

Review

Advancements in microbial-mediated radioactive waste bioremediation: A review

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ABSTRACT

The global production of radioactive wastes is expected to increase in the coming years as more countries have resorted to adopting nuclear power to decrease their reliance on fossil-fuel-generated energy. Discoveries of remediation methods that can remove radionuclides from radioactive wastes, including those discharged to the environment, are therefore vital to reduce risks-upon-exposure radionuclides posed to humans and wildlife. Among various remediation approaches available, microbe-mediated radionuclide remediation have limited reviews regarding their advances. This review provides an overview of the sources and existing classification of radioactive wastes, followed by a brief introduction to existing radionuclide remediation (physical, chemical, and electrochemical) approaches. Microbe-mediated radionuclide remediation (bacterial, myco-, and phycoremediation) is then extensively discussed. Bacterial remediation involves biological processes like bioreduction, biosorption, and bioprecipitation. Bioreduction involves the reduction of water-soluble, mobile radionuclides to water-insoluble, immobile lower oxidation states by ferric iron-reducing, sulfate-reducing, and certain extremophilic bacteria, and in situ remediation has become possible by adding electron donors to contaminated waters to enrich indigenous iron- and sulfate-reducing bacteria populations. In biosorption, radionuclides are associated with functional groups on the microbial cell surface, followed by getting reduced to immobilized forms or precipitated intracellularly or extracellularly. Myco- and phycoremediation often involve processes like bio-sorption and bioaccumulation, where the former is influenced by pH and cell concentration. A Strengths, Weaknesses, Opportunities, and Threats (SWOT) analysis on microbial remediation is also performed. It is suggested that two research directions: genetic engineering of radiation-resistant microorganisms and co-application of microbe-mediated remediation with other remediation methods could potentially result in the discovery of in situ or ex situ microbe-involving radioactive waste remediation applications with high practicability. Finally, a comparison between the strengths and weaknesses of each approach is provided.

1. Introduction

Since the inception of nuclear fuel utilization for electricity production in 1954, approximately 394,130 tonnes of heavy metals (t HM) of spent fuel have been generated globally from nuclear power plants (NPPs), accounting for about 99% of the total, along with non-power reactors used for isotope production, research, and other purposes, constituting roughly 1% of the total (International Atomic Energy Agency [IAEA], 2022). The volume of radioactive spent fuel and other radioactive wastes, including worn-out reactor components contaminated with radioactive substances, resulting from nuclear reactor operations (IAEA, 2022) is expected to rise in the future due to the increasing number of nations adopting or planning to adopt nuclear

power to reduce reliance on fossil-fuel-generated energy in recent years. Among these nations are Belarus (Nazarov et al., 2020), Ghana (Agyekum et al., 2020), Egypt, Jordan, Turkey, United Arab Emirates (Hickey et al., 2021), Bangladesh (Ahmed et al., 2020), Sri Lanka (Jayasinghe and Gunasekara, 2021), and others. As predicted by the United Nations Department of Economic and Social Affairs, Population Division UN DESA (2022), these regions are likely to experience growing populations toward 2050, leading to increased demands for alternative energy sources to meet rising energy needs.

Isotopes have found extensive applications in various fields, including medicine for diagnosing and treating health issues such as cancers (Pei et al., 2021), paleoenvironmental science, archaeology, and art, where radiocarbon dating is used to estimate the age of samples

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Table 1
Radionuclides that are commonly found in radioactive wastes from various sources.

Source of Discharge	Form of Discharge	Radionuclide	Half-Life ^{1-7, 9-18, 22-29}	Source of Discharge	Form of Discharge	Radionuclide	Half-Life ^{1-7, 9-18, 22-29}				
A. Nuclear re-processing plant (condition uranium and plutonium to oxides, treat and condition wastes produced by the former, etc.)	Effluent ¹	U-238	4,468,300,000 y			Cm-243	29 y				
		U-235	703,810,000 y			Sr-90	29 y				
		I-129	16,140,000 y			Cm-244	18 y				
		U-234	250,000 y			Pu-241	14 y				
		Tc-99	210,000 y			H-3	12 y				
		Pu-239	24,000 y			Kr-85	11 y				
		Pu-240	6600 y			Eu-154	9 y				
		C-14	5730 y			Eu-155	5 y				
		Am-241	430 y			Pm-147	3 y				
		Ni-63	100 y			Sb-125	3 y				
		Pu-238	88 y			Cs-134	2 y				
		Cs-137	30 y			Cm-242	163 d				
		Sr-90	30 y			Y-90	64 h				
		Cm-244	18 y			Np-239	57 h				
		Pu-241	14 y			Sn-121	26 h				
		H-3	12 y			Am-242	16 h				
		Eu-154	9 y			Ba-137m	3 min				
		Co-60	5 y			C. Uranium mining and milling site	Mill tailings ¹⁸	U-238	4,468,300,000 y		
		Sb-125	3 y					U-235	703,810,000 y		
		Cs-134	2 y					U-234	250,000 y		
		Ru-106	1 y					Th-230	75,584 y		
		Mn-54	312 d					Ra-226	1600 y		
		Ce-144	284 d					Rn-222	4 d		
		Co-57	272 d					D. Nuclear weapon production site (leakage from storage site)	Solid/liquid waste leakage to river ¹⁹	I-131	22 y
		Zn-65	244 d							Zn-65	244 d
		Co-58	71 d							Sc-46	84 d
		Zr-95	65 d							Cr-51	28 d
		Sr-89	51 d							P-32	14 d
Nb-95	35 d	Y-90	64 h								
Y-90	64 h	Np-239	57 h								
Pr-144	17 min	As-76	26 h								
Rh-106	30 s	Na-24	15 h								
B. Nuclear power plant	Spent nuclear fuel ⁸	Pu-242	370,000 y	E. Hospital (radioisotope medicinal application)	Effluent (e.g., urine) ^{12, 20-21}					Ga-72	14 h
		U-234	250,000 y			Mn-56	3 h				
		Tc-99	210,000 y			Sr-89	51 d				
		Pu-239	24,000 y			Re-186	91 h				
		Cm-245	8500 y			Ga-67	78 h				
		Am-243	7400 y			Zr-89	78 h				
		Pu-240	6600 y			In-111	67 h				
		Am-241	430 y			I-133	21 h				
		Am-242m	150 y			Cu-64	13 h				
		Sm-151	90 y			I-123	13 h				
		Pu-238	88 y			Tc-99m	6 h				
		Sn-121m	55 y			F-18	110 min				
Cs-137	30 y	Ga-68	68 min								

^a s: second; min: minute; h: hour; d: day; y: year.

^b The superscripts of numbers are in-text citations, corresponding to: 1. Organisation for Economic Co-operation and Development (2003); 2. Andersen et al. (2004); 3. Asaro et al. (1960); 4. Keenan (1959); 5. Salutsky and Kirby (1955); 6. Snyder and Beard (1968); 7. Kweon et al. (2014); 8. Forström (2012); 9. Zunkley et al. (1996); 10. Ma and Hooda (2010); 11. Fitzsimmons and Mausner (2015); 12. Usuki et al. (2016); 13. Rama Sastry et al. (1964); 14. Hoeschele et al. (2007); 15. García-Toraño et al. (2018); 16. Fasching et al. (1970); 17. Jaffey et al. (1971); 18. Abdelouas (2006); 19. Hu et al. (2010); 20. Signoriello et al. (2022); 21. Krawczyk et al. (2013); 22. Cheng et al. (2013); 23. Laing and Ferguson (1958); 24. Moghaddam-Banaem et al. (2012); 25. Matuszek and Sugihara (1961); 26. Kraushaar et al. (1956); 27. Sathoff et al. (1963); 28. Nairne et al. (2015); 29. Bailey et al. (2021).

containing organic or inorganic carbon (Hajdas et al., 2021). Isotopes are also vital for hydrological studies (assess groundwater contamination sources and mechanisms, study groundwater circulation, and more) (Nadaradjan et al., 2023). Some of these applications, particularly those in the medical field, may contribute increasingly to the discharge of radioactive wastes due to the continuous introduction of novel radioisotope medical applications (de Nardo et al., 2022; Xia et al., 2021), which may require greater isotope production from non-power reactors for clinical trials and medical use.

While the radioactive wastes stemming from reactor operations are projected to rise, historical discharges to the environment, including accidents like nuclear power plant crises and non-accident events such as nuclear weapon development and testing during the Cold War, continue to accumulate in the surroundings. Steinhauser et al. (2013) conducted a study indicating that within one year of the Fukushima

Daiichi Nuclear accident 2011, soil located 0.88 km from the nuclear power plant (NPP) showed a maximum of 1,790,000 Bq/kg of cesium-137 (Cs-137) and 1,070 Bq/kg of strontium-90 (Sr-90), while ground 4.3 km from the plant exhibited 2,740,000 Bq/kg of Cs-137 and 232 Bq/kg of Sr-90. The Chernobyl NPP accident, one of the most severe NPP incidents to date, led to soil contamination of 1,239,000 Bq/kg of Cs-137 and 420,000 Bq/kg of Sr-90 at a distance of 4 km from the NPP, and 74,000 Bq/kg of Cs-137 and 36,000 Bq/kg of Sr-90 at a distance of 5 km from the plant (measurements taken within ten years after the incident) (Konoplyova et al., 1993; Malek et al., 2002; Victorova et al., 2000). The Cs-137 and Sr-90 detected in these contaminated areas were about 3.2×10^5 to 1×10^6 times and 11 to 1.5×10^3 times more concentrated than those measured in non-radioactive-contaminated soils. To illustrate, 2.7 Bq/kg Cs-137 as measured in Al-Negila, Egypt, 5.32 Bq/kg Cs-137 as measured in Kizilirmak Delta, Turkey, 5.67 Bq/kg

Cs-137 as measured in Jeju Island, Korea, and 0.7–20.9 Bq/kg Sr-90 as measured in Lithuania's unspecified national parks (Ariman and Gümüş, 2018; Gudeliënė et al., 2006; Kang et al., 2020; Monged et al., 2022). Additionally, Crowley and Ahearne (2002) reported that the United States (U.S.)'s nuclear weapon production from the Second World War until the Soviet Union's dissolution may have resulted in the contamination of approximately 29 million m³ of soil and sediment and 4.7 billion m³ of surface and groundwater.

Radioactive wastes pose significant risks to human health, both upon acute and chronic exposure. Acute radiation exposure (dose threshold: ≥ 1000 mSv) can cause severe deoxyribonucleic acid (DNA) damage, leading to cell death and dysfunction of tissues or organs when numerous distortions remain unrepaired or are misrepaired and accumulate in cells (Anzai et al., 2011; World Health Organization [WHO], 2016). On the other hand, chronic radiation exposure may result in malignancies like leukemia, often manifesting years or even decades after the exposure (Anzai et al., 2011). Chronic exposure to radiation from radioactive wastes may also negatively impact wildlife, leading to increased oxidative stress (Einor et al., 2016), disruption of DNA structure, chromosomal abnormalities (Lourenço et al., 2016), and molecular-level mutations (Møller and Mousseau, 2015). Birds living in areas contaminated by the Chernobyl fallout also reported having reproductive problems such as aspermy and sperm motility decrement (Møller et al., 2014). Furthermore, Geras'kin et al. (2008) linked increased radioactivity in habitats to changes in community structure and loss of biodiversity. Given these adverse consequences, proper management and remediation of radioactive wastes are crucial to minimize associated health and environmental hazards.

Radioactive wastes can undergo various treatment methods, such as physical, chemical, electrochemical, or biological means. Physical remediation options include soil washing (Kim et al., 2007), evaporation (Deng et al., 2022; Hou et al., 2022; Yu et al., 2020), membrane separation (Chen et al., 2021; Liu et al., 2019; Yu et al., 2018), and others. Chemical treatment involves processes like ion sorption by natural or engineered adsorbents (Wu et al., 2022; Zhang et al., 2021) and precipitation (Jiao et al., 2021; Ouyang et al., 2019). In electrochemical remediation, radioactive wastewater is subjected to electrochemical mineralization (Lv et al., 2021), electrosorption (Yu et al., 2022), or other processes to remove the radionuclides. While many studies have been conducted to develop novel or improve existing physical and chemical remediation approaches for radioactive waste treatment, they have certain limitations. Physical remediation performance can be influenced by fouling, and extra resources are required to maintain the system (Liu et al., 2020). A sophisticated setup may also be involved (Hou et al., 2022) and, therefore, can be costly. Natural chemical adsorbents often exhibit low adsorption capacities, slow kinetics, and poor stability (Sheng et al., 2017), while engineered adsorbents, with higher capacities, can be expensive (Kadadou et al., 2023). Electrochemical remediation, when applied to highly contaminated sites, may result in high energy costs due to its performance dependence on the applied voltage (Adeola et al., 2022). In contrast, bioremediation is considered an effective, economical, eco-friendly, and easy-processing approach for the removal of radioactive waste (Kadadou et al., 2023).

Among the various forms of bioremediation (bacterial remediation, mycoremediation, phycoremediation, and phytoremediation) (Manobala et al., 2021; Saleh et al., 2017; Song et al., 2019; Yuan et al., 2022), bacterial, myco-, and phycoremediation have captured the attention of researchers. Unlike phytoremediation, these methods do not necessitate additional safeguards against animals that might consume the plants and transfer radionuclides assimilated in the plants through the food chain (Kadadou et al., 2023). Continuous efforts have resulted in numerous studies aiming to identify novel microbial strains capable of remediating radioactive wastes while tolerating the radiotoxicity and chemotoxicity associated with contaminated environments. Additionally, researchers have explored introducing novel radionuclide remediation abilities into existing microbial strains using recombinant

techniques. Some researchers are also intrigued by the potential of co-applying microbes with other remediation methods, such as adsorption by natural adsorbents (Akhtar et al., 2009; Bai et al., 2014; Ding et al., 2019a; Kolhe et al., 2020; Wang et al., 2010), to further enhance remediation efficiency. Nonetheless, there remains a scarcity of comprehensive systematic reviews that focus on microbe-mediated radioactive waste remediations and their recent advances in applications for radioactive waste remediation.

Accordingly, this article aims to report findings on microbe-mediated bioremediation approaches, including bacterial, myco-, and phycoremediation, and delve into the mechanisms involved and the factors influencing their remediation performance. Additionally, a SWOT analysis will be conducted to assess the advantages, limitations, opportunities, and threats associated with microbe-mediated remediation. Prior to the bioremediation discussion, a summary will be provided on the sources and classification of radioactive waste, as well as an overview of the physical, chemical, and electrochemical treatments used to manage radioactive waste.

2. Radioactive wastes: classification and sources

The International Atomic Energy Agency (IAEA) (2009) categorized radioactive wastes into six classes: exempt waste (EW), very short-lived waste (VSLW), very low-level waste (VLLW), low-level waste (LLW), intermediate-level waste (ILW), and high-level waste (HLW), where each class differs in terms of the activity content (total activity, specific activity of a radionuclide, or activity concentrations present per volume of radioactive waste) of the waste. Radionuclides are the primary components within the radioactive wastes contributing to the wastes' radioactivity. Table 1 summarizes the radionuclides commonly found in radioactive wastes from various sources and their physical half-lives.

Exempt waste contains concentrations of radionuclides that are too low to harm human health or the environment. Consequently, its disposal does not require regulatory control, irrespective of whether it is recycled or buried in conventional landfills (IAEA, 2009). The waste may constitute materials resulting from the demolition of nuclear installations during decommissioning, like pipes and concrete (Darda et al., 2021), as well as naturally occurring radioactive materials (NORM) containing wastes generated by various industries, including aluminium production from bauxite, fertilizer production from phosphate ore, and hard coal mining (Leopold and Wiegand, 2008). The activity concentrations of radionuclides in these wastes fall below the thresholds set by the IAEA (2014), typically ranging from 100 to 1×10^7 Bq/kg depending on the radionuclide. For example: 1) red sludge from aluminium oxide extraction from bauxite, containing radium-226 (Ra-226) at 190 Bq/kg and lead-210 (Pb-210) at 330 Bq/kg (Tulcea, Romania), 2) phosphogypsum, a byproduct of phosphate ore processing for fertilizer production, with less than 200 Bq/kg of activity from radionuclides in both the uranium and thorium decay series (Belgium), and 3) surface sediments at sites where radionuclide-rich groundwater is pumped during underground coal mining, with Ra-226 and Pb-210 concentrations below 1,000 Bq/kg (Ruhr, Germany) (Leopold and Wiegand, 2008), can be classified as exempt wastes.

Very short-lived waste (VSLW) comprises radioactive waste containing radionuclides with very short half-lives (typically up to a few years) and activity concentrations higher than the clearance levels established by IAEA (IAEA, 2009). Due to the potentially harmful effects of VSLW's high activity concentration, it is mandatory to store such waste until the radionuclides decay to a point where their activities fall below the clearance level. At that stage, the resulting low-activity waste can be managed as exempt waste. VSLW is commonly associated with 'monoisotopic' radioactive waste originating from medical, industrial, or research applications (Burcl, 2013; Darda et al., 2021). For example, any wastes contaminated by cobalt-60 and iridium-192, which have half-lives of 5.27 years (Ma and Hooda, 2010) and 74.2 days (Turrel and Koblik, 1983), respectively, during cervix cancer brachytherapy

Table 2
Various physical remediation methods for treating radioactively contaminated soil and other solid wastes.

Physical Remediation Methods	Target Radionuclide	Parameter	Outcome
(A) Detector-based separation^{1,2} <ul style="list-style-type: none"> The excavated soil is transported to a chamber containing a detector to measure its radioactivity level, and the soil is separated via segregation gates based on the detector's readings and preset threshold radioactivity. Ultimately, the more radioactive parts of the soil are segregated from those less radioactive. 	U	Threshold gamma activity for separation: 30 pCi/g	96.8% volume reduction ³
(B) Soil washing by particle size-based separation⁴ <ul style="list-style-type: none"> Finer soil particles, i.e., silts and clays, hold higher concentrations of radionuclides as they have a significantly greater surface area for radionuclide absorption than larger particles of the same volume. Clay minerals also pose a high cation exchange capacity. Thus, they can adsorb more cationic radionuclides. Water action is applied to deagglomerate and wash the finer particles from the larger ones. 	Cs-137	Particles of size <200 mesh (<75 μm) are classified as silt and clay fraction	The silt and clay fraction (~25.0% of raw soil) separated contained 50.0% of the total Cs-137 activity in the raw soil ⁵
(C) Soil washing by magnetic separation^{6,7} <ul style="list-style-type: none"> The contaminated soil is mixed with water. The slurry formed is subjected to a magnetized medium, e.g., polyethylenimine (PEI)-coated magnetite (Fe₃O₄) nanoparticles or ferromagnetic filter wires, which can adsorb negatively-charged clay minerals from other soil particles. An external magnetic field is applied to separate the magnetized medium-clay mineral complexes from the treated soil. 	Cs-137	<ul style="list-style-type: none"> PEI-coated Fe₃O₄ nanoparticles with inclusion rate: 1.2% of the contaminated soil (mass ratio) Mesh filter (0.075 mm) 	Separation of Cs-bound fine particles (<0.0750 mm) from the raw soil resulted in 89.6% radioactivity reduction ⁶
	Cs-137	<ul style="list-style-type: none"> Superconducting high gradient magnetic separation Ferromagnetic filter wire diameter: 0.10 mm 20–75 μm clay particles 	33.0% radioactivity reduction efficiency ⁷
(D) Soil washing by dense medium and gravitational separation^{8,9} <ul style="list-style-type: none"> The contaminated soil is mixed with a heavy liquid solution, e.g., sodium polytungstate (SPT) solution, of a specific density x and centrifuged. The pellet represents the soil fraction with a density $> x$ while the supernatant represents the soil fraction with a density $< x$. 	Cs-137	<ul style="list-style-type: none"> 1 g soil:30 mL SPT_{2.4 g/cm³} solution Centrifugation: 10 min at 3000 rpm 	Cs-137 concentration in the soil fraction with $>2.40 \text{ g/m}^3$ density was reduced by 12.5–74.4% of the bulk samples ⁸
	<ul style="list-style-type: none"> Mn-54 Fe-55 Co-60 Eu-152 Eu-154 	<ul style="list-style-type: none"> 1 g concrete powder:20 g SPT_{2.7 g/cm³} solution Centrifugation: 3 min at 300 g Feed sand particle size: 75–500 μm 	The concrete sand was separated to higher-density mafic minerals and lower-density felsic minerals, where the former held more radioisotopes. ⁹
(E) Soil washing by flotation separation^{10,11} <ul style="list-style-type: none"> A surfactant, e.g., ethylhexadecyldimethyl-ammonium-bromide (EDAB) and hydrophobic silica (HPOS) nanoparticles, is added to render the fine soil particles hydrophobic. The surfactant-soil mixture is added to a container filled with water and air bubbles, where the bubbles are generated by adding frother, e.g., methyl isobutyl carbinol (MIBC), etc. The hydrophobic particles get attached to the bubbles and floated to the water surface. 	Cs-137	<ul style="list-style-type: none"> 0.18 mM EDAB 116.67 μL MIBC/g solids 	About 75.0% recovery efficiency of Cs-contaminated clay particles from a mixture of Cs-contaminated and pristine clays was achieved ¹⁰
	Cs-137	0.04 g HPOS nanoparticles/g soil	99.5% separation efficiency of fine silt and clay from a Cs-contaminated soil containing sand, clay, and silt ¹¹

^aThe superscripts of numbers are in-text citations, corresponding to: 1. Bayliss and Langley (2003); 2. Roybal et al. (1998); 3. Cummings and Booth (1996); 4. Eagle et al. (1993); 5. Anderson et al. (1999); 6. Kim et al. (2020b); 7. Nishimoto et al. (2021); 8. Yamasaki et al. (2022); 9. Hong and Um (2023); 10. Zhang et al. (2019); 11. Kim et al. (2021a).

(Srivastava et al., 2022), can be managed as VSLW.

Very low-level waste (VLLW) refers to radioactive waste with activity concentrations slightly exceeding the clearance levels (IAEA, 2009), which are typically around a few 10,000 Bq per kilogram of waste (Bonin, 2010). Generally, VLLW contains very few amounts of long-lived radionuclides (Burel, 2013). Due to its relatively low hazard, VLLW can be disposed of at surface landfill facilities, and there is no requirement for high-level confinement (IAEA, 2009). A significant volume of VLLW is generated during the operation, maintenance, and decommissioning of nuclear facilities, taking the form of materials such as rubble, concrete, soil, steel, thermal insulators, and others (Burel, 2013; Darda et al., 2021; IAEA, 2009). Additionally, mining and processing of minerals or ores, such as uranium mining and milling, also produce VLLW in the form of fine-grained slurry mine tailings (Brun-Yaba et al., 1996; Harpy et al., 2020; Ramadan et al., 2022). To illustrate, mine tailings generated by uranium ore processing sites in Allouga, Egypt, contained uranium-238 and radium-226 with a mean activity concentration of 2071.8 Bq/kg and 6783.63 Bq/kg, respectively (Harpy et al., 2020), exceeding the clearance level (1,000 Bq/kg) for naturally-occurring radionuclides (IAEA, 2014).

Low-level waste (LLW) encompasses a wide range of radioactive wastes, spanning from wastes with activity concentrations just above those of VLLW to wastes with significantly higher radioactivity concentrations, with the upper limit set at 3.7×10^8 Bq/kg (Bonin, 2010). These wastes may consist of short-lived radionuclides with high radioactivity concentrations, long-lived radionuclides with low activity contents, or a combination of both (IAEA, 2009). LLW is produced by various sources, including industries, hospitals, and the nuclear fuel cycle, in the form of injections, clothing, laboratory animal carcasses and tissues, filters, reactor water treatment residues, and others (Darda et al., 2021; IAEA, 2009). The U.S. Department of Energy (DOE) also reported producing LLW containing transuranic radioisotopes (radioisotopes of elements with an atomic number higher than that of uranium), primarily plutonium, with concentrations of 3.7×10^5 – 3.7×10^6 Bq/kg during isotope separation and enrichment, reactor operation, and others (Leroy et al., 2006). Typically, low-level wastes are stored in near-surface disposal facilities, which can vary in depth, reaching depths up to 30–100 m below the surface. The depth of containment is contingent upon the concentrations of short- and long-lived radionuclides present in the wastes (Darda et al., 2021; Forsström, 2012; IAEA,

Table 3
Chemical remediation of nuclear wastewater via adsorption and precipitation.

Chemical Remediation Methods	Target Radionuclide	Materials (Adsorbent/Chemical) with Parameter	Outcome
(A) Adsorption ¹⁻⁵ <ul style="list-style-type: none"> Natural or synthetic adsorbents are used to adsorb radionuclides from the wastewater. Radionuclide-adsorbent complexes can be separated from the treated wastewater by a permanent magnet, centrifugation, etc. Desorption agent(s) can be used to elute the radionuclides adsorbed. 	Sr(II)	Magnetic zeolite/Fe ⁰ <ul style="list-style-type: none"> 1:1 (w/w) zeolite:iron C_i = 200 mg/L Sr(II) Adsorbent dosage = 5 g/L pH 6.0 T = 25 °C t = 60 min 	Adsorption capacity = 32.83 mg/g ¹
	Sr(II)	Magnetic hexamethylene diamine tetramethylene phosphonic acid (HDTMP)-hydroxyapatite nanoparticles (HD-MHAP)	<ul style="list-style-type: none"> Adsorption capacity = 320.7 mg/g Removal efficiency = 80.20%²
	Co(II)	<ul style="list-style-type: none"> 10% HDTMP doping dosage C_i = 200 mg/L Sr(II)/Co(II) 25 mg adsorbent:50 mL Sr(II)/Co(II) pH 7.0 T = 25 °C t = 24 h 	<ul style="list-style-type: none"> Adsorption capacity = 172.3 mg/g Removal efficiency = 42.70%²
	U(VI)	Polyethylene polyamine/polydopamine modified carboxylated chitosan aerogel (PEPA/PDA-CMCS) <ul style="list-style-type: none"> 3.5% CMCS; 0.7 mg/mL PDA; 2.0% PEPA, 250 µL 30% glutaraldehyde Adsorbent dosage = 1 g/L Simulated nuclear feed liquid with 0.05 mmol/L for each ion: U(VI), La(III), Ni(II), Co(II), Gd(III), Pr(III), Ce(III) pH 3.0 	<ul style="list-style-type: none"> Saturated adsorption capacity = 467.7 mg/g 99.92% U(VI) concentration reduction in simulated nuclear wastewater³
	U(VI)	Phosphate-based hypercrosslinked polymer-dibenzyl phosphate (HCP-DBP) <ul style="list-style-type: none"> 1:1 aryl phosphate: crosslinking polymer C_i = 150 mg/L U(VI) Adsorbent dosage = 0.33 g/L pH 7.0 T = 40 °C 	Adsorption capacity = 302.0 mg/g ⁴
U(VI)	Amidoxime-based collagen fiber (AO-CF) <ul style="list-style-type: none"> C_i = 2.5 mmol/L U(VI) Adsorbent dosage = 0.8 g/L pH 5.0 T = 45 °C 	Adsorption capacity = 0.8770 mmol/g (234.2 mg/g) ⁵	
(B) Precipitation ⁶⁻⁸ <ul style="list-style-type: none"> A chemical is added to the radioactive wastewater to co-precipitate the targeted radionuclide. While both adsorption and precipitation may involve the same mechanism, i.e., adsorption, the former does not produce sludge or insoluble minerals like the latter. 	Ra-226	Barite (barium sulfate-containing mineral) <ul style="list-style-type: none"> Initial radioactivity level = 189 Bq T = 25 °C 	<ul style="list-style-type: none"> Ra-226 co-precipitated by barite = 184.0 Bq Ra-226 yield = 97.10%⁶
	Cs(I)	Nickel-potassium ferrocyanide (KNiFC) <ul style="list-style-type: none"> 1.33:1 Ni(NO₃)₂:K₄Fe(CN)₆ C_i = 1000 mg/L Cs(I) 0.20 g/0.30 g Ni(NO₃)₂ in 100 mL CsNO₃ pH 7.0 T = 25 °C 	Cs(I) removal rate ⁷ <ul style="list-style-type: none"> 0.20 g Ni(NO₃)₂ = -90.00% 0.30 g Ni(NO₃)₂ = -100.0%
	Cs(I) (Cs-137)	Aluminium-potassium ferrocyanide (AlKFC) <ul style="list-style-type: none"> 0.01 M K₄[Fe(CN)₆] with 8 g/L Al₂(SO₄)₃ Initial radioactivity level = 103807.44 Bq/L t = 7 days Steady stirring for 30 min 	<ul style="list-style-type: none"> Final radioactivity level = 21.50 Bq/L Removal efficiency = 99.98%⁸

a The superscripts of numbers are in-text citations, corresponding to: 1. Shubair et al. (2022); 2. Lu et al. (2023); 3. Huang et al. (2023); 4. Yue et al. (2023); 5. Tang et al. (2023); 6. Ouyang et al. (2019); 7. Jiao et al. (2021); 8. Sopapan et al. (2023).

2009). The robust containment of LLW, particularly those with higher activities, can last for several hundred years, usually at least 300 years, approximately ten times the half-lives of Sr-90 and Cs-137 (Forsström, 2012). Bonin (2010) assumed that such containment periods are sufficient for all the radioactivity, particularly those in LLW with no long-lived radionuclides, to disappear.

Radioactive wastes containing long-lived radionuclides with concentrations higher than those in LLW, possessing an overall higher radioactivity concentration than LLW, or having both characteristics, fall under the classification of intermediate-level waste (ILW) (Burcl, 2013; IAEA, 2009). While Bonin (2010) defined ILW as radioactive wastes with radioactivity ranging between 3.7×10^8 Bq/kg and 3.7×10^{11} Bq/kg, this range is not universally agreed upon and may vary

among disposal facilities (Burcl, 2013; IAEA, 2009). ILW may contain alpha radiation-emitting long-lived radionuclides that might not decay to an activity concentration level sufficiently low for near-surface disposal within the period when regulatory controls can be relied upon, given that the half-lives of these radionuclides may last up to millions of years (Bonin, 2010; Burcl, 2013). Due to the presence of such long-lived radionuclides, these wastes, generated in significant amounts during the reprocessing of nuclear power plants' spent fuel and primary nuclear waste treatment, necessitate a higher degree of containment and isolation from the environment and accidental human intrusions (Burcl, 2013). Disposal facilities for ILW are typically located tens to a few hundred meters below the surface, constructed in silos, caverns, or developed from drift mining into mountainsides (IAEA, 2011).

High-level wastes (HLW) consist of radionuclides with activity concentration levels (10^4 – 10^6 TBq/m³) that are sufficiently high to generate large amounts of heat (>2 kW/m³) through radioactive decay for up to centuries (Burcl, 2013; IAEA, 2009). In addition to emitting dangerous levels of heat, HLW also releases lethal radiation doses. According to the United States Nuclear Regulatory Commission [NRC] (2002), spent nuclear fuel, a typical form of HLW, emits radiation doses of more than 20,000 rem/h (200 Sv/hour) a meter away, even after its removal from the reactor for a decade. Such radiation doses are significantly greater than those required to cause instant incapacitation and death in humans upon exposure (500–5000 rem, equivalent to 5–50 Sv) (NRC, 2002; Speight, 2020). Furthermore, HLW may contain significant amounts of long-lived radionuclides; for instance, plutonium-239, a common transuranic radioisotope found in spent fuel, has a half-life of 24,000 years (NRC, 2019). Given these hazardous characteristics of HLW (including spent fuel, conditioned waste from spent fuel reprocessing, vitrified spent fuel, and others), HLW should be stored in geological formations that are at least several hundred meters below the surface, with additional cooling (IAEA, 2009).

These radioactive wastes with activity content surpassing clearance levels are stored in facilities. However, they may also end up in the environment due to unintentional discharges during NPP accidents, nuclear weapon development and testing (Hu et al., 2010), leakage from disposal facilities (Andersen et al., 2004), or be left at abandoned uranium and thorium ore mining and milling sites (Burcl, 2013). In any case, these wastes can be subjected to remediation efforts to mitigate the radio- and chemotoxicity of radionuclides.

3. Radioactive waste remediation: approaches

3.1. Physical remediation

Radionuclide-contaminated soil, particularly that originating from decommissioned nuclear power plant (NPP) sites, is frequently subjected to remediation through excavation. During this process, the contaminated soil is replaced with non-contaminated soil, and the excavated soil undergoes treatment to reduce its volume, remove radionuclides, or is disposed of at a radioactive waste disposal site (Yoon et al., 2021a). Various physical remediation methods, such as detector-based separation and soil washing utilizing particle separation techniques (size-based, gravity, magnetic, and flotation separation), are commonly employed to treat the excavated soil (Bayliss and Langley, 2003; Eagle et al., 1993; Kim et al., 2020b; Yamasaki et al., 2022; Zhang et al., 2019). Table 2 provides a description of the mentioned processes. Additionally, incineration can be utilized to treat low-level radioactive solid wastes (IAEA, 1992).

On the other hand, liquid radioactive waste, particularly liquid organic radioactive wastes (e.g., vacuum pump oils produced from nuclear research centers' activities, extraction solvents used during uranium extraction operations, and others), can also undergo incineration to achieve significant volume reduction. However, this process generates secondary wastes such as radioactive ash and volatile organic waste. Additional filtration and containment measures are necessary to prevent the ash from escaping into the environment and to control the formation of unconfined explosive gas and vapor mixtures when the volatiles encounter the atmosphere (IAEA, 1992). Jiao et al. (2023) established coprecipitation flotation with potassium nickel hexacyanoferrate as the Cs-137 co-precipitant and cetyltrimethylammonium bromide (CTAB) as the surfactant, achieving a high Cs(I) removal rate of up to 97.8% from 1.0 mg/L simulated radioactive wastewater at pH 7.0. Moreover, mechanical- and solar-driven evaporation processes for radioactive wastewater, including those with high salinities, have been investigated to reduce the wastewater volume (Deng et al., 2022; Hou et al., 2022; Yu et al., 2020). Furthermore, membrane separation technology has been studied to remove radionuclides from wastewater through mechanisms such as forward and

Table 4

Electrochemical processes involved in radioactive wastewater remediation.

Electrochemical Remediation Methods	Target Radionuclide	Electrodes Used and Parameter	Outcome
(A) Electrodialysis ¹	Co(II)Sr(II) Cs(I) [−]	Titanium coated with ruthenium • Cation-exchange membrane: CM-2; anion-exchange membrane: ASE • CoCl ₂ , SrCl ₂ , CsCl, and KI (1 mmol/L each) with 3.5 wt% NaCl in diluate chamber; deionized water in concentrate chamber • V = 6 V • Volume ratio of initial concentrate chamber solution to initial diluate chamber solution = 1:40	Max concentration times, C _{max} /decontamination efficiency, DE: 1 • Co(II): C _{max} = 9.50 × ; DE = 96.2% • Sr(II): C _{max} = 20.1 × ; DE = 97.6% • Cs(I): C _{max} = 9.90 × ; DE = 95.6% • I [−] : C _{max} = 32.5 × ; DE = 95.6% Overall wastewater volume reduction = 80.5%
(B) Electrosorption (capacitive deionization system) ^{2,3}	Cs(I)	Cathode: copper hexacyanoferrate Anode: porous carbon • V = 1.4 V • Feed solution: mixed solution with 100 mg/L of Cs(I), Sr(II), and Na(I) • pH 5.6	• Max electrosorption capacity of Cs (I) = 397 mg/g • High Cs(I) selectivity: Cs(I) (162 mg/g) > Sr (II) (33.4 mg/g) > Na(I) (10.7 mg/g) 2
• Comprises of two electrodes in contact with wastewater to be treated. • Voltage is applied to generate an electrical potential difference across the electrodes and drive the migration of ions in the wastewater to electrodes.	Sr(II)	Cathode: aryl diazonium salt (ADS) modified porous activated carbon (SPAC) Anode: porous activated carbon • V = 1.2 V • Feed solution: 30 mg/L SrCl ₂	Sr(II) electrosorption capacity: 3 SPAC (33.1 mg/g) > porous activated carbon (16.1 mg/g) > cheese-like activated carbon (10.2 mg/g)
(C) Electrochemical mineralization ⁴	U(VI)	Cathode: graphite Anode: iron • V = 36 V • Feed solution: pH 2.78 simulated uranium-containing wastewater • Uranium separation from iron: heat the precipitate at >300 °C for 4 h, and add 60 g/L sulfuric acid to the cooled product and stand for 2 h at 25 °C	>80.0% uranium was recovered as triuranium octoxide (U ₃ O ₈) ⁴
• The targeted radionuclide is concentrated and co-precipitated with iron as magnetite at the cathode. • The radionuclide can be separated from the iron by heating at high temperature to form solid oxides, where the oxides are dissolved by acids while the iron remains insoluble.			

a The superscripts of numbers are in-text citations, corresponding to: 1. Li et al. (2022); 2. Lee et al. (2022); 3. Xiang et al. (2022); 4. Lv et al. (2021).

reverse osmosis (Chen et al., 2021; Liu et al., 2019).

3.2. Chemical remediation

Various chemical remediation methods are available for the treatment of radioactive wastewater, including adsorption by natural or synthetic adsorbents and precipitation (Kadadou et al., 2023). Table 3 provides examples of adsorbents and chemicals used in recent studies for the adsorption and precipitation processes. Wet oxidation is another applied technique, where iron (or metals like copper and manganese) and hydrogen peroxide react to produce radicals with an extremely high oxidation potential, and these radicals are then employed to oxidize organic contaminants, decomposing organic radioactive wastes such as used ion-exchange resins and decontamination liquids containing organic chelating agents from nuclear power plants, spent nuclear fuel reprocessing solvents, and others (Walling et al., 2021). Moreover, volume reduction of bulky radioactive combustible wastes can be achieved through acid digestion using concentrated strong acids like sulfuric acid at high temperatures, along with an oxidizing agent such as hydrogen peroxide or nitric acid, where this process breaks down the chemical bonding of the contaminants (Kobayashi et al., 1980).

3.3. Electrochemical remediation

Radioactive waste can be remediated through various electrochemical processes, such as electrodialysis, electrosorption (or capacitive deionization), and electrochemical mineralization (Li et al., 2022; Lv et al., 2021; Xiang et al., 2022). While all mentioned methods involve a pair of electrodes in their setup, the way radionuclides get separated from the radioactively polluted water varies. An electrodialysis setup contains alternating anion- and cation-exchange membranes between the anode and cathode. When an electrical current is applied to drive the electrodiffusion of ions in the radioactive water across the membranes, the radionuclides get trapped in concentrate chambers between the alternating cation- and anion-exchange membranes (Li et al., 2022). In contrast, rather than being accumulated in concentrate chambers, positively-charged radionuclides are adsorbed onto the negatively-charged cathode and vice versa for anionic radionuclides in electrosorption (Lee et al., 2022; Xiang et al., 2022). As for electrochemical mineralization, radionuclides in the wastewater are accumulated at the cathode and co-precipitated with another metal (Lv et al., 2021). Table 4 provides setups of these remediation methods and their respective efficiency. Additionally, electrochemical treatment can also be applied to radioactively contaminated soil (Bayliss and Langley, 2003).

4. Bioremediation of radioactive waste

Biological approaches for radioactive waste treatment involve the use of microorganisms, such as bacteria, fungi (mycoremediation), algae (phycoremediation) or plants (phytoremediation) to remediate radionuclide-contaminated wastewater or soil (Gul et al., 2022; Manobala et al., 2021; Saleh et al., 2017; Song et al., 2019; Yuan et al., 2022).

4.1. Bacterial remediation

Bacteria can remediate radioactive wastes via various microbial processes, including bioreduction, biosorption, and bioprecipitation (biomineralization).

4.1.1. Bioreduction

Certain radionuclides, such as uranium (U), chromium (Cr), technetium (Tc), and neptunium (Np), can exist in multiple oxidation states. Their oxidized forms (U: U(VI); Cr: Cr(VI); Tc: Tc(VII); Np: Np(V)) are relatively water-soluble, making them mobile in natural water systems like groundwater when discharged into the environment, compared to their reduced forms (U: U(IV); Cr: Cr(III); Tc: Tc(IV); Np: Np(IV)) (Roh et al., 2015; Tomaszewski et al., 2017). The enzymatic reduction of soluble radionuclides to insoluble forms by certain bacteria offers a promising approach for radionuclide immobilization. This process restricts the movement of radionuclides in water bodies, minimizing their exposure to living organisms. Bioreduction activities have been observed in various bacterial genera, including dissimilatory ferric iron-reducing bacteria (e.g., *Geobacter* spp. and *Shewanella* spp.) (Jeon et al., 2004; Lloyd et al., 2003; Marshall et al., 2006; Orellana et al., 2013; Renshaw et al., 2005), sulfate-reducing bacteria (e.g., *Desulfovibrio* spp. and *Desulfosporosinus* spp.) (de Luca et al., 2001; Payne et al., 2002; Rittmann et al., 2002; Suzuki et al., 2004), anaerobic extremophiles (e.g., thermophilic *Thermoanaerobacter* sp. and haloalkaliphilic *Halomonas* sp.) (Khijniak et al., 2003; Madden et al., 2012), and many others (Table 5).

Electron donors play a crucial role in both direct and indirect radionuclide bioreduction (Fig. 1), and different electron donors supplied might affect bacterial radionuclide reduction differently. De Luca et al. (2001) reported that *Desulfovibrio fructosovorans* achieved significantly higher Tc(VII) removal (92%) when hydrogen (H₂) was supplied as the electron donor compared to organic electron donors like pyruvate, lactate, fructose, formate, and fumarate (3–26% removal). However, while organic electron donors did not efficiently support radionuclide removal in *D. fructosovorans*, they proved effective in driving satisfactory radionuclide bioreduction in other bacteria. For instance, Marshall et al. (2009) found that both H₂ and acetate could drive U(VI) reduction in *Anaeromyxobacter dehalogenans* (with over 90% removal for both electron donors). However, the acetate-driven U(VI) reduction had slower rates than the H₂-driven counterpart. This rate difference might be attributed to the subcellular localization of enzymes catalyzing the process (H₂-oxidizing hydrogenase located in the periplasm, whereas acetate-oxidizing dehydrogenase or acetyltransferase situated in the cytoplasm) (Marshall et al., 2009) or the variation in free energy (ΔG°) produced during the oxidation of the electron donor (H₂ oxidation generating greater ΔG° than acetate oxidation, and the additional energy was sufficient to fuel subsequent U(VI) reduction) (Wu et al., 2006a). Apart from electron donors, electron shuttling agents like anthraquinone-2,6-disulfonate (AQDS) can also facilitate enzymatic radionuclide reduction. AQDS may aid in electron transfer from metal-reducing bacteria to radionuclides adsorbed to solid surfaces, particularly those in micropore form that may be enzymatically inaccessible, thereby facilitating radionuclide reduction (Jeon et al., 2004).

Following the initial studies on the roles of various electron donors in radionuclide reduction, in situ biostimulations have been carried out at radioactively contaminated sites. Biostimulation, as defined by North et al. (2004), involves adding exogenous nutrients (such as H₂ or organic electron donors in the case of bacterial radionuclide bioreduction) to enhance the population or activity of native microbial communities for bioremediation purposes. Williams et al. (2011) observed rapid U(VI) removal from radioactively contaminated groundwater at the United States Department of Energy (DOE) Integrated Field Research Challenge (IFRC) site within 2–10 days after adding acetate to the oxygenation-minimized groundwater. The U(VI) concentration decreased from 0.8 to 1.2 μM to levels below the dictated uranium mill tailing remedial action (UMTRA) limit (0.18 μM) in all monitoring wells, and certain wells reached levels below 0.126 μM , which is the United

Table 5

Performance of bacteria species in remediating radionuclides and heavy metals associated with radioactive wastes.

Bacteria Species	Target Contaminant	Mechanism	Parameter	Performance	Reference
<i>Microbacterium testaceum</i> (MT), <i>Bacillus coagulans</i> (BC), <i>Cellulosimicrobium cellulans</i> (CC)	Ce(III) Co(II)	Biosorption	pH 4.5, 25 °C, 2-h incubation with 2.8 mmol/L bacterial biomass	MT: 68.1 BC: 55.1 CC: 73.8 MT: 49.6 BC: 49.2 CC: 34.1	Elgarahy et al. (2023)
<i>Microbacterium</i> sp. Be9	U(VI)	Biosorption and bioprecipitation	28 °C, 48-h incubation, no exogenous phosphates	88.0*	Martínez-Rodríguez et al. (2023)
<i>Bacillus thuringiensis</i> X-27	U(VI)	Bioprecipitation	pH 5.0, 30 °C, 96-h incubation	96.4*, 29.5	Zhu et al. (2023)
<i>Pseudomonas fluorescens</i>	U(VI)	Biosorption and bioprecipitation	30 °C, 50-h incubation	79.2-97.8*	Zheng et al. (2022)
<i>Kocuria rosea</i>	U(VI)	Biosorption and bioprecipitation	pH 5.0, 30 °C, 6-h incubation	~90.0*	Zhou et al. (2022)
<i>Bacillus</i> sp. ZJ-3	U(VI)	Bioprecipitation	pH 7.0, 30 °C, 108-h incubation with 10 mM glycerol-3-phosphate	~80.0*	Zhong et al. (2021)
<i>Shewanella putrefaciens</i>	U(VI)	Biosorption, bioprecipitation, and bioreduction	pH 7.0, 30 °C, 48-h incubation, anoxic	92.3*	Huang et al. (2020)
<i>Pseudomonas aeruginosa</i>	Cr(VI)	Biosorption and bioreduction	pH 5.5, 45 °C, 108-h incubation with 4.0 g/L glucose	100*	Li et al. (2020)
<i>Stenotrophomonas</i> sp. Br8 CECT 9810	U(VI)	Bioprecipitation and biosorption	pH 6.3, 28 °C, 48-h incubation with 5 mM glucose-2-phosphate	94.7*, 373	Sánchez-Castro et al. (2020)
<i>Streptomyces</i> sp. CuOff24	Sr(II)	Biosorption	RT, 12-h incubation with 1 g/L extracellular polymeric substance	92.0*	Kamala et al. (2019)
<i>Bacillus</i> sp. dw-2	U(VI)	Bioprecipitation	pH 7.0, 48-h incubation with 20 mM sodium glycerophosphate	~93.0*	Tu et al. (2019)
<i>Kocuria</i> sp.	U(VI)	Biosorption and bioprecipitation	pH 5.0, 25 °C, 4-h incubation with 20 mg/L initial U concentration	98.0*	Wang et al. (2019)
<i>Deinococcus radiodurans</i> R1	I ⁻	Biosorption	1-h incubation in synthetic urine	>99.0*	Choi et al. (2017)
<i>Pseudomonas aeruginosa</i> PAO1	Cs(I)	Biosorption	pH 6.0, 30 °C, 24-h incubation	76.1*	Kang et al. (2017)
<i>Acidovorax facilis</i>	U(VI)	Biosorption and bioprecipitation	pH 5.0, 24-h incubation with 0.1 mM initial U concentration	~100*, 140	Gerber et al. (2016)
<i>Bacillus</i> sp. dwc-2	U(VI)	Biosorption and bioprecipitation	pH 3.76, 12.69-h incubation with 8.90 g/L biomass dosage and 9.83 mg/L initial U concentration	86.4*	Zhao et al. (2016)
<i>Microvirga aerilata</i> LM	Pb(II)	Bioprecipitation	5-h incubation on ice, 50 Gy/h γ -radiation	~70.0*	Luo et al. (2014)
<i>Geobacter sulfurreducens</i>	U(VI)	Bioreduction and bioprecipitation	30 °C, 4-h incubation with 5 mM acetate, anoxic	83.0*	Orellana et al. (2013)
<i>Halomonas</i> sp. SR4	Sr(II)	Bioprecipitation	25 °C, 7-d incubation with 100 mg/L initial Sr(II)	~80.0*	Achal et al. (2012)
<i>Thermoanaerobacter</i> sp. TOR-39	U(VI)	Bioreduction and bioprecipitation	pH 7.8, 65 °C, 20-d incubation followed by storage at RT anaerobically for 3 y	96.4-100*	Madden et al. (2012)
<i>Anaeromyxobacter dehalogenans</i> 2CP-C	U(VI)	Bioreduction and bioprecipitation	pH 7.0, 30 °C, 72-h incubation with H ₂ /10 mM acetate, anoxic	~93.0-100*	Marshall et al. (2009)
	Tc(VII)		pH 7.0, 30 °C, 144-h incubation with H ₂ /10 mM acetate, anoxic	~95.0-99.0*	
<i>Shewanella oneidensis</i> MR-1	U(VI)	Bioreduction and bioprecipitation	30 °C, 48-h incubation with 10 mM lactate, anoxic	~90.0*	Marshall et al. (2006)
<i>Thermoterrabacterium ferrireducens</i>	U(VI)	Bioreduction and bioprecipitation	pH 7.0, 65 °C, 68-h incubation with 3 mL/L glycerol and 0.2 g/L yeast extract, anoxic	80.0-90.0*	Khijniak et al. (2005)
<i>Geobacter sulfurreducens</i>	U(VI)	Bioreduction and bioprecipitation	pH 7.0, 30 °C, 24-h incubation with 10 mM acetate, anoxic	100*	Renshaw et al. (2005)
<i>Geobacter sulfurreducens</i>	U(VI)	Bioreduction	pH 6.8, 22 °C, 30-d incubation with 0.1 mM AQDS added at 16 d, anoxic	~91.0*	Jeon et al. (2004)
<i>Salmonella subterranea</i>	U(VI)	Bioreduction	pH 6.8, 4-h incubation with 20 mM acetate, anoxic	0.140 [#]	Shelobolina et al. (2004)
<i>Arthrobacter</i> (98 % <i>A. ilicis</i>)	U(VI)	Bioprecipitation	pH 4.0, 1-h incubation	36.9	Suzuki & Banfield (2004)
<i>Arthrobacter</i> (99 % <i>A. ramosus</i>)	U(VI)	Bioprecipitation		41.0	
<i>Desulfosporosinus</i> spp. (strain DSM765 and P3)	U(VI)	Bioreduction and bioprecipitation	pH 7.0, 24-h incubation without bicarbonate ion and sodium chloride, anoxic	> 90.0*	Suzuki et al. (2004)
<i>Halomonas</i> spp.	Tc(VII)	Bioreduction and bioprecipitation	pH 10.0, 30 °C, 2-month incubation with acetate, lactate, formate, ethanol, methanol (10 mM each), anoxic	80.0*	Khijniak et al. (2003)
<i>Geobacter sulfurreducens</i>	U(VI)	Bioreduction	30 °C, anoxic, with H ₂ /20 mM acetate	0.560-0.570 [#]	Lloyd et al. (2003)
<i>Desulfovibrio desulfuricans</i> G20	U(VI)	Bioreduction	pH 7.0, 2.5-h incubation with H ₂ /10 mM (pyruvate/lactate), anoxic	1.50-2.20 [#]	Payne et al. (2002)
<i>Desulfovibrio</i> consortium (<i>D. desulfuricans</i> , <i>D. gigas</i> , <i>D. vulgaris</i> -resembled strain)	Np(V)	Bioreduction and bioprecipitation	25 °C, 17-d incubation with 12.3 mM H ₂ added 4 d after Np addition, anoxic	~99.0*	Rittmann et al. (2002)
<i>Pseudomonas fluorescens</i>	Np(V)	Biosorption	pH 7.0, 25 °C, 4-h incubation	15.0-20.0*	Songkasiri et al. (2002)

(continued on next page)

Table 5 (continued)

Bacteria Species	Target Contaminant	Mechanism	Parameter	Performance	Reference
<i>Desulfovibrio fructosovorans</i>	Tc(VII)	Bioreduction and bioprecipitation	pH 5.5, 30 °C, 24-h incubation with 20 min H ₂ supply, anoxic	92.0*	de Luca et al. (2001)
<i>Microbacterium flavescens</i> JG9	Pu(IV)	Intracellular bioaccumulation (no precipitation)	21-23 °C, 10-h incubation	2.50 × 10 ⁻⁴ #	John et al. (2001)
<i>Deinococcus radiodurans</i> R1	Tc(VII)	Bioreduction	21-d incubation with 0.1 mM AQDS, anoxic	82.5-98.4*	Fredrickson et al. (2000)
	U(VI)			88.7-97.9*	
	Cr(VI)			72.0*	

^a Abbreviations: RT: room temperature; AQDS: anthraquinone-2,6-disulfonate; H₂: hydrogen gas.

^b Symbol representing each unit for different expressions of bacteria's performance:

* Percentage removal (%)

Rate of reduction (mmol/h/g dry weight biomass)

Rate of reduction (μM/mg protein/min)

˘ Biosorption/bioprecipitation capacity (mg/g dry biomass)

˘ Biosorption/bioprecipitation capacity (mmol/g dry weight)

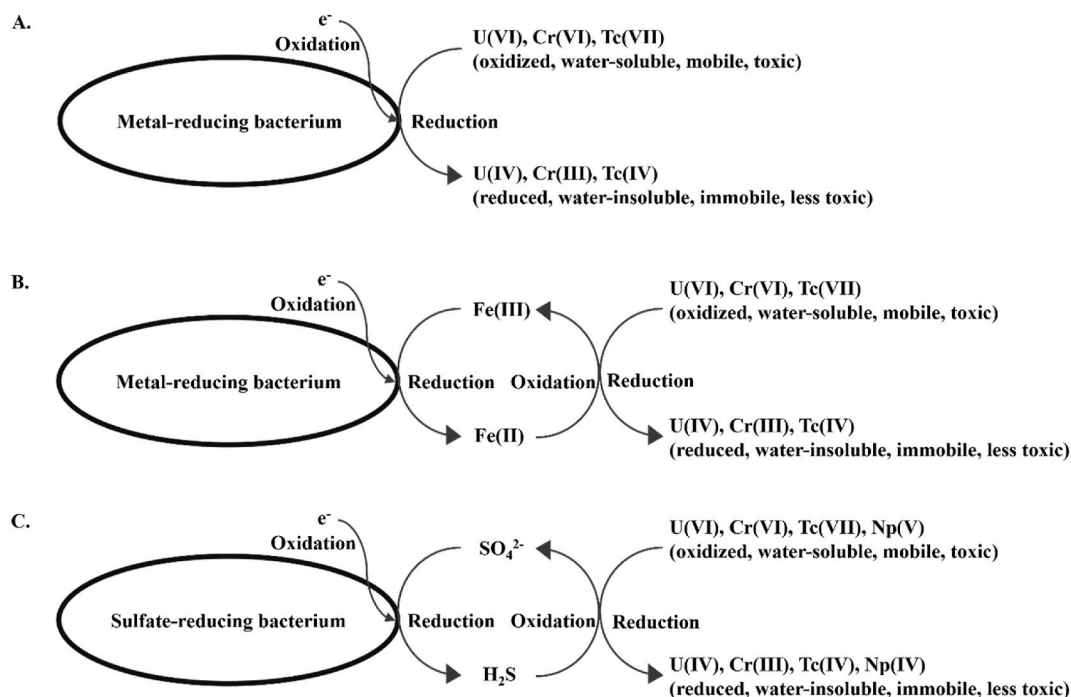


Fig. 1. Summary of radionuclide bioreduction pathways driven by metal- and sulfate-reducing bacteria: (A) Direct bioreduction of radionuclides by metal-reducing bacteria; (B and C) Indirect bioreduction of radionuclides with (B) metal-reducing and (C) sulfate-reducing bacteria (Adapted from Kumar et al., 2007).

States Environmental Protection Agency (EPA) prescribed drinking water standard (Williams et al., 2011). The low U(VI) levels could be maintained as long as acetate was supplied. Anderson et al. (2003) also observed a reduction in U(VI) levels below the UMTRA limit within 50 days after injecting acetate into neutral U-contaminated groundwater at a DOE site. Both studies resulted in *Geobacter* population enrichment. Istok et al. (2004) successfully stimulated U(VI), Tc(VII), and nitrate (NO₃⁻) reduction in an acidic (pH 3.3–6.8) aquifer co-contaminated with NO₃⁻ by enriching the indigenous metal-reducing *Geobacter*. However, U (VI) reduction required two or more electron donors (acetate, glucose, and ethanol) to be initiated.

In addition to in situ remediation, ex situ biostimulation can be conducted to identify indigenous bacterial species with radionuclide bioreduction capabilities. Safonov et al. (2018) identified various denitrifying bacteria (*Rhodanobacter*, *Acidovorax*, *Thermomonas*, *Rhizobium*, *Pseudomonas*, *Brevundimonas*, *Ensifer*, and *Simplicispira*) that can remove NO₃⁻ from radioactive groundwater co-contaminated with NO₃⁻ by adding acetate, sucrose, or milk whey to the groundwater samples,

and suggested that denitrification should precede radionuclide reduction since NO₃⁻ appeared to be preferentially reduced over U(VI). Some observations showed that U(VI) reduction did not occur in sediments contaminated with both U(VI) and NO₃⁻ until the NO₃⁻ was removed (Elias et al., 2003; Wu et al., 2006b). Suzuki et al. (2003) collected U-contaminated water and sediment from an inactive open-pit U mine and incubated the samples anaerobically with organic compounds. As a result, indigenous sulfate-reducing *Desulfohalobium* and fermentative *Clostridium* dominated the microbial populations in the incubated samples and removed approximately 98.5% of U(VI) within a month. Furthermore, anaerobic incubation of highly saline (10 × more saline than typical seawater) U-contaminated groundwater, collected from an aquifer at a U mine tailings site along with acetate led to the enrichment of bacteria closely related to *Desulfohalobium* and *Pseudomonas* that are possibly salt-tolerant species from these genera that can reduce U(VI) (Nevin et al., 2003). This discovery highlights the potential role of bacteria in remediating radioactive wastewater under harsh abiotic conditions.

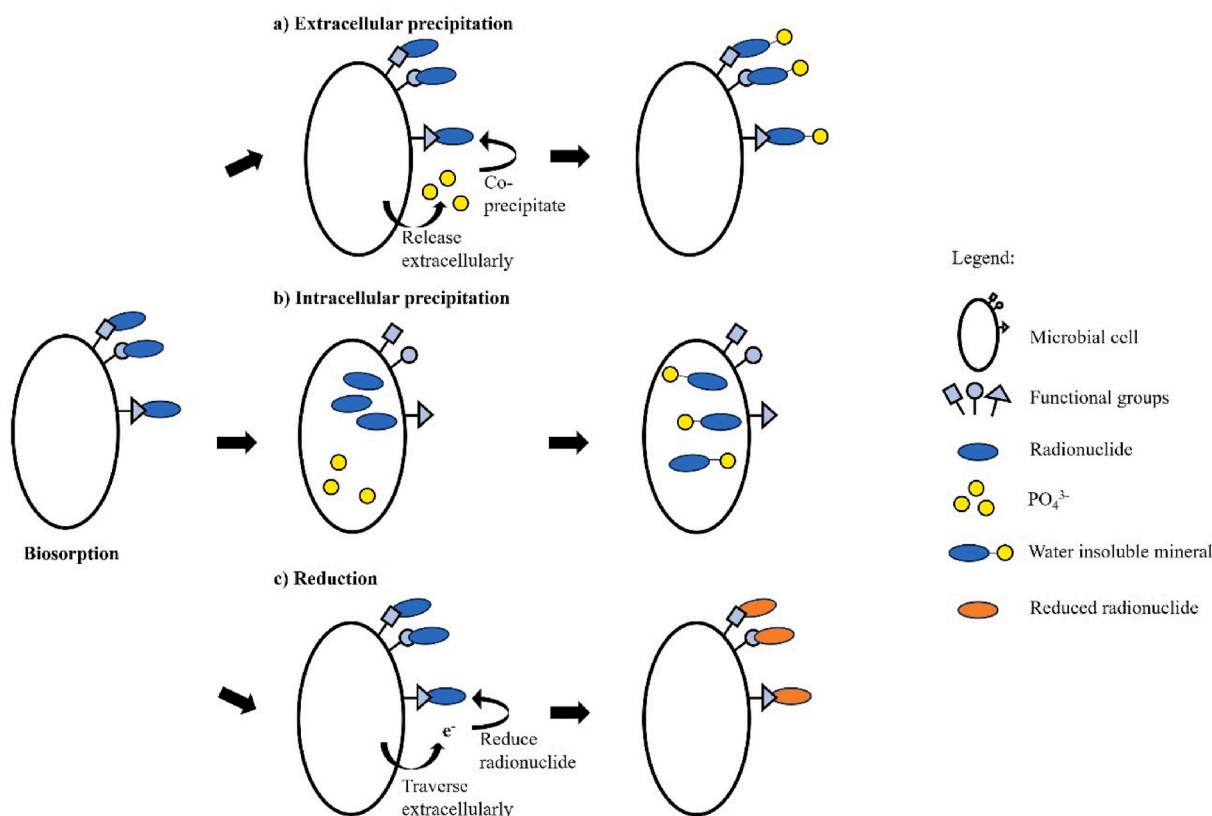


Fig. 2. Possible pathways of a radionuclide associated with the microbial cell surface.

4.1.2. Biosorption

Biosorption is a remediation process where target contaminants are associated with the cell surface. Researchers have reported successful adsorption of radionuclides from radioactively contaminated water by both Gram-positive and Gram-negative bacteria (Table 5). *Pseudomonas* (Kang et al., 2017; Li et al., 2020; Songkasiri et al., 2002; Zheng et al., 2022), *Stenotrophomonas* (Sánchez-Castro et al., 2020), *Acidovorax* (Gerber et al., 2016), and *Shewanella* (Huang et al., 2020), were among the Gram-negative bacteria reported exhibiting radionuclide biosorption activity, and *Streptomyces* (Kamala et al., 2019), *Kocuria* (Wang et al., 2019; Zhou et al., 2022), *Bacillus*, *Microbacterium*, and *Cellulomicrobium* (Elgarahy et al., 2023; Zhao et al., 2016) for Gram-positive bacteria.

Bacteria can adsorb radionuclides through functional groups on their cell wall. According to Wang et al. (2019), *Kocuria* sp.'s cell surface contains phosphorus (P) functional groups ($-P=O$), carbonyl groups ($-C=O$), hydroxyl groups ($-OH$), and carboxyl groups ($-COOH$) that may interact with U(VI) through complexation. The adsorbed U(VI) rapidly accumulates on the cell surface, providing nucleation sites for precipitation with phosphate (PO_4^{3-}). Krawczyk-Bärsch et al. (2015) suggested that phosphates released by bacterial cells from cellular polyphosphate in response to uranium stress led to the formation of water-insoluble uranium phosphate minerals extracellularly. Similar observations were made by Zhou et al. (2022) in *Kocuria rosea*, where P functional groups, $-OH$, $-COOH$, and amino groups ($-NH_2$) were also involved in U(VI) biosorption. Nevertheless, U biomineralization occurred both extracellularly (U(VI) reacted with PO_4^{3-} , hydroxide ions (OH^-), and hydrogen ions (H^+) to form chernikovite) and intracellularly (U(VI) migrated to the inner cell and reacted with PO_4^{3-} to produce U-P compounds) (Zhou et al., 2022). Likewise, in *Pseudomonas fluorescens*, Zheng et al. (2022) observed the involvement of $-OH$, $-COOH$, $-NH_2$, and P-containing groups (phosphoric acid groups) in U biosorption, followed by extracellular bioprecipitation of chernikovite at the cell

surface.

In addition to complexation via functional groups on the cell surface, bacterial radionuclide biosorption can also occur through functional groups present in extracellular polymeric substances (EPS) secreted by bacteria. This phenomenon has been observed in species like *Shewanella putrefaciens* (Huang et al., 2020) and the strontium ion-tolerant marine actinobacterium *Streptomyces* sp. CuOff24 (Kamala et al., 2019). The EPS contains carbohydrate functional groups, including $-C=O$, $-OH$, and others, which play a crucial role in radionuclide sorption (Kamala et al., 2019). The secretion of EPS by microbes serves as a protective mechanism against toxic substances, similar to the release of phosphates (Huang et al., 2020). By adsorbing or complexing with metallic ions, functional groups in EPS can significantly reduce their toxicity, as the toxicity of free metallic ions is largely determined by their activity (Hsieh et al., 1994).

The bacterial biosorption process can be biphasic, consisting of a rapid first phase where most of the radionuclides are adsorbed to the cells, followed by a slow second phase. In this second phase, the functional groups available for biosorption become limited and saturated, eventually reaching a sorption equilibrium state, as observed in studies by Gerber et al. (2016), Wang et al. (2019), and Zheng et al. (2022). Once immobilized by biosorption, the radionuclides may undergo several pathways (Fig. 2): 1) precipitation or mineralization extracellularly, as discussed earlier; 2) migration and accumulation intracellularly, where they associate with cytoplasmic polyphosphates to form inclusions or precipitates, as observed in studies by Gerber et al. (2016), Zhao et al. (2016), and Zhou et al. (2022); 3) reduction, with the potential involvement of EPS in facilitating electron transfer for bio-reduction to occur, as highlighted in the research by Li et al. (2020). Therefore, it is inferred that bacteria, at least some species, can remediate radionuclides in more than one way, where the ability arises from various protective mechanisms against toxic compounds they possess (see Fig. 2).

Table 6

Percentage of radionuclide removal or biosorption capacity of various fungi involved in radioactive waste (radionuclides or radioactive waste-associated heavy metals) remediation.

Fungus Species	Target Contaminant	Mechanism	Parameter	Performance	Reference
<i>Penicillium piscarium</i> (dead biomass)	U(VI)	Biosorption	pH 3.5, 25 °C, 1-h incubation with 0.01 g biomass/mL and 10–100 mg/L uranium nitrate	93.2–97.5*	Coelho et al. (2020)
<i>Aspergillus niger</i> (active)	U(VI)	Biosorption	pH 5.5, 25 °C, 1-h incubation with 0.01 g biomass/mL and 1–100 mg/L uranium nitrate	38.0–92.2*	
<i>Aspergillus niger</i> (inactive)	U(VI)	Biosorption	pH 5.0, 30 °C incubation with 50 mg/L U(VI) and 0.4 g biomass	44.2*, 50.7 ^ˆ , 82.9*, 83.4 ^ˆ	Ding et al. (2019b)
<i>Mucor circinelloides</i> (sporangiospores)	U(VI)	Biosorption, bioaccumulation, bioreduction and bioprecipitation	pH 6.0, 25.85 °C, 48-h incubation	166 ^ˆ	Song et al. (2019)
<i>Rhodotorula taiwanensis</i> MD1149	Hg(II), Cr(VI)	Biosorption	pH 2.3 with the presence of Hg(II), Cr(III) and Cr(VI) and 36 Gy/h	Thrive and produce biofilm	Tkavc et al. (2018)
<i>Saccharomyces cerevisiae</i> (active)	U(VI)	Biosorption and bioprecipitation	pH 5.0, 4-h incubation with 10 mg/L U(VI) and 25–200 mg/L biomass	8.00–38.6*, 1.90–3.20 ^ˆ	Wang et al. (2017)
<i>Saccharomyces cerevisiae</i> (inactive)				69.0–85.6*, 4.30–94.9	

a Symbol representing each unit for different expressions of fungi’s performance.

* Percentage removal (%).

^ˆ Biosorption capacity (mg/g).

Table 7

Performance of microalgae species in remediating radionuclides and radioactive waste-associated heavy metals.

Microalga Species	Target Contaminant	Mechanism	Parameter	Performance	Reference
<i>Microcystis aeruginosa</i>	U(VI)	Biosorption	pH 5.0, 25.05 °C, 12-h incubation with 50 mg/L U(VI) and 0.1–0.9 g/L biomass	42.6–49.7*, 31.0–213 ^ˆ	Yuan et al. (2022)
<i>Haematococcus</i> sp. Sogang	Cs(I)	Bioaccumulation	48-h incubation with 1×10^6 cells in 200 mL 5 Bq/mL Cs(I)	53.7*	Kim et al. (2021b)
<i>Haematococcus</i> sp. Goyang				51.6*	
<i>Haematococcus lacustris</i>				34.5*	
<i>Parachlorella</i> sp. AAl	U(VI)	Biosorption	pH 9.0, 25 °C, 60-h incubation with 20 mg/L U(VI) and 2×10^7 cells	95.6*	Yoon et al. (2021b)
<i>Desmodesmus armatus</i> SCK	Cs(I)	Bioaccumulation and biosorption	pH 7.0, 25 °C, 24-h incubation with K ⁺ -starved cells, 600 μmol/L Cs(I) and volatile fatty acids	49.8*, 299 ^ˆ	Kim et al. (2020a)
			pH 7.0, 25 °C, 24-h incubation with K ⁺ -starved cells and 600 μmol/L Cs(I)	26.8*, 162 ^ˆ	
			pH 7.0, 25 °C, 24-h incubation with K ⁺ -starved cells and 100 μmol/L Cs(I)	99.3*, 99.3 ^ˆ	
			pH 7.0, 25 °C, 24-h incubation with 100 μmol/L Cs(I)	86.3*, 86.3 ^ˆ	
<i>Haematococcus</i> sp.	Cs(I)	Biosorption	pH 5.0, 48-h incubation with 1.5 Bq/mL Cs(I) and 4.14×10^5 cells/mL	88.0*	Kim et al. (2020c)
<i>Haematococcus</i> sp.	Co(II)		pH 5.0, 48-h incubation with 1.5 Bq/mL Co(II) and 2.67×10^6 cells/mL	62.3*	
<i>Vacuoliviride crystalliferum</i> NIES-2860			pH 5.0, 48-h incubation with 1.5 Bq/mL Co(II) and 6.43×10^5 cells/mL	19.5*	
<i>Desmodesmus armatus</i> SCK	Cs(I)	Bioaccumulation	pH 7.0, 25 °C, 7-d incubation with 100 μmol/L Cs(I)	0.0639 ^ˆ	Kim et al. (2019)
<i>Haematococcus pluvialis</i> CCAP 34/7	Cs(I)	Biosorption and bioaccumulation	25 °C, 20-d incubation with 370 Bq/mL Cs(I) in 3 mM NaHCO ₃	~95.0* (in 2 d)	Lee et al. (2019)
<i>Chlorella vulgaris</i> UTEX 26				~90.0* (>10 d)	
Eustigmatophycean nak 9	Cs(I)	Biosorption	8-d incubation with 2.2 ng/mL Cs(I)	~90.0*	Fukuda et al. (2014)
<i>Batrachospermum virgato-decaisneanum</i> NIES-1458				~38.0*	
<i>Chloroidium saccharophilum</i> NIES-2352				~22.0*	
<i>Stigonema ocellatum</i> NIES-2131	Sr(II)		8-d incubation with 7.1 ng/mL Sr(II)	~41.0*	
<i>Oedogonium</i> sp. nak 1001				~36.0*	
<i>Nostoc commune</i> TIR 4	I ⁻		8-d incubation with 5.9 μg/mL I ⁻	~66.0*	
<i>Scytonema javanicum</i> NIES-1956				~62.0*	
<i>Stigonema ocellatum</i> NIES-2131				~49.0*	
<i>Ophiocytium</i> sp. nak 8				~42.0*	

a Symbol representing each unit for different expressions of microalgae’s performance.

* Percentage removal (%).

^ˆ Removal capacity (mg/g).

^ˆ Removal capacity (mmol/g dry cell weight).

^ˆ Removal capacity (μmol/L)amount of astaxanthin (Kim et al., 2021b).

4.1.3. Bioprecipitation (biomineralization)

Bioprecipitation can occur both extra- and intracellularly, with cellular phosphates playing a vital role in the process. Phosphate solubilizing bacteria (PSB), such as *Microbacterium* sp. Be9, have been identified as capable of accumulating and precipitating uranium intracellularly in the presence of cellular phosphates (Martínez-Rodríguez et al., 2023). However, Martínez-Rodríguez et al. (2023) made an interesting discovery that in the presence of exogenous P sources, Be9 could also solubilize phosphates from uranium phosphates that were precipitated abiotically by these external sources. This caused the re-solubilization and re-release of U in its toxic hexavalent form back into the environment. Apart from exogenous P sources, the presence of strong organic ligands, such as citrate and oxalate, in the environment can also influence uranium phosphate bioprecipitation. These strong ligands may compete with cellular phosphates to form complexes with U (VI), thereby hindering U biomineralization. The degree of hindrance might be further exacerbated when uranium is present in the form of uranyl ions (UO_2^{2+}), as UO_2^{2+} has been reported to exhibit a high affinity for organic ligands (Tu et al., 2019). These findings underscore the importance of surveying the chemical components of the environment and possibly conditioning the environment accordingly to ensure an optimum bacterial bioprecipitation remediation strategy.

4.2. Mycoremediation

Applications of fungi (active or dead cell biomass) in remediating radionuclides and heavy metals associated with radioactive wastes have also been reported, where biological processes like biosorption and bioaccumulation are involved (Table 6).

The efficiency and biosorption capacity of radionuclide by active or dead fungal cell biomass can be influenced by pH. pH plays a crucial role in determining the speciation of the sorbate (radionuclides) and the surface charge of the biosorbent (fungal cells) in the aqueous medium (Sert et al., 2008). For example, U exhibits different dominant species at different pH levels. In highly acidic environments ($\text{pH} \leq 4$), the toxic, water soluble, positively charged uranyl ion (UO_2^{2+}) is the primary form. In mildly acidic environments ($\text{pH} 5\text{--}6$), positively charged U hydroxide species [$(\text{UO}_2)_3(\text{OH})_5^+$] dominates. Under mildly alkaline conditions ($\text{pH} 7\text{--}9$), soluble, negatively-charged uranyl carbonates [$\text{UO}_2(\text{CO}_3)_3^{4-}$ and $(\text{UO}_2)_2\text{CO}_3(\text{OH})_3$] are abundant (Kolhe et al., 2020).

Fungal biomass, whether in the form of cells or sporangiospores, typically carries negative surface charges over a wide pH range ($\text{pH} > 3$) (Coelho et al., 2020; Song et al., 2019). In mildly acidic environments, the positively charged U species can be effectively adsorbed to the negatively charged biomass through electrostatic attraction (Song et al., 2019). However, highly acidic environments may lead to suboptimal biosorption as the increased abundance of H^+ ions at lower pH levels competes with UO_2^{2+} for anionic surface binding sites on the fungal biomass (Kolhe et al., 2020; Sar and D'Souza, 2002). Similarly, alkaline environments may result in suboptimal radionuclide removal as the repulsion between the anionic fungal surface charges and the negatively charged U species could hinder the biosorption processes (Coelho et al., 2020). Hence, pH 5–6 (mildly acidic environment) may result in optimum biosorption activity in fungi (Ding et al., 2019b; Song et al., 2019; Wang et al., 2017).

Moreover, the concentration of the biosorbent can also influence biosorption efficiency and capacity. Wang et al. (2010) observed that the percentage removal of U increased with higher biosorbent dosages, likely due to the greater surface area and availability of binding sites for U biosorption (Esposito et al., 2001). In contrast, the biosorption

capacity of fungal biomass decreased as the biosorbent dose increased. This reduction could be attributed to a "screen" effect created by the concentrated biomass, blocking the surface binding sites and limiting access to radionuclides (Wang et al., 2010). Similar relationships between biosorbent dose and capacity were also found in other microbial biosorbents (Kolhe et al., 2020; Yuan et al., 2022; Zhao et al., 2016).

Radionuclides associated with the fungal biomass may then undergo extracellular precipitation in the presence of cellular phosphates (Wang et al., 2017) or experience intracellular reduction before interacting with oxygen-containing functional groups to form more stable compounds (Song et al., 2019).

4.3. Phycoremediation

4.3.1. Microalgae

Microalgae have also been studied for their potential in remediating radionuclides and radioactive waste-associated heavy metals (see Table 7).

Haematococcus, a unicellular green microalga, has been found to exhibit radioactive metal removal activity. Lee et al. (2019) conducted investigations on the cesium-137 (Cs-137) biosorption ability of different life cycles of *Haematococcus pluvialis*: palmella (green cells with lost flagella, derived from the previous flagellated cell life cycle, with an expanded cell size), aplanospores (red cysts developed from palmella under continuous environmental stress, while accumulating astaxanthin) (Boussiba and Vonshak, 1991), and intermediate cells transitioning from palmella to aplanospores. Among these life forms, red cysts showed the highest Cs-137 removal efficiency (~95%) within 48 h. Besides, a significant decrease in cellular potassium (K) in red cysts as Cs concentration increased was observed, possibly due to Cs^+ and K^+ being chemically similar (Lee et al., 2019). Combining the fact that both ions are chemically similar and other microalgal species like *Chlorella emersonii* (Avery et al., 1992) have been reported to actively take up Cs^+ through K^+ -transporting systems, Lee et al. (2019) suggested that K transporters played a role in *H. pluvialis* red cysts' Cs uptake. Additionally, the accumulation of astaxanthin in aplanospores appeared to induce the expression of K^+ -transporting proteins (Kim et al., 2006), which explains the higher Cs^+ removal activity in aplanospores compared to palmella (Lee et al., 2019) and the greater accumulation of Cs^+ in *Haematococcus* strains that accumulated a greater amount of astaxanthin (Kim et al., 2021b).

Apart from *Haematococcus*, K^+ -transporting systems were also responsible for Cs^+ uptake in *Desmodesmus armatus*, *Chlorella vulgaris*, and *Chlamydomonas reinhardtii* (Kim et al., 2020a). The variation in affinity towards Cs^+ among the different types of K^+ -transporting systems possessed by these microalgae could also influence the Cs^+ removal capacity (Kim et al., 2020a). Therefore, further research exploring the effectiveness of identified K^+ -transporting systems in the microbial Cs^+ uptake should be conducted, as it may help identify novel or existing microbes with high potential for remediating radioactive Cs.

4.3.2. Macroalgae

Marine macroalgae (seaweeds), especially Phaeophyta (brown seaweeds), Chlorophyta (green seaweeds), and Rhodophyta (red seaweeds), have also seen potential in radionuclide removal (Senthilkumar et al., 2018).

Sargassum seaweeds are one of the brown macroalgae most commonly studied. Fresh *Sargassum horneri* biomass managed to remove strontium (Sr), manganese (Mn), and cobalt (Co) simultaneously in multi-nuclide contamination scenarios (Wang et al., 2021). At the low

contamination scenario ($Sr_{\text{initial}} = 20 \text{ mg/L}$; $Mn_{\text{initial}} = 100 \text{ }\mu\text{g/L}$; $Co_{\text{initial}} = 100 \text{ }\mu\text{g/L}$), the biomass removed 32.0% Sr, 91.9% Mn, and 63.9% Co from the solution, and 31.3% Sr, 50.2% Mn, and 29.4% Co from the solution simulating high contamination scenario ($Sr_{\text{initial}} = 20 \text{ mg/L}$; $Mn_{\text{initial}} = 1,000 \text{ }\mu\text{g/L}$; $Co_{\text{initial}} = 1,000 \text{ }\mu\text{g/L}$) (Wang et al., 2021). Beside Sr, Mn, and Co, *Sargassum* also reportedly adsorbed U(VI) and Th(IV). *Sargassum aquifolium* powder had a maximum sorption capacity of 20.43 mg/g and 24.13 mg/g for U(VI) and Th(IV), respectively, when 0.1 g of the powder was applied to treat 40.0 mg/L U(VI) and Th(IV) mono-nuclide solutions for 4 h at pH 3.0, 25 °C (Albayari et al., 2023). Other brown seaweeds with radionuclide biosorption ability included *Sargassum glaucescens*, *Cystoseira indica* (adsorbed Cs) (Dabbagh et al., 2008), *Laminaria japonica*, *Laminaria digitata* (adsorbed Sr-85, Cs-134, Ra-226, and Am-241) (Pohl and Schimmack, 2006), *Padina pavonia* (adsorbed U(VI)) (Aytas et al., 2014), and many more.

As the polysaccharide universal across brown macroalgae, alginic acid could be found abundant in the cell wall matrix, mucilage, or intercellular substances, accounting for 10–40% of the dry weight of the macroalgae (Davis et al., 2003). The alginate polymer comprises carboxylic groups – brown macroalgae's most abundant acidic functional group in general – that contribute to brown macroalgae's satisfactory metal biosorption capacity (Davis et al., 2003). Being the second most abundant acidic functional group in brown seaweeds, sulfonic acid groups carried by fucoidan play a less significant role in biosorption, except when the process happens at low pH (Senthilkumar et al., 2018). The fact that carboxylic groups can adsorb metal, e.g., U, at high pH, whereas sulfonic acid groups become the primary functional group involved in metal adsorption at low pH (Banerjee et al., 2022) suggests the potential of brown seaweeds' wide applications in remediating broad ranges of radionuclide-contaminated aqueous environments with a good removal capacity.

Ulva green seaweeds were shown to hold potential as biosorbents for several radionuclides. To illustrate, Vijayaraghavan and Joshi (2014) reported maximum biosorption capacities of grounded *Ulva* sp. for Cr (III) and Mn(II) as 150.3 mg/g and 58.8 mg/g, respectively, at pH 4.5 mono-nuclide Cr(III) and Mn(II) solutions. Live *Ulva prolifera* was capable of adsorbing an average of 0.4475–3.349 mg/g Cs and 3.183–7.730 mg/g Co from single-radionuclide sterilized seawater with $Cs_{\text{initial}} = 10\text{--}100 \text{ mg/L}$ and $Co_{\text{initial}} = 10\text{--}100 \text{ mg/L}$, respectively (Wang et al., 2022). Nevertheless, green macroalgae's biosorption abilities are less studied when compared to brown macroalgae. This trend was likely because brown seaweeds tend to outperform green seaweeds in metal biosorption for all metals studied (Senthilkumar et al., 2018), owing to green seaweeds, e.g., *Ulva*, having lesser negatively charged binding groups on their cell wall (Wang et al., 2022).

Gracilaria spp. (*G. changii*, *G. corticata*, *G. edulis*, *G. salicornia*, etc.) (Dabbagh et al., 2018; Hashim and Chu, 2004), *Kappaphycus alvarezii* (Praveen and Vijayaraghavan, 2015) are among the red seaweeds studied for their potential for radionuclide and heavy metal biosorption. Similar to green seaweeds, red seaweeds received less attention than brown seaweeds due to their generally relatively poor performance in metal biosorption (Vijayaraghavan and Yun, 2008), which is contributed by their lesser cationic functional groups (Senthilkumar et al., 2018).

4.4. Phytoremediation

Phytoremediation involves the application of plants to remove radionuclides from radioactively contaminated environments. The commonly adopted mechanisms include phytoextraction and phytostabilization (Gul et al., 2022). In phytoextraction, plants take up

radionuclides from polluted soil or water via their roots and transfer the contaminants to aerial parts, e.g., shoot, leaf, and stem, through the xylem for accumulation (Gul et al., 2022). *Cucumis sativus* L. (cucumber) (Ali et al., 2022), *Arabidopsis halleri* (rockcress) (Burger et al., 2019), and *Vetiveria zizanioides* L. Nash (vetiver grass) (Pentyala and Eapen, 2020) are among the species reported to exhibit phytoextraction of radionuclides, including Cs, Sr, and U. In contrast to phytoextraction, phytostabilization does not focus on accumulating radionuclides in plant tissues (Singh et al., 2022). Instead, the radionuclides are immobilized and sequestered near the roots (rhizosphere), where the root system retains the radionuclides by reducing water infiltration (Singh et al., 2022). An example of plants capable of carrying out phytostabilization is *Miscanthus × giganteus* (giant miscanthus), which can stabilize cadmium and mercury in soil (Zgorelec et al., 2020), where mercury is a heavy metal that might be associated with radioactive waste (Brim et al., 2000).

5. Strengths, weaknesses, opportunities, and threats (SWOT) analysis of microbe-mediated radionuclide bioremediation

5.1. Strengths

Remediation methods that are used conventionally in radionuclide and radioactive waste-associated heavy metal removal, including membrane separation (ultrafiltration, microfiltration, nanofiltration, and reverse osmosis), chemical precipitation, electro dialysis, and other electrochemical remediations, might remove the target contaminants incompletely from radioactively contaminated water, especially when the concentrations of these contaminants in the water are below 100 mg/L (Kanamarlapudi et al., 2018; Yang et al., 2011). On the other hand, numerous microorganisms have shown satisfactory radionuclide removal performance ($\geq 80\%$ removal) when being applied to treat radioactively contaminated water with dilute radionuclides. For example, *Kocuria* sp. (98% U removal from 20 mg/L U) (Wang et al., 2019), dead *Penicillium piscarium* biomass (93.2–97.5% U removal from 10 to 100 mg/L U) (Coelho et al., 2020), Eustigmatophycean nak 9 (90% Cs^+ removal from 0.0022 mg/L Cs^+) (Fukuda et al., 2014) (Tables 5–7). Furthermore, unlike conventional treatments, microbe-mediated bioremediation does not generate as much sewage sludge (Chojnacka, 2010), making it a 'cleaner' treatment (Kadadou et al., 2023).

Microbe-mediated bioremediation can also have low production costs. For instance, inexpensive microbe sorbents like yeasts can be obtained directly in large amounts at almost zero cost from fermentation industries such as brewery and winery industries because these industries produce yeasts as byproducts or wastes (Wang and Chen, 2006). While some may doubt that cultivating microbes specifically for applications in remediating radioactive waste will result in high production costs, several studies have claimed that such cultivations did not increase the cost as bacteria and yeasts are easy to grow in low-cost growth media (Choudhary et al., 2019; Kapoor and Viraraghavan, 1995). Some researchers have also developed growth media for cost-effective and optimum industrial cultivation of microbes. For example, Holland et al. (2006) optimized a phosphate-free morpholinepropanesulfonic acid-based medium consisting of only one carbon source, two amino acids, four vitamins, and salts that can support rapid *Deinococcus radiodurans* R1 growth (< 7 h doubling time, depending on the components used). Microbial bioremediations may also have lower operation costs as they do not require sophisticated devices and have lower energy and reagent requirements than other remediation techniques (Kanamarlapudi et al., 2018). Biostimulation requires a continuous supply of carbon source(s) serving as electron donor(s) to maintain the

microbial bioreduction activity (Williams et al., 2011). Nevertheless, some carbon sources typically used in biostimulation, e.g., acetate and glucose, are cheap industrial wastes (Dubey et al., 2015) and can be collected at low prices.

Besides, microbial remediation is eco-friendly and does not increase the chemical oxygen demand (COD) and biochemical oxygen demand (BOD) of the wastewater treated, particularly when dead microbial biomass is applied because they do not metabolize nutrients (Rezaei, 2016). The non-requirement on nutrients and negligible impact on COD and BOD indicated that there is no need for nutrient input, maintenance, and continuous COD and BOD monitoring when dead microbial biomass is applied as biosorbent to treat radioactively polluted effluents, making it more economical and cost-effective (Moffat, 1995; Rezaei, 2016). Moreover, dead microbial cells' biosorption capacity is also unaffected by the radionuclides' toxicity (Rezaei, 2016).

5.2. Weaknesses

Active microbial cells appeared to have poorer radionuclide biosorption ability than dead biomass. As the initial concentration of U(VI) in radioactive wastewater increased from 10 to 100 mg/L, the biosorption capacity of active *Aspergillus niger* reduced from about 45 to 15 mg/g, which was lower in general than that of heat-killed *A. niger* (reduction from approximately 82.5 to 25 mg/g) (Ding et al., 2019b). Wang et al. (2017) also observed lower U(VI) biosorption capacity by metabolically active cells. Ding et al. (2019b) suggested that the overall lower adsorption capacities of active *A. niger* was related to the stress that fungi experienced under high U(VI) concentrations. Besides biosorption-based microbes, numerous microbes relying on metabolic activities (bioreduction, bioprecipitation, and bioaccumulation) to remediate radionuclides have also been reported to have their growth inhibited by radionuclides of certain concentrations. These microbes include *Stenotrophomonas* sp. Br8 (minimum inhibitory concentration (MIC): ~950 mg/L U) (Sánchez-Castro et al., 2020), *Acidovorax facilis* (MIC: ~50 mg/L U) (Gerber et al., 2016), *Halomonas* sp. SR4 (MIC: ~440 mg/L Sr) (Achal et al., 2012), *Thermoterrabacterium ferrireducens* (no growth observed at 1,190 mg/L U) (Khijniak et al., 2005), *Desulfovibrio* spp. (~60 mg/L Np-237 reduced the bacterial growth by ~80%) (Rittmann et al., 2002), and many more. Growth inhibition of these microbes at relatively high radionuclide concentrations may limit their practical application because growth inhibition can result in slower metabolism (Choramo, 2022) and even cell death, making the metabolism-dependent bioremediation processes less efficient or unable to carry out.

Besides limited applications for treating concentrated radionuclides, microbes, particularly those that function as radionuclide biosorbents, also have shorter shelf lives than synthetic sorbents like ion exchange resins (Volesky, 2007). Shorter shelf lives may increase the labor required to acquire new batches of microbe sorbents for replacement, leading to a higher operation and maintenance cost (Holmes et al., 2021).

Furthermore, if living microbial cells are used, it may be essential to supply external energy sources like molasses containing sucrose to the cells (Aksu and Dönmez, 2005). Biostimulation also requires continuous supplies of carbon sources as electron donors to make the indigenous microbial radionuclide bioreduction activities persist (Williams et al., 2011), which indicated that the microbial remediation involving living cells can be hard to self-sustain.

5.3. Opportunities

Two research directions covering the 1) genetic modification and 2) co-application with other remediation approaches are believed to expand microbe-mediated bioremediation's applications.

5.3.1. Genetic engineering

Genetic engineering of radiation-resistant microorganisms can be conducted to introduce novel radionuclide bioremediation abilities to the engineered microbes. To illustrate, *Deinococcus radiodurans* is a highly radiation-resistant bacterium that can thrive in the presence of radiation level (60 Gy/h) that exceeds typical radiation levels detected in radioactive waste sites (Brim et al., 2000) without experiencing suppression on its ability to express recombinant genes (Lange et al., 1998). Appukuttan et al. (2006) expressed a nonspecific acid phosphatase-encoding gene, *phoN*, isolated from an Indian *Salmonella enterica* serovar Typhi isolate in *D. radiodurans* R1. The engineered bacterium was able to supply cellular phosphates for U(VI) bioprecipitation, and it efficiently precipitated >90% of the U(VI) within 6-h incubation in a 0.8 mM uranyl nitrate solution. Recombinant *D. radiodurans* R1 with *phoN* also successfully mineralized U(VI) from 1 mM uranyl nitrate solution (Misra et al., 2012). The engineered bacterium was able to retain its novel bioremediation activity after being exposed to 6 kGy of cobalt-60 γ -rays, making it a potential candidate for in situ remediation of uranium from low-level nuclear wastes (Appukuttan et al., 2006).

Besides *phoN*, nickel/cobalt transporter (NiCoT) genes have also been introduced to *D. radiodurans* R1 to allow the bacteria to synthesize NiCoT for Co-60 uptake from radioactive wastes. Gogada et al. (2015) constructed two recombinant *D. radiodurans*: one inserted with NiCoT-encoding gene isolated from *Rhodopseudomonas palustris* CGA009 (*ncxA*) and another with NiCoT gene from *Novosphingobium aromaticivorans* F-199 (*nvoA*). Both engineered strains could take up >60% of Co-60 from simulated spent decontamination effluent containing a trace amount of targeted Co and significantly higher concentrations of non-targeted metallic ions, indicating the high specificity of Co-60 uptake ability introduced to *D. radiodurans*. A novel *D. radiodurans* strain that can form biofilm was also engineered by Manobala et al. (2021), and the lyophilized biofilm removed up to about 80% of the U(VI) from solutions containing initial U(VI) concentrations of 100 and 1,000 mg/L.

Given these research outputs, it is then suggested that identified radiation-resistant microbes should be studied for their genetic transformability. This would enable the production of recombinant microbes with novel or multiple radionuclide removal capabilities, making them highly practical for in situ low-level radioactive waste remediation. These specially engineered microbes can thrive under radiation exposure and effectively carry out biological processes to remove radionuclides from polluted sites, as opposed to experiencing suppressed growth and metabolism due to radionuclide-associated radiotoxicity, chemotoxicity, or both.

5.3.2. Co-application with other remediation strategies

The immobilization of fungal cells on physical adsorbents like alginate beads has been applied for contaminant removal from aqueous solutions (Kaygusuz et al., 2014; Zhou et al., 2014, 2015). This co-application's potential for removing radionuclide from radioactive wastewater has also been studied. Wang et al. (2010) immobilized *Aspergillus fumigatus* spores on calcium alginate beads for U adsorption, where the alginate beads served as both U adsorbent and a supporting structure to trap the spores in position for higher U biosorption. Results

showed that the *A. fumigatus*-immobilized beads removed 85.8% of U(VI) from a solution with an initial U(VI) concentration of 100 mg/L within 60 min, which was four times more efficient than blank Ca-alginate beads (20.7%). Ding et al. (2019a) introduced a novel Ca-alginate-immobilized *Aspergillus niger* microsphere (AAM) with a superb thorium(IV) (Th(IV)) biosorption capacity (303.95 mg/g at pH 6, 40 °C). More importantly, the alginate-immobilized AAM can be recycled for new rounds of remediation by desorbing the Th(IV) from the sorbent via hydrochloric acid (HCl) and rinsing the Th(IV)-unloaded AAMs with deionized ultrapure water. Ding et al. (2019a) reported that the alginate-immobilized AAMs retained around 80% Th(IV) biosorption efficiency after six cycles of biosorption-desorption tests, thereby suggesting the novel sorbent's potential economical and environmentally friendly application for Th removal from radioactive wastes. *Yarrowia lipolytica* (a tropical marine yeast)-immobilized Ca-alginate beads were also reported to exhibit a high U(VI) biosorption activity with 53.09–73.10% for initial U(VI) concentrations ranging from 11.9 to 238 mg/L, which was at least two times as efficient as blank beads (Kolhe et al., 2020). Improvement of radionuclide biosorption capacity upon immobilization to alginate beads was also observed in *Trichoderma harzianum* (Akhtar et al., 2009) and *Rhodotorula glutinis* (Bai et al., 2014).

Furthermore, co-applying electrochemical and microbial approaches in removing radionuclides from radioactively contaminated sites is also possible. Vijay et al. (2020) established a microbial fuel cell (MFC) with graphite felt cathode and anode, with denitrifying microbial consortia added to the cathode. Through phosphatase produced by the consortia, phosphates were released from glycerol-3-phosphate and combined with U(VI) to form uranyl phosphate minerals. Simultaneously, nitrate was also reduced to nitrogen gas at the cathode as the cathode continuously received electrons sourced from the oxidation of acetate at the anode. Thakare et al. (2021) also summarized numerous plant-microbe remediation approaches that potentially remove radionuclides from the soil.

These studies demonstrated that the co-applications of microbial and other remediation strategies produce favorable outcomes, i.e., enhanced removal ability for a particular radionuclide or the removal of multiple contaminants in a single remediation approach. More research focusing on the effective co-application of microbe-mediated remediation with other approaches should be conducted to discover more remediation combinations with high practicability for both in situ and ex situ radioactive waste treatment.

5.4. Threats

According to Kuppusamy et al. (2015), it is uncertain what form of interactions may occur between indigenous microbial communities and

foreign microbes introduced to the radioactively polluted site for bioremediation purposes. Introduction of *Sphingobium chlorophenicum* C3R to agricultural soil microcosms co-contaminated with polycyclic aromatic hydrocarbons (PAHs) and metals to degrade phenanthrene had resulted in reduced populations of various native PAH degrading bacteria, including the indigenous *Sphingomonas* by out-competing them (Colombo et al., 2011). Similarly, the foreign *Penicillium oxalicum* XD-3.1 that Olicón-Hernández et al. (2021) introduced to remove pharmaceutical active compounds from hospital wastewater also led to decreased autochthonous bacteria and fungi populations. These studies indicated the possibility of indigenous microbes' survival suppression by microbes introduced to remove radionuclides, thereby adversely impacting the biodiversity. Therefore, thorough research regarding the interaction between radionuclide-treating foreign live microorganisms and native microbial communities should be conducted to ensure the microbial remediation is suitable for a particular radioactively contaminated site. Otherwise, biostimulation or other remediation strategies that do not negatively affect autochthonous microbial populations would need to be reconsidered.

Horizontal gene transfer can occur through transformation from a microbe, which can be either alive or dead and lysed, to another (Lorenz and Wackernagel, 1996; Overballe-Petersen and Willerslev, 2014), implying that foreign genes carried by non-indigenous radionuclide-removing microbes that one introduced to remediate a radioactively contaminated location might be picked up by native microbe species, affecting the local gene pool. Horizontal gene transfer can occur with a relatively high possibility in the environment. Thus, the microbes to be introduced should not carry any possibly dangerous genes, such as genes encoding virulence factors (Lorenz and Wackernagel, 2014). Besides, a suicide system where the killing gene targets the DNA itself can also be established in genetically engineered microorganisms (GEMs) if GEMs are to be added to the polluted site. To illustrate, Ahrenholtz et al. (1994) introduced a nuclease gene (isolated from *Serratia marcescens*) with its leader-coding sequence deleted to *Escherichia coli* and was regulated by lambda p_1 promoter. Upon thermoinduction of the promoter at 42 °C, the nuclease's intracellular DNase and RNase activities were activated, and the nuclease disintegrated the transformed *E. coli*'s DNA. Consequently, the gene transfer from GEMs to other microbe species can be prevented (Ahrenholtz et al., 1994). It is suggested that research into such biological containment systems (controlled suicide process) and their efficacy and effectiveness in preventing horizontal gene transfer from GEMs to other microbes may reduce the potential risks like genetic swamping (Frankham, 2019) associated with the introduction of GEMs to remediate radionuclide-contaminated sites. Fig. 3 summarizes the SWOT analysis for microbe-mediated radioactive waste remediation.

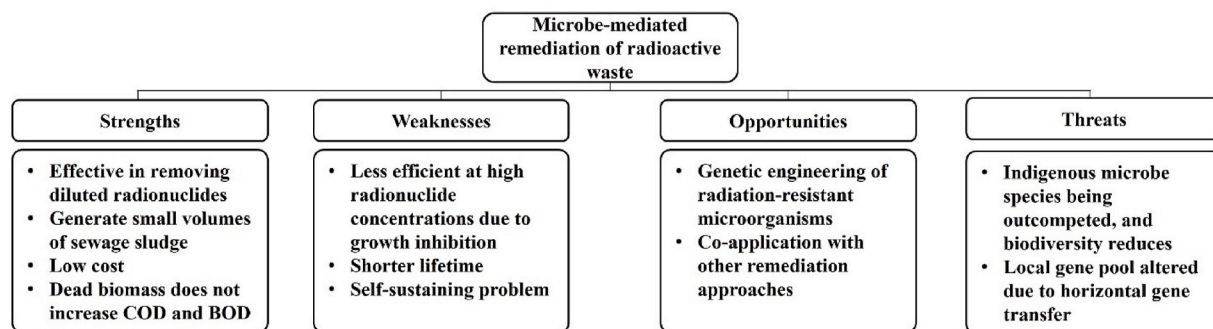


Fig. 3. SWOT analysis for microbe-mediated remediation of radioactive waste.

Table 8

The strengths and weaknesses among the physical, chemical, electrochemical, and biological remediation approaches.

	Physical remediation ¹⁻¹⁴	Chemical remediation ¹⁵⁻²⁰	Electrochemical remediation ²¹⁻²³	Biological remediation	
				Microbe-mediated remediation	Phytoremediation ²⁴⁻²⁶
Strengths	<ul style="list-style-type: none"> Radioactive waste volume reduction. Eco-friendly.^a Cost-effective.^a Allow the recovery of radionuclide from radioactive waste. 	<ul style="list-style-type: none"> Radioactive waste volume reduction. Eco-friendly.^d Cost-effective.^d Allow the recovery of radionuclide from radioactive waste. Relatively simple to implement.^e 	<ul style="list-style-type: none"> Radioactive waste volume reduction. Eco-friendly. Allow the recovery of radionuclide from radioactive waste. 	<ul style="list-style-type: none"> Eco-friendly. Cost-effective. Allow the recovery of radionuclide from radioactive waste. Relatively simple to implement. Effective in removing diluted radionuclides. Dead biomass does not increase biological oxygen demand (BOD) and chemical oxygen demand (COD). 	<ul style="list-style-type: none"> Eco-friendly. Cost-effective. Allow the recovery of radionuclide from radioactive waste. Relatively simple to implement.
Weaknesses	<ul style="list-style-type: none"> Pose environmental risk.^b High operational cost.^c Equipment and setup involved are more sophisticated. 	<ul style="list-style-type: none"> Pose environmental risk.^f High operational cost.^f Limited applicability and removal efficiency by natural adsorbents. 	<ul style="list-style-type: none"> High operational cost. More difficult to implement. Equipment and setup involved are more sophisticated. 	<ul style="list-style-type: none"> Methods involve the use of live cells are less efficient at high radionuclide concentrations due to growth inhibition. Shorter lifetime. Self-sustaining problem. 	<ul style="list-style-type: none"> Require additional nutrient supply or natural or synthetic chelators. Longer turnover time. Radionuclides accumulated in the plants may enter the food chain. Limited efficiency at high radionuclide concentrations.

* The superscripts of numbers are in-text citations, corresponding to: 1. Bayliss and Langley (2003); 2. Chen et al. (2021); 3. Deng et al. (2022); 4. Eagle et al. (1993); 5. Hong and Um (2023); 6. Hou et al. (2022); 7. IAEA (1992); 8. Kim et al. (2020b); 9. Merrington (2011); 10. Nishimoto et al. (2021); 11. Roybal et al. (1998); 12. Yamasaki et al. (2022); 13. Yu et al. (2020); 14. Zhang et al. (2019); 15. Huang et al. (2023); 16. Kadadou et al. (2023); 17. Kobayashi et al. (1980); 18. Tang et al. (2023); 19. Walling et al. (2021); 20. Yue et al. (2023); 21. Lee et al. (2022); 22. Li et al. (2022); 23. Lv et al. (2021); 24. Fatima et al. (2017); 25. Mustafa and Hayder (2021); 26. Singh et al. (2022).

^a Includes methods such as soil washing by dense medium and gravitational separation and solar-driven evaporation of high-salinity radioactive seawater.

^b Includes methods such as incineration of liquid radioactive waste.

^c Includes methods such as detector-based separation of radioactive soil, soil washing by magnetic separation, soil washing by flotation separation, mechanical-driven evaporation of radioactive wastewater, and incineration of liquid radioactive waste.

^d Includes chemical remediation methods like radionuclide adsorption from radioactive waste by natural adsorbents.

^e Particularly adsorption of radionuclides by adsorbents and precipitation of radionuclides with chemicals.

^f Includes chemical remediation methods such as wet oxidation and acid digestion of solid radioactive waste.

6. Comparison among the physical, chemical, electrochemical, and biological remediation approaches

In general, physical, chemical, and electrochemical remediation approaches are capable of reducing the volume of radioactive waste, encompassing both solid forms, such as radioactively contaminated soil, concrete, and ion-exchange resins, and liquid forms, including radioactive seawater and wastewater, significantly decrease the space required to store the waste (Bayliss and Langley, 2003; Chen et al., 2021; Eagle et al., 1993; IAEA, 1992; Kim et al., 2020b; Kobayashi et al., 1980; Li et al., 2022; Nishimoto et al., 2021; Walling et al., 2021; Yamasaki et al., 2022; Yu et al., 2020; Zhang et al., 2019).

However, unlike biological approaches, which are generally more environmentally friendly and cost-effective, the environmental impact and costs associated with physical and chemical approaches may differ significantly depending on the specific techniques employed. For instance, physical methods such as soil washing by dense medium and gravitational separation (Hong and Um, 2023; Yamasaki et al., 2022) and solar-driven evaporation of high-salinity radioactive seawater (Deng et al., 2022) are considered more environmentally friendly. The former generates a small quantity of secondary waste (e.g., the heavy liquid solution such as sodium polytungstate solution used in the soil washing can be reused) while the latter is powered by a clean energy source, i.e., solar power. Similarly, chemical methods like radionuclide adsorption by natural adsorbents (Kadadou et al., 2023) are also environmentally friendly as the adsorbents are biodegradable. The minimal additional costs incurred to treat secondary waste produced, the reusability of chemicals involved, as well as the readily availability of power source and adsorbents used make these techniques cost-effective.

Conversely, some physical and chemical remediation approaches, like the incineration of liquid radioactive waste (IAEA, 1992), wet

oxidation (Walling et al., 2021), and acid digestion (Hong and Um, 2023), are less environmentally friendly as they generate considerable amounts of secondary wastes, including radioactive ash, volatile organic wastes (from incineration), secondary iron hydroxide precipitate effluent (from wet oxidation), and acidic waste solutions (from acid digestion), all of which require further processing and lead to high operational costs. Furthermore, co-precipitation of radionuclides with nickel-potassium ferrocyanide (Sopapan et al., 2023), while effective in removing radionuclides such as Cs(I) at concentrations up to 1,000 mg/L, raises concerns due to the toxicity and high cost associated with the use of nickel. Additionally, physical methods like detector-based separation of radioactive soil (Bayliss and Langley, 2003; Roybal et al., 1998), soil washing by magnetic separation (Eagle et al., 1993), soil washing by flotation separation (Merrington, 2011), and mechanical-driven evaporation of radioactive wastewater (Hou et al., 2022), also have high operational costs. The detector-based separation incurs high costs both on-site, due to high commissioning and dismantling costs, and off-site, due to the transportation of contaminated soil to fixed centralized plants. The use of materials and equipment such as magnetic nanocomposites and magnetic separators in soil washing by magnetic separation (Eagle et al., 1993; Kim et al., 2020b), various chemicals like surfactants and frothers in soil washing by flotation separation (Merrington, 2011), and the sophisticated system required for mechanical-driven radioactive wastewater evaporation and its requirement for a constant electrical input (Hou et al., 2022), all contribute to the high operational costs and can limit the upscaling of these remediation methods.

As in the case of electrochemical remediation, the techniques employed are generally considered more environmentally friendly compared to some of the physical and chemical remediation methods discussed as they do not generate as significant quantities of solid or

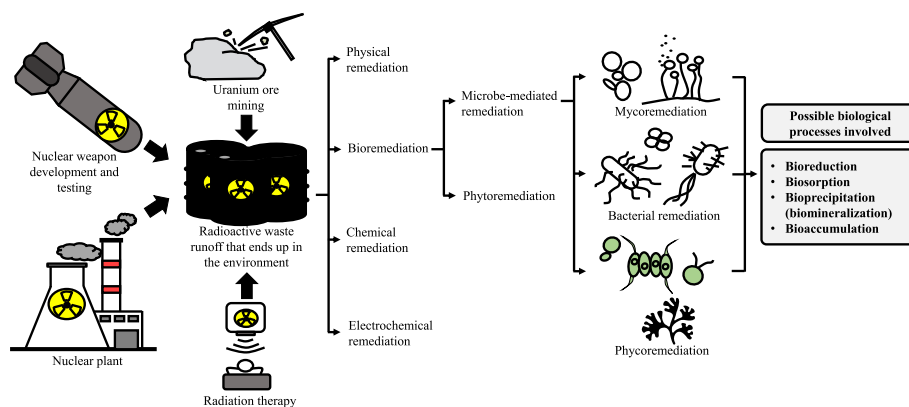


Fig. 4. The summary of radioactive remediation review.

liquid secondary wastes (Lv et al., 2021). However, these methods may not be as cost-effective as bioremediation approaches due to the costly materials required to perform electrochemical remediation (e.g., membranes used in electrodialysis) and the limited availability of low cost yet high-performance alternatives (Li et al., 2022). Additionally, the efficiency of certain methods, i.e., electrosorption, is strongly dependent on the applied voltage (Lee et al., 2022), implying the need for a substantial electrical input to achieve effective radionuclide removal.

All variations of radioactive waste remediation strategies contain specific techniques that allow the recovery of radionuclides from the radioactive waste, which were demonstrated in several studies. Physical methods include soil washing by dense medium and gravitational separation (Hong and Um, 2023), chemical methods involve the adsorption of radionuclides by synthetic adsorbents or modified natural adsorbents, like polyethylene polyamine/polydopamine-modified carboxylated chitosan aerogel (Huang et al., 2023), phosphate-based hyper-crosslinked polymer-dibenzyl phosphate (Yue et al., 2023), and amidoxime-based collagen fiber (Tang et al., 2023), and electrochemical methods such as electrochemical mineralization (Lv et al., 2021). Biological methods such as biosorption by microbes like *Trichoderma harzianum* (Akhtar et al., 2009) and phytoextraction by hyper-accumulator plants (Singh et al., 2022) have also been shown to allow radionuclide recovery from radioactive waste. Nevertheless, biological and certain chemical (particularly adsorption and precipitation) approaches are relatively simple to implement, as they do not necessitate the use of complex equipment or any sophisticated setup.

Table 8 summarizes the strengths and weaknesses associated with each radioactive waste treatment strategy. Each remediation approach cannot be fully replaced by others or get omitted, as each has its distinct strengths and weaknesses, making them suitable for application in different scenarios depending on factors including the amount and characteristic of radioactive waste to be treated, the suitability for the radioactive waste to be treated on- or off-site, and budgetary constraints. While microbe-mediated remediation is generally more environmentally friendly and cost-effective, processes like bioreduction and bioprecipitation by live microbial cells may be less effective in removing radionuclides when the radionuclides' concentrations are high because such conditions can suppress their growth and result in slower metabolism (Choramo, 2022) and even cell death. In such cases, besides biosorption by dead microbial cells, alternative approaches like chemical adsorption and physical separation techniques may be applied for more efficient radionuclide removal. By carefully considering the strengths and limitations of each approach, one can employ the most effective and appropriate methods for specific scenario, and may even co-apply multiple strategies to enhance the overall efficiency of the remediation process, as discussed in 5.3.

7. Conclusion and recommendations

This paper summarized the sources and classification of radioactive wastes and provided a brief overview of existing radionuclide remediation approaches, including physical, chemical, and electrochemical methods. Microbe-mediated bioremediation includes bacterial remediation, mycoremediation, and phytoremediation. Bacteria can remediate radioactive waste with various biological processes, including bio-reduction, biosorption, and bioprecipitation (biomineralization). In bioreduction, dissimilatory ferric iron-reducing, sulfate-reducing, and certain extremophilic bacteria can reduce water-soluble, mobile radionuclides to lower oxidation forms, which are water-insoluble and immobile, thereby restricting radionuclides' movement in water bodies. Knowing that electron donors play a vital role in kickstarting the bio-reduction process, various electron donors have also been added directly to radioactively contaminated sites to enrich indigenous ferric iron- and sulfate-reducing bacteria populations to initiate in situ remediation. Biosorption involves the association of radionuclides with functional groups on the bacterial cell surface or extracellular polymeric substances secreted by bacteria, where these radionuclides may then get reduced or precipitated extracellularly or intracellularly. Mycoremediation frequently involves biosorption, and its efficiency is affected by pH and cell concentration. In terms of phytoremediation, microalgae with the red cyst life form exhibited the highest radionuclide biosorption activity compared to other life forms, i.e., palmella and flagellated cell, possibly due to the high amount of astaxanthin it accumulated, which induces the expression of K^+ -transporting proteins that also actively take up other radionuclides like Cs^+ . Macroalgae-based phytoremediation was also briefly reviewed. Fig. 4 summarized the contents of this review.

The SWOT analysis of microbe-mediated remediation found that its strengths include being highly effective in removing radionuclides with diluted concentrations from radioactively polluted waters, generating small volumes of sewage sludge, low production and operation costs and not increasing the COD and BOD of treated water. Microbe-mediated remediation also contains drawbacks like shorter shelf lives, self-sustaining problems, and reduced efficiency in high radionuclide concentrations due to growth inhibition. It is suggested that 1) genetic modification of radiation-resistant microbes and 2) co-application of microbe-mediated remediation with other remediation strategies might lead to more opportunities for discovering more practical in situ and ex situ microbe-involving radionuclide removal applications. Besides, research regarding the interaction between indigenous microbial communities and foreign microbes to be introduced for remediation purposes and biological containment systems should be conducted to prevent the threats associated with microbial remediation, including decreased biodiversity due to native microbes being outcompeted and introduction of foreign genes via horizontal gene transfer that affects gene pool. It is recommended that more research that venture into the

genetic engineering of radiation-resistant microorganisms, co-application of microbial remediation with other remediation approaches, interaction between foreign radionuclide-remediating microbes and indigenous communities, and development and refinement of biological containment systems should be conducted to unlock greater potentials of microbe-mediated remediation while minimizing the potential risks that it may bring upon to the environment.

Additionally, a comparison between the strengths and limitations of each approach (physical, chemical, electrochemical, biological: microbe-mediated and plant-mediated) is also provided. By considering the pros and cons of each approach, it is possible to co-apply non-microbe-mediated remediation strategies with microbe-mediated methods when the radionuclide removal efficiency of microbe-mediated methods is limited by scenarios, like high radionuclide concentrations, to enhance the overall efficiency of the radionuclide remediation process.

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CRedit authorship contribution statement

Jin Ping Tan: Writing – original draft, Visualization, Conceptualization. **Christal Winona Clyde:** Writing – review & editing, Visualization, Conceptualization. **Chuck Chuan Ng:** Writing – review & editing, Validation, Supervision, Project administration, Funding acquisition, Conceptualization. **Swee Keong Yeap:** Supervision, Project administration, Funding acquisition. **Chean Yeah Yong:** Writing – review & editing, Supervision, Funding acquisition.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

No data was used for the research described in the article.

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