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ORIGINAL ARTICLE

Biosorption of arsenic through bacteria isolated from Pakistan

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Abstract

The aim of this study was to isolate arsenic-resistant bacteria and to further exploit it for remediation purposes. In the present study, we have isolated arsenic-resistant strain from ground water of Pakistan AT-01. The strain was cultivated at 37 °C in Luria Bertani broth supplemented with different concentrations of arsenate and arsenite. The minimum inhibitory concentration of arsenic against the bacterial isolate was 7 g/L (7000 mg/L) for arsenate and 1.4 g/L (1400 mg/L) for arsenite salt. The bacterial isolate was also characterized both on molecular and biochemical basis. The isolated strain belonged to the *Pseudomonas aeruginosa*. The high resistance against arsenic offered by the bacteria was exploited further for bioremediation purposes. The bacterial biomass generated from AT-01 strain was able to efficiently remove arsenic with 98% efficiency. Arsenic contamination of ground water is a widespread worldwide problem. The present study shows the potential of high arsenic-resistant bacteria for efficient arsenic removal.

Keywords Arsenic resistance · Minimum inhibitory concentration · Biomass · Bioremediation · Arsenic-resistant bacteria (ARB)

Introduction

Arsenic contamination in groundwater has affected over 300 million people globally (Bibi et al. 2016). The simultaneous incessant poisonings with arsenic are a concern in various Southeast Asian countries (Ayoob and Gupta 2006). Arsenic readily changes its oxidation states or chemical form through environment-associated chemical and biological reactions (Huisman et al. 2011). It exists in different oxidation states of −3, 0, 3, and 5. Exposure to arsenic above the threshold level, i.e., 0.05 ppm (μg/ml), is reported as potential threat to various organs as liver, kidney, pancreas, cardiovascular, and reproductive system (Straub et al. 2007). Drinking water and food are main sources of arsenic exposure to animals and humans.

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Different methods and techniques are employed for removal of arsenic contamination from drinking water. These methods are coagulation, membrane technology, ion exchange method, nanoparticles, phytoremediation, and biological removal (Ng et al. 2004). Microorganisms use the toxic contaminants as energy source for their metabolism process and convert them into less toxic compounds. Biological removal of metal and non-metal contamination from drinking water is preferred over other chemical process because it is cheap, easily available, cost-effective, and has almost no side effects and by-products (Qin et al. 2006; Andreoni et al. 2012). Microbes remediate arsenic through various different mechanisms like oxidation, reduction, methylation, and intracellular bioaccumulation (Satyapal et al. 2016). District Vehari is at a serious health risk as 95% of the ground water samples are documented to be unfit for drinking purpose. Moreover, the level of arsenic is exceeding the World Health Organization (WHO) permissible limit (arsenic 10 μg/L) (Shahid et al. 2017). In the aquifers of Vehari, the concentration of arsenic has been reported to be approximately 12–156 μg/L (ppb (μg/L)) (Farooqi et al. n.d.).

Different arsenic-resistant bacteria were isolated which can withstand high concentration of arsenic and can be used for bioremediation but limited work has been done for the removal of arsenic using arsenic-resistant bacteria. In this study, arsenic-resistant bacteria were isolated from Vehari district

of Pakistan. The bacteria were characterized and the extent of resistance was determined. Furthermore, bacterial biomass from this bacterium was used for bioremediation of arsenic from water.

Materials & methods

Study area & sampling

The present study was conducted in Vehari, Punjab, Pakistan. Vehari (which was already reported for arsenic contamination (Shakoor et al. 2015)) is situated within 30.0452° N and 72.3489° E at an altitude of 135 m with a total area of 12 km^2. Ground water sample was collected from hand pump after 5 min flushing of wells in order to ensure fresh sample collection. The sample was taken in a pre-sterilized plastic tube and kept at 4 °C until further use.

Physio-chemical analysis of water sample

After collection of water sample, physio-chemical analysis was performed to measure and check the pH, electrical conductivity, total dissolved solid (TDS), acidity, alkalinity, hardness, and salts contents in the water. pH and electrical conductivity were measured using a pH meter (Hanna Instrument model 8519, Italy) and digital conductivity meter (E.C. meter Hach-44600-00, USA) respectively. Turbidity (NTU) was determined by a turbidity meter (Lamotte, Model 2008, USA). Total dissolved solids (TDS) were measured in mg/L by using a TDS meter (Model 2540C, standard method, 1992). Total alkalinity was determined using acid titration, with methyl-orange as an indicator (2320, standard method, 1992). Calcium hardness, Mg hardness, and total hardness, as CaCO3, were determined as reported previously (Frank 2017). Chloride was estimated by potentiometric titration with a standard 0.0141N silver nitrate solution (standard method, 1992). Sulfate ions were precipitated as barium sulfate in an acidic medium with barium chloride. The absorption of light by this precipitated suspension was measured by spectrophotometer (SulfaVer4 (Hach-8051) at 420 nm (APHA, 22nd Edition). For cations and heavy metals, the analysis was done by using flame atomic absorption spectrophotometer (AAS Vario 6, Analytik Jena AG) method (Kopp and McKee 1979). Samples were left unfiltered in order to provide accurate values.

Isolation of arsenic-resistant bacteria

Ground water was serially diluted up to 10^{-5} and was inoculated in 50 mL LB broth containing sodium arsenate (100 ppm (100 μg/ml)) and sodium arsenite (20 ppm (20 μg/ml)). The inoculated medium was incubated at 37 °C for 24 h and bacterial growth was measured at regular intervals using spectrophotometer. After 24 h of incubation, 10 μL of bacterial culture was spread on LB agar plates and incubated at 37 °C for overnight. Bacterial colonies were observed the following day on the agar plates.

Growth curves of bacterial strain

The isolated bacterial strain along with already isolated AT-01 and *E. coli* Dh5-Alpha (as reference/control) were cultured in different concentrations of arsenate and arsenite to determine the resistivity of the strains. The bacterial strains were inoculated in 10 mL LB broth containing sodium arsenate (100 ppm (μg/ml), 300 ppm (μg/ml), 500 ppm (μg/ml), 700 ppm (μg/ml), 1000 ppm (μg/ml), 3000 ppm (μg/ml), 5000 ppm (μg/ml) and 7000 ppm (μg/ml)) and sodium arsenite (20 ppm (μg/ml), 60 ppm (μg/ml), 100 ppm (μg/ml), 140 ppm (μg/ml), 200 ppm (μg/ml), 600 ppm (μg/ml), and 1000 ppm (μg/ml) and 1400 ppm (μg/ml)) separately and incubated in shaking incubator at 37 °C for 24 h. Optical density (at 600 nm) was measured for the strains at regular interval of time using spectrophotometer. The same process was repeated on LB agar plates containing arsenate and arsenite for the bacterial strains along with *E. coli* (DH5-Alpha) as a control. The inoculum was serially diluted and poured onto the plates and incubated at 37 °C for 24 h. Minimum inhibitory concentration (MIC) is the lowest concentration at which the heavy metal like arsenic in this case can inhibit the growth of the test strain was determined from the growth curves. Moreover, the maximum tolerable concentration (MTC) of the heavy metal that has no effect on the growth of our isolated strain was also determined.

LB agar plates

LB agar plates were prepared with different concentrations of arsenate and arsenite salts. The three strains were taken from their exponential phase when their OD was around 0.6 and serially diluted. These dilutions were plated on LB agar with arsenic salts and incubated at 37 °C for 24 h. The growth on the plates was compared.

Molecular and biochemical characterization of the bacterial strain/identification

For identification and characterization of the isolated bacterial strain, different tests were performed including gram staining, triple sugar iron (TSI) tests, fermentation of carbohydrates, catalase/oxidase, and other biochemical tests. The strain was further characterized by PCR using genus and specie specific primers targeting the *oprI* and *oprL* genes as documented previously (Douraghi et al. 2014).

Table 1 Analysis report of Vehari ground water

Serial no.	Water quality parameters	Result
Physical parameter		
1.	Color	Colorless
2.	pH	7.68
3.	Electrical conductivity (μS/cm)	1044
4.	Turbidity (NTU)	11.20
Chemical parameters		
5.	Alkalinity (as $CaCO_3$) (ppm (μg/ml))	352
6.	Bicarbonate (ppm (μg/ml))	352
7.	Calcium (ppm (μg/ml))	13
8.	Carbonate (ppm (μg/ml))	BDL (below detection limit)
9.	Chloride (ppm (μg/ml))	36
10.	Hardness (calcium and magnesium carbonates) (ppm (μg/ml))	130
11.	Magnesium (ppm (μg/ml))	24
12.	Potassium (ppm (μg/ml))	7.6
13.	Sodium (ppm (μg/ml))	160
14.	Sulfate (ppm (μg/ml))	80
15.	TDS (total dissolved solids) (ppm (μg/ml))	574
Trace and ultra-trace elements		
16.	Arsenic (ppb (μg/L))	123.4

Sequence and phylogenetic analysis

The 16S rRNA sequence of the isolated AT-01 strain was amplified by the primers listed in supplementary Table 1. For the calculation of phylogenetic tree, MSA was carried out using MUSCLE (http://www.ebi.ac.uk/Tools/msa/muscle/). MSA is performed to analyze and find evolutionary relationships through homology between the sequences. Point mutations and insertion or deletion (indel) events were detected It was subjected for the calculation of the phylogenetic tree through MEGA 7.06. The evolutionary history was inferred using the neighbor-joining method (Saitou and Nei 1987). The optimal tree with the sum of branch length = 0.179 is shown. The percentage of replicate trees in which the associated taxa clustered together in the bootstrap test (500 replicates) is shown next to the branches (Kumar et al. 2016). The tree is drawn to scale, with branch lengths in the same units as those of the evolutionary distances used to infer the phylogenetic tree. The evolutionary distances were computed using the p-distance method (Felsenstein 1985; Nei and Kumar 2000) and are in the units of the number of base difference per site. The analysis involved 15 nucleotide sequences. All positions containing gaps and missing data were eliminated.

Bacterial biomass

The arsenic-resistant bacterium (ARB) biomass was obtained by inoculating 50 mL of inoculum in 1 L of LB Broth media. This was incubated at 37 °C for 18 to 24 h. Then, the broth culture was centrifuged at 4000 rpm for 10 min. Then, it was washed several times with deionized water. Then the pellet was incubated in oven at 70° C and the ARB biomass was collected in powder form. This process was repeated 15 times to obtain sufficient amount of bacterial biomass.

Remediation experiment

The remediation of arsenic was performed with the ARB biomass through biosorption. It is a physicochemical process in which the heavy metals or contaminants binds to the cellular structure of a biomass. Of arsenic solution, 10,000 ppb (μg/L) was used as initial concentration of arsenic. The ARB biomass was used in concentration of 1 g/L.

The ARB biomass was mixed with 10,000 ppb (μg/L) of arsenic solution at pH 7 and was incubated at room temperature within shaking incubator. The incubation time was ranging from 0.5 to 2 h. After incubation, the solution was filtered for the removal of ARB biomass. The final concentration of arsenic in the filtrate after incubation was determined via spectrophotometric method. For the spectrophotometric method following steps were carried out. 0.5 mL of concentrated H_2SO_4 was added and mixed to 1 mL of the filtrate (treated solution) followed by addition of 2 mL of 2% potassium iodate with shaking. Afterwards, 0.5 mL of hydrogen peroxide was added and mixed followed by the addition of 2 mL chloroform. The solution was left for 15 min and as a result pink color was produced. The absorbance of the pink portion of the solution was checked at 515 nm and was compared with the standard curve. (Sandhu 1976). The standard curve was made with known concentrations of arsenic ranging from 100 to 10,000 ppb (μg/L) using the abovementioned spectrophotometric method. The obtained values were also confirmed by atomic absorption spectrophotometer. The remediation efficiency was determined by using the following formula:

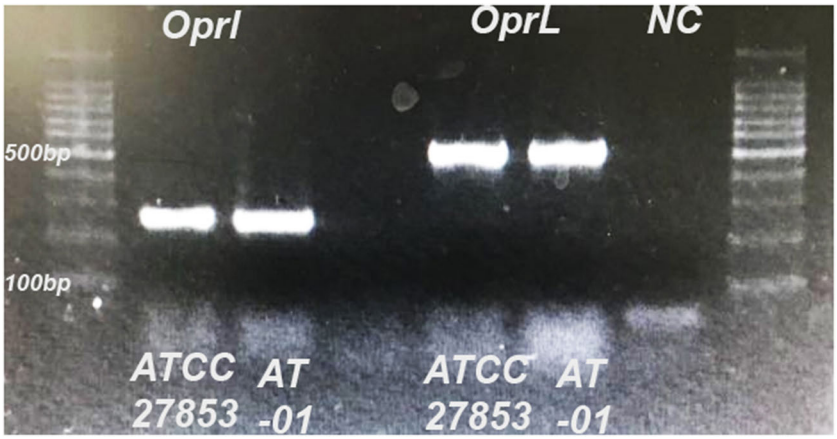

Fig. 1 PCR amplification of *oprI* and *oprL* genes yielding amplification product of 249 bp and 504 bp

$$E = \left(\frac{C_i - C_f}{C_i}\right) * 100$$

where C_i is the initial concentration of the metallic ion (mg L^{-1}), C_f is the final concentration of the metallic ion (mg L^{-1}), m is the mass of the biosorbent in the reaction mixture (g), and V is the volume of the reaction mixture (L) (Barros Júnior et al. 2003).

Results

Physio-chemical analysis of water sample

The ground water sample was collected from Vehari, Pakistan. The temperature of the samples ranged from 28 to 31 °C, pH was 7.68.The physio-chemical properties of the water sample are presented in Table 1. These characteristics show slightly alkaline conditions that might be causing arsenic release from soil to the ground water.

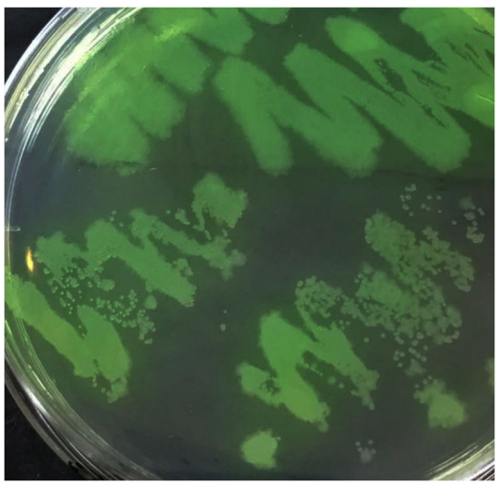

Fig. 2 Growth of AT-01 on *Pseudomonas aeruginosa* selective cetrimide agar

Molecular and biochemical characterization of the bacterial strain

The isolated strain named AT-01 was biochemically characterized by performing different tests. The bacterial strain is Gram negative, rod-shaped aerobic bacterium. The bacterial isolate had ability to degrade hydrogen peroxide with the help of catalase enzyme. The bacterium was not able to convert urea into ammonia by urease enzyme. The detailed result of the biochemical characterization is given in the Supplementary Table 1 that confirmed the isolated strain was *Pseudomonas aeruginosa*. Furthermore, genus and species specific primers targeting the *oprI* and *oprL* genes were used. Primers targeting the *oprL* gene can effectively identify the specie of *Pseudomonas* as documented previously (Douraghi et al. 2014). *oprI* and *oprL* are peptidoglycan outer membrane lipo-proteins. *oprI* gene is conserved in fluorescent pseudomonads and, hence, can identify strains belonging to *Pseudomonas* genus. The *oprL* gene is a *Pseudomonas aeruginosa* specie specific gene (Douraghi et al. 2014). Amplification of the *oprI* and *oprL* genes yielded 249 and 504 bp products confirming that the isolated strain is *Pseudomonas aeruginosa*. The amplification was also compared to *Pseudomonas aeruginosa* ATCC27853 *strain* that was used as a positive control (Fig. 1). Furthermore, the isolated strain was also grown on the specific media for *Pseudomonas aeruginosa* selective cetrimide agar. Growth on this agar further validates that our isolated strain is *Pseudomonas aeruginosa* (Fig. 2). Moreover, we amplified the 16S rRNA of the AT-01 strain by using primers listed in Supplementary Table 1 followed by sequencing. The 16S rRNA sequence of the isolated AT-01 strain was aligned by using MUSCLE and phylogenetic tree was constructed by employing neighbor joining method. Hence, the isolated strain AT-01 showed strong resemblance with the *Pseudomonas aeruginosa* strains (Fig. 3).

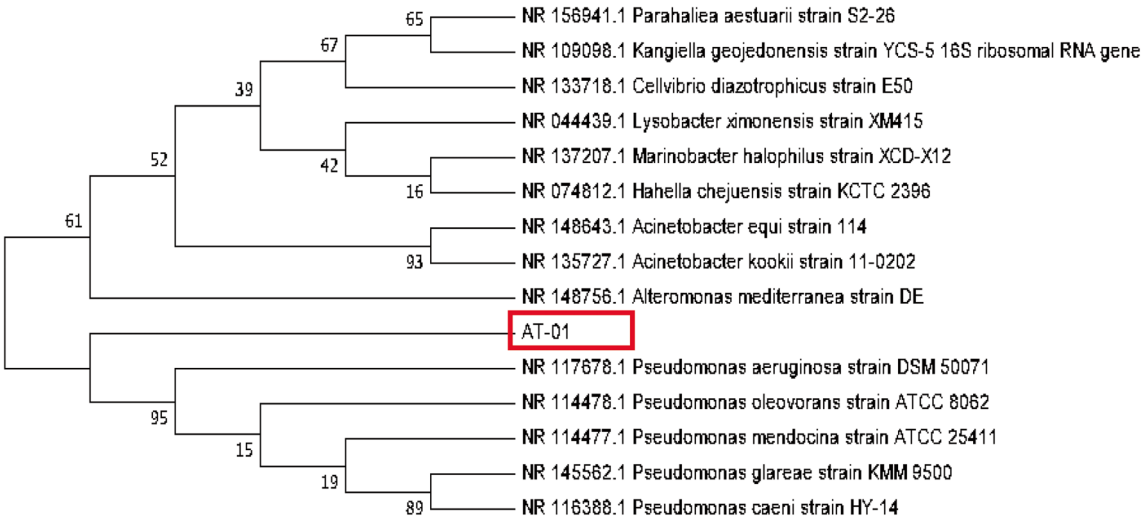

Fig. 3 Phylogenetic tree of the isolated AT-01 strain demonstrating its resemblance to *Pseudomonas aeruginosa* specie

Minimum inhibitory concentration and maximum tolerable concentration of bacterial strains

Minimum inhibitory concentration (MIC) is the lowest concentration at which arsenic can inhibit the growth of the AT-01 strain. We have also determined the maximum tolerable concentration (MTC) of the arsenic pausing, no threat to the growth of our isolated strain. The bacterial resistance to arsenic was determined by incubating the strain AT-01 with LB broth medium containing sodium arsenate (100–7000 ppm (μg/ml)) and sodium arsenite (20–1400 ppm (μg/ml)). From the recorded optical density, growth curves were plotted and presented in Fig. 4.

The lag phase of arsenic-resistant bacteria AT-01 extended on average from 0 to 4 h in normal and stressed condition, i.e., in the presence of arsenate and arsenite. The exponential (log) phase determined the growth of bacteria and can be seen from the figure that without arsenic salts the growth (OD) is higher

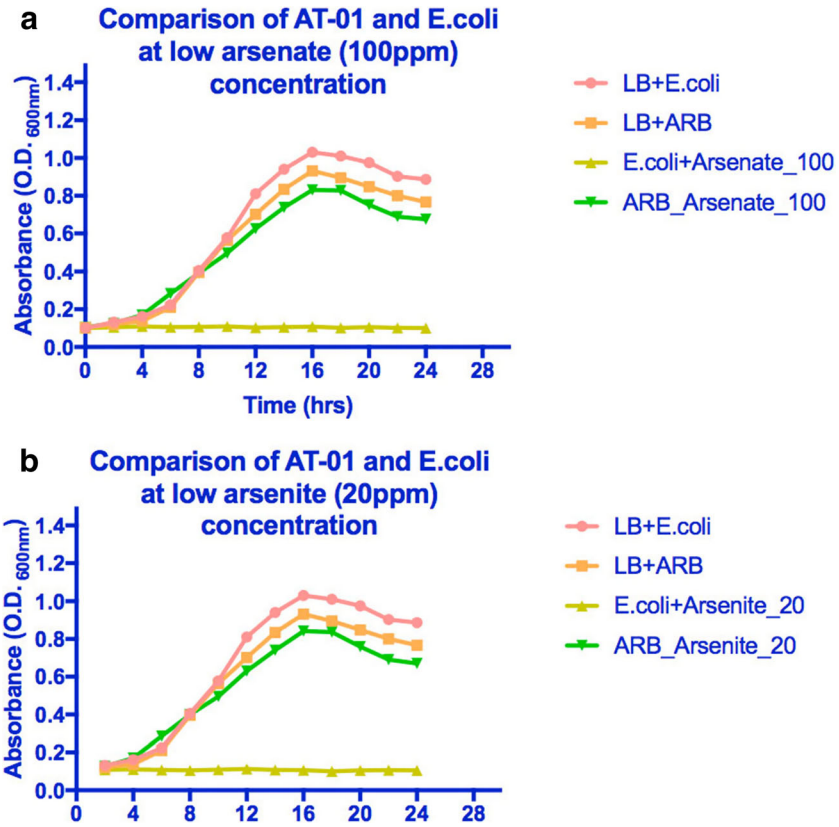

Fig. 4 Growth curve of AT-01 strain at low concentration of arsenate (**a**) and arsenite (**b**) 100–700 ppm (μg/ml) and 20–140 ppm (μg/ml) respectively. (**c**) and (**d**) represent growth curve of AT-01 at high concentration of arsenate and arsenite 1000–7000 ppm (μg/ml) and 200–1400 ppm (μg/ml) respectively. The minimal inhibitory concentration of AT-01 was found to be 7000 ppm (μg/ml) for arsenate and 1400 ppm (μg/ml) for arsenite. The strain was grown in plain LB as a control

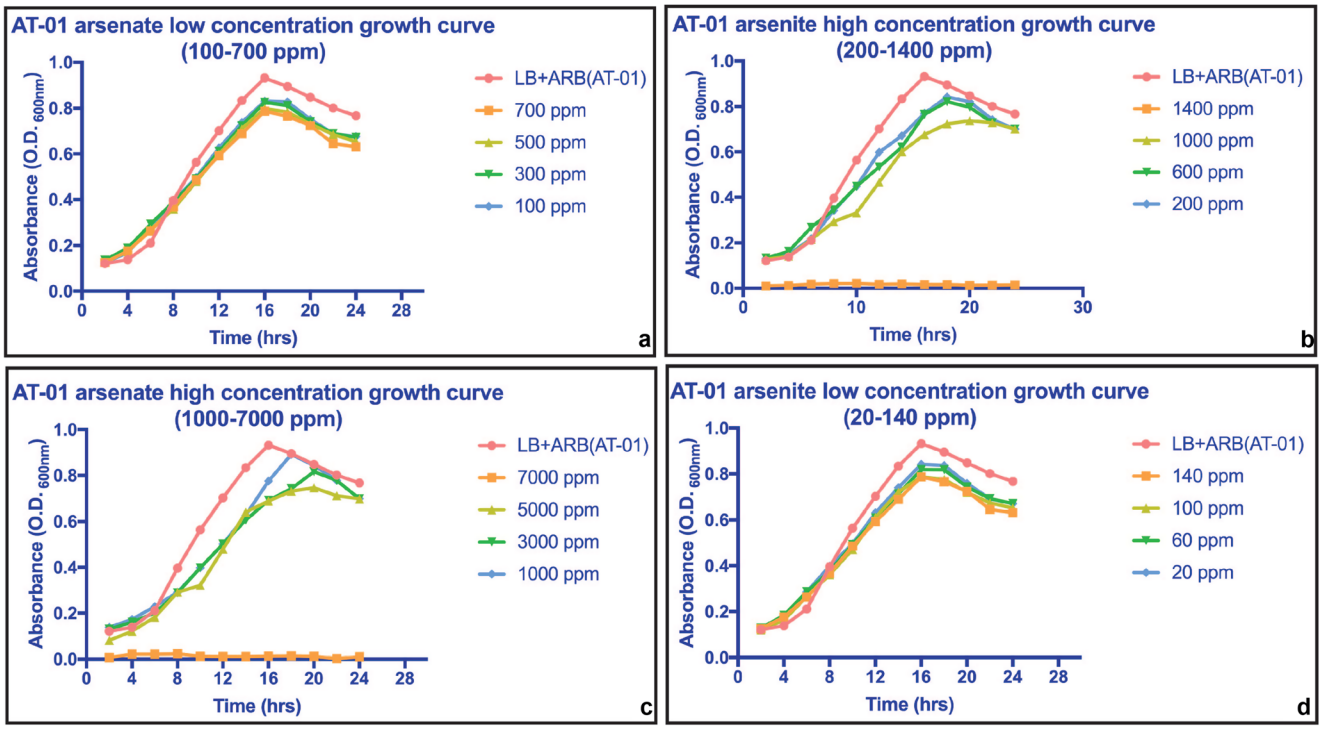

Fig. 5 Comparison of growth curves of AT-01 and *E. coli* strains at low concentration of arsenate (**a**) and arsenite (**b**) 100–700 ppm (μg/ml) and 20–140 ppm (μg/ml) and high concentration of arsenate and arsenite 1000–7000 ppm (μg/ml) and 200–1400 ppm (μg/ml) respectively. *E. coli* was used as a negative control as the *E. coli* strain DH5-α was not able to survive even at the low concentration of arsenate 100 ppm (μg/ml) and arsenite 20 ppm (μg/ml) respectively

when compared to stressed conditions. The exponential phase is followed by 2 h stationary phase which lead to decline phase after 18–20 h. Growth pattern observed for the bacterial strain in LB broth and with arsenic (arsenate & arsenite) salts is similar. The only difference is between the OD's which are higher when grown in plain LB. The minimum inhibitory concentration of the isolated strain was 7000 ppm (μg/ml) arsenate and 1400 ppm (μg/ml) arsenite at which no growth was observed. The maximum tolerable concentration of the isolated strain was 5000 ppm (μg/ml) arsenate and 1000 ppm (μg/ml) arsenite Fig. 4.

We also used a *Pseudomonas aeruginosa* ATCC27853 strain along with *E. coli* as a negative control and compared its growth with the isolated strain AT-01. Interestingly, the reference strain and *E. coli* was not able to survive even the lowest concentration of arsenate (100 ppm (μg/ml)) and arsenite (20 ppm (μg/ml)). Since both the strains were sensitive to arsenic, therefore, we were unable to determine maximum tolerable concentration and minimum inhibitory concentration, Figs. 5 and 6.

Bacterial growth on LB plates

The effect of arsenic on bacterial growth was further confirmed by growing the three strains AT-01, and *E. coli* and the ATCC27853 strain were grown on LB agar plates (Figs. 7 and 8). In the plain LB agar plates, both the strains grow well at all the dilutions. *E. coli* and *Pseudomonas aeruginosa* reference strain *ATCC 27853* were not able to grow at any concentration of arsenate and arsenite salts in LB agar. AT-01 was unable to grow at 7000 ppm (μg/ml) arsenate and 1400 ppm (μg/ml) arsenite salts which define their minimum inhibitory concentration validating the results obtained in the LB broth.

Remediation of arsenic by AT-01 (ARB) biomass

The remediation of arsenic by ARB biomass via biosorption and efficiencies of arsenic remediation at different time intervals are shown in Fig. 9. As mentioned in the methodology, the initial concentration of arsenic in case of remediation experiment was 10,000 ppb (μg/L). There was significant reduction in the level of arsenic by treating with ARB biomass as shown in Fig. 9. The amount of arsenic was reduced from 10,000 to 928 ppb (μg/L) upon 30 min of incubation. This was further reduced by increasing the incubation time and with 2 h of incubation, the arsenic amount was reduced to 208 ppb (μg/L).

The percentage of arsenic left after treatment with the bacterial biomass was determined by dividing the amount of arsenic left to the initial starting concentration of arsenic, i.e., 10,000 ppb (μg/L). The obtained value was multiplied by 100 for percentage calculation. The arsenic remediation efficiency

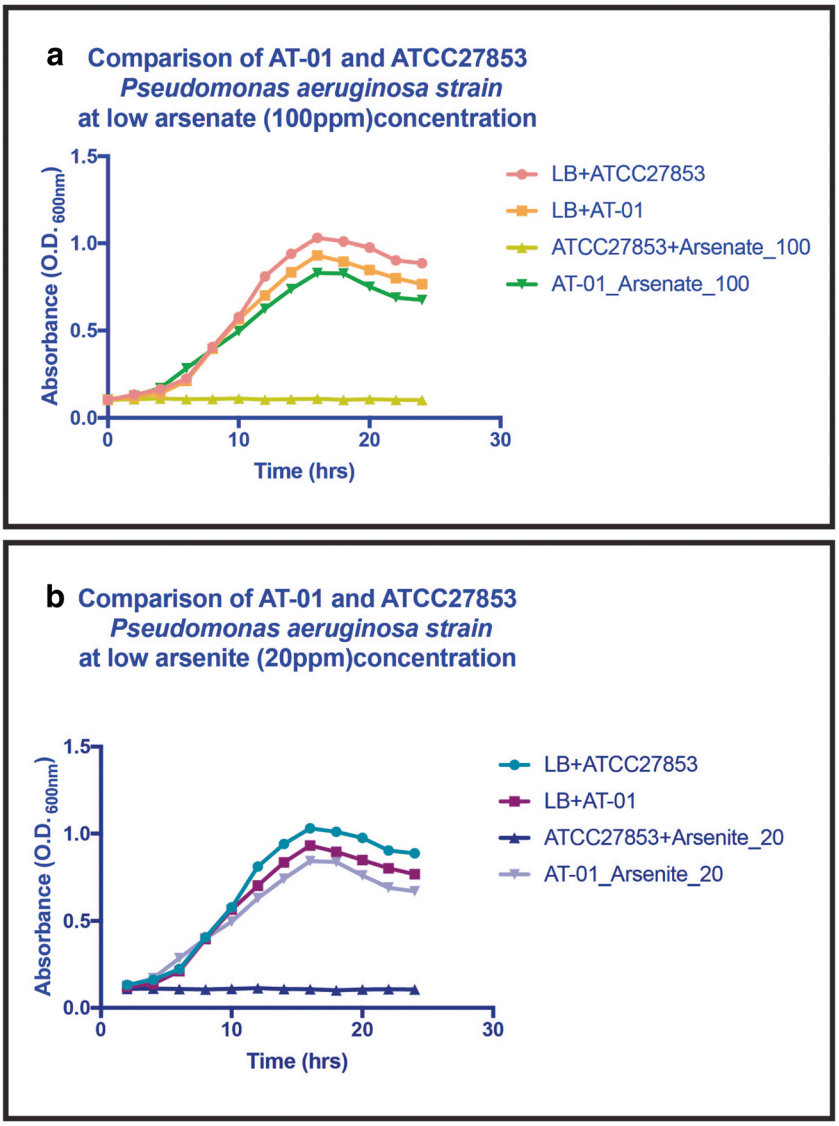

Fig. 6 Comparison of growth curves of AT-01and reference ATCC27853 strain at low concentration of arsenate (**a**) and arsenite (**b**) 100–700 ppm (μg/ml) and 20–140 ppm (μg/ml) and high concentration of arsenate and arsenite 1000–7000 ppm (μg/ml) and 200–1400 ppm (μg/ml) respectively. *E. coli* was used as a negative control as the ATCC27853 *Pseudomonas aeruginosa* strain was not able to survive even at the low concentration of arsenate 100 ppm (μg/ml) and arsenite 20 ppm (μg/ml) respectively

was determined by subtracting the percentage of arsenic left from 100. The arsenic remediation efficiency by ARB biomass is shown in Fig. 9. There was about 90.72% of arsenic remediation by ARB biomass within 30 min. This remediation efficiency was increased with increasing the incubation time. Maximum remediation occurs at 2 h of incubation. The remediation efficiency at 2 h of incubation was 97.92%.

Discussion

Exposure to heavy metal contamination above threshold level has posed significant threat to public health and has hazardous effect on microbes. In order to combat such harsh environment, microbes have evolved various defense mechanisms resulting in heavy metal resistant microbes (Prabhakaran et al. 2016). Microbial transformation of arsenic (AsIII and AsV) has a potential for arsenic removal from contaminated water (Kruger et al. 2013). Bacteria survive in aquatic system by employing different arsenic detoxification mechanisms. Different studies have reported isolation of arsenic-resistant bacteria from contaminated water bodies (Goswami et al. 2015; Pepi et al. 2007).

In Pakistan, arsenic is reported in ground water mostly in Punjab and Sindh areas along the Indus river basin. The levels of arsenic in ground water vary in different areas and range from 1.5 to 201 μg L^{-1} (Shakoor et al. 2015; Farooqi et al. 2009). In a recent study, arsenic level in the ground water was determined from five different areas of Punjab (Chichawatni, Multan, Bahawalpur, Vehari, and RahimYarKhan), Pakistan. The mean arsenic concentration in these areas was found to be 120, 72, 53, 22, and 9 μg/L respectively. The highest arsenic concentration was found in Chichawatni, approximately 206 μg/L (Shakoor et al. 2018). Different researchers have postulated various mechanisms regarding release of arsenic in the ground water. Oxidation of arsenic-containing minerals

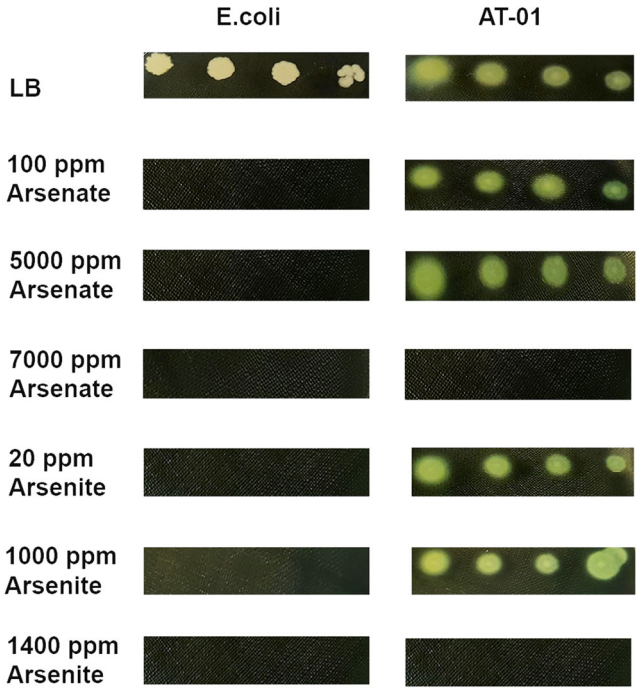

Fig. 7 Growth comparison of AT-01 and *E. coli* on LB agar plate

or its desorption due to saline conditions and alkaline pH are few possible mechanisms of its release into the aquifers. These probable mechanisms are associated with arid and semi-arid areas like Pakistan (Shakoor et al. 2018; Scanlon et al. 2009; Currell et al. 2011; Shaheen et al. 2016). In this study, we have taken water sample from district Vehari. The physio-chemical characteristics of the sample showed a similar pattern of

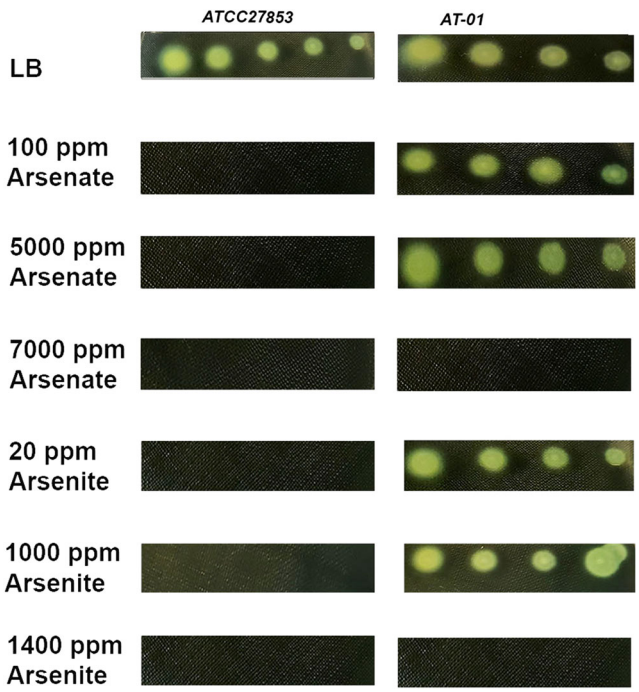

Fig. 8 Growth comparison of AT-01 and the reference ATCC27853 strain on LB agar plate

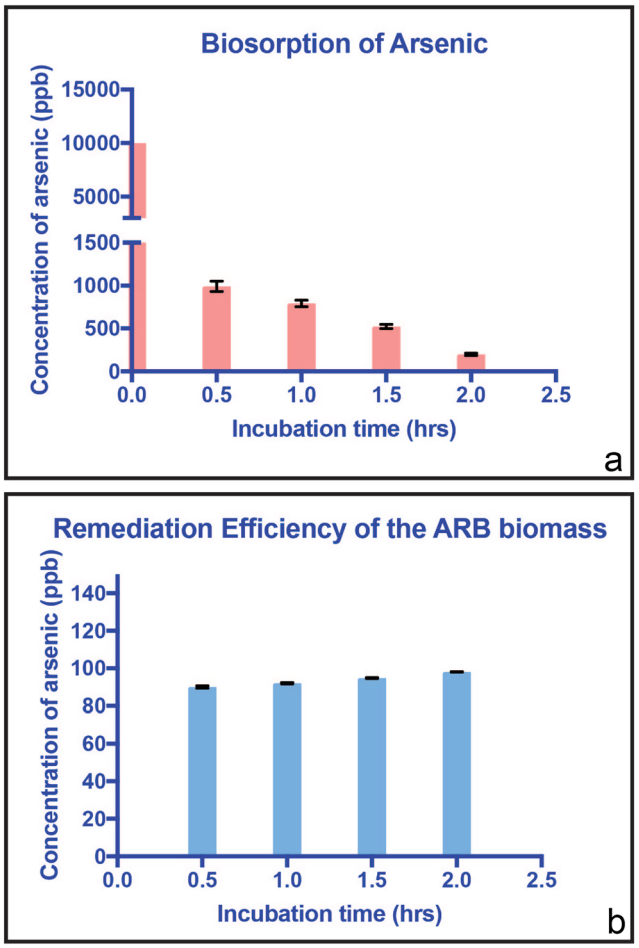

Fig. 9 Bioremediation of arsenic **a**. Biosorption of arsenic at different time intervals **b**. Bioremediation efficiency of the ARB biomass

arsenic, bicarbonate, and alkalinity. We propose that under alkaline conditions arsenic might be undergoing oxidative dissolution. Moreover, due to warm weather conditions in the district Vehari so evaporation of water might be another contributing factor in raising the concentration.

In the present study, we have isolated a highly resistant bacterial strain against arsenic from the contaminated groundwater of Vehari district. The ground water of Vehari district is highly contaminated with arsenic (Farooqi et al. n.d.). The isolated bacterial strain AT-01 belongs to genus *Pseuodomonas*. For specie specific identification, we have amplified the DNA using specie specific primer targeting the *oprL* gene. *oprI* and *oprL* are peptidoglycan-associated outer lipoproteins. *Opr I* gene is specific for genus *Pseudomonas* yielding 249 bp product size whereas *opr L* is specific for aeruginosa specie with an amplification product of 504 bp. These genes have been documented previously to identify different *Pseudomonas* isolates from cystic fibrosis patients (Douraghi et al. 2014).

The biomass of arsenic-resistant bacteria has been reported for arsenic adsorption. Biosorption through different bacterial

biomasses has been reported previously like *Bacillus cereus* has the adsorption capacity of 32.42 mg/g for arsenite at pH 7.5 with biomass dose of 6 g/L (Giri et al. 2013). Different species belonging to genus *Pseudomonas* have been documented previously to be associated with arsenic resistance (Joshi et al. 2008; Fernández et al. 2016). The reduction of arsenate (As V) to arsenite (As III) is another metabolic pathway mediated mechanism of arsenic resistance in *Pseudomonas* species (Pepi et al. 2011). In this study, we found that our isolated strain AT-01 belongs to *Pseudomonas aeruginosa* type can resist 5000 ppm (μg/ml) of arsenate and 1000 ppm (μg/ml) arsenite. Previous reports have demonstrated the effective use of *Pseudomonas aeruginosa s*trains for adsorption of heavy metal ions such as Cd (II) and Pb (II) (Anikó et al. 2014). The positively charged heavy metals tend to bind to negatively charged cell wall components or other extracellular proteins. Therefore, we incubated the dried biomass of the isolated AT-01 strain with a known concentration of arsenic for this purpose. The biomass showed 90.72% remediation of arsenic within 30 min. The remediation efficiency of arsenic was enhanced with increasing incubation time. The remediation efficiency was enhanced to 98% upon 2 h of incubation. We propose that the biomass of this bacterium can be used as a remediation tool for the removal of arsenic from water without the need of harboring any metabolic pathway. The biomass of this bacterium comprising mainly the cell wall components was used for the remediation of arsenic from water.

The present study provides the first evidence of arsenic resistant bacteria isolated from Vehari with significant potential of bioremediation. Therefore, we can conclude that the arsenic-resistant bacteria isolated in this study was *Pseudomonas aeruginosa* which can thrive and resist higher concentration of arsenic salts, i.e., up to 5000 ppm (μg/ml). The results showed that the bacteria have evolved some mechanism to tolerate high concentrations of arsenic, thus, can efficiently remove arsenic from contaminated water.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest

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