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Lipid Accumulation Staining

Evaluation of the Heavy Metal Bioremediation Potential of Indigenous Bacteria Isolated from Textile Effluents in Kano, Nigeria

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ABSTRACT

This study aimed to assess the capability of indigenous bacteria isolated from textile effluents to bio-remove heavy metals at the Challawa industrial site in Kano, Nigeria. Physicochemical analyses of the textile effluent samples were conducted, and bacteria were isolated based on their physical, biochemical, and molecular traits. Bio-removal studies were conducted following standard procedures. The physicochemical parameters, including conductivity, total suspended particles, total dissolved solids, turbidity, dissolved oxygen, and biochemical oxygen demand, were found to have exceeded the WHO limits. Heavy metals were also found to be at initial concentrations significantly exceeding the WHO limits, namely: cadmium 0.12 ± 0.09 to 0.11 ± 0.6 mg/L, nickel 0.125 ± 0.04 to 0.103 ± 0.09 mg/L and lead 0.019 ± 0.10 to 0.018 ± 0.05 mg/L, except for zinc (0.468 ± 0.10 to 0.62 ± 0.08 mg/L), which was within limits. After a 21-day bio-removal period, the concentrations decreased significantly: cadmium (0.008 ± 0.02 to 0.006 mg/L), nickel (0.072 ± 0.15 to 0.045 ± 0.10 mg/L), lead (0.013 ± 0.02 to 0.010 ± 0.08 mg/L), and zinc (0.259 ± 0.15 to 0.072 ± 0.09 mg/L). These bacterial isolates achieved biomass reductions for cadmium (50.00%, 36.36%, 27.27%), nickel (56.31%, 50.50%, 42.40%), zinc (44.66%, 37.87%, 28.00%), and lead (47.37%, 33.33%, 31.58%), respectively. The study showed bio-removal after 21 days could be effective, signifying the possibility of applying bacteria in reducing environmental pollutants. This is a cost-effective and environmentally friendly approach to pollution reduction and enhancing ecosystem sustainability.

INTRODUCTION

Fast industrialization and population growth have led to more pollution, where industries discharge untreated effluents containing colorants, heavy metals, and various organic and inorganic substances into natural water systems [1]. Environmental pollution now presents significant threats to all human and animal life [2]. Textile industries, in particular, generate high quantities of effluents with high concentrations of harmful substances, which can lead to dissolved oxygen depletion and disrupt ecosystems [3,4]. Industrial effluent from the textile industry is improperly treated and disposed of in Nigeria, where it leads to severe environmental contamination [5]. Major cities such as Kano, Kaduna, Lagos, and Port Harcourt experience challenges related to pollution, as it involves the discharge of untreated effluents into rivers, which affects water quality and public health [6]. These metals in wastewater may increase fertility in both the sediment and in the water column, potentially leading to eutrophication. Eventually, this can then lead to oxygen depletion in open water, promote algal blooms, and result in the death of aquatic organisms [7]. Despite their costs, physical and chemical techniques may not always guarantee that pollutants are eliminated.

A variety of physicochemical methods have been employed to mitigate the detrimental effects of heavy metal pollutants in the environment, including electrocoagulation [8], electrowinning [9], membrane filtration, ion exchange, precipitation, soil replacement, and the application of activated carbon [10,11]. Although these physical-chemical methods are

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effective, they are expensive and produce secondary pollution; the use of bioremediation offers several advantages over these traditional remediation methods. Conventional approaches often involve high expenses, substantial disturbances, and the generation of secondary contaminants. In contrast, bioremediation aligns with principles of sustainability and environmental responsibility, as it harnesses the natural processes of the environment to restore areas affected by pollution [11]. This method provides an eco-friendly and economically viable means for heavy metal detoxification in effluents. Bioremediation has emerged as a pivotal approach for tackling environmental pollution caused by various contaminants, owing to its ecological sustainability and cost-effectiveness [11,12].

In bioremediation, microorganisms can utilize pollutants as nutrients, thereby reducing heavy metal concentrations in a process known as bio-removal [13,14]. Numerous microorganisms exhibit specific physiological traits, including the capacity for bioaccumulation and phosphatase-mediated biocatalysis, which make them valuable agents for mitigating heavy metal contamination in wastewater [15]. Furthermore, scientific studies have confirmed that living microorganisms, including bacteria, fungi, and microalgae, are highly effective as bioremediation agents. This is largely due to their exceptional ability to tolerate high levels of heavy metals, a critical factor contributing to their success [10], [12].

Most microorganisms employ two primary mechanisms during bioremediation: either sequestering or immobilizing metals or enhancing the solubility of the metals. Additionally, some organisms oxidize or reduce heavy metals into less toxic forms. This can occur in both aerobic and anaerobic environments, though aerobic conditions have been shown to be more efficient and faster compared to anaerobic settings [10]. Several bacterial isolates reported for their ability to remove heavy metals in wastewater include *Acinetobacter* sp., *Arthrobacter* sp., *P. aeruginosa, Brevibacterium iodinium, Comamonas* sp., *Bacillus pumilus, Bacillus cereus, Flavobacterium* sp., *Micrococcus* sp. and *Pseudomonas putida* [10,15,16]. Among these, *Klebsiella species* have shown particularly strong potential for the heavy metal bioremediation. This improved performance arises from their resistance and removal capabilities. Furthermore, these organisms serve as effective biosorbents for heavy metal ions in highly polluted environments, owing to their adaptability to extreme conditions. For instance, Chakraborty et al. [18] reported that *Klebsiella* sp. TIU20 achieved removal efficiencies of 72.5% for cadmium (Cd), 28% for chromium (Cr), and 25.7% for lead (Pb). Similarly, Zafar et al. [19] highlighted the potential of *Klebsiella pneumoniae* strain PBS3A2 in mitigating lead (Pb) toxicity, underscoring its applicability in heavy metal bioremediation. This biosorption mechanism, believed to have evolved as a resistance strategy, positions organisms as promising candidates for treating industrial wastewater contaminated with heavy metals, such as textile effluents [16].

Kano State is the Nigeria's second most populous urban center and a hub of commercial activity often termed the "Centre of Commerce,", which hosts Challawa Industrial Estate, one of the nation's largest industrial complexes. This estate accommodates numerous industries, such as the textiles and tanneries, which are important contributors to environmental pollution. Textile industries, in particular, often rely on water-intensive processes that generate substantial volumes of wastewater. This effluent is characterized by elevated levels of color, odor, pH, chemical oxygen demand (COD), and biochemical oxygen demand (BOD), rendering it highly toxic if discharged untreated into the environment. Building on the established role of microbial bioremediation in addressing industrial pollutants, this study aims to evaluate the heavy metal bioremediation potential of indigenous bacteria isolated from textile effluents in Kano, Nigeria. By focusing on these indigenous bacteria, this study contributes to the novel insights into microbial remediation strategies for sustainable wastewater management in Nigeria.

MATERIALS AND METHODS

Study area

The Challawa Industrial Estate is located in Kano State, Nigeria. It is geographically positioned at coordinates 11.986987°N, 8.547297°E. The estate is bounded by Panshekara Town to the north, Yadanko Village to the south, Kumbotso Town to the east, and Zawachiki Town to the west. The industrial park hosts a diverse range of industries, including textiles, pulp and paper, beverages, ceramics, water bottling, and tanneries, which contributes significantly to the local economy.

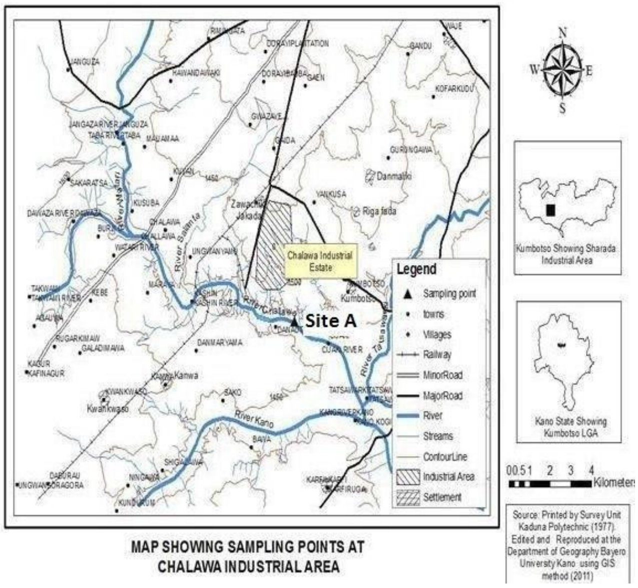

Fig. 1. Sampling point at Challawa Industrial Estate of Kumbotso Local Government Area of Kano State

Sample collection

Samples were aseptically collected at three locations around 10:00 am weekly for 3 weeks from the Kano Industrial Estate (Challawa) to prevent contamination. Each textile effluent sample was taken from the effluents' flow routes and from the industrial facilities' discharge pipes. There were three (3) sterile two-liter (2 L) plastic containers with screw caps for each location. The samples were then transported to Bayero University's Microbiology Laboratory in Kano within 2 hours of collection for the analysis of various physicochemical parameters, and the concentrations of heavy metals were kept at a temperature of about 4 °C. Subsequently, 500 mL sterile reagent bottles were filled with 250 mL of sample liquid. The bacteria were isolated from these samples came from these samples [17].

Analysis of physicochemical parameters of the textile effluents

In conformity with widely recognized protocol and standard methodologies for the examination of water and wastewater, all samples were analyzed for physicochemical parameters [18].

Those variables measured included: pH, temperature, color, odor, dissolved oxygen, biochemical oxygen demand, turbidity, electrolytic conductivity, total suspended solids, and total dissolved solids. Those measurements were done using a pH /Temperature meter (an analytical instrument pH-25), and a Hanna water quality meter (Model HI 9813 - 6).

Determination of heavy metal concentration (zinc, cadmium, lead, and nickel) from textile effluents

A 10:1 mixture of sulfuric and nitric acids was used as a digesting agent. The analytical procedure to measure heavy metals was carried out according to the method adopted by [19] to calculate the concentration of Zn, Cd, Pb, and Ni. After distributing one hundred (100) milliliters of the effluent sample, ten milliliters of concentrated HNO_3 were added. The mixture was digested on a hot plate and then filtered through Whatman 125mm filter paper to remove the insoluble material. An atomic absorption spectrophotometer of the model Bulk Scientific VGP 210 was used to measure the quantities present in the digest of Zn, Cd, Pb, and Ni.

Bacterial isolation and characterization

The bacteria present in the effluents were isolated through the serial dilution method, as described by [17]. On the nutrient agar medium, the isolated bacterial strains were grown. Dilutions were prepared to facilitate straightforward numerical handling, allowing easier computation of the number of cells per milliliter in the original sample. Ten-fold and successive ten-fold dilutions were employed [20]. The bacterial isolates were identified based on their morphological and biochemical characteristics using Gram's stain, citrate, catalase, MRVP, indole, starch hydrolysis, and oxidase reactions [18].

Bio-removal of heavy metals from textile effluents using selected bbacterial sspecies

Inoculum preparation

Each bacterial species was cultured in nutrient broth for 24 hours at 37 ºC. Following centrifugation, each bacterial pellet was resuspended in 50 mL of sterile saline solution containing 0.85% NaCl and standardized to achieve a uniform density of approximately 10^8 CFU/mL.

Heavy metals bio-removal studies

Heavy metal removal experiments were conducted in 250 mL Erlenmeyer flasks containing 100 mL of textile effluent. The pH of the effluent was adjusted to 7.0 using NaOH and H_2SO_4, after which the flasks were autoclaved at 121 °C for 15 minutes to ensure sterility. Following autoclaving, 5 mL of the bacterial inoculum from each isolate was aseptically added to the flasks. The flasks were incubated at 37 °C for a period of 21 days. During this time, the pH of the effluent in each flask was measured every 48 hours to monitor changes and provide growth kinetics. Additionally, heavy metal content in the supernatant was analyzed using atomic absorption spectroscopy (AAS) [19].

Sample digestion and heavy metals analysis

The standard protocol provided by [21] was followed for the heavy metal analysis. 2 mL of the treated effluent was transferred into a boiling tube and mixed with 10 mL of a triple acid solution. The triple acid solution consisted of a 9:2:1 ratio of HNO_3, H_2SO_4, and $HClO_4$. The mixture was heated until the effluent became colorless, indicating complete digestion. The digested sample was then filtered using Whatman No. 1 filter paper to remove any particulate matter.

The filtrate was analyzed for heavy metal content using an Atomic Absorption Spectroscopy Double Beam Spectrophotometer (Model: SL 176). The percentage of removal was calculated based on the following equation:

$$\%Removal = \frac{Initial\ amount - final\ amount}{initial\ amount} x100$$

Molecular identification of the most potent bio-removal bacteria

Following the isolation of several bacteria, the most effective bacteria that removed Cd, Ni, Pb, and Zn were confirmed using molecular methods. In this work, DNA was extracted using the Quick-DNA ™ Microprep Kit according to the manufacturer's instructions. After the extraction, polymerase chain reaction (PCR) amplification was conducted using a Primus 25 advanced thermo cycler (Techne Techgene FTGENE5D USA) based on the method described by [22], utilizing a pair of forward 27F (AGAGTTTGATCCTGGCTCAG) and reverse 1492R primers (TACGGYTACCTTGTTACGACTT). The following were the PCR conditions: Denaturation processes include initial denaturation at 95°C for five minutes, 30 cycles at 94 was sequenced by Sanger Sequencing (Inqaba Biotec™). The DNA sequence was trimmed using Chromas software to obtain a consensus sequence, which was identified through BLAST on the NCBI database (https://blast.ncbi.nlm.nih.gov/Blast.cgi?PROGRAM=blastn). Sequence alignment was conducted using MEGA 11 software, and a phylogenetic tree was constructed using the same software [23].

RESULTS AND DISCUSSION

Physicochemical parameters of textile effluents

Table 1 represents the physicochemical parameters of the textile effluents before and after bio-removal by the bacterial isolates. The mean temperature values before treatment ranged from 28.00 ± 0.03°C to 29.00 ± 0.12°C, while after treatment, the mean temperatures decreased to a range of 26.65 ± 0.02 °C to 27.54 ± 0.09 °C. The initial pH values range from 11.00 ± 0.26 to 11.25 ± 0.10, while the pH values after removal range from 6.54 ± 0.21 to 6.70 ±0.11. This corroborates the work of [24], which shows that the initial pH of untreated effluent was alkaline. The electrical conductivity values were high, ranging from 5400 to 5530 µS/cm, while TSS ranged from 965 to 1140 mg/L across various samples. Total dissolved solids were equally high, with ranges between 2680 to 2830 mg/L, which is relatively high above the discharge limits. These increased values suggest that there is a high presence of dissolved materials, which reduces light penetration in water, thus decreasing photosynthetic activity.

The dissolved oxygen (DO) values ranged from 1.9 ± 0.03 mg/L to 3.1 ± 0.09 mg/L, while the biochemical oxygen demand (BOD) values ranged from 0.4 ± 0.01 mg/L to 1.2 ± 0.06 mg/L. The relatively low DO concentrations observed may be attributed to reduced photosynthetic activity, likely caused by elevated total dissolved solids (TDS) levels. Elevated electrical conductivity (EC) values at various sampling points were likely due to the presence of chemical salts in the effluent used for textile dyeing. The levels of both organic and inorganic compounds in the effluent may also be linked to the chemicals utilized in textile processing. Additionally, high temperatures were found to reduce gas solubility in water, which, in turn, contributed to higher BOD and chemical oxygen demand (COD) values, indicating an increased load of organic pollutants in the effluents.

Table 1. Physicochemical analysis of textile effluents before and after bio-removal.

Parameters	Before	Sample 1 After	%	Before	Sample 2 After	%	Before	Sample After	%
Temperature (°C)	29.00 ± 0.12	27.54 ± 0.09	5.03	28.00 ± 0.03	26.65 ± 0.02	4.82	29.00 ± 0.12	26.71± 0.10	4.61
pH	7.00 ± 02	6.54 ± 0.21	6.57	7.00 ± 02	6.70 ±0.11	4.29	7.00 ± 02	6.68 ± 0.9	4.57
E/C (mS/cm)	5530.00 ± 2.21	4255.02 ± 0.54	23.05	5490.00 ± 3.21	4472.05 ± 3.10	18.54	5400.00 ± 1.30	4750.02± 1.90	12.03
DO (mg/L)	2.2 ± 0.13	1.32 ± 0.08	40.00	3.1 ± 0.09	2.20 ± 0.11	29.03	1.9 ± 0.03	1.4 ± 0.13	26.32
BOD (mg/L)	0.4 ± 0. 01	0.21 ± 0.06	47.50	1.2 ± 0.06	0.90 ± 0.02	25	0.6 ± 0.07	0.4 ± 0.01	33.33
TSS (mg/L)	1140.00 ± 2.95	225.01 ± 1.87	80.26	1020.00 ± 2.10	431.01 ± 2.11	57.74	965.00 ± 1.92	420.04 ± 1.98	56.48
TDS (mg/L)	2680.00 ± 3.54	1355.06 ± 3.21	49.44	2830.00 ± 3.11	1455.08 ± 2.90	48.59	2720.00 ± 2.96	1460.06± 2.20	46.32

Note: The data represent the average results of three separate experiments, each performed in triplicate, along with their corresponding standard deviations. Key: % = percentage, pH= Negative logarithm to base 10 of hydrogen ion concentration. Temp = temperature of effluent. DO= Dissolved oxygen, BOD = Biological oxygen demand. E/C = Electrical conductivity, TSS = Total suspended solids. TDS = Total dissolved solids. Mg/L = Milligram per liter, (MS/cm) = Micro Semen per centimeter

Heavy metal concentrations (zinc, cadmium, lead and nickel)
The heavy metal analysis of the textile effluents indicated the presence of cadmium, nickel, lead, and zinc, which would be components of several chemical solutions utilised in various types of the textile industry. The concentration of cadmium (Cd) ranged from 0.011 ± 0.06 mg/L to 0.012 ± 0.09 mg/L. Sample 1 exhibited the highest cadmium concentration at 0.112 ± 0.09 mg/L, followed by samples 2 and 3, which had concentrations of 0.11 ± 0.06 mg/L. For nickel (Ni), the concentrations ranged between 0.103 ± 0.09 mg/L and 0.125 ± 0.04 mg/L. Sample 3 showed the highest nickel concentration at 0.125 ± 0.04 mg/L, while sample 2 had a concentration of 0.120 ± 0.07 mg/L. Sample 1 recorded the lowest nickel concentration at 0.103 ± 0.09 mg/L. Zinc (Zn) concentrations ranged from 0.062 ± 0.08 mg/L to 0.468 ± 0.10 mg/L. Sample 1 had the highest zinc concentration at 0.468 ± 0.10 mg/L, followed by sample 3 (0.100 ± 0.13 mg/L). Sample 2 had the lowest zinc concentration at 0.062 ± 0.08 mg/L. For lead (Pb), concentrations ranged from 0.018 ± 0.05 mg/L to 0.019 ± 0.10 mg/L. Samples 1 and 3 recorded the highest lead concentration at 0.019 ± 0.10 mg/L, while sample 2 had the lowest concentration at 0.018 ± 0.05 mg/L (**Table 2**). This analysis showed that Zinc (Zn) was the most abundant heavy metal among the three analysed, with the highest concentration observed in sample 1, followed by sample 3, and the lowest concentration in sample 2. Nickel (Ni) was present at the highest concentration in sample 3, while cadmium (Cd) was found at elevated levels in sample 1. Lead (Pb) concentrations were higher in samples 1 and 3 compared to sample 2. Fluctuations in heavy metal concentrations in this study may be attributed to different types of dyestuffs employed in textile production.

Table 2. Heavy metal concentrations in textile effluents.

Heavy metals	Sample 1	Sample 2	Sample 3	Permissible limits mg/L
Cadmium (Cd) mg/L	0.012 ± 0.09	0.011 ± 0.06	0.011 ± 0.06	0.003
Nickel (Ni) mg/L	0.103 ± 0.09	0.120 ± 0.07	0.125 ± 0.04	0.02
Zinc (Zn) mg/L	0.468 ± 0.10	0.062 ± 0.08	0.100 ± 0.13	3
Lead (Pb) mg/L	0.019 ± 0.10	0.018 ± 0.05	0.019 ± 0.11	0.015

Note: The data represent the average results of three separate experiments, each performed in triplicate, along with their corresponding standard deviations. Key: mg/L = Milligram per liter, Sample = Textile effluent, Cd = Cadmium, Ni = Nickel, Zn = Zinc, Pb = Lead

Isolation and screening of heavy metal-resistant bacteria
The effluent samples were cultured on nutrient agar and selective media to get bacterial isolates. The characterization of the isolates was done according to their morphological and biochemical properties [17]. The identified bacterial strains were denoted as MHK1, MHK2, and MHK3. These isolates showed varying potentials for reducing heavy metal concentrations.

MHK1 exhibited the highest reduction efficiency, decreasing cadmium (Cd) by 50.00%, nickel (Ni) by 56.31%, zinc (Zn) by 44.66%, and lead (Pb) by 47.37%. MHK2 also showed significant reduction capabilities, lowering cadmium by 36.36%, nickel by 50.00%, zinc by 33.87%, and lead by 33.33%. In comparison, MHK3 displayed moderate reduction efficiency, with cadmium decreasing by 27.27%, nickel by 42.40%, zinc by 28.00%, and lead by 31.58%. This shows that MHK1 has the highest remediation potential for heavy metals in the textile effluents.

Determination of kinetic growth
Fig. 2 shows the growth kinetics of the bacterial isolates. The results of the kinetic growth study indicate the variation in alkalinity (pH) over a 48-hour interval for three effluent samples (Sample 1, Sample 2, and Sample 3) during a 20-day (480-hour) period of removal studies. Sample 1 ranged between 6.54 ± 0.20 to 7.90 ±0.67, day 8 (192 hours) presents the maximum alkalinity pH of 7.90 ± 0.6 then day 6 (144 hours) and day 10 (240 hours) (7.87 ± 0.62 and 7.89 ± 0.76) respectively while day 20 (480 hours) had the lowest value of 6.54 ± 0.20. Sample 2 varied between 6.70 ± 0.10 to 8.10 ± 0.32, day 8 (192 hours) exhibited the highest alkalinity pH of 8.10 ±0.32 followed by day 10 (240 hours) and day 12 (288 hours) (8.09 ± 0.24 and 8.05 ± 0.15) respectively, while day 20 (480 hours) registered the lowest value of 6.70 ± 0.10. Sample 3 ranged from 6.68 ± to 7.92, day 8 (192 hours) observed the highest alkalinity pH of 7.92 ± 0.43 followed by Day 10 (240 hours) and Day 6 (144 hours) (7.90 ± 0.34 and 7.88 ± 0.65) respectively, day 20 (480 hours) recorded the least value of 6.68 ± 0.11. All three samples showed a similar trend, with pH values reaching a peak around day 8 (192 hours), and then gradually declining toward the end of the monitoring period (day 20 or 480 hours) (**Fig. 2**). The efficiency of bioremediation for various heavy metals such as Pb (II), Cd (II), Cr (VI), Ni (II), and Cu (II), was examined within a pH range of 5 to 9 and the shows that the highest bioremediation effectiveness was observed at a pH of 7 [25]. This is also in tandem with the findings of [26], which states that competition between cations and protons for binding sites often results in reduced biosorption of metals such as Cu, Cd, Ni, Co, and Zn at lower pH levels.

Bio removal of zinc, cadmium, lead, and nickel
The bacterial isolates used for the bio-removal of the heavy metals in the textile effluents included MHK1 for sample 1, MHK2 for sample 2, and MHK3 for sample 3. There was a reduction in pH of the three effluents after 21 days. The pH of Sample 1 decreased from 7.00 ± 0.20 – 6.54 ± 0.20, the pH of Sample 2 decreased from 7.00 ± 0.20- 6.70 ± 0.10, and pH of the Sample 3 decreased from 7.00 ± 0.20 – 6.68 ± 0.11 after 21 days.

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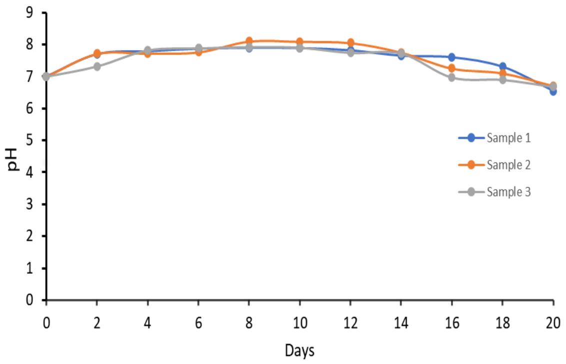

Fig. 2. The kinetic growth of the bacterial isolates.

Table 3 represents the heavy metal concentrations of the effluents before and after bio-removal. All the physicochemical parameter concentrations were reduced after the bio-removal study. Reductions of 23.05%, 18.54% and 12.03% were recorded for Electrical Conductivity (EC) of the effluent, respectively. Total suspended solids of sample 1 sample 2 and sample 3 were reduced by 80.26% 57.74% and 56.48% respectively. The total dissolved solid value had a reduction of 49.44%, 48.59%, and 46.32% in sample 1, sample 2, and sample 3, respectively. A reduction of 40.00%, 29.03%, and 26.32% was noted for DO in sample 1, sample 2, and sample 3 effluent, respectively. A reduction of 47.50%, 25%, and 33.33% was noted for BOD in sample 1, sample 2, and sample 3 effluent, respectively. These results indicated that MHK1 showed a considerable decrease in the physicochemical parameters and heavy metal concentrations in textile effluents. This implies that the bacteria were efficient in removing pollutants without impacting the aquatic environment.

Table 3. Heavy metal concentrations in textile effluents before and after bio-removal.

Bacterial isolates	*Klebsiella pneumoniae*			*Bacillus cereus*			*Escherichia coli*		
Heavy metal	Before	Sample 1After	%	Before	Sample 2 After	%	Before	Sample 3 After	%
Cd mg/L	0.012 ± 0.09	0.006 ± 0.08	50.00	0.011 ± 0.06	0.007 ± 0.09	36.36	0.011 ± 0.06	0.008 ± 0.02	27.27
Ni mg/L	0.103 ± 0.09	0.045 ± 0.10	56.31	0.120 ± 0.07	0.060 ± 0.11	50.00	0.125 ± 0.04	0.072 ± 0.15	42.40
Zn mg/L	0.468 ± 0.10	0.259 ± 0.15	44.66	0.062 ± 0.08	0.041 ± 0.12	33.87	0.100 ± 0.13	0.072 ± 0.09	28.00
Pb mg/L	0.019 ± 0.10	0.010 ± 0.08	47.37	0.018 ± 0.05	0.012 ± 0.10	33.33	0.019 ± 0.10	0.013 ± 0.02	31.58

Note: The data represent the average results of three separate experiments, each performed in triplicate, along with their corresponding standard deviations. Key: Sample = Textile Effluents, % = Percentage, Cd = Cadmium, Ni = Nickel, Zn = Zinc, Pb = Lead

Molecular characterization

Following isolation and characterization based on morphological and biochemical characteristics, the strain exhibiting the highest removal efficiency, designated as MHK1, was identified. **Fig. 3** shows the amplified region of the 16S RNA gene through gel electrophoresis. The 16S rRNA gene sequence of MHK1 (607 bp) was subjected to BLAST analysis on the NCBI database, which revealed a similarity of 98.09% with *Klebsiella pneumoniae* strain KSK23. Phylogenetic analysis further demonstrated that MHK1 clusters most closely with *Klebsiella pneumoniae* (**Fig. 4**). These findings confirm that MHK1 is a strain of *Klebsiella pneumoniae*.

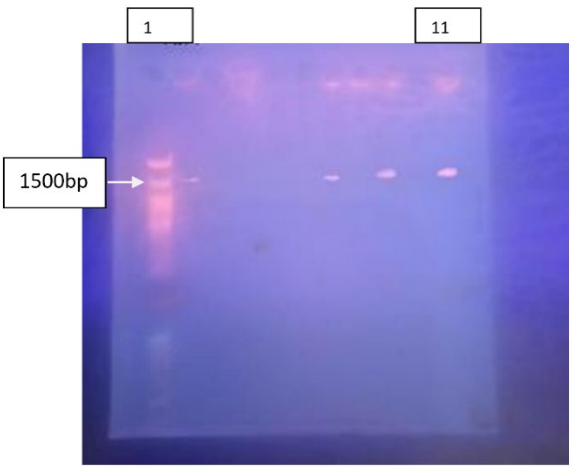

Fig. 3. Agarose gel electrophoresis of *Klebsiella pneumoniae* MHK1

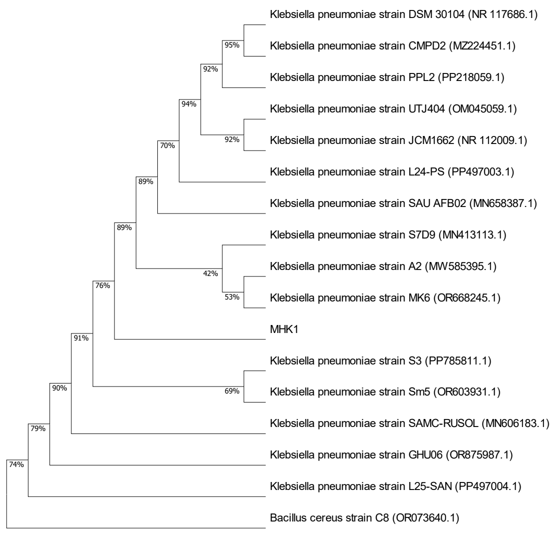

Fig. 4. Phylogenetic tree of *Klebsiella pneumoniae* MHK1 with other related bacterial species

CONCLUSION

This study highlights the environmental threat posed by untreated textile effluents in Nigeria, due to the elevated concentrations of heavy metals such as cadmium, nickel, and lead, which were found to exceed WHO permissible limits. The successful reduction of these metals by the indigenous bacterial strains, particularly *Klebsiella pneumoniae* MHK1, after 21 days of treatment demonstrates not only the bio-removal potential of indigenous bacteria but also the feasibility of adopting low-cost, sustainable biotechnological solutions for industrial wastewater management in developing countries. These findings have broader implications for public health and environmental protection policies, especially in industrial hubs like Kano State, where regulatory enforcement may be limited. By providing evidence that local microbial resources can effectively remediate hazardous effluents, this study supports the integration of bioremediation strategies into national pollution control frameworks. Moreover, it opens avenues for future research to optimize microbial consortia, scale up treatment systems, and explore the genetic and metabolic pathways involved in heavy metal resistance and removal.

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