

Review article

Microorganisms in Arsenic Bioremediation with Special Reference to Filamentous Fungi: A Review

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Received : August 2, 2017; revised : September 5, 2017; accepted: September 12, 2017

Abstract: Arsenic pollution of groundwater and other water sources is one of the biggest environmental disasters people are facing presently. About 140 million people around the world have been exposed to arsenic contaminated groundwater. Due to its carcinogenic and toxic effects to human and animal's health, remediation of arsenic-contaminated water has become of prime importance. The north-eastern region of India has long been reported with high level of arsenic contaminations. In a preliminary survey conducted in Arunachal Pradesh (Papum Pare and Lower Subansiri districts) and Assam (North Lakhimpur district), arsenic contamination was detected as high as 193.2 ppb which is manifold higher than the arsenic maximum contaminant level (MCL) set by Environmental Protection Agency (EPA) for public water supplies (10 ppb). Therefore, this paper is presented with the aims to review current status of arsenic pollution worldwide with reference to north eastern India along with various arsenic remediation techniques currently available (conventional and modern). In recent years, bioremediation techniques have emerged out as modern innovative technologies for the removal of arsenic from aqueous system. Many highly arsenic resistant microbes (bacteria, fungi and algae) have been reported with high arsenic tolerance capacity and/or ability to oxidize arsenite to less toxic forms. Among them, fungi could be potential agent of arsenic bioremediation due to their mycelia nature, high growth capacities and production of variety of enzymes, however less emphasis has been given to this group of organisms. Thus this review mainly emphasises the role of filamentous fungi as an effective agent and potentially be used for the bioremediation of arsenic from arsenic contaminated ground water.

Key words: Algae, Arsenic pollution, Bacteria, Bioremediation, Filamentous fungi

Introduction

Arsenic (As) is a toxic element widely distributed in nature. Arsenic pollution is currently a major environmental problem because metal ions persist in the environment due to their non-degradable nature posing threat to both soil and water resources at potentially harmful levels. In natural waters, it exists in both inorganic and organic forms. The inorganic salts of arsenic i.e. arsenite As(III) and arsenate As(V) are its most toxic forms than organic forms (Srivastava *et al.*, 2011). The

detection of arsenic has threatened the use of groundwater as major source of drinking water throughout the globe (Bundschuh *et al.*, 2015). In addition to other forms, the oxides of arsenic are most hazardous for human health. Their origin in soil, water and air is due to various natural processes (Matschullat, 2000; WHO, 2001; Bhattacharya *et al.*, 2002). These forms are extracted into the groundwater as components of geologic formations. As (III) is dominant in

more reduced condition whereas As(V) is dominant in oxidizing environment. The toxicity and bioaccumulation tendency of arsenic in the environment is a serious threat to human health. Skin lesions, rhagades, damaged mucous membrane, digestive, respiratory, circulatory and nervous systems and skin cancers etc. are some of the serious health hazards caused due to arsenic poisoning. The current evidence indicates that arsenic increases the risk of skin, liver and lung cancers (Wang *et al.*, 2001; Biswas *et al.*, 1998). It is also associated with non-cancer health effects such as diabetes, hypertension and other cardiovascular events. Although limited, the existing evidence also suggests that arsenic may have adverse reproductive effects in humans (Hopenhayn, 2006). Due to hazardous effects of arsenic on human systems, the World Health Organization (WHO) in 1993 and the United States Environmental Protection Agency (US EPA) in 2001 reduced the limit of arsenic in drinking water from 50 µg/L to 10 µg/L (Pokhrel and Viraraghavan, 2006).

Sources of arsenic pollution

Arsenic is the 20th most abundant element on earth. It is released into the environment in many ways such as weathering and volcanic eruptions, and may be transported over long distances as suspended particulates and aerosols through water or air due to various natural processes. Arsenic emission from industrial activity also accounts for widespread contamination of soil and groundwater environment (Jacks and Bhattacharjee, 1998; Juillot *et al.*, 1999). Arsenic is extracted into the groundwater as components of geologic formations. Arsenic emissions to the atmosphere are reported on global, regional and local scales (Nriagu, 1988; Pacyna, 2001). Once introduced into the atmosphere (including lithosphere and hydrosphere), it may circulate in natural ecosystems for a long time depending on the prevailing geochemical environments (Boyle, 1973; Yan chu, 1994).

Arsenic in soil

Arsenic is known to occur naturally in the earth's crust, metal ores (soils and rocks) and sediments both in organic and

inorganic forms (Stolz *et al.*, 2006) in detectable quantities. It may also occur as sulphides, oxides or salts of sodium, copper and iron among others (Tsai and Singh, 2009; Rosen and Liu, 2009). No clearly defined relationship exists between the arsenic content of soils and the parent material or climatic conditions under which the soils were formed. Extensive use of arsenate in copper smelting industries, metallurgical activities, pigments and insecticides are the major sources of arsenate in soil and natural waters (Bhargavi and Savitha, 2014).

Arsenic availability in ground waters

The arsenite and arsenate forms of arsenic occur most commonly in the aquatic environment. Arsenic is found in aqueous environments predominantly in oxidation states as 3+[As(III), arsenite] and 5+[As(V), arsenate]. The other oxidation states of arsenic include 0 (arsenic), 3⁻ (arsine) (Jiang *et al.*, 2014). Arsenic contaminated water may also contain it in acid forms i.e. arsenous acid and arsenic acid or their derivatives. These acid forms are soluble forms of arsenic near neutral pH. These compounds are extracted from the underlying rocks that surround the aquifer. Arsenic reaches into groundwater during weathering of rocks and minerals followed by subsequent leaching and run-off. The main factors responsible for controlling arsenic speciation in groundwater include redox potential (Eh), adsorption/desorption, precipitation/dissolution, arsenic speciation, pH, presence and concentration of competing ions, biological transformation, etc. (Ghosh, 2010).

Arsenic pollution worldwide

Natural arsenic pollution is a global phenomenon. Occurrence of high concentrations of arsenic in environment has been recognized as a major public health concern in several parts of the world. The World Health Organization (WHO) in 2001 estimated that about 130 million people worldwide are exposed to arsenic concentrations above 50 µg/L whereas about 140 million people have been exposed to more than 10 µg/L arsenic in drinking water (Bagchi, 2007). Elevated arsenic level has been documented in 70 countries on all

continents, except Antarctica. A total 42 countries have been detected with high arsenic pollution in which Asian countries are most affected. They are Bangladesh, China, India, Italy, Japan, Nepal, Taiwan, Thailand, Vietnam, Poland, Finland, Hungary, Spain, Canada, USA, Chile, Argentina, Mexico and New Zealand etc. (Jiang *et al.*, 2014; Shankar *et al.*, 2014). An estimated 36 million people in the Bengal Delta are at risk from drinking arsenic-contaminated water (Nordstrom, 2002). In Bihar, India, 40 % of 3000 tube wells exceed the WHO limit of arsenic levels and that 12% contained water at more than 20 times the limit (Pearce, 2003). WHO has described the situation of Arsenic contamination in Bangladesh as the largest poisoning of a population in history (Shankar *et al.*, 2014).

Arsenic contamination - Indian Scenario

In India, arsenic contamination of ground water was first detected in West-Bengal. After that many states, like Jharkhand, Bihar, and Uttar Pradesh in flood plain of the Ganga River; Assam and Manipur in flood plain of the Brahmaputra and Imphal rivers, and Rajnandgaon village in Chhattisgarh have chronically been reported with arsenic contaminated ground water. Ganga-Meghna-Brahmaputra basin of India is one of the major arsenic-contaminated hotspot in the world (Bhattacharya *et al.*, 2010). Nine districts in West Bengal have arsenic levels in groundwater above the WHO maximum permissible limit (Chowdhury *et al.*, 2000). Among the contaminated districts, Nadia district is severely affected with high level of arsenic contamination and large area coverage (Bhattacharya *et al.* 2009). The total number of arsenic-affected people in the country is about 1.48 crore as of March, 2017 as per current affairs (gktoday.in/tags/arsenic-contamination link).

Arsenic in north east India

Recent detection of arsenic in groundwater from large areas of north eastern region of India has ranged the bell of concern for millions of people. It was detected in parts of Assam, Arunachal Pradesh, Manipur, Meghalaya, Mizoram, Nagaland Tripura and Sikkim (Singh, 2004; Devi *et al.*, 2009). A report (2007) of north eastern Regional Institute of Water and Land

Management (NERIWALM) mentions arsenic levels in Assam, Manipur, Tripura and Arunachal Pradesh above 300 parts per billion (ppb). The 28,181 nos. of water sources located in Assam have been found contaminated with arsenic, iron and fluoride inorganic materials, followed by 2,931 in Tripura, 566 in Arunachal Pradesh, 136 in Nagaland, 124 in Meghalaya, 76 in Sikkim, 37 in Manipur and 26 in Mizoram (Devi *et al.*, 2009). In Assam (Chakraborti *et al.*, 2004), maximum arsenic concentration ($490 \mu\text{g/L}$) was observed in Jorhat (Titabor, Dhakgorah, Selenghat and Moriani blocks), Dhemaji (Sissiborgoan and Dhemaji blocks), Golaghat district (Podumani block) and Lakhimpur (Boginodi and Lakhimpur blocks) and Nagaon (Singh, 2004). In flood plain area of Assam viz. Barpeta, Dhemaji, Dhubari, Darrang, and Golaghat, the arsenic was found in between 100-200mg/L. In Manipur, arsenic was reported only in Kakching block area of Thoubal district. As per Agartala news (2007) arsenic (65-444mg/L) was found in Jirania and Bishalgarh in West Tripura district, Salema, Halahali, Kamalpur, Joynagar in Dhalai district, Sanitala, Rajbari, Dharmanagar, Kailishahar, Kanchanpur and Jampui in North Tripura district. In Nagaland, 7 locations in Mokokchung and 5 locations in Mon districts were detected with arsenic in groundwater (Singh, 2004). In Arunachal Pradesh, six districts were detected with arsenic contamination and maximum arsenic (618 mg/L) was found in part of Midland block of Dibang valley district. However, as per the investigation conducted in 2011 (Table 1), Papum Pare district of Arunachal Pradesh (which was earlier detected with high arsenic) was found safe from arsenic contamination, as compared to its neighbouring district of Assam i.e. North Lakhimpur where very high arsenic was detected in ground water.

Arsenic removal from ground water

The available arsenic removal methods may be grouped as:

1. Chemical/Physical Treatment Technologies (precipitation oxidation coagulation/filtration, adsorptive media, ion exchange, reverse osmosis),
2. Biological Treatment Technologies (using biological sulfate reduction to precipitate arsenic and heavy metals),

3. Modern Treatment Technologies (electrochemical arsenic remediation (ECAR), regenerating adsorptive media (AM), nanomaterials based arsenic removal system) and
4. Bioremediation Technologies (using microorganisms and plants).

In addition, three alternate arsenic removal technologies are also used viz. (i) Removal system based on alum and iron coagulation; (ii) Removal system based on sorptive filtration using iron coated sand filter, and (iii) Removal system based on sorptive filtration using gravel bed containing iron sludge (Reinsel, 2015) including sludge disposal.

Bioremediation of arsenic using microorganisms

Bioremediation is considered as an alternative processing technology for removing the arsenic ions from polluted area. It is the technique of reducing or converting environmental pollutants into less toxic forms via naturally living organisms/ their systems. Microorganisms reduce the toxicity of contaminants by using them in their metabolic processes as energy source. Many naturally occurring microorganisms such as bacteria, algae and fungi or even a few plant species hold potentials to degrade or detoxify hazardous ingredients. They have developed different mechanisms like arsenite methylation, arsenite oxidation, arsenic volatilization etc. to transform more toxic arsenite to less toxic arsenate (Qin et al., 2006).

Bacteria

A large number of bacteria have been reported to remove arsenic from liquid medium (Table 2). The bacteria strains with the ability to resist arsenic include *Acinetobacter*, *Agrobacterium*, *Alcaligene*, *Alkaliphilus oremlandii*, *Arthrobacter*, *Bacillus*, *Bosea*, *Bradyrhizobium*, *Citrobacter*, *Chromobacter*, *Clostridium*, *Cupriavidus*, *Desulfomicrobium*, *Enterobacter*, *Escherichia coli*, *Microbacterium oxydans*, *Ochrobactrum anthropi*, *Polyphusa peniculus*, *Pseudomonas*, *Psychrobacter rhodobium*, *Rhodococcus*, *Sinorhizobium*, *Staphylococcus*, *Sulfurospirillum*, *Thiobacillus*, *Vibrio*, *Wolinella*, an arsenic reducing bacteria from *Flavobacterium-*

Cytophaga group and a ferum reducing bacteria such as *Geobacter* species (Lim et al., 2014; Mumford et al., 2012; Liao et al., 2011). However, most of the bacteria could remove arsenic contents comparatively in lower percentages. *Lactobacillus casei* (DSM20011) can remove only 7.8±1.7% to 38.1±9.0% of As(V) from liquid medium (Halttunen et al., 2007). A special type of enzyme 'arsenic oxidase' present in the protoplasm of arsenic oxidizing bacteria is responsible to oxidize arsenite to arsenate (Andreoni et al., 2012). Bacterial strain named *Aneurinibacillus aneurinilyticus* can remove 51.99% of arsenite and 50.37% of arsenate from liquid medium (Dey et al., 2016). A number of *Bacilli* have been used by many researchers (Dey et al., 2016; Lim et al., 2014; Ghodsi et al., 2011) that could remove maximum 51-60 % of arsenic. The bacterial strain *Pseudomonas putida* (MTCC 1194), *Ralstonia eutropha* (MTCC 2487) could remove 60% and 67 % arsenic respectively (Mondal et al., 2008). Iron oxidizing bacteria was removing 80 % of arsenic from aqueous medium (Katsoyiannis et al., 2002). A genetically engineered *Escherichia coli* expressing ArsR gene could remove 98% of arsenic. ArsR is a metalloregulatory protein, which offers high affinity and selectivity toward arsenite, was overexpressed in *Escherichia coli* and resulted in elevated levels of arsenite bioaccumulation but also a severe reduction in cell growth (Kostal et al., 2004), however; bacterial strain *Marinomonas communis* has removed 100% arsenic i.e. 45% in cytosol fraction, and 55% in membrane-associated fraction (Takeuchi et al., 2007). *Sulfurospirillum arsenophilum* and *Chrysiogenes arsenatis* are notable arsenic metabolising bacteria. Instead of respiring with oxygen, *Chrysiogenes arsenatis* respire using the most oxidized form of arsenic, arsenate hence can be used in bioremediation (Afkar, 2012).

Algae

A few species of algae such as *Ankistrodesmus convolutes*, *Chlorella vulgaris*, *Euglena gracilis*, *Fucus gardneri*, *Lessonia nigrescens*, *Spirulina platensis* etc. have also been reported to possess bioremediation capabilities (Table 2). The species *Tetraselmis chuil* (Irgolic et al., 1977) has shown high tolerance

against arsenic. *Ankistrodesmus convolutes*, *Chlorella vulgaris*, *Euglena gracilis*, *Scenedesmus bijuga*, *Spirulina platensis* and a mixed culture of *Oscillatoria-Lyngbya* could remove 43% to 64% of arsenic after 21 days of incubation from liquid medium at 0.1mg/L concentration (Samal et al., 2004). Other algal species include *Ankistrodesmus convolutes*, *Euglena gracilis*, *Fucus gardneri*, *Lessonia nigrescens*, *Spirulina platensis* which have removed 43 – 64 % of arsenic at 0.1mg/L concentration, *Spirulina platensis* after 21 days of incubation. *Scenedesmus abundans* could remove high (70%) arsenic contents (Jahan et al., 2006). *Lessonia nigrescens* (Hansen et al., 2006) have maximum removal capacities of 45.2 mg/g (pH = 2.5), 33.3 mg/g (pH = 4.5), and 28.2 mg/g (pH = 6.5). A *Chlorella* strain could also remove 50% of arsenite from a solution (Beceiro-Gonzalez et al., 2000). *Fucus gardneri*, a brown algae, was also found to be arsenic tolerant (Lim et al., 2014). *Spirogyra hyalina* dried biomass was used as biosorbent for removal of arsenic (Kumar and Oommen, 2012).

Fungi

Fungi are being increasingly investigated for mycoremediation of arsenic due to their abilities to remove, sequester, and/or detoxify arsenic by more efficient and environmentally sound methods than traditional metal remediation and by other microorganisms (Singh, 2006). Owing to specific properties of fungi, like their mycelia nature, high growth capacities and production of variety of enzymes also make them a potential agent of arsenic bioremediation; however less emphasis has been paid to this group of organisms. The ability of filamentous fungi to volatilize arsenic is demonstrated by many scientists as compare to unicellular yeasts. Fungi, being able to grow in diverse environments and have tolerated high amount of arsenic, showed fast growth with high cell wall binding and high metal uptake capacities (Visoottiviset and Panviroj, 2001; Vala and Sutariya, 2012; Rodriguez et al., 2013). Zafar et al. (2007) have observed that fungi are able to tolerate, biosorb and detoxify metals by several mechanisms including valence transformation, extra and intracellular precipitation and active uptake. Recently Lim et al. (2014) has reported

arsenic resistant filamentous fungi *Fomitopsis pinicola*, *Penicillium gladioli*, *Fusarium oxysporum meloni* and *Scopulariopsis koningii*.

Many fungal species are capable of transforming inorganic arsenic compounds, arsenite and arsenate by biomethylation, into methylated arsenic species such as monomethylarsonic acid (MMA), dimethylarsinic acid (DMA), trimethylarsine (TMA), and trimethylarsine- oxide (TMAO). Three of these, MMA, DMA and TMAO, are less toxic than arsenate, arsenite and TMA (Visoottiviset and Panviroj, 2001). *Penicillium* and *Aspergillus* spp. are the two most metal tolerant species, showing enhanced growth even at high concentrations (2000 mg/L) of heavy metals (Valix et al., 2001). A number of arsenic resistant *Penicilli* with 24 different fungi and 7 bacterial species were isolated from arsenic contaminated ground waters (Shrivastava et al., 2012). The main factors that affect biovolatilization by fungi such as cultivation time, chemical treatment, live/dead cells etc. A strain of *Aspergillus candidus* isolated from waters of Bhavnagar coast, Gulf of Cambay, west coast of India could remove high amount of arsenic after 3 days cultivation (Vala, 2010). It was amounted in *Scopulariopsis brevicaulis* which was able to biovolatilize 0.007-0.014mg/L of arsenic after 7 days cultivation from culture medium (Pearce et al., 1998). The filamentous fungi *Penicillium coffeae* was able to remove 66.8 % of arsenic from liquid medium after treating with alkali (Bhargavi and Savitha, 2014). Arsenic removal using modified fungal biomass of *Aspergillus niger* (ATCC # 11414) treated with iron oxide (Pokhrel and Viraraghavan, 2006) could remove up to 95% As(V) and 75% As(III). Not only live fungal cells but autoclaved fungal mats collected as waste product during black tea fermentation have also been used to remove arsenic after pre-treatment with FeCl₃. It had removed 100% of As(III) and Fe(II) after 30 min and 77% of As(V) after 90 min contact time (Murugesan et al., 2006). Iron oxide coated fungal biomass of *Paecilomyces* species collected from polluted air with industrial vapors in Mexico could remove 8.4% of arsenic after 24 hours incubation at pH 7 (Rodriguez et al., 2013). Fungal biomass of 10 out of 15

fungal strains collected from agricultural soils of West Bengal, India removed 10.92% to 65.81% arsenic biologically from the liquid medium (Srivastava *et al.*, 2011). Two arsenic resistant strains of *Aspergillus flavus* and *A. niger* were isolated from polluted sites of Kolkata were capable of removing 50%-76% of arsenic from different arsenic enriched medium (Mukherjee *et al.*, 2013). *Aspergillus niger*, *Trichoderma viride*, and *Penicillium glabrum* (Urik *et al.*, 2007) have removed 0.010 to 0.067 $\mu\text{g/L}$ arsenic whereas *Penicillium purpurogenum* could remove 3.4 mg/g of arsenic (Say *et al.*, 2003). Cernansky *et al.*, 2009 showed that fungal species *Neosartorya fischeri*, *Aspergillus clavatus* and *Aspergillus niger* isolated from soil samples of a mining site highly contaminated with arsenic can volatile approximately 23% of arsenic from all arsenic rich culture medium. *Aspergillus clavatus* had biovolatilized 20-22.1% of arsenic (Urik *et al.*, 2007). Edvantoro *et al.*, (2004) used augmentation of particular arsenic volatilizing fungal strains (*Penicillium* sp. and *Ulocladium* sp.) for bioremediation of cattle-dip site soils contaminated with arsenic. After chemical modification, *Penicillium chrysogenum* reported with higher removal ability as compare to unmodified biomass (Loukidou *et al.*, 2003). Three filamentous fungi viz. *Aspergillus niger*, *Serpula himantioides* and *Trametes versicolor* were investigated for their potential abilities to accumulate (and possibly solubilize) arsenic from an agar environment consisting of non-buffered mineral salts media amended with 0.2, 0.4, 0.6 and 0.8% (w/v) arsenopyrite (FeAs₂). Arsenic solubilisation was observed in order of *A. niger* > *S. himantioides* > *T. versicolor* with *T. versicolor* as the most effective species. *Aspergillus niger*,

Serpula himantioides and *Trametes versicolor* accumulate and solubilize high arsenic from agar media with non-buffered mineral salts (Adeyemi, 2009). Four out of fifteen fungal species isolated from arsenic-contaminated agricultural fields in West Bengal, India, the isolates of *Westerdykella*, *Trichoderma*, *Rhizopus* and *Lasiodiplodia* improved soil nutrient content and enhanced plant growth in arsenic infested area when inoculated in plants. The use of these fungi has been recommended as bio-inoculants for plant growth

promotion and improved soil properties in arsenic-contaminated agricultural soils (Edvantoro *et al.*, 2004; Srivastava *et al.*, 2012). Higher As bioaccumulation and biovolatilization has been observed in seven fungal strains, *Aspergillus oryzae*; three *Fusarium* spp., *Aspergillus nidulans*, *Rhizomucor variabilis* and *Emericella* sp. These strains have shown significant plant growth promotion in some plants species (Singh *et al.*, 2015). Other highly arsenate tolerant strains (up to 10000 mg/L) belong to *Rhizopus*, *Microdochium*, *Chaetomium*, *Myrothecium*, *Stachybotrys*, *Rhizomucor*, *Fusarium*. The arsenic resistant fungal species of *Penicillium*, *Aspergillus*, *Neosartorya* and *Gliocladium reseau* and a yeast *Candida humicola* have also been reported as potential agents

Table 1. Arsenic concentration in ground water samples

Sl. No.	Site of collection	Coordinates	Arsenic concentration (ppb)
No.		Latitude	Longitude
North Lakhimpur District, Assam			
1.	Gandhalipar1	N27°11.293'	E94°02.366' 193.2
2.	Khanajan	N27°13.335'	E94°01.739' 161.1
3.	Nalkata1	N27°16.846'	E94°03.592' 103.55
4.	Gosaipathar 1	N27°00.976'	E93°54.176' 44.9
5.	Bogolijan,	N27°08.710'	E93°45.031' 23.95
6.	Lakhimpur Town	N27°14.446'	E94°04.969' 20.63
7.	Bihlanganiya	ND	ND 09.92
8.	Gandhalipar 2	N27°22.1258'	E93°44.726' 7.76
9.	Rangajan	N27°00.070'	E93°53.290' 4.88
10.	Nalkata 2	N27°15.159'	E94°04.698' 2.58
11.	Kachikata	N27°00.210'	E93°53.974' <2.0
12.	Gosaipathar 2	N27°02.038'	E93°53.512' <2.0
13.	Rajbari Tinali	N27°01.818'	E93°52.944' <2.0
Arunachal Pradesh			
14.	Doimukh 2, Papum Pare	N27°08.710'	E93°45.031' 23.04
15.	NERIST 1, Nirjuli, Papum Pare	ND	ND 09.92
16.	Doimukh 3, Papum Pare	N27°08.750'	E93°45.141' 9.8
17.	Doimukh 6, Papum Pare	N27°08.437'	E93°45.979' 4.71
18.	Yazali 2, Lower Subansiri	N27°32.495'	E93°48.932' 2.44
19.	Yazali 1, Lower Subansiri	N27°27.44'	E93°45.855' <2.0
20.	Zero 1, Lower Subansiri	N27°35.745'	E93°50.370' <2.0
21.	Zero 2, Lower Subansiri	ND	ND <2.0
22.	Zero 3, Lower Subansiri	N27°07.785'	E93°44.303' <2.0
23.	NERIST (5 spots) Nirjuli, Papum Pare	N27°07.590'	E93°44.218' <2.0
24.	Doimukh 1, Papum Pare	N27°08.664'	E93°45.216' <2.0
25.	Doimukh 4, Papum Pare	N27°08.663'	E93°45.274' <2.0
26.	Doimukh 5, Papum Pare	N27°08.545'	E93°45.810' <2.0

ND- Not detectable

Table 2. Microorganisms showing arsenic bioremediation ability

Group of microorganisms	Arsenic removal/tolerance capacity	Reference
Bacteria	Bacteria	Bacteria
<i>Aneurinibacillus aneurinilyticus</i> , <i>Bacillus</i> sp.	Removed 51.99% and 51.45% of arsenite and 50.37% and 53.29% arsenate respectively	Dey et al., 2016
<i>Alkalilimnicola ehrlichii</i> , <i>Bacillus sphaericus</i>	Arsenite oxidation by <i>ArxA</i> and resistance by <i>ars C</i> genes	Lim et al., 2014
<i>Bacillus selenitireducens</i> , <i>Chrysiogenes arsenatis</i> , <i>Pyrobaculum arsenaticum</i> , <i>Sulfurospirillum arsenophilum</i> , <i>Sulfurospirillum barnesii</i>	Reduced arsenate to arsenite under anaerobic condition	Afkar, 2012
<i>Bacillus macerans</i> , <i>B. megaterim</i> , <i>Corynebacterium vitarium</i>	Removed 60%, 38% and 43% arsenite	Ghods et al., 2011
<i>Neisseria mucosa</i> , <i>Rahnella aquatilis</i>	Reduced arsenate and selenate	Youssef et al., 2009
<i>Bacillus indicus</i> <i>Pseudomonas putida</i> <i>Ralstonia eutropha</i>	Removed 61%, 60% and 67% arsenic from liquid medium	Mondal et al., 2008
<i>Lactobacillus casei</i> DSM20011	Removed arsenate (7.8 % to 38.1 %)	Halttunen et al., 2007
<i>Marinomonas communis</i>	100% arsenic removal (45% in cytosol and 55% in membrane)	Takeuchi et al., 2007
<i>Escherichia coli</i> containing <i>ArsR</i> gene	Removed 98% arsenite	Kostal et al., 2004
Iron oxidizing bacteria	Removed 80% Arsenic	Katsoyiannis et al., 2002
<i>Alcaligenes faecalis</i> , <i>Comamonas terrae</i>	Oxidized Arsenite to arsenate	Philips and Taylor, 1976; Chitpirom et al., 2009
Algae		
<i>Spirogyra hyalina</i>	Highest arsenic uptake peak at 40 mg/L in 120 min	Kumar and Oommen, 2012
<i>Chlorella vulgaris</i> Beijerinck var. <i>vulgaris</i>	Removed 40 ppm arsenic	Maeda et al., 2006
<i>Lessonia nigrescens</i>	Removed 45.2, 33.3 and 28.2 mg/g arsenic at 2.5, 4.5, 6.5 pH respectively	Hansen et al., 2006
<i>Scenedesmus abundans</i>	Removed 70% of arsenic	Jahan et al., 2006
<i>Ankistrodesmus convolutus</i> , <i>Chlorella vulgaris</i> , <i>Euglena gracilis</i> , <i>Oscillatoria-Lyngbya</i> mixed culture, <i>Scenedesmus bijuga</i> , <i>Spirulina platensis</i>	Removed arsenic 64%, 50%, 47%, 45% and 43% each respectively	Samal et al., 2004
<i>Chlorella</i> sp.	Removed 50% arsenite	Beceiro-Gonzalez et al., 2000
Fungi		
<i>Chaetomium</i> sp. <i>Fusarium</i> sp. <i>Microdochium</i> sp. <i>Myrothecium</i> sp. <i>Rhizomucor</i> sp. <i>Rhizopus</i> sp. <i>Stachybotrys</i> sp.	Tolerates high arsenate (upto 10 000 mg/L)	Singh et al., 2015
<i>Penicillium coffeae</i>	Arsenic adsorption with alkali treated biomass (66.8 %) and with untreated dead biomass (22.9 % - 60.2 %)	Bhargavi & Savitha, 2014
<i>Aspergillus flavus</i> , <i>Aspergillus niger</i>	Removed 50% - 76 % arsenic	Mukherjee et al., 2013
<i>Paecilomyces</i> sp.	Removed 8.4 % of arsenic at pH 7	Rodriguez et al., 2013
<i>Westerdykella</i> sp. <i>Trichoderma</i> sp., <i>Rhizopus</i> sp. <i>Lasiodiplodia</i> sp.	Used as bioinoculants in arsenic-contaminated agricultural soils	Srivastava et al., 2012
<i>Aspergillus flavus</i> <i>Rhizopus</i> sp.	Higher biomass accumulation in As(III) medium	Vala and Sutariya, 2012
<i>Aspergillus</i> (3 species), <i>Neocosmospora</i> (2 species) <i>Penicillium</i> spp. (2 species), <i>Rhizopus</i> sp., <i>Trichoderma</i> sp., <i>Mycelia sterila</i>	Remove 10.92 to 65.81% arsenic	Srivastava et al., 2011
<i>Aspergillus candidus</i>	Removed high amount of arsenic	Vala, 2010
<i>Aspergillus niger</i> , <i>Serpula himantoides</i> <i>Trametes versicolor</i>	Accumulated and solubilize arsenic from agar media with non-buffered mineral salts	Adeyemi, 2009
<i>Aspergillus clavatus</i> <i>Aspergillus niger</i> <i>Neosartorya fischeri</i>	Each volatilized 23% arsenic	Cernansky et al., 2009
<i>Aspergillus niger</i>	Removed 0.010 - 0.06750 β g/L arsenic	Urik et al., 2007
<i>A. niger</i> A	Biovolatilized arsenic (25.2 - 26.8%)	
<i>A. niger</i> B	Biovolatilized arsenic (9.2 - 10.3%)	
<i>Aspergillus clavatus</i>	Biovolatilized arsenic (20 - 22.1%)	
<i>Penicillium glabrum</i>	Biovolatilized arsenic (25.2 - 26.2 %)	
<i>Trichoderma viride</i>	Biovolatilized arsenic (4 - 9.3%)	
<i>Aspergillus niger</i> treated with iron oxide	Removed 95% As(V) and 75% As(III)	Pokhrel and Viraraghavan, 2006
<i>Penicillium</i> sp. <i>Ulocladium</i> sp.	Bioremediation of arsenic contaminated soils	Edvanto et al., 2004

<i>Penicillium chrysogenum</i>	Higher arsenic removal by chemically modified than unmodified fungal biomass	Loukidou et al., 2003
<i>Penicillium purpurogenum</i>	Removed 3.4 mg/g of arsenic	Say et al., 2003
<i>Scopulariopsis brevicaulis</i>	Biovolatilized 0.007–0.014 mg/L of arsenic	Pearce et al., 1998

for arsenic removal through bioaccumulation and biovolatilization (Visoottiviseth and Panviroj, 2001; Urik et al., 2007; Cernansky et al., 2009; Srivastava et al., 2011). Table 2 indicates different fungal species with their arsenic removal/tolerance capacities described so far. The experiments conducted in our lab with 10 arsenic tolerant filamentous fungi (Pramanik et al., 2016) isolated from arsenic contaminated groundwater showed that all were able to remove high Arsenic contents (>80%). Out of 10 fungi, 6 could remove > 85% of Arsenite and 9 could remove >85% of arsenate from the arsenic enriched liquid media (data not published).

Conclusions

Recent research focussed in this review suggests that the problem of arsenic contamination of drinking water is assuming global dimensions and posing serious threat to human health. In India, northeast region have long been reported with arsenic contaminated ground water above WHO's permissible limit (10 µg/L). In spite of various precautionary and corrective measures adopted so far, new arsenic affected areas have been regularly reported. The control strategies (conventional and modern), thus, need to be reviewed and strengthened by schematic scientific patronages. Microorganisms such as bacteria, algae and fungi have shown great affinity towards many heavy metals including arsenic and reported as arsenic tolerant/resistant hence being utilised effectively for arsenic bioremediation. Although bacteria have developed various mechanisms to grow in arsenic environment, filamentous fungi have shown more promising results to remove arsenic from liquids. This review, hence, highlights the contributions of the various filamentous fungi which are potentially effective and readily available for arsenic removal. Many of these fungi are ubiquitous in nature hence can be easily accessed and utilized, however; a sustainable approach still needs to be developed in the form of an

appropriate, economic and easy 'ready to use' technology for arsenic removal by these fungi.

Acknowledgement

The fund received in the form of research grant under DBT-Twining project (BT/19/NE/TBP/2010 dated 25/02/2011) is thankfully acknowledged.

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