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Microbiological methods for removing heavy metals from saline water‑based drilling mud: challenges and practical strategies

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Abstract

This study highlights the potential of *Aquibacillus halophilus* and microbial treatments to address water quality issues in drilling operations, ofering promising avenues for mitigating heavy metal contamination and reducing total water hardness to achieve sustainable development goal (SDG) 6 (Clean Water and Sanitation) and SDG 14 (Life Below Water). *Aquibacillus halophilus* exhibited rapid growth and remarkable water quality enhancement capabilities. Its robust growth at pH 7 suggests minimal interference from in situ bacteria, thereby preserving the optimal pH level for drilling operations and promoting sustainable water resource management. Keeping the total hardness below 800 ppm is essential to utilize water in well-logging effectively. *Aquibacillus halophilus* offers an alternative approach to hardness reduction that reduces reliance on chemical additives such as caustic soda. By incorporating *Aquibacillus halophilus*, a conventional treatment requiring 100% caustic soda to treat 19,100 ppm hardness, it can be reduced to 19.37%, obtaining a remarkable 80% reduction in material consumption. This reduction facilitates wastewater reuse significantly, promotes resource efficiency, and is consistent with SDG 6. In addition to reducing well-logging costs, the microbial technique safeguards the environment by addressing heavy metal contamination, which aligns with SDG 14's objective of protecting aquatic life.

Keywords Bioremediation · Heavy metal removal · Hawizeh Marshes · Total hardness · Sustainable development goal · Water-based drilling mud

Introduction

The efficacy of the drilling operation is highly dependent on the drilling fuid, which accounts for 15–18% of the total costs associated with oil well drilling (Ytrehus et al. [2023](#page-10-0)). Based on their chemical properties and phase type, drilling fuids are typically classifed as water-based or oil-based (Karakosta et al. [2021](#page-9-0); Alkalbani and Chala [2024\)](#page-7-0). Various compounds, polymers, and additives are combined with drilling mud to maintain properties such as mud weight, gel strength, viscosity, and fltration (Inemugha et al. [2019](#page-9-1);

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Abed and Rasaei [2024](#page-7-1); Davoodi et al. [2024](#page-8-0); Li et al. [2024](#page-9-2)). The pH range for water-based mud (WBM) drilling is typically between 7 and 12 but may vary depending on well conditions (Blkoor et al. [2021\)](#page-8-1). It is essential to maintain the proper pH to prevent chemical reactions that can contribute to corrosion and ensure clay stabilization (Ahmed et al. [2021\)](#page-7-2). The pH level has a direct impact on the solubility, reactivity, and efectiveness of various additives used in drilling mud formulations (Bardhan et al. [2024](#page-8-2); Fadairo and Oni [2024](#page-8-3)). Deviations from the optimal pH range can impair the performance of these additives, potentially resulting in reduced drilling fuid properties and compromised wellbore stability (Kong et al. [2024;](#page-9-3) Zhendong et al. [2024](#page-11-0)). Therefore, maintaining the appropriate pH in drilling mud additives is critical. Inorganic compounds, especially heavy metals, in the water that return from the oil well to separate cuttings are considered an environmental risk (Hu et al. [2024](#page-9-4)). The concern stems from the potential for these inorganic compounds to have negative ecological and human health consequences due to their persistence and inherent toxicity (Gautam et al. [2023](#page-8-4); Sharma et al. [2023;](#page-10-1) Verma et al. [2023](#page-10-2);

Wu et al. [2024](#page-10-3)). Heavy metals, such as lead, cadmium, and mercury, have the ability to accumulate in aquatic ecosystems, resulting in long-term environmental consequences (Brindhadevi et al. [2023\)](#page-8-5). The discharge of water containing elevated levels of heavy metals into natural water bodies endangers aquatic organisms and, as a result, disrupts the ecological balance. Furthermore, heavy metals can bioaccumulate in the food chain, eventually reaching human populations via consuming contaminated aquatic organisms (Dong and Li [2024](#page-8-6); Saidon et al. [2024](#page-10-4)). The intricate interplay of inorganic compounds, particularly heavy metals, highlights the importance of stringent environmental management practices and regulatory frameworks to mitigate the potential negative impacts associated with them (Edo et al. [2024](#page-8-7)).

Several methods can be used to recover and remove heavy metals from polluted environments. These techniques include oxidation and reduction methods, various flters, electrochemical techniques, evaporation, ion exchange, demineralization, and deionization (Alkhadra et al. [2022](#page-7-3); Balasubramaniam et al. [2022;](#page-8-8) Antony et al. [2024](#page-7-4); Tang et al. [2024](#page-10-5)). The low cost and signifcant ecological benefts are two of the most signifcant advantages of microbial methods for managing the concentration of heavy metals in polluted environments (Kurella et al. [2023](#page-9-5); Tedesco et al. [2024\)](#page-10-6). The use of microbial methods completely depends on the situation of a polluted environment, which leads to using in-situ and ex-situ bioremediation methods (Ciampi et al. [2024](#page-8-9); Yuan et al. [2024\)](#page-10-7).

Advanced techniques such as atomic absorption spectrometry (AAS) and inductively coupled plasma mass spectrometry (ICP-MS) aid in detecting trace amounts of heavy metals in biological and environmental samples (Halmi et al. [2019](#page-8-10); Jenikova et al. [2022](#page-9-6)).

Bacteria can readily absorb minerals due to the structure of their cell surface. During ion absorption, various elements, including nitrogen, oxygen, sulfur, and phosphorus, may also be absorbed into bacteria's cell walls (Mu et al. [2021;](#page-10-8) Baek et al. [2022\)](#page-7-5). In most instances, the absorption of nitrogen, oxygen, sulfur, and phosphorus has no direct efect on the absorption of heavy metals. These elements' absorbing mechanisms and pathways are distinct and independent. Nitrogen, oxygen, sulfur, and phosphorus are essential nutrients for biological systems and are typically absorbed by plants, microorganisms, and other organisms via specifc metabolic and nutrient absorption processes (Guerra-Renteria et al. [2019;](#page-8-11) Li et al. [2020\)](#page-9-7). Microorganisms have negatively charged groups, such as phosphoric acid anions and carboxyl anions, on the surface of their cell walls. In addition, heavy metals contain cationic groups that enable them to interact with the bacteria cell wall. Microorganisms accumulate heavy metal ions through adsorption, which does not require metabolic energy. However, the absorption process in living cells depends on energy metabolism (Hu and Chen [2023](#page-9-8)). Table S1 lists several studies regarding mineral absorption by microorganisms. On the other hand, incorporating lemon oil or ethylenediaminetetraacetic acid (EDTA) can increase mineral elimination rates by 26.3–31.5% if absorption is slow and inefficient (Ning et al. 2019). In other words, this indicates that lemon and EDTA can be used as a catalyst to increase bacterial cell wall mineral absorption.

However, when essential nutrients are introduced, the cell's capacity to absorb metal ions increases. During the catabolism and anabolism processes, bacteria produce organic acids that dissolve heavy metals in the soil, immobilizing heavy metals on the cell walls of bacteria. According to several studies, microorganisms can produce organic acids and dissolve Cd in the soil if given sufficient nutrients and energy. Specifcally, the study found that the rate of leaching was only 9% in the absence of nutrients, but it increased to 36% when glucose and other nutrients were added (Chang et al. [2019](#page-8-12); Ke et al. [2020](#page-9-9); Sharma [2021\)](#page-10-10). *Citrobacter* can absorb signifcant quantities of toxic metals by producing unbound inorganic phosphate and can absorb minerals differently (Liu et al. [2023](#page-9-10); Shiri-Yekta et al. [2023\)](#page-10-11). Through oxidation–reduction reactions, the ability of microbes to metabolize heavy metals is signifcantly enhanced when heavy metals combine with oxygen or hydrogen. Prokaryotic microorganisms, for instance, can alter the oxidation state of heavy metals, thereby modifying their properties and rendering them less toxic. Aerobic microorganisms can convert Hg^{2+} to Hg^{0} before evaporation (Merkel et al. [2021](#page-10-12); Sazykin and Sazykina [2023\)](#page-10-13). Although Hg(0) is fairly toxic, it cannot compare to the toxicity of $Hg(II)$ and $Hg(I)$, which can produce organomercury compounds such as methylmercury and dimethylmercury. This and similar compounds enter the trophic chain and can bioaccumulate, accumulating mercury in fsh (Wang et al. [2022](#page-10-14)). *Corynebacterium* can absorb and reduce Cr^{6+} in water to Cr^{3+} . The Cr^{6+} form is more toxic than the Cr^{3+} form. It was discovered that the toxicity of Cr^{3+} is due to its specific antagonism with iron absorption (DesMarias and Costa [2019](#page-8-13); Liu et al. [2022](#page-9-11)). *Bacillus licheniformis R08* uses the same process to convert Pb^{2+} to Pb^0 (Margaryan et al. [2021](#page-9-12); Zhang et al. [2022\)](#page-11-1). Rapidly manifesting symptoms result from exposure to organic lead, which is likely more toxic than inorganic lead due to its lipid solubility. Organic lead compound poisoning symptoms primarily afect the central nervous system and include insomnia, delirium, cognitive deficits, tremors, hallucinations, and convulsions (Frolova et al. [2021](#page-8-14)).

Previous research (Rasti et al. [2021\)](#page-10-15) investigated the organic matter discharged in the drilling area of the Hawizeh Marshes using in-situ bacteria. In this study, the intention was to reduce the total hardness of the drilling field by decreasing the amount of heavy metals in the water-based drilling mud through halophilic bacteria. It is the first time this species of bacteria has been used to reduce total hardness. Additionally, using this species in ex-situ bioremediation processes represents a pioneering effort in the field. The ultimate objective is to reduce the cost of sustainable forestry and protect the environment from hazardous chemicals. This research was conducted under laboratory conditions to prevent variations in pH, temperature, and mineral composition, which might otherwise lead to alterations in environmental conditions. The investigation was undertaken over a period spanning from 2021 to 2023 within the confines of the Hawizeh Marshes.

Materials and methods

Sampling

A sample was collected from the water tank just prior to reinjection into the well in the Hawizeh Marshes region of southwest Iran, in close proximity to Ahwaz, during the continuous circulation of water-based mud (Fig. [1](#page-2-0)). Sampling was conducted in the best possible way, with the assistance of personnel from the drilling company and the use of containers supplied by the drilling company. The characteristics of the drilling fluid employed in the well up to 2771 m are displayed in Table S2.

Addition of microorganisms to wastewater

In this investigation, four bacterial species were isolated in situ and added individually to wastewater. Each bacterium was collected and transferred to blood agar for growth. A bacterial sample was obtained in the incubator after 24 h at 35 °C to create McFarland standards. Then, 0.15 mL of McFarland standards were combined with 10 mL of wastewater and incubated for one week. The same procedure was applied to *Aquibacillus halophilus*, procured from a bacterial culture collection*.* It was added separately to serum physiology to create the McFarland standard, then added to efuent and incubated for seven days. Seven days later, a visual inspection revealed that the water's color had changed. In the subsequent phase, the test was repeated while increasing the volume of wastewater and bacteria to 200 mL and 10 mL, respectively. After a week, the color changes of the wastewater were recorded. After two days, the evidence showed that no in situ bacteria could thrive in the moderately halophilic medium.

Why is *Aquibacillus halophilus* **chosen?**

The contaminated environment must be assessed before choosing bacteria from outside the environment. Based on several factors, including the minerals present in the drilling fluid and the location of the reservoir, which is

Fig. 1 The location of the sampling site (31.576849, 47.746551) is in the Hawizeh Marshes (Rasti et al. [2021\)](#page-10-15)

surrounded by saline water, it is hypothesized that only halophilic species can survive in this environment. This hypothesis was further supported by the inductively coupled plasma-optical emission spectrometry (ICP-OES) results and total hardness measurements. *Aquibacillus halophilus* is a unique bacterium that can survive in highly salty environments. Based on the ICP-OES results and total hardness, an ex-situ bacterium, *Aquibacillus halophilus,* was chosen to thrive in wastewater because it can thrive in NaCl concentrations, with optimal growth occurring at 10% (w/v) NaCl. According to Amoozegar et al. ([2014b,](#page-7-6) [a](#page-7-7)), the ideal temperature and pH for its proliferation are 35 °C and 7.0, respectively (Amoozegar et al. [2014a](#page-7-7); [b](#page-7-6)). *Aquibacillus halophilus* is a gram-positive, moderately halophilic bacterium. The bacterial cells are rod-shaped and motile and can produce oval endospores in non-swollen sporangia. The strain number is IBRC-M 10775, obtained from the Iranian Biological Resource Center. Amoozegar et al. ([2014a](#page-7-7); [b\)](#page-7-6) isolated and identified this species from the Aran-Bidgol hypersaline lake in central Iran, which contains NaCl, $Na₂SO₄$, $MgCl₂$, and $MgSO₄$ with traces of carbonate ion (Amoozegar et al. [2014a,](#page-7-7) [b](#page-7-6); Lee and Whang [2019\)](#page-9-13).

ICP‑OES analysis

750 mL of water-based drilling fluid was sent to the Meyar Danesh Pars laboratory for ICP-OES and hardness measurements. Those experiments were done to determine the total concentration of heavy metals and to identify and quantify heavy metals from the investigated emplacement. The ICP (atomic emission spectrometer) laboratory of Danesh Pars is outfitted with a Perkin Elmer Company Optical Emission Spectrometer 8300 and a Sherwood Company flame photometer. The ASTM D1976 standard has been applied to identify water minerals.

Results and discussion

Sampling location

The investigation of the well in the Hawizeh Marshes is of utmost importance due to the profound environmental changes induced by drilling campaigns. This investigation was initiated because the oil company responsible for these activities recognized the need to address and mitigate the damages caused to the area.

Microbiological techniques for removing heavy metals from water‑based drilling slurry

A case study on in situ bacteria will help us comprehend the diversity of bacteria and their ability to absorb heavy metals. Combining in-situ and ex-situ bacteria is indeed advantageous. Our team identifed the four species of bacteria detected in a water-based drilling mud using a blood agar medium (Rasti et al. [2021\)](#page-10-15). They were added separately to the drilling wastewater, but the appearance of the wastewater samples did not alter. In contrast, heavy metals such as lead, iron, and manganese were found in water-based drilling sludge (Fig. [2\)](#page-3-0). The total hardness of the water-based drilling slurry was determined to be 19,100 parts per million (ppm). High salinity levels in the mud are responsible for the inability of four in-situ bacterial species to thrive in drilling wastewater. Due to membrane lysis or rupture, these in-situ bacteria cannot grow in water with high salinity (Sharghi et al. [2014](#page-10-16)). However, previous research has demonstrated that these bacteria could thrive in a blood culture medium, indicating they may be pathogenic.

Fig. 2 The heavy metals detected in wastewater before bioremediation with halophilic bacteria (*Aquibacillus halophilus*)

Fig. 3 The color of the effluent changed **a** before *Aquibacillus halophilus* treatment and **b** one week after *Aquibacillus halophilus* treatment

Saline water surrounds subterranean petroleum reserves (Agapkin and Kotov [2021;](#page-7-8) Ershaghi [2024\)](#page-8-15). Drilling water has a high salinity, and the presence of halophilic bacteria is indicative of a salient medium (Rezaei Somee et al. [2018](#page-10-17); Rezaei Somee et al. [2021;](#page-10-18) Gorriti et al. [2023](#page-8-16); Novák et al. [2023](#page-10-19)). A medium with a moderate salinity level was used to identify halophilic bacteria in wastewater. In contrast, after one week of *Aquibacillus halophilus* bacterium addition, the hue of the effluent changed to a milky white (Fig. [3](#page-4-0)). The density of bacteria in the effluent was high, *Aquibacillus halophilus* did not interact with other bacteria, and it can proliferate without restriction. The four in situ bacteria can only grow on blood agar because they defend themselves against salty conditions within the cell. Drilling mud samples containing heavy metals and bacteria were sent to the Meyar Danesh Pars laboratory to determine the wastewater's metal content and total hardness. As anticipated, the microorganisms were highly efective at absorbing heavy metals. The variations in minerals are depicted in Table [1.](#page-4-1) The initial concentration of 18 of the 30 identifed heavy metals has signifcantly changed or decreased. In addition, the total hardness indicated after treatment was 3700 ppm. The measured total hardness was substantially lower than its original value of 19,100 ppm before bioremediation. Despite a substantial decrease in water hardness, the current total hardness is still above the standard limit for wastewater reuse. Following regulations, the total hardness should preferably be less than 800 ppm.

The most signifcant alterations in elemental concentrations are delineated in Fig. [2.](#page-3-0) These elements are categorized into three main groups. The frst group comprises Ni, Mo,

Table 1 The percentage changes and the element concentrations in ppm before and after bacteria treatment

| Number | Elements | Concentration before treatment (ppm) | Concentration after treatment (ppm) | Changes in percentage $(\%)$ |
|----------------|----------------|--|---|------------------------------------|
| 1 | Na | 3.01 | 1.52 | 49.50 |
| $\overline{2}$ | B | 2.19 | 0.28 | 87.21 |
| 3 | Si | 1.65 | 0.18 | 89.09 |
| $\overline{4}$ | W | 0.9 | 0.87 | Unchanged |
| 5 | Sb | 0.87 | 0.29 | 66.66 |
| 6 | S | 0.79 | 0.19 | 75.94 |
| 7 | As | 0.7 | 0.33 | 52.85 |
| 8 | Ca | 0.69 | 0.14 | 79.71 |
| 9 | K | 0.66 | 0.16 | 75.75 |
| 10 | Al | 0.58 | 0.41 | 29.31 |
| 11 | C _d | 0.54 | 0.32 | 40.74 |
| 12 | Cu | 0.52 | 0.52 | Unchanged |
| 13 | Bi | 0.48 | 0.28 | 41.66 |
| 14 | Fe | 0.43 | 0.35 | 18.60 |
| 15 | Se | 0.43 | 0.38 | 11.62 |
| 16 | Pd | 0.42 | 0.42 | Unchanged |
| 17 | Mn | 0.34 | 0.27 | 20.58 |
| 18 | Ti | 0.33 | 0.33 | Unchanged |
| 19 | Sn | 0.33 | 0.33 | Unchanged |
| 20 | Mg | 0.32 | 0.15 | 53.12 |
| 21 | Cr | 0.32 | 0.31 | Unchanged |
| 22 | Zn | 0.31 | 0.26 | 16.12 |
| 23 | Co | 0.29 | 0.29 | Unchanged |
| 24 | Be | 0.29 | 0.29 | Unchanged |
| 25 | La | 0.28 | 0.28 | Unchanged |
| 26 | Sr | 0.27 | 0.11 | 59.29 |
| 27 | Mo | 0.25 | 0.25 | Unchanged |
| 28 | \mathbf{V} | 0.25 | 0.25 | Unchanged |
| 29 | Ni | 0.24 | 0.24 | Unchanged |
| 30 | Ba | 0.22 | 0.11 | 50 |

The elements whose concentrations have decreased drastically are highlighted in bold

La, Co, Cr, and Cn, exhibiting relatively minor fuctuations. Nevertheless, certain studies have demonstrated that Pseudomonas species, such as Pseudomonas aeruginosa, possess a notable capacity to absorb Cr and Ni, reaching up to 30% and 90%, respectively (Oves et al. [2023;](#page-10-20) Priyadarshanee and Das [2024](#page-10-21)). The second group encompasses Mn and Se, demonstrating more pronounced variations than the preceding category. Notably, Pseudomonas spp. exhibit commendable capabilities in heavy metal absorption. Additionally, Bacillus spp., including Bacillus selenitireducens, represents a viable option for the absorption of Mn and Se (Guo et al. [2023](#page-8-17); Huang et al. [2023](#page-9-14); Oves et al. [2023](#page-10-20)). Finally, the third

group consists of Bi, Cd, K, As, Sb, Si, and Na, which are signifcantly reduced after the addition of A. halophilus. A. halophilus, a halophilic bacterium, has demonstrated remarkable ability to remove a wide range of heavy metals, including Bi, Cd, K, As, Sb, and Si, despite their disparate chemical properties and periodic table group afliations.

There are several reasons why *A. halophilus* did not exhibit a signifcant impact on the absorption of heavy metals, specifcally Ni, Mo, La, Co, Cr, and Cn. Firstly, the type of bacteria and the metal ions in issue have a signifcant impact on the capacity of microorganisms to sequester heavy metals. It's possible that *A. halophilus*, an acinetobacter species, lacks binding sites or specialized transporters that effectively interact with these particular heavy metals. Because metal absorption selectivity is frequently speciesspecifc, *A. halophilus's* cellular processes and innate metabolic pathways may be to blame for the absence of reactions seen (Abd Elnabi et al. [2023;](#page-7-9) He et al. [2023](#page-8-18); Liu et al. [2024\)](#page-9-15). Secondly, it's possible that the experiment's heavy metal concentrations weren't ideal for getting *A. halophilus* to react noticeably. The degree of tolerance that diferent microbial species show for heavy metals varies, and concentrations that are too low might not cause a noticeable bioaccumulation reaction (Chakravorty et al. [2023;](#page-8-19) Huang et al. [2024\)](#page-9-16). Conversely, concentrations that exceed the organism's tolerance threshold might lead to toxicity, hindering the absorption process (Chakravorty et al. [2023](#page-8-19); Khalid et al. [2023](#page-9-17); Zhou et al. [2023](#page-11-2); Alabssawy and Hashem [2024](#page-7-10)). Furthermore, the kinds of heavy metals tested and their chemical composition could be quite important. Heavy metals, in particular oxidation states or chemical forms, are frequently the target of microbial resistance mechanisms; if the metals were supplied in a form that *A. halophilus* could not access, this could account for the observed lack of efect (Yin et al. [2019](#page-10-22); Jeyakumar et al. [2023](#page-9-18); Mansoor et al. [2023\)](#page-9-19). In conclusion, the interaction of microbial physiology, heavy metal concentrations, and chemical forms has a signifcant impact on the efectiveness of *A. halophilus* in absorbing heavy metals. Based on Fig. [2](#page-3-0) and Table [1](#page-4-1), this bacterium can selectively interact with and sequester heavy metals by using diferent mechanisms, such as biosorption and bioaccumulation, regardless of chemical group diferences. In terms of environmental cleaning, *A. halophilus* can be used in bioremediation strategies, providing a sustainable and environmentally friendly solution. Economically, using such ex-situ microbial methods is consistent with cost-efective and efficient remediation strategies.

Detection of heavy metals via ICP‑OES analysis

The signifcant change in ICP-OES results demonstrates the superior performance of *Aquibacillus halophilus* bacteria. This study was conducted in a laboratory setting to

specifcally assess the impact of *Aquibacillus halophilus* on wastewater while mitigating the infuence of any external factors (e.g., changes in pH, temperature, and mineral composition). Table [1](#page-4-1) indicates that approximately thirty elements were analyzed by ICP-OES before and after treatment, except sodium, which has an inverted process. The alterations fall into three categories: Elements such as Na, B, Si, Sb, S, As, Ca, K, Mg, Sr, Zn, and Ba make up the frst group with the most signifcant concentration reduction. The second group includes elements such as Al, Cd, Bi, Fe, Se, and Mn, whose concentrations have decreased to a lesser extent. The fnal group consists of elements whose concentrations have not altered, including W, Cu, Pd, Ti, Sn, Cr, Co, Be, La, Mo, V, and Ni. Table [1](#page-4-1) demonstrates that the colloidal solution of these substances may have a negative ionic charge, preventing their absorption by bacterial cell walls. Either they are independently negatively charged or associated with negatively charged elements such as iron, oxygen, and chloride.

Although Ca and Mg elements typically govern total hardness (Kozisek [2020](#page-9-20); Dey et al. [2024\)](#page-8-20), numerous minerals in this drilling waste, including Na, Fe, and Mn, have afected the hardness of the water. After treatment with *Aquibacillus halophilus*, total hardness is observed to decrease signifcantly. After bacterial treatment, the total hardness dropped from 19,100 to 3700 ppm, a tremendous and signifcant reduction of more than 15,400 ppm. It demonstrates *that Aquibacillus halophilus* absorbs most minerals as a source of energy. This bacterium provides distinct advantages to this research over another research. While the optimal growth conditions for *Aquibacillus halophilus* are 35 °C and pH 7, this bacterium has a broad temperature range and can grow between 15 and 45 °C and pH 5 and 10 (Amoozegar et al. [2014a;](#page-7-7) [b\)](#page-7-6). Due to its wide temperature range, this bacterium can be utilized in various climates. The ability of this halophilic bacterium to substantially reduce the concentration of 18 heavy metals is a key diference between it and other species, as shown in Table [1](#page-4-1). In addition, this halophilic bacterium substantially infuenced the concentrations of Ca and Mg, resulting in decreases of 79% and 53%, respectively, and contributing to a remarkable reduction in total hardness. Notably, none of the microorganisms listed in Table [1](#page-4-1) discovered in water samples could signifcantly reduce the total hardness at this level.

Assessing the growth efect of *Aquibacillus halophilus* **on the ecosystem of Hawizeh Marshes**

Bacteria play a crucial role in our environment, infuencing various ecosystems and important processes (Singh and Yadav [2021;](#page-10-23) Chen et al. [2024\)](#page-8-21). Understanding their behavior and distribution is vital for monitoring environmental health, identifying potential hazards, and ensuring the well-being of ecosystems and human populations (Gomte et al. [2024;](#page-8-22) Liao [2024;](#page-9-21) Mauck et al. [2024](#page-10-24)). Therefore, the primary conclusion derived from this discussion is that purifying and recycling effluent is the most effective way to aid the Hawizeh Marshes ecosystem. Improper management has caused the Hawizeh Marshes to dry completely (Hasab et al. [2020\)](#page-8-23). According to Hasab et al. [\(2020](#page-8-23)), the average salinity in this region has increased to over 1800 mg/L, creating a high probability for the proliferation and spread of *Aquibacillus halophilus* bacteria. Due to the presence of carotenoids, increased salinity can result in a higher concentration of bacteria, causing the water to appear orange (Lazrak et al. [2024\)](#page-9-22). In contrast, as salinity decreases, bacteria cannot obtain the necessary resources, leading to the lysis process in which bacterial cell walls rupture and bacteria are annihilated (Dawson et al. [2023\)](#page-8-24). In other words, the proliferation of *Aquibacillus halophilus* can be efectively monitored and controlled through the control of wastewater salinity. In contrast, *Aquibacillus halophilus* is typically regarded as nonpathogenic or opportunistic. It is a halophilic bacterium typically found in saline environments, such as salt lakes and salted edibles. Typically, it is not associated with human or animal disease transmission (Amoozegar et al. [2014a;](#page-7-7) [b](#page-7-6)). Therefore, it can be concluded that this bacterium not only aids in removing heavy metals from the environment but also poses no hazard to the environment or human health. On the other hand, the ability of halophilic bacteria to remove and reduce harmful chemicals from petroleum felds has also been investigated and reported recently. Table [2](#page-6-0) presents a compilation of numerous examples of such research.

The various varieties of halophilic bacteria that are efective at removing and reducing toxic substances under

varying salinity conditions are detailed in Table [2.](#page-6-0) However, it should be noted that in this type of study, a number of variables, including oxygen concentration, pH, pressure, and temperature, play signifcant roles due to the variation in sampling depth and geological formation (Bailey and Ahmadi [2014;](#page-8-25) Matthiesen et al. [2015](#page-9-23); Houben et al. [2018](#page-9-24); Riedel [2019](#page-10-25)). It is evident from Table [2](#page-6-0) that the majority of research conducted in recent years has been devoted more to eliminating specifc kinds of distinct toxic substances like sulfide and phosphate (Ahmed et al. [2019](#page-7-11)). Moreover, findings from Table [2](#page-6-0) indicate that halophilic bacteria can efectively grow and operate in diverse geological formations, demonstrating their ability to thrive across a broad range of geological conditions for the management of hazardous substances. Based on the literature review, so far, no study has found or reported on reducing the salinity of water-based mud using microbial methods. Conversely, most are focused on eliminating corrosive toxic substances (Table [2](#page-6-0)). Comparing the removal of toxic substances presented in Table [2](#page-6-0) to the ability to reduce a wide variety of heavy metals (or total water hardness), as shown in Table [1,](#page-4-1) which highlights the signifcance of this research, is of equal importance. By implementing this research, we can make signifcant strides toward attaining sustainable development goals 6 and 14 in this region. The quality of the water utilized in the region will not only be improved through the purification of water sources but the fragile ecosystem, which has been deteriorating, can also be revitalized and restored.

Table 2 An overview of research on halophilic bacteria in the petroleum feld for the removal of toxic substances

| Bacteria species | Toxic substance | Geological formation | Salinity level |
|---|--|---|--|
| Desulfobacterota, Halanaerobiaeota, Sinergis- tota, Pseudomonadota, and Bacillota | Sulfide | Carbonate | 6–11% salinity (Kadnikov et al. 2023) |
| Pseudodesulfovibrio thermohalotolerans sp. Nov. | Sulfate | Sediment | 3% salinity (Gaikwad et al. 2023) |
| Exiguobacterium mexicanum (Em), Terribacil- <i>lus saccharophilus</i> $ZY-1(Ts)$, and <i>Staphylo-</i> coccus warneri (Sw) | Petroleum hydrocarbon or crude oil | Sand body | 84%, 50%, and 6% salinity, respectively (Su et al. 2023) |
| Marinobacter hydrocarbonoclasticus strain MAM1 (MF716467), Marinobacter sp. strain MAM2 (MF716468), and Marino- bacter hydrocarbonoclasticus strain MAM3 (MF716469) | PAHs | Briny water and sediment 4% salinity (Jamal 2020) | |
| Halanaerobium, Desulfovermiculus, Halo- monas, and Marinobacter | Nitrate | Shale oil field | 0.6–3.6 mEq of NaCl (An et al. 2017) |
| Marinobacter sp. GN001 (KY818661), and Geobacillus sp. TK004 | Sulfide | Sandstone | 0.52 ± 0.04 mEq of NaCl (Okpala et al. 2017) |
| Serratia sp. SL-12 | Phosphate | Soil | 15% salinity (Singh and Jha 2016; Kapadia et al. 2022) |

Conclusion and future perspectives

In accordance with the goals of SDG 6 (Clean Water and Sanitation) and SDG 14 (Life Below Water), the study's fndings indicate that *Aquibacillus halophilus* exhibited a remarkable ability to efectively mitigate a broad spectrum of heavy metals while concurrently reducing the total hardness. The bacterium demonstrated its effectiveness in enhancing water quality despite its rapid growth. In addition, the robust growth of *Aquibacillus halophilus* at pH 7 indicates that the in-situ bacteria did not interfere with this halophilic bacteria while maintaining a stable pH level of 7, which is optimal for drilling operations. For effective reutilization in the well-logging feld, the water's total hardness must remain below 800 ppm. If not, additional chemical substances, such as caustic soda, would be required to reduce the overall hardness. However, using *Aquibacillus halophilus* provides an alternative strategy, potentially reducing reliance on such chemicals. Consider a scenario where, conventionally, 100% of materials are required to remediate effluent with a hardness of 19,100 ppm. By incorporating *Aquibacillus halophilus*, it is possible to reduce material consumption and total hardness by approximately 80%, as only 19.37% of materials, such as caustic soda, are needed to efectively treat water with a signifcantly reduced total hardness down to 3700 ppm. Water use in well logging is subject to certain restrictions, one of which pertains to the maximum allowable hardness of the water used during drilling operations, which is 800 ppm. With the dramatic reduction of water hardness from 19,100 ppm to below 3700 ppm, attaining a total hardness level of less than 800 ppm makes reusing wastewater signifcantly easier. This reduction indicates a signifcant decrease in the resources required for the purifcation process, contributing to SDG 6 by promoting sustainable practices and resource efficiency. In line with SDG 14's objective to preserve aquatic life, this microbial technique reduces well-logging costs and protects the environment from hazardous substances, particularly heavy metals.

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Declarations

Conflict of interest The authors declare that they have no known competing fnancial interests or personal relationships that could have appeared to infuence the work reported in this paper.

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