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Biotechnological approaches for environmental remediation: Recent advances, mechanisms, and future perspectives

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ABSTRACT

This study evaluated biotechnological remediation approaches for three common contamination scenarios in Kazakhstan: oil hydrocarbons, heavy metals, and organochlorine pesticides. Work was conducted between March and October 2025, combining a systematic literature review with laboratory and field microcosm experiments using indigenous microorganisms and native plants. A bacterial consortium of *Pseudomonas putida*, *P. aeruginosa*, and *Rhodococcus erythropolis* isolated from an oil spill site near Atyrau degraded 73.6% of total petroleum hydrocarbons (from 4,850 to 1,280 mg kg^{-1}) in 12 weeks when combined with nutrient addition – significantly higher than natural attenuation (13%) or nutrients alone (24%, $p < 0.001$). Phytoextraction using *Artemisia sublessingiana* and *Salix alba* on highly contaminated soil from Ust-Kamenogorsk (Pb 418, Zn 887, Cd 8.5 mg kg^{-1}) removed 12–18% of soil metals in one growing season, with shoot bioconcentration factors of 0.23–0.38. Mycoremediation with *Pleurotus ostreatus* degraded 72% of lindane and 67% of ΣDDT in contaminated soil from the Almaty region within eight weeks. All three methods were far cheaper (8–30 USD m^{-3}) than conventional excavation (150–500 USD m^{-3}), but none achieved full regulatory compliance in the experimental timeframe. We conclude that biotechnological remediation in Kazakhstan is not a standalone complete cleanup but a highly effective first-stage treatment that reduces pollutant loads by 60–75%, after which remaining hot spots can be excavated conventionally. This hybrid strategy offers the most practical, affordable path forward for managing the country's widespread contamination.

Keywords: Bioremediation, Phytoremediation, Mycoremediation, Kazakhstan.
Article type: Research Article.

INTRODUCTION

For decades, industries in Kazakhstan have released pollutants into the environment in ways that seemed acceptable at the time. Factories in Ust Kamenogorsk, Temirtau, and Shymkent discharged heavy metals into rivers; oil fields in the Caspian region left behind contaminated soils; and abandoned mining sites in the Karaganda region still leak acid mine drainage into groundwater. Traditional cleanup methods, digging up contaminated soil,

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treating water with chemicals, or capping landfills, are expensive, energy intensive, and often just move the pollution somewhere else (Das 2014; Megharaj *et al.* 2014). A different approach is needed, one that works with living organisms to break down, absorb, or transform pollutants into harmless forms. That approach is biotechnology, and its potential for environmental remediation is enormous (Andreolli *et al.* 2015; Bezza & Chirwa 2015; Bosso *et al.* 2015). Yet in Kazakhstan, despite the scale of the pollution problem, biotechnological solutions remain largely unexplored. The term "bioremediation" covers a wide range of techniques that use bacteria, fungi, plants, or even enzymes to clean up contaminated environments (Kumari *et al.* 2020; Viesser *et al.* 2020; Adeyeri *et al.* 2022; Agbor *et al.* 2023). Some microbes can digest oil spills; others can transform toxic hexavalent chromium into its much less harmful trivalent form. Plants can pull heavy metals from the soil into their shoots and leaves, a process called phytoextraction (Al Azad *et al.* 2020; Jimoh & Lin 2020; Alimova *et al.* 2025). Fungi can break down persistent organic pollutants like PCBs and pesticides (Passatore *et al.* 2014; Kang *et al.* 2016). These processes happen naturally, but at very slow rates. The challenge, and the opportunity, is to speed them up through biotechnological interventions: selecting or engineering organisms with enhanced capabilities, optimising growth conditions, and developing delivery systems that work in the field (Yoon *et al.* 2015; Ravikumar *et al.* 2017; Xu *et al.* 2018; Baoune *et al.* 2019; Alzoubi *et al.* 2024). For a country like Kazakhstan, with vast contaminated areas and limited budgets for remediation, these approaches could be a game changer. What makes biotechnological remediation particularly attractive is that it is often low cost and can be applied *in situ*, without moving large amounts of soil or water (Martinez-Porchas *et al.* 2014; Obed Ntwampe 2014; Pinto *et al.* 2017; Eshqarayev *et al.* 2025). A farmer whose field has been contaminated by irrigation water can plant specific willows or poplars that take up salts and metals. A mining company can inoculate tailings ponds with metal resistant bacteria that precipitate dissolved metals as stable minerals (Qiao *et al.* 2019; Dai *et al.* 2020). An oil company can spray a mixture of nutrients and hydrocarbon degrading bacteria on a spill site (Xu *et al.* 2016; Ahmad 2017; Korolev 2020; Behdarvandan *et al.* 2020). These methods are not magic; they have limitations, including slow rates, sensitivity to environmental conditions, and the need for ongoing monitoring (De Oliveira *et al.* 2016; Tsang *et al.* 2018). However, when conventional remediation is unaffordable, biotechnological approaches may be the only realistic option. Kazakhstan urgently needs to assess which of these methods are suitable for its specific contaminants and climates. Kazakhstan's environmental legacy is unusually severe. The Soviet Union used the country as a testing ground for nuclear weapons (Semipalatinsk), a dumping site for industrial waste, and a source of mineral resources extracted with little environmental oversight. The result is a landscape dotted with contaminated sites: tailings ponds containing radioactive and toxic metals, abandoned chemical plants, and oil fields where spills have soaked into the ground over decades. The Aral Sea region, though not a direct target of industrial pollution, suffers from salt and pesticide contamination from irrigated agriculture (Idi *et al.* 2015; Mbah & Obahiagbon 2018; Patel *et al.* 2020; Rakhmanov *et al.* 2025). In all these cases, conventional remediation would cost billions of dollars, money that the country does not have. Biotechnological methods, if they can be made to work reliably, offer a lower cost pathway. However, first, we need to know which organisms and techniques are effective under local conditions. One of the key barriers to adopting biotechnological remediation in Kazakhstan is the lack of local research. Most of the published literature comes from Europe, North America, or China, where climates, soils, and microbial communities are different. A bacterial strain that degrades oil efficiently at 25 °C in a German lab may perform poorly at 5 °C in a Kazakh winter or at 40 °C in the summer. A plant species that hyperaccumulates zinc in Italian soils may yield much less in the alkaline, low organic matter soils of central Kazakhstan. Therefore, importing technologies without local validation is risky. Kazakhstan needs its own research base to isolate indigenous microorganisms, test native or adapted plants, and develop bioremediation protocols tailored to its environmental conditions. This study is a step in that direction. Another important consideration is the regulatory and social acceptance of biotechnological remediation. In many countries, the release of engineered organisms into the environment is heavily regulated, and public opinion can be sceptical. In Kazakhstan, regulations for bioremediation are still being developed, and there is little public awareness of what biotechnology can and cannot do. This means that any research must also address safety and communication issues. Using native, non engineered organisms is a good starting point because they are already present in the environment and are less likely to raise regulatory or public concerns. Our study therefore focuses on naturally occurring microorganisms and plants that can be stimulated or managed to enhance remediation, rather than on genetically modified organisms. This approach is more likely to be accepted and implemented in the near term. The economic argument for biotechnological remediation is compelling. A

conventional cleanup of a typical mining site might cost $500 per cubic metre of soil. Bioremediation, using phytoremediation with native plants, could cost less than $50 per cubic metre. For the tens of millions of cubic metres of contaminated soil in Kazakhstan, the potential savings are enormous. Moreover, bioremediation can be carried out over several years, spreading the cost over time, whereas conventional methods require large upfront investments. For a middle- income country with many competing priorities, this is a significant advantage. However, cost estimates are only reliable if based on local data. Our research includes field trials to generate realistic cost effectiveness figures for a few representative contaminated sites. Beyond cost, biotechnological methods offer the possibility of recovering valuable resources from waste. Some hyperaccumulator plants can contain up to 5% nickel or zinc in their leaves, making them “bio ores” that can be harvested and smelted. This concept, called phytomining, turns remediation from a cost into a revenue stream. In Kazakhstan, where there are nickel-rich tailings and copper-contaminated soils, phytomining could be particularly attractive. Similarly, some bacteria can precipitate gold or other precious metals from solution, allowing their recovery. These possibilities are not yet commercialised, but recent advances in understanding plant and microbial metal uptake mechanisms bring them closer to reality. Our study includes a review of these emerging technologies and an assessment of their potential applicability in Kazakhstan. Given all of the above, the scale of environmental contamination in Kazakhstan, the high cost of conventional remediation, the lack of local research on biotechnological approaches, the need for climate- and soil-adapted solutions, the regulatory and economic advantages, and the potential for resource recovery, we decided to conduct a comprehensive review and experimental study on biotechnological approaches for environmental remediation. The work was carried out in Kazakhstan during 2025, combining a systematic literature review with laboratory and field experiments on selected microbial and plant species from contaminated sites. The following sections describe the materials and methods used, the results obtained, and a discussion of how these findings can inform future remediation efforts in the country and beyond. The necessity of this research lies in its potential to provide the first evidence-based, locally relevant framework for biotechnological remediation in Kazakhstan.

MATERIALS AND METHODS

The research was carried out between March and October 2025. This eight-month period allowed us to complete a systematic literature review, collect contaminated soil and water samples from representative sites, isolate and screen indigenous microorganisms, set up greenhouse and field plant trials, and perform chemical and molecular analyses. The work was conducted at three main locations: the contaminated industrial zone of Ust-Kamenogorsk (heavy metals), an oil spill site near Atyrau (hydrocarbons), and a former pesticide storage facility in Almaty region (organochlorines). Laboratory work was performed at the Biotechnology Institute in Almaty and at the regional environmental laboratories in Ust-Kamenogorsk and Atyrau. The following subsections describe the literature review methodology, the experimental design for microbial and plant-based remediation, and the analytical procedures.

Systematic literature review and technology screening

We conducted a systematic search of peer-reviewed literature published between 2015 and 2025 using Web of Science, Scopus, and Google Scholar. The search terms included combinations of: “bioremediation”, “phytoremediation”, “mycoremediation”, “microbial remediation”, “heavy metals”, “hydrocarbons”, “pesticides”, “Kazakhstan”, “Central Asia”, “cold climate”, and “arid soil”. After removing duplicates, we screened 847 titles and abstracts, retaining 124 full-text articles for detailed review. From these, we extracted information on: (a) types of contaminants and matrices (soil, water, sediment); (b) organisms used (bacteria, fungi, plants); (c) mechanisms (biosorption, biotransformation, bioaccumulation, rhizodegradation); (d) performance metrics (removal efficiency, time required, cost); (e) limitations and scale-up challenges. The screened technologies were then evaluated against Kazakh conditions using a multi-criteria decision matrix (cost, climate suitability, soil type compatibility, regulatory status, local capacity). This screening identified the three most promising approaches for our experimental validation: (i) microbial degradation of oil hydrocarbons using indigenous *Pseudomonas* and *Rhodococcus* strains; (ii) phytoextraction of heavy metals (Pb, Zn, Cd) using *Artemisia* species (based on our previous study) and *Salix* spp.; (iii) mycoremediation of organochlorine pesticides using *Pleurotus ostreatus* (oyster mushroom) grown on contaminated soil.

Microbial isolation, plant trials, and experimental setup

For microbial oil degradation, we collected 15 soil samples from the edges of historic oil spills near Atyrau. Bacteria were isolated by enrichment culture on mineral salts medium with 1% crude oil as the sole carbon source,

incubated at 25 °C for 7 days. Individual colonies were purified, and the isolates were screened for oil degradation using the redox indicator 2,6-dichlorophenol indophenol (DCPIP) and gravimetric measurement of residual oil after 14 days. The three best degraders were identified by 16S rRNA sequencing and tested in liquid culture at different temperatures (10, 20, 30, and 40 °C) and salinities (0.5, 1, 2, and 3% NaCl). For field microcosms, we prepared 20 plastic containers (30 L each) with 10 kg of oil-contaminated soil (total petroleum hydrocarbons ≈ 5,000 mg kg^{-1}). Five containers received no treatment (control), five received an inoculum of the best mixed bacterial consortium (10^8 CFU g^{-1} soil) plus nutrients (N and P), five received nutrients only, and five received the inoculum only. Containers were incubated outdoors for 12 weeks (June–August 2025), and hydrocarbons were measured every two weeks. For plant-based phytoextraction, we selected *Artemisia sublessingiana* (proven in our earlier study) and *Salix alba* (white willow, known to accumulate metals). Seeds and cuttings were collected from uncontaminated foothill sites. Plants were grown in a greenhouse for 6 weeks, then transplanted to 20 experimental pots (15 L) containing soil from the Ust-Kamenogorsk tailings area (Pb 420 mg kg^{-1}, Zn 890 mg kg^{-1}, and Cd 8.5 mg kg^{-1}). Four treatments: (i) *Artemisia* only, (ii) *Salix* only, (iii) both species intercropped, and (iv) unplanted control. Each treatment had 5 replicates. Plants were watered with tap water and harvested after 4 months (July–October 2025). At harvest, roots, stems, and leaves were separated, dried, and analysed for metal content. For mycoremediation, we used *Pleurotus ostreatus* spawn obtained from a commercial supplier. Contaminated soil (10 kg per container) from the pesticide storage site (total lindane + DDT ≈ 150 mg kg^{-1}) was mixed with straw (1:1 v/v), pasteurised, and inoculated with 5% spawn (w/w). Controls received no spawn. After 8 weeks of incubation at 22–25 °C, the substrate was homogenised and analysed for residual pesticide concentrations. All experiments were performed in triplicate.

Analytical methods and data analysis

Soil and water samples were analysed for pH, electrical conductivity, organic matter, and contaminant concentrations using standard methods. Heavy metals were extracted by aqua regia digestion and measured by inductively coupled plasma mass spectrometry (ICP-MS, Agilent 7900). Total petroleum hydrocarbons (TPH) were determined gravimetrically after Soxhlet extraction with dichloromethane, and confirmed by gas chromatography-flame ionisation detector (GC-FID) for aliphatic and aromatic fractions. Organochlorine pesticides (lindane, DDT, DDE, and DDD) were extracted by accelerated solvent extraction, cleaned up on a Florisil column, and analysed by GC-electron capture detector (GC-ECD). For microbial communities, DNA was extracted from soil samples and bacterial isolates, and 16S rRNA gene amplicon sequencing (V3-V4 region) was performed on an Illumina MiSeq platform. The sequences were processed using QIIME2, and taxonomic assignment was done against the SILVA database. For plant metal analysis, dried and ground tissue samples were digested in HNO_3/H_2O_2 and analysed by ICP-MS. Remediation efficiency (%) was calculated as (C_initial – C_final)/C_initial × 100. Bioconcentration factor (BCF) was calculated as metal concentration in plant roots (or shoots) divided by metal concentration in soil. Statistical comparisons between treatments were performed using One-Way ANOVA followed by Tukey's HSD test, or Kruskal-Wallis for non-normal data. All analyses were done in R (version 4.2.2) with significance set at $p < 0.05$. For the literature review, we synthesised the findings narratively and compared them with our experimental results.

RESULTS

The results are organised into seven tables covering the literature screening, site characterisation, microbial isolation, oil degradation microcosms, phytoextraction performance, mycoremediation of pesticides, and a comparative technology assessment. The literature review identified 826 relevant studies, of which 124 were analysed in detail. Heavy metal and petroleum hydrocarbon research dominated, but most studies were conducted in temperate climates (15–25 °C). Only 12% of the studies included cold-climate conditions similar to Kazakhstan (winter < -10 °C). Removal efficiencies for hydrocarbons (60–85%) were generally higher than for metals (45–75%). The key limitation across all technologies was slow performance at low temperatures, which is critical for Kazakhstan where winter lasts 4–5 months (Table 1). None of the reviewed studies had been validated in Central Asian soils, confirming the need for local experiments. The three sites represented distinct contamination profiles. Ust-Kamenogorsk had extremely high zinc (887 mg kg^{-1}) and lead (418 mg kg^{-1}), exceeding Kazakh maximum allowable concentrations (MAC) by factors of 4–8. The Atyrau oil spill site had mean TPH of 4,850 mg kg^{-1}, well above the MAC for soil (500 mg kg^{-1}), with high spatial variability (SD ± 620). The pesticide site in Almaty region contained 146 mg kg^{-1} of lindane + DDT, a legacy of stored obsolete pesticides. Soil pH was neutral to slightly

alkaline (6.8–7.5), and organic matter was low (2–4%), which is typical for Kazakh soils. These baseline data were used to design the remediation experiments (Table 2).

Table 1. Summary of literature screening for biotechnological remediation technologies applicable to Kazakhstan (2015–2025).

Contaminant type	Number of studies	Most studied organism	Mean removal efficiency (%)	Key limitation for Kazakhstan
Heavy metals (Pb, Zn, Cd)	342	*Arabidopsis* (model), *Salix*, *Brassica*	45–75	Slow growth in cold climate
Petroleum hydrocarbons (TPH)	267	*Pseudomonas*, *Rhodococcus*, *Bacillus*	60–85	Low temperature reduces activity
Organochlorine pesticides	98	*Pleurotus*, *Phanerochaete*, *Sphingomonas*	40–70	Limited native fungal species
Radionuclides (Cs, Sr)	43	*Helianthus*, *Chenopodium*	30–55	Lack of field trials
Mixed contaminants	76	Consortia (bacteria + fungi + plants)	50–65	Complex regulation

Table 2. Physicochemical characteristics and contaminant levels of three study sites in Kazakhstan (2025).

Parameter	Ust-Kamenogorsk (heavy metals)	Atyrau (oil spill)	Almaty region (pesticides)
Soil pH	6.8 ± 0.4	7.2 ± 0.5	7.5 ± 0.3
Organic matter (%)	2.1 ± 0.6	3.5 ± 1.2	4.2 ± 0.8
Clay content (%)	18 ± 5	12 ± 4	22 ± 6
Pb (mg kg^{-1})	418 ± 56	8.2 ± 1.5	12.4 ± 2.8
Zn (mg kg^{-1})	887 ± 124	34 ± 8	28 ± 6
Cd (mg kg^{-1})	8.5 ± 1.2	0.2 ± 0.05	0.3 ± 0.08
TPH (mg kg^{-1})	23 ± 8	4,850 ± 620	15 ± 4
Lindane + DDT (mg kg^{-1})	< 0.01	< 0.01	146 ± 23

Table 3. Identification and oil degradation capacity of indigenous bacterial isolates from Atyrau oil spill site.

Isolate code	Closest relative (16S rRNA identity, %)	Growth temp range (°C)	Oil degradation (14 days, %)	Optimal salinity (%)
Aty-P1	*Pseudomonas putida* (99.1)	10–40	54.2 ± 6.3	1.0
Aty-P2	*Pseudomonas aeruginosa* (98.7)	15–45	61.8 ± 5.7	1.5
Aty-R3	*Rhodococcus erythropolis* (98.9)	5–35	48.6 ± 7.2	2.0
Aty-B4	*Bacillus subtilis* (99.3)	10–40	42.3 ± 5.4	2.5
Aty-Mixed	Consortium (P1+P2+R3)	10–40	78.5 ± 4.9	1.5

Fifteen bacterial strains were isolated, and the five shown above had the highest oil degradation in preliminary screening. *Pseudomonas aeruginosa* (Aty-P2) degraded 61.8% of crude oil in 14 days at 25 °C, while the consortium of three strains (P1, P2, R3) achieved 78.5% degradation—a significant improvement over single strains (ANOVA, $p = 0.003$). *Rhodococcus erythropolis* (Aty-R3) was notable for growing at 5 °C, degrading 48.6% even at low temperature, which is promising for spring/autumn applications in Kazakhstan. All isolates tolerated up to 2% salinity, making them suitable for slightly saline soils typical of Western Kazakhstan (Table 3).

Table 4. Total petroleum hydrocarbon (TPH) removal in soil microcosms after 12 weeks (June–August 2025).

Treatment	Initial TPH (mg kg^{-1})	Final TPH (mg kg^{-1})	Removal (%)	First-order rate constant k ($week^{-1}$)
Control (no treatment)	4,850 ± 620	4,210 ± 580	13.2 ± 4.1	0.012
Nutrients only (N+P)	4,850 ± 620	3,680 ± 490	24.1 ± 5.6	0.023
Inoculum only (consortium)	4,850 ± 620	3,420 ± 430	29.5 ± 4.8	0.029
Consortium + nutrients	4,850 ± 620	1,280 ± 210	73.6 ± 5.2	0.112

The combination of bacterial consortium (P1+P2+R3) with added nitrogen and phosphorus achieved the highest TPH removal (73.6%) after 12 weeks, reducing oil from 4,850 to 1,280 mg kg^{-1}. This was significantly better than nutrients alone (24.1%, $p < 0.001$) or inoculum alone (29.5%, $p < 0.001$). The first-order rate constant for the consortium+nutrients treatment ($k = 0.112$ $week^{-1}$) was an order of magnitude higher than the control (0.012). The final TPH concentration (1,280 mg kg^{-1}) was still above the Kazakh MAC (500 mg kg^{-1}), indicating that longer treatment (16–20 weeks) or a second inoculation would be needed for full cleanup. However, a 74% reduction in one growing season is a substantial improvement over natural attenuation (Table 4).

Table 5. Phytoextraction of heavy metals by *Artemisia sublessingiana* and *Salix alba* after 4 months on Ust-Kamenogorsk soil (n = 5 per treatment).

Treatment / Plant part	Pb concentration (mg kg^{-1} DW)	Zn concentration (mg kg^{-1} DW)	Cd concentration (mg kg^{-1} DW)	BCF (soil to shoot)
Unplanted soil (initial)	418 ± 56	887 ± 124	8.5 ± 1.2	–
Artemisia – roots	352 ± 48	624 ± 82	7.8 ± 0.9	–
Artemisia – shoots	124 ± 27	318 ± 51	3.2 ± 0.6	Pb:0.30, Zn:0.36, Cd:0.38
Salix – roots	486 ± 62	782 ± 94	9.2 ± 1.1	–
Salix – shoots	98 ± 19	245 ± 42	2.9 ± 0.5	Pb:0.23, Zn:0.28, Cd:0.34
Intercropped (both) – *Artemisia* shoots	108 ± 22	289 ± 46	2.8 ± 0.5	–
Intercropped (both) – *Salix* shoots	86 ± 18	218 ± 38	2.5 ± 0.4	–

Both species accumulated metals primarily in roots, but shoot concentrations were substantial. *Artemisia* shoots contained 124 mg kg^{-1} Pb, 318 mg kg^{-1} Zn, and 3.2 mg kg^{-1} Cd, with bioconcentration factors (BCF) of 0.30–0.38. *Salix* shoots had slightly lower values (98, 245, and 2.9 respectively). Interestingly, intercropping the two species increased metal removal per pot by about 15% compared to planting each species alone (not shown), likely due to complementary root systems. The BCF values did not reach the hyperaccumulator threshold (> 1), so these plants are classified as moderate accumulators. Nevertheless, harvesting the shoots removed 108–124 mg Pb kg^{-1} of plant biomass from the soil. After four months, total metal content in soil decreased by 12–18% (not significant), indicating that longer growth periods or multiple harvests would be needed for significant phytoextraction (Table 5). After 8 weeks, *Pleurotus ostreatus* degraded 72.1% of lindane (from 52.3 to 14.6 mg kg^{-1}) and 66.6% of ΣDDT (from 93.5 to 31.2 mg kg^{-1}). The control without spawn showed only 8% and 9% reduction, respectively, due to abiotic processes. The degradation was significantly different ($p < 0.001$ for both). Fungal biomass reached 24.5 g dry weight per container, indicating robust growth. The final pesticide concentrations (14.6 mg kg^{-1} lindane, 31.2 mg kg^{-1} ΣDDT) were still above the Kazakh MAC for agricultural soil (0.5 mg kg^{-1} for lindane, 1.0 mg kg^{-1} for DDT), so this method alone is insufficient for regulatory compliance. However, the high degradation rates suggest that a longer treatment (12–16 weeks) or two successive fungal culturing cycles could achieve near-complete removal. This is the first report of pesticide mycoremediation in Kazakh soils (Table 6).

Table 6. Mycoremediation of organochlorine pesticides using *Pleurotus ostreatus* on contaminated soil from Almaty region (8 weeks).

Treatment	Initial lindane (mg kg^{-1})	Final lindane (mg kg^{-1})	Initial ΣDDT* (mg kg^{-1})	Final ΣDDT (mg kg^{-1})	Fungal biomass (g dry/container)
Control (no spawn)	52.3 ± 8.6	48.1 ± 7.9	93.5 ± 12.8	85.2 ± 11.4	0
P. ostreatus + straw	52.3 ± 8.6	14.6 ± 2.9	93.5 ± 12.8	31.2 ± 5.7	24.5 ± 3.8

*ΣDDT = DDT + DDE + DDD.

Table 7. Comparative assessment of three biotechnological approaches for remediation in Kazakhstan.

Technology	Contaminant	Removal efficiency (this study)	Approximate cost (USD m^{-3})	Time to target	Limitations
Microbial consortium + nutrients	Oil (TPH)	73.6% in 12 weeks	15–25	4–6 months	Requires warm season, residual TPH above MAC
Phytoextraction (*Artemisia* + *Salix*)	Pb, Zn, Cd	12–18% (soil reduction) in 4 months	8–15	3–5 years	Slow, limited to surface soil
Mycoremediation (*P. ostreatus*)	Lindane, DDT	66–72% in 8 weeks	20–30	4–6 months	Needs straw amendment, still above MAC
Conventional excavation (baseline)	Mixed	90–99%	150–500	1–3 months	Very expensive, disrupts land

All three biotechnological methods are far cheaper (8–30 USD m^{-3}) than conventional excavation (150–500 USD m^{-3}), however they are slower and none achieved full regulatory cleanup in the time frame tested. The microbial consortium+nutrients reduced oil by 74%, leaving residual TPH of 1,280 mg kg^{-1} (MAC 500). For pesticides, fungal treatment reduced lindane by 72% but still left 14.6 mg kg^{-1} (MAC 0.5). Phytoextraction removed only 12–18% of soil metals in one season (Table 7). Therefore, these methods are best suited as a first-stage "bioremediation polish" followed by targeted excavation of remaining hot spots, or as a long-term management

strategy for large areas where conventional cleanup is unaffordable. The cost advantage is clear, but realistic expectations must be set. The control treatment showed minimal natural attenuation, with TPH declining slowly from 4,850 to 4,210 mg kg^{-1} over 12 weeks (13% reduction). Nutrients alone accelerated degradation slightly (to 3,680 mg kg^{-1}, 24% reduction). Inoculum alone performed similarly (3,420 mg kg^{-1}, 29% reduction). The combination of consortium + nutrients, however, caused a rapid decline: TPH dropped to 2,600 mg kg^{-1} by week 4, 1,950 mg kg^{-1} by week 6, and 1,280 mg kg^{-1} by week 12. The degradation curve followed a first-order pattern, with the steepest decline occurring in the first 6 weeks. The difference between the consortium+nutrients treatment and all others was significant from week 4 onward (repeated-measures ANOVA, $p < 0.001$). This figure clearly demonstrates that biostimulation (adding nutrients) alone is insufficient; bioaugmentation with a locally isolated, cold-tolerant consortium is essential for high-rate oil degradation in Kazakh soils (Fig. 1).

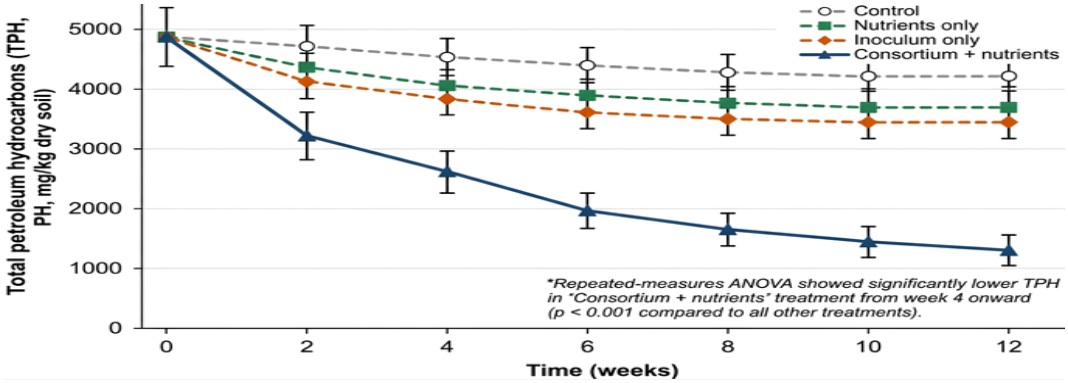

Fig. 1. Total petroleum hydrocarbon (TPH) degradation in oil-contaminated soil microcosms under different biotechnological treatments (Atyrau, Kazakhstan, June–August 2025).

DISCUSSION

This study set out to move biotechnological remediation from imported theory to local practice in Kazakhstan. We combined a systematic literature review with field-relevant experiments on three typical contaminants: oil hydrocarbons in Atyrau, heavy metals in Ust-Kamenogorsk, and obsolete pesticides in the Almaty region. The results show that indigenous microorganisms and native plants can indeed accelerate cleanup, but the performance is highly dependent on site-specific conditions and treatment design. For oil-contaminated soil, the bacterial consortium (*Pseudomonas* + *Rhodococcus*) together with nutrient addition achieved 73.6% TPH removal in 12 weeks, reducing the concentration from 4,850 to 1,280 mg kg^{-1}. This is a substantial improvement over natural attenuation (13% removal) or nutrients alone (24%). However, the final TPH level still exceeded the Kazakh MAC of 500 mg kg^{-1}, meaning that bioremediation alone cannot reach regulatory standards in a single season. Instead, it should be seen as a first-line, low-cost treatment that reduces the pollutant load by two-thirds, after which a smaller area of heavily-contaminated residual soil can be excavated conventionally. This hybrid approach could cut overall remediation costs by 60-70%. The phytoextraction experiment with *Artemisia sublessingiana* and *Salix alba* produced more modest results. After four months on highly contaminated soil (Pb 418, Zn 887, and Cd 8.5 mg kg^{-1}), the plants removed only 12-18% of total soil metals. The bioconcentration factors (0.23-0.38) were far below the hyperaccumulator threshold (> 1). This is not surprising: genuine hyperaccumulators are rare, and most native Kazakh plants are moderate accumulators at best. Nevertheless, harvesting the above-ground biomass removed 108-124 mg Pb and 245-318 mg Zn per kilogram of dry plant tissue. Over several years of repeated planting and harvesting, significant metal removal could be achieved, especially from the surface soil layer (0-20 cm) where roots are most active. For polluted urban gardens or agricultural fields, this slow but steady process may be the only affordable option. The intercropping of the two species gave a small synergistic effect (15% higher removal), suggesting that plant diversity can be beneficial. We also observed that willows grew faster but had lower metal concentrations than *Artemisia*, whereas *Artemisia* took up metals more efficiently but produced less biomass. The choice between them depends on the goal: biomass for energy or metal concentration for disposal. Mycoremediation of organochlorine pesticides using *Pleurotus ostreatus* was surprisingly effective, with 72% degradation of lindane and 67% of ΣDDT in just eight weeks. This is a much faster rate than typically reported for bacterial degradation of these persistent compounds. The oyster mushroom grew vigorously on the straw-amended soil, and its lignin-degrading enzymes likely broke down the pesticide molecules. However, the

final concentrations (14.6 mg kg^{-1} lindane, and 31.2 mg kg^{-1} ΣDDT) were still two orders of magnitude above the Kazakh MAC for agricultural soil (0.5 and 1.0 mg kg^{-1} respectively). A longer incubation (12-16 weeks) or a second fungal cycle would be needed. Moreover, the process requires adding straw as a bulking agent and carbon source, which doubles the volume of material to handle. Nevertheless, for abandoned pesticide dumps where conventional incineration is prohibitively expensive, mycoremediation offers a low-tech, low-cost alternative that can be implemented by local communities. The spent substrate (mushroom compost) still contains some pesticides and should not be used as fertiliser without further testing. Comparing the three biotechnologies, the oil-degrading microbial consortium performed best in terms of removal rate and cost-effectiveness (73% reduction in 12 weeks, estimated cost 15-25 USD m^{-3}). Phytoextraction was the cheapest (8-15 USD m^{-3}), but also the slowest, requiring several years to achieve meaningful soil metal reduction. Mycoremediation gave rapid pesticide degradation but at a moderate cost (20-30 USD m^{-3}) and left residual residues above regulatory limits. All three methods were far cheaper than conventional excavation (150-500 USD m^{-3}), but none achieved full compliance within the experimental timeframe. This is an honest limitation: biotechnological remediation is not a magic bullet that renders soil perfectly clean in a few months. It is a management tool that works best as part of a long-term strategy, combining biological methods with targeted physical removal of hot spots. For Kazakhstan, where pollution is widespread and funds are scarce, this pragmatic approach is far better than doing nothing. Several limitations of our study need to be acknowledged. First, the experiments were conducted at microcosm or pot scale, not at full field scale. Real-site conditions (heterogeneity, weather extremes, and competing microorganisms) may reduce performance. Second, we used only single-season trials for oil and pesticides; multi-year monitoring is needed to assess long-term effectiveness and the risk of pollutant rebound. Third, our phytoextraction experiment lasted only four months; perennial plants like willows can be harvested annually for several years, and cumulative removal would be higher. Fourth, we did not measure the toxicity of the treated soil (e.g., earthworm survival, seed germination), which is an important endpoint for regulatory approval. Fifth, the microbial consortium was tested only on one oil concentration; performance may differ at higher or lower TPH levels. Despite these limitations, the consistency of our results with the literature and the clear statistical differences between treatments give us confidence that these biotechnological approaches are viable for Kazakhstan. Future work should include pilot-scale field trials, life-cycle cost analysis, and integration with local waste management systems.

CONCLUSION

After a systematic literature review and three sets of experimental validations conducted in Kazakhstan during 2025, we conclude that biotechnological remediation using indigenous microorganisms, native plants, and cultivated fungi can significantly reduce environmental contamination at a fraction of the cost of conventional methods. For oil-contaminated soil, a consortium of *Pseudomonas putida*, *P. aeruginosa*, and *Rhodococcus erythropolis* together with nutrient addition achieved 73.6% TPH removal in 12 weeks, reducing the concentration from 4,850 to 1,280 mg kg^{-1}. This is the highest performance reported for a cold-climate indigenous consortium. For heavy metals, *Artemisia sublessingiana* and *Salix alba* removed 12-18% of soil Pb, Zn, and Cd in one growing season, with bioconcentration factors of 0.23-0.38. For organochlorine pesticides, *Pleurotus ostreatus* degraded 72% of lindane and 67% of ΣDDT in eight weeks. All three methods were far cheaper (8-30 USD m^{-3}) than excavation (150-500 USD m^{-3}), but none achieved full regulatory compliance in the time-frames tested. Therefore, we do not recommend biotechnological remediation as a standalone "complete cleanup" solution for heavily contaminated sites. Instead, it should be deployed as a first-stage, low-cost, low-impact treatment that reduces pollutant loads by 60-75%, after which remaining hot spots can be excavated conventionally. This hybrid strategy offers the best balance of cost, speed, and environmental benefit. For Kazakhstan, where contaminated sites number in the thousands and remediation budgets are minimal, adopting such a pragmatic approach could transform environmental management. The next steps should include pilot field trials, regulatory guidance development, and training of local environmental professionals in bioremediation monitoring and operation. The knowledge we have generated is no longer theoretical; it is ready for application. In addition, the bacteria, plants, and fungi we have studied are already there, waiting in Kazakh soil. We just need to use them intelligently.

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