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Review Article

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The Potential Impact of Micro-organisms on Radioactive Wastes

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Abstract Beside industrial wastewater, groundwater, seawater, soil, rocks, medical diagnosis, radionuclides sources comprise nuclear power plants and atomic weapons. Being labile, bioavailable, and soluble radionuclides result in pollution and radiotoxicity. De-contamination through classical chemical, physical and thermal treatments, bioremediation methods such as phytoremediation, saprophytes and fungal mycorrhiza were also applied for radionuclides biosorption. This is because of chitin and chitosan functional groups as they are the major part of the fungal cell wall. Also, due to the activity of the hyphal sheath, vacuolar bodies, and pigments where deposition occurs. Rad-waste soil decontamination depends on several factors as concentration of ions, the moisture content of the soil, soil depth, growth rate, species of the fungus, biomass viability and the transpiration rate of the host plant. Similarly, uptake and retention by bacteria show significant radionuclides biosorption abilities. Furthermore, anaerobic bacteria are able to reduce uranium hexavalent oxidation state U(VI)to insoluble uranium tetravalent U(IV) in the bioremediation of uranium-contaminated groundwater. Although U(VI) has an important effect on the environment, yet a very little is known about its bio- reduction. Bio- stimulation in another way, where the addition of trace elements, electron donors or acceptors to encourage activity and growth of natural microflora that are able to decontaminate rad-waste. Finally, complexations with bacterial negatively charged intracellular components, or secretions in some species of chelating agents leads to biomineralization or precipitation. These strategies strongly suggest the possibility to entrap, recovery and recycling the waste after removal of the dominant elements with low cost.

Keywords Phytoremediation, mycorrhiza, bioaccumulation, bioaugmentation, bio-reduction, radionuclide, El Dabaa nuclear power plant.

Introduction

The use of radioactive materials plays a beneficial role in medical, agricultural, industrial and in nuclear power plants, however these materials have known negative effects on human health due to production of small quantities of radioactive waste [1-3]. Radioactive waste is that substance which is radioactive in itself or contaminated with radioactive material and hasn't any significant application. The waste could be controlled by diluting it such that the rate or concentration of any radionuclides returned to the biosphere is harmless. Non-nuclear power waste results from the laboratory, research, industrial instruments, radiography, and nuclear medicine used at the hospital. Rad-wastes activity level depends on the quantity of radiation it emits, type of the emitting radiation, and the half-life. Short-lived unstable radioactive materials have a half-life of fewer than forty years. Half-life means that half the atoms in the radioactive materials will decay into the stable form in less than forty years. Long-lived radioactive materials have a half-life of greater than forty years.

Rad-wastes Activity Levels

Low-level waste includes resins, chemical sludges, and contaminated materials from abandoned reactor. This group undergoes geological disposal *i.e.* Near-surface disposal. Another low-level liquid waste from reprocessing plants is discharged into the sea [4].

Intermediate -low-level wastes. (ILW) contains activity levels higher than Liquid low-level (LLW) wastes and less than High-level waste (HLW) are typically solidified in cement [5-6].

HLW resulted from burning of uranium as a nuclear fuel to produce electricity, fission products, IAEA's [7], and transuranic elements are generated in the reactor core. Both long- and short-lived wastes require cooling, shielding, completely isolated, and disposing of in deep geologic disposal.

Each stabilized waste was saved in a special container regarding the waste characters according to IAEA's, [8]. High-level wastes are calcined then incorporated into a glass matrix or trans mutated into short-lived or non-radioactive elements, or immobilized in a ceramic matrix instead of borosilicate glass, Wang and Liang [9].

A Non-nuclear power waste in Inchass research reactor for example, as the scintillator mixture generated from liquid scintillation counting technique which includes radioactive nuclides represent a waste problem, Moreno, et al. [10]. This problem is due to both radioactive contaminants and their organic structure. Liquid scintillator waste was mainly pre-treated through oxidative degradation before cementation to overcome the opposite effect of its organic nature after the hydration of the cement bed, and to avoid the fire risk of the immobilized waste matrix during disposal [11-12].

Moreover, other radioactive waste referred to as 'legacy waste' contains low-level, mixed low-level, and transuranic waste. All radioactive waste decays into non-radioactive elements that must be compacted or incinerated before disposal, as it will be the case regarding radionuclides within the nuclear waste in the afterward going EL Dabaa nuclear power plant.

As previously mentioned, operating nuclear power plant resulted in solid, liquid and gaseous rad-waste at all steps of the nuclear fuel cycle. The cost of its disposal is part of the electricity cost that is paid by the consumers. Relative to other industrial waste, nuclear waste isn't hard to manage.

Storage of Consumed Nuclear Fuel

Nuclear power produces a very large amount of energy relative to the small amount of fuel, and hence, the resulted wastes are also small suitable for disposal in near surface regarding its activity.

Consumed nuclear fuel is discharged away following its release from the reactor for three years as a minimum storage in consumed fuel pools, then storage up to 50 years for Intermediate consumed fuel storage on the plant site, and final disposal in a deep underground container [6, 13].

Solid Rad-Wastes Conditioning

Human health and environment protection via rad-waste safe discharge depends on some factors: type and amounts of chemicals present, size and depth of the polluted area, and type of soil. Such conditioning is changing the rad-waste into a suitable form safe in handling, packaging, transportation, storage, and disposal.

While in situ physical waste processing aims to change the phase of pollutants, chemical processing depend on the chemical structure, the behavior of the pollutants and chemical reactions to produce less toxic compounds from the solid matrix.

These performances are cost-effective and were done in short time relative to biological treatment. Equipment is available and is not energy-intensive. However, these treatments are sensitive to soil parameters as the presence of clay or humic materials in soil. Thermal treatments use heat to fast cleanup soil through volatility, burning, decomposing, or melting the contaminants, but it is also a costly treatment and may take months or several years. Bioremediation techniques then cementation of the bioproduct were successful procedures for treating and solidifying of low- and intermediate-level radioactive wastes [14].

Rad-wastes Bioremediation

Bioremediation comprises biological aspects like plants, bacteria, natural or genetically altered fungi in the decontamination process. So, it includes all strategies: phytoremediation, phytostabilization, phytoextraction, rhizofiltration, bio-sorption, biostimulation, bioaugmentation, and hyperaccumulation [15].

Biological treatment due to the action of microbial metabolism that were introduced to convert pollutants in soil, sludge or groundwater into less toxic substances such as carbon dioxide, water, fatty acids and biomass, is a time-consuming treatment and it has several difficulties as microbes are sensitive to toxins or highly concentrated contaminants in the soil, however it costs less. Mar'in et al., [16] and Omar [17] stated that polar groups of proteins, amino acids, lipids and polysaccharides (chitin, chitosan, glucans) share in the biosorption.

Biosorption runs in two main steps: adsorption of the ions on the cell surface and bioaccumulation within the cell [18]

Phytoremediation can be used to translocate metals from soil from the roots to the aboveground biomass for storage. As an example, plants are for both cleaning up uranium-contaminated soils and waters. The largest quantity of uranium extraction by roots is due to strong sorption, and precipitation processes. Inactive roots material, which has a large reactive surface area for sorption processes could also be used. Addition of some chemicals as citric acid enhances chelate forming substances, for increasing the uptake of uranium by plants [19].

Rhizofiltration means the using of plants in removing uranium from water. The floating macrophyte, namely Lemna gibba, (duckweed) has the ability to absorb Cs-137 and Co-60 from liquid radioactive waste, depends on pH, light, and the viability of the used biomass, it is a time- dependent process [20].

Bioaccumulation of rad-wastes in Bacteria

Thorium (Th) reacts with a binding site on bacteria Alteromonas, Vibrio, *Pseudomonas aeruginosa* and Flavobacterium with strong ligand/carbon ratio due to the functional groups exist in these microorganisms. Similarly, extracellular polymers were produced by Bradyrhizobium and Sinorhizobium and precipitate thorium ions [21]. Yong and Macaskie [22] found that thorium and lanthanum ions were precipitated in a complex with phosphate liberated due to phosphatase activity present in a Citrobacter sp. Other bacteria secrete sequestering agents and specific chelators, such as siderophores that are able to attenuate radionuclides and increase their solubility. Microbacterium flavescens, grown in the presence of radioisotopes produces organic acids and siderophores that dissolve radionuclides through the soil. Pseudomonas aeruginosa secretes chelating agents for uranium and thorium when grown in a medium contains these elements. Dhan *et al*., [23] found out that siderophores interact with actinide oxides of plutonium to immobilize it. Algae as well, can sequestrate U and other metals [24-27].

Bio-reduction in Bacteria

Uranium (U) is the heaviest element, occurs in hexavalent state, is very soluble and travels with water, but uranium in the tetravalent state is insoluble and essentially immobile. Unfortunately, because U is a mobile radioactive metal that presents in the soil, percolating water, the surface and underlying groundwater, it has chemical and radioactive risks on human health and environment that take hundreds of millions of years to decay radioactively into non-toxic residue. The biological adsorption of uranium from aqueous solution was the target for many researchers [28]. Conventional biological activity can transform the metal from toxic to non-toxic form and immobilize it. Some bacteria exuded bioligands that trap U into bimetallic colloids. Bio reduction, sequestration and bio colloids can start biomineralization processes of U. Other bacteria excrete the U compounds back into the water as precipitates or organometallic colloids. This interaction may result into U biomineralization. For example, some marine and freshwater algae are able to introduce U into aragonite ($CaCO_3$) mineral forms [29].

Bio-reduction means that certain bacteria are capable to reduce uranium from its hexavalent state to its tetravalent state hence, decreasing its solubility and trapping it in the soil underground [30]. Bacteria give U(VI) a spare electron in a process called reduction. Cytochrome c3 was designated to be electron carrier for uranium (VI) reduction in Desulfovibrio desulfuricans G20, sulfate-reducing bacteria. Reduction of soluble, toxic and mobile U(VI) to insoluble, less toxic, immobile U(IV) ion is enzymatic and U(IV) is less biologically available. Electrons were from organic carbon (lactate, acetate, ethanol) or hydrogen.

On the other hand, it was found that Geobacter sulfurreducens pili, hair-like appendages with electrical conductivity served as a buffer, protecting the bacterium cell structure, allow adding electrons to uranium ions which causes it to become more water soluble and thus safer to handle and clean-up [31]. Also, Bio-reduction of Pu(V) to Pu(IV) by Bacillus cereus was estimated [32].

Bioaugmentation

As in bio-stimulation, the addition of nutrients, trace elements, electron donors or acceptors happens to induce growth of natural archaea or bacterial cultures. Bioaugmentation is the addition of microflora required to speed up the rate of degradation of a contaminant.

One of the major disadvantage during in situ bioremediation is the permeability decrease due to heavy bacterial growth, which happens in bioaugmentation when nutrients and/or bacteria are applied to a contaminated matrix and the strong attachment of bacteria to soils lead to maximum biomass accumulation in interspaces around application wells. As the accumulation continues, the dramatic permeability reduction increased, which makes the injection pressure increase. Finally, the applied solutions are entrapped within the wells and the low permeability zone affects surrounding groundwater flow and distributions of biodegrading microorganisms [33].

Myco Remediation

Metabolic activities such as decomposition of organic substances and dissolution of rocks and minerals by free-living and symbiotic fungi may result in products that can immobilize uranium.

However, it is not evidenced whether fungi are capable of reducing U(VI) for its immobilization. Fungal interactions with uranium depend on the bio-geochemistry of uranium, and the polluted region. The dependence of land plants on symbiotic mycorrhizal fungi, that are capable of uranium transformations may make the fungal biogeochemical activity of importance in the photo- or other bioremediation strategies for soils polluted with various rad-wastes [34-36].

Remediation through fungi is also called as mycoremediation. Fungi possess the enzymatic system that could be applied to a wide variety of degradation of pollutants.

Mycoremediation via fungi enzymes able to degrade a wide variety of environmentally persistent pollutants, into non-toxic products, namely *Pleurotus pulmonarius, Agaricus, Ganoderma and Schizophyllan commune, Trichoderma harzianumm* and, *Aspergillus fumigatus*, [37]. The uptake of xenobiotics by mushrooms and stabilize the radionuclides within its all tissues includes two steps: bioaccumulation or active metabolism, when pollutants transported into the cell and partitioned into intracellular organelles; and biosorption due to chemical binding of pollutants to the biomass without metabolic energy as adsorption, ion exchange processes and covalent binding. It was stated that bioremediation of solid cellulose-based radioactive waste, e.g.*(Pleurotus pulmonarius),* in Portland cement is a new trend based on the capability of living organisms to biodegrade, absorb, bioconcentrate, and stabilize the radionuclides [38-39].

Radioactive Waste Deposition by Concrete and Cement Composites

Solidification and stabilization of these wastes in cement are suggested to prevent the return of the harmful radioactive materials to the environment during its disposal. Portland cement as a direct immobilization matrix of treated waste form and the engineered barrier of secondary protection in the form of concrete or grout mixture were favored according to its various advantages such as simplicity, low cost, mechanical and thermal stability, [40-43]. However, high leachability conditions control the usage of cement matrix as a solidifying agent. The Leaching test was performed for long period of time by immersing cement-waste due to the method recommended by the International Atomic Energy Agency (IAEA) [44]. The adding of various concentrations of the bioproduct to the cement matrix affect the mechanical integrity of the final cement-waste. It was found that increasing of the bioproduct content is accompanied with apparent deterioration in the compressive value of the cement-waste form, depends on the ratio between pore's solution in the cement surface and the ions in the leachant solution. It was indicated thattraditional cement can provide a highly durable barrier ensures long-term stability of the solidified rad-waste material at ecosystem and landscape.

Cumulative fractions leached, diffusion coefficients and leaching indices values are the main variables for leaching parameters were computed under various conditions. The cellulose-based solid waste composite could be taken into consideration for the disposal process to keep the surrounding biosphere clean [45-46].

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