively [3]. A recent report uses the synthetic acid mine drainage containing 20 ppm As along with Fe, Cu, Zn, Ni, Mg and Al [4].

Considering the health impact of the arsenic poisoning in water the maximum contaminant level (MCL) of arsenic in drinking water has been reduced to 10 ppb by many countries. Similarly, the permissible limit of arsenic in industrial effluents is also low (0.2 ppm). For Cu, Zn, Fe and Mn these values are 3.0, 5.0, 3.0 and 0.2 ppm, respectively [5].

Amongst various arsenic removal methods, the bio removal process using immobilized whole bacterial cells has attracted more research interest in recent years [6]. Some of the bacteria having arsenic removal capability are *Alcaligenes faecalis*, *Agrobacterium tumafecians*, bacteria *NT26*, *Bacillus indicus*, *Bacillus subtilis*, *Corynebacterium glutamicum*, *Desulfovibrio desulfuricans*, *Galleonella ferrigunea*, *Leptothrix ocracia*, *Pseudomonas putida*, *Pseudomonas arsenitoxidans*, *Ralstonia picketti*, *Thiomonas ynys1*, *Thiobacillus ferroxidans*, etc. [7–17]. Amongst these arsenic bacteria the *D. Desulfuricans*, *G. ferri-*

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gunea, *L. ocracia*, *R. picketti*, *T. ynys1*, etc., have been exploited recently to remove arsenic in bio-column reactor [4,13,14,17]. Different types of bacteria have different types of gene. Although all the arsenic bacteria can survive in arsenic atmosphere, the bacteria type which reduces As(V) to As(III) and accumulate As(III) is specifically termed as arsenic resistant bacteria [12]. Arsenic resistant bacteria normally contain arsR and arsC gene in either plasmid or chromosome or in both and produce arsenic regulatory ArsR protein and arsenate reductase enzyme [10,12]. ArsR has specific active sites for accumulating As(III) on the cell surface [10]. Recently, arsR-arsC gene cluster has been observed in *R. eutropha CH34* [10], which is also known as *R. eutropha MTCC 2487* [18]. This strain can produce ArsR protein and arsenate reductase enzyme [10]. However, the arsenic removal by this strain is not yet demonstrated [10].

In a bio-column reactor bio-layer is developed on the solid support inside the reactor and it works as a bio-filtration unit [4]. This bio-filtration technique can be applied for treating contaminated ground water if the bacteria is indigenous to the ground water and can proved to be a cheaper option for treating arsenic contaminated industrial effluents also. For arsenic resistant bacteria like *R. eutropha*, the arsR protein of bacterial mass can capture As(III) or can convert As(V) to As(III) followed by its adsorption on the protein of the bacterial cell surface [10,19]. *R. eutropha MTCC 2487*, isolated from Zn factory wastewater, can also remove Cd, Co, Hg, Ni and Zn along with arsenic from contaminated water [18]. Hence, the use of these bacteria for the treatment of industrial effluents/acid mine drainage containing these metals appears to be promising.

In batch study the process parameters like temperature, pH, oxidation–reduction potential (ORP), dissolved oxygen (DO), etc., are optimized, kinetic and equilibrium data are generated which can be applied to design and operate column reactor. However, without study in a column reactor the suitability of a technology cannot be established. The removal efficiency of the pollutants in a column study depends upon the empty bed contact time and bed height of the reactor. In a bio-column reactor the pH, ORP and DO of the treated water varies with time and along the height of the reactor, even when these values in inlet water are kept constant. Another important advantage of bio-column reactor is that it does not require regeneration of the adsorbent bed. However, backwashing within a certain interval is essential for the effective operation of the bio-column reactor.

In the present study, the treatment of a synthetic acid mine drainage containing As, Fe, Mn, Cu and Zn in a bio-column reactor for 15 days has been carried out. *R. eutropha MTCC 2487* has been immobilized over GAC (conditioned in metal solution) to produce bio-column reactor. The effects of empty bed contact time (EBCT), bed height and backwashing of the exhausted bed on the arsenic concentration of the treated water have been discussed. Removal of arsenic species at a constant EBCT and bed height has been discussed. The change of pH, ORP and DO with time has also been presented. It also discusses the removal of the Fe, Mn, Cu and Zn at the minimum EBCT value.

2. Materials and methods

The source of microorganism, its acclimatization to the heavy metal environment, experimental setup and experimental procedure are discussed as follows.

2.1. Source of organism

R. eutropha MTCC 2487 species was obtained from Institute of Microbial Technology, Chandigarh, India.

2.2. Acclimatization

The acclimatization of *R. eutropha MTCC 2487* in arsenic and metal environment was performed as follows.

The revived culture was first grown in nutrient broth (NB) media (13 g/l) [18] in a 250 ml conical flask. After 48 h significant bacterial growth was observed in the flask. Appropriate quantity of stock solution of arsenic was added into the flask containing NB to get a concentration of 1 mg/l of arsenic. It was kept aside, initially growth of *R. eutropha MTCC 2487* was inhibited and log phase started after 10 h. Thereafter, the arsenic was periodically added in increments of 1 mg/l in a series of 250 ml flasks till the arsenic concentration in the growth media reached 25 mg/l. The concentrations of Fe, Mn, Cu and Zn in the solution were also increased to 10, 2, 5 and 10 mg/l, respectively, in the similar manner. The NB content was decreased and arsenic and other metal content of the media were increased over a period of around one and half months. For inoculums, a further sub culturing was done and all the inoculums transfers were done in exponential phase (DO value ~0.6 at 600 nm). The temperature was maintained at $29 \pm 1\ ^{\circ}C$.

2.3. Experimental setup

The experiments related to the removal of arsenic and other metals were conducted in a bioreactor column constructed of SS pipe. The schematic diagram of the experimental setup is shown in Fig. 1, which is consisted with bio-column reactor, mixing chamber fitted with stirrer, feed tank, peristaltic pump, compressor, steam generator, rotameters and filter units for water and air. The reactor assembly is a close circuit unit. The column reactor had a working height of 100 cm, an internal diameter of 8 cm and a net empty working volume 5.03 ± 0.002 l. It was equipped with a total of four equidistant ports (excluding inlet and out let) of 1.25 cm diameter for collecting liquid samples along the height of the reactor. The top and bottom portions were connected with the main column by two flange joints, supported on SS screen (mesh no.: 16 BSS, width aperture: 1.00 mm). The final pore volume (void space) of the reactor was between 1796 and 1885 ml. The reactor was filled with granular activated carbon (GAC). The particle size and bulk density of the GAC were 2–4 mm and 40 g/100 ml, respectively. Before use, GAC was purified by soxhlet extraction with acetone/n-heptane (50:50, v/v) for 24 h and then dried [20]. Some physical properties of GAC are shown in Table 1.