Sahel Journal of Life Sciences FUDMA (SAJOLS)
June 2025 Vol. 3(2): 483-490
ISSN: 3027-0456 (Print)
ISSN: 1595-5915 (Online)
DOI: *https://doi.org/10.33003/sajols-2025-0302-55*

Research Article

Isolation and Identification of Bacteria Isolated from Saline Soil of Dan Hassan in Kura, Kano State

***M. Ibrahim[1], A. H. Kawo[2], S. Yahaya[2] and B. M. Aminu[2]**

[1]Department of Soil Science, Faculty of Agriculture, Bayero University, PMB 3011, Kano, Nigeria
[2]Department of Microbiology, Faculty of Life Sciences, Bayero University, PMB 3011, Kano, Nigeria
****Corresponding Author's email*: mibrahim.ssc@buk.edu.ng; Phone: +2348039331570**

ABSTRACT
Soil salinity is a global challenge in agriculture for crop production, and its widespread occurrence is becoming alarming due to poor irrigation management, excessive fertilizer and pesticide application. This research aimed to isolate and identify bacteria capable of tolerating soil salinity stress. Soil samples were collected from Dan Hassan in Kura LGA, Kano state. Some of the physicochemical properties of the soil sample were determined by using the standard method. The bacteria isolated were identified by cultural, biochemical, and molecular techniques. The pH, EC, nitrogen, phosphorus, and potassium were 9.48, 11.56dsm^{-1}, 0.30 %, 24.52 mg kg-1, and 0.19 cmol kg-1, respectively. The bacteria isolated and identified were bacilli, and gram-positive and gram-negative. Their 16S rRNA sequences, when BLAST, were found to be *Bacillus Subtilis* IM-BUK-3, which was newly deposited in NCBI, *Sphingobacterium psychroaquaticum* MOL, and *Paenochrobactrum glaciei* PHBR-1 with accession numbers PV716630, FJ156081, and LC557044.1, respectively. The bacteria isolated can be subjected to trials to ascertain their potential to alleviate salt stress and improve plant growth.

Keywords: Bacterial isolates; Identification; Isolation; Saline Soil; Salt Tolerance

Citation: Ibrahim, M., Kawo, A.H., Yahaya, S., & Aminu, B.M. (2025). Isolation and Identification of Bacteria Isolated from Saline Soil of Dan Hassan in Kura, Kano State. *Sahel Journal of Life Sciences FUDMA*, 3(2): 483-490. DOI: *https://doi.org/10.33003/sajols-2025-0302-55*

INTRODUCTION

Most biotic factors like pathogenic infections, salinity stress, drought, and extremely high temperatures can reduce the growth and yield of any crop and can significantly retard its production. Soil salinity is among many stress factors responsible from hindering crop productivity across the world. It effects on seed germination, plant growth and development, and reproductive success eventually leading to a decline in agricultural crop growth and yield (Ke *et al.,* 2020; Kumar and Sharma, 2020).

Large arable land is left unproductive, which can cause substantial economic losses and food insecurity as well as the livelihoods of millions of people who depend on agriculture for their living. Major effects of soil salinity on agricultural lands are particularly high in arid and semi-arid regions, where limited rainfall and high evaporation rates contribute to the buildup of salts in the soil. In such environments, the disadvantage of soil salinity is more complex due to poor drainage systems, poor managements and inappropriate application of inorganic fertilizer and pesticides, which impede the leaching of salts from the soil, exacerbating salinity levels over time (FAO, 2020).

Plant growth promoting rhizobacteria (PGPR) reduce the effect of abiotic and biotic stresses, which may be directly and indirectly and can support plant growth and yield (Albdaiwi *et al.,* 2019). Salt tolerant PGPR lower salinity damages in several crops wherein salt stress is significantly reduced, which by so doing improves agricultural output in terms of better plant

growth, crop health and productivity in soil with extreme saline condition (Kapadia *et al.,* 2022; Khumairah *et al.,* 2022; Reang *et al.,* 2022).

The applications of PGPR may be effective in the case of biostimulation by inclusion of both abiotic and biotic systematic tolerance in plants (Singh *et al.,* 2015). In extreme salinity conditions, various PGPR can regulate and stimulate plant growth through biostimulantion of growth promoting properties. Many reports recorded an investigation and suggest that such bacterial isolates belong to the following genera: *Azospirillum*, *Alcaligenes*, *Arthrobacter*, *Acinetobacter*, *Bacillus*, *Burkholderia*, *Bradyrhizobium*, *Enterobacter*, *Erwinia*, *Flavobacterium*, *Pseudomonas*, *Rhizobium*, and *Serratia* (Akram *et al.*, 2016).

Considering the distribution and diversity of PGPR that are exceptional to the mangrove region, there are need to find out more about the native PGPR found in arid and semi-arid region of Nigeria, identify them and find out their capacity to withstand salt tolerance and produce growth promoting properties. By so doing, PGPR can be a key microbe in alleviating salt stress and plant growth and yield. This study aims to identify the bacterial species with abilities to tolerate salinity and produce certain properties that can enhance the growth and yield of crops.

MATERIALS AND METHODS

Collections of Soil Sample

The soil sample was collected from Dan Hassan town in Kura Local Area Government of Kano state, Nigeria ($11^0$46.17'' N and longitudes 08^o 25' 49" E), during the summer of June 2024. Twenty (20) soil sample cores were randomly collected with sterile soil auger at a depth of 10cm, and bulk to three (3), stored in a new polyethylene bag and transported to the Centre for Dryland Agriculture, Bayero University, Kano for processed and kept for further analysis.

Physicochemical Properties of Saline Soil

Determinatin of Soil pH in H_20 (1:2.5)

Ten (10g) of air-dried soil sample was weighed into a beaker, and 25ml of distilled water was added and stirred for 30 minutes using a glass rod for an interval of 10 minutes. The suspension was allowed to settle for 5 minutes. The pH meter (Janway, P757s) was calibrated using buffer 4 and 7, and the pH of the soil was then recorded (Eno *et al.,* 2009).

Electrical Conductivity (EC)

A beaker containing 10g of air-dried soil sample was mixed with 50ml of distilled, stirred for 30mins, and the suspension was allowed to settle. The EC meter (Janway, 4520) was calibrated with 0.01M KCl at 25^{o}C temperature and the EC of the soil sample was recorded (Danbarati, 2016).

Determination of Total Nitrogen

One (1g) of soil sample was digested with a catalyst (K_2SO_4 and $CuSO_4$) container concentrated sulphuric acid in a ratio of 1:10. The digest was then distillated, where the distillate was collected with 2% boric acid and titrated with standard acid solution (0.01N HCl) (Eno *et al.,* 2009).

Determination of Phosphorus Content

The Olsen method was the method adopted for the determination of available phosphorus in the soil sample (Olsen *et al.,* 1954). The soil sample was extracted using sodium carbonate, and the extract was mixed with a colour developer. The solution was allowed to stand for 30 minutes for proper colour development, and the absorbance was recorded at 883nm using a spectrophotometer. A set of standards of 0.2, 0.4, 0.6, 0.8, and 1.0ppm was prepared, and the concentration was extrapolated from the graph of the calibration curve.

Exchangeable Potassium (K)

The exchangeable K of the soil was extracted with 1M ammonium acetate (1M NH_4OAC) solution. Air-dried soil sample (5g) was weighed into a plastic container, shaken for 30minutes, and filtered. The filtrate was read using a flame photometer (Jenway, PEP7) to determine the exchangeable K. A standard of 0, 10, 20, 30, 40, and 50ppm was prepared to extrapolate potassium (Anderson and Ingram, 1998).

Isolation of Bacteria from Soil Sample

One (1g) of air-dry soil sample was weighed, and 9ml of distilled water and diluted logarithmically up to 10^{-9}. A prepared nutrient agar supplemented with NaCl (5%) and incubated at 37°C for 72 h. A pure culture was made and each colony was identified by cultural and biochemical test (Danbarati *et al.,* 2016).

Morphological Characterization of the Bacterial Isolates

Gram staining

A bacterial colony was picked to form a thin smear where primary reagents (Gram's iodine), decolourizer (alcohol, 95%), and counter stain (safranin) were applied, and bacteria were observed to be either

Gram-positive or negative. Gram-positive organisms would retain the primary stain while Gram-negative bacteria took up the secondary (counter) stain (Todar *et al.,* 2005).

Characterization of the Bacterial Isolates

Catalase Test

A pinch of bacterial colony was smeared on a grease-free glass slide, 2.0ml of hydrogen peroxide was added on the colony. The presence of bubbles indicated a positive reaction while absence of bubble indicated a negative reaction (Bhattacharya *et al.,* 2002).

Urease Test

Urea agar medium was inoculated with fresh bacterial culture and incubated at 37ºC for 48 hours. After incubation, observed changes of colour from light orange to pink showed a positive result (Kummerer, 2004).

Oxidase Test

A bacterial colony was smeared on a grease-free glass slide, and 1% aqueous solution of tetramethyl-p-phenylenediamine hydrochloride was added. It was then observed within 5 seconds, and purple colour recorded as a positive result (Chesebrough, 2003).

Citrate Utilization Test

This was carried out by inoculating the test organism in a test tube containing Simon's citrate medium, which was incubated at 37 °C for 24 hours. The development of deep-blue colour after the incubation period indicated a positive result (Udeani *et al.,* 2009).

MR-VP Test

A quantity (5.0 ml) of MR-VP broth was inoculated with the bacterial colony and incubated 37ºC for 72 hours, after which 1.0 ml of the broth was transferred into a small test tube. Three drops of methyl red solution were added. A red colour development on the addition of the indicator signified a positive methyl red test, while a yellow colour signified a negative test. To the rest of the broth in the original tube, five drops of 4% potassium hydroxide (KOH) was added, followed by fifteen (15) drops of 5% -naphtol in ethanol. No colour change indicated a VP negative test (Dubey, 2002).

Indole Test

A bacterial colony was incubated in 1% tryptophan broth in a test tube and incubated at 37°C for 48 hours. Then, 1.0 ml of chloroform was added to the broth. The test tube was then shaken gently, and then 2 ml of Kovac's reagent was added and shaken gently, and allowed to stand for twenty (20) minutes. The formation of red colouration at the top layer indicated a positive test, while yellow colouration indicated a negative test (Udeani *et al.,* 2009)

Molecular Characterization

Identification of efficient bacterial isolates with salt tolerance was performed on the basis of the nucleotide sequence of 16S rRNA at the Centre for Dryland Agriculture molecular laboratory. The total genomic DNA of the most potent bacterial isolates was extracted by the following procedure and confirmed using agarose 1% (w/v) gel. After incubation, the bacterial colony was Scraped from plate into a 1.5 ml tube containing 800ul CTAB and 100 uL of 10% SDS, vortex for 30 seconds, incubate at 60°C for 30 minutes, cooled at room temperature and 600 uL of chloroform: isoamyl alcohol (24:1) was mixed to form an emulsion and then centrifuge at 12,000 rpm at 4°c for 5 minutes. It was then transferred to the top aqueous phase into a new microcentrifuge tube, 0.6 volume of ice-cold isopropanol (2/3 volume of aqueous recovered), and 100 µL of 5M NaCl was then added. It was precipitated at -20°C for 15-20 and left overnight at 4°C. After it was removed from the ice, it was then centrifuged at 12000 rpm for 10 minutes. The supernatant was discarded, and the DNA was washed twice with ice-cold 70% ethanol, removed without drying the DNA pellet by leaving the tube open at room temperature for 20 minutes. The DNA pellet in <100 µL TE solution (PH 8) was removed by placing it in a water bath at 50°c with periodic shaking for 2 hours. Using 16S rRNA Universal primers (27F AGAGTTTGATCMTGGCTCAG and 1492R TACGGYTACCTTGTTACGACTT), gene fragments of isolates was amplified in a MJ Research PTC-225 Peltier Thermal Cycler (Osborn *et al.*, 2005). For each isolate, PCR will be performed in a final volume of 50 µL containing PCR amplification buffer (1X), Taq DNA polymerase (2.5 U), dNTPs (4 mM), primers (0.4 µM) and template DNA (4 ng). Amplification conditions were initial denaturation at 94°C for 3 min, 30 cycles at 94°C, 30 s at 52°C and 90 s at 72°C, followed by a final 7 min extension at 72°C. PCR products was purified by using Montage PCR Clean up kit (Millipore) and Sequencing reactions was performed using a ABI PRISM BigDyeTM Terminator Cycle Sequencing Kits with AmpliTaq DNA polymerase (FS enzyme) (Applied

Biosystems). The sequence similarity searches were done using the BLAST program available in NCBI database (Orthan and Gulluce, 2015). Alignment of sequences was carried out with CLUSTAL W program version 2.1. Phylogenetic trees were constructed using the neighbour joining method. Tree files were generated by MEGA 11. bootstrap analysis (Tamura *et al.,* 2021).

Data Analysis

The statistical analysis was carried out using Microsoft Office Excel 2019 and Genstat software 17th. Means were compared using analysis of variance. Mean values were expressed as mean ± SD. Duncan's multiple range test compared values with a significance level of $p < 0.05$

RESULTS

The soil sample was found to have a pH and electrical conductivity (EC) of 9.48 and 11.56 dSm^{-1} respectively. Whereas the total nitrogen, phosphorus, and potassium recorded moderate values of 0.30%, 24.54 mg kg^{-1}, and 0.19 cmol kg^{-1}, respectively (Table 1).

Table 1: Some Physicochemical Properties of Saline Soil at Dan Hassan

Parameters	Values
pH	9.48 ± 0.123
EC dSm^{-1}	11.56 ± 0.644
N %	0.30 ± 0.228
P $mgkg^{-1}$	24.52 ± 4.219
K $cmolkg^{-1}$	0.19 ± 0.116

Characterization of the Bacterial Isolates

From our investigation, all the bacterial isolates were found to be rod-shaped, where *Bacillus subtilis* and *Bacillus cereus* are Gram positive with both having terminal and central spores, respectively. *Sphingobacterium psychroaquaticum* and *Paenochrobactrum glaciei* were non-sporeforming and also gram negative (Table 2).

Biochemical Characterization of the Bacterial Isolates

In this investigation, *B subtilis* and *B cereus* were found to be Methyl red (MR), Voges-Proskauer (VP), indole production, urease negative, while citrate, catalase, and oxidase were positive. In the case of *Sphingobacterium psychroaquaticum* and *Paenochrobactrum glaciei* were found to be MR-VP, citrate, and urease negative, with only indole production was recorded positive for *Paenochrobactrum glaciei* (Table 3).

Molecular Characterizations

Figure (1) show the polymerase chain reaction (PCR) amplification product of extracted DNA sequences of 16S rRNAof the most potent bacterial isolates in terms of salt tolerance which had an amplicon size of 1500bp. The sequences of the bacterial isolates were Blast in NCBI gene bank, where *Sphingobacterium psychroaquaticum* MOL and *Paenochrobactrum glaciei* PHBR-1 with accession numbers and percentage identities of FJ156081 (89.55%) and LC557044.1(85.96%), respectively. A new strain of was deposited in the NCBI server and tagged as *B subtilis* IM-BUK-3 given an accession number of PV716630.

Table 2: Morphological Characteristics of Bacterial Isolates

1	Gram Staining	+	+	-	_
2	Cell Shaped	Rod	Rod	Rod	Rod
3	Spore	Terminal	Central	Non	Non
4	Colony Colour	Creamy	Whitish	Yellowish	Yellowish
	Identified Organism	*Bacillus* subtilis	*Bacillus cereus*	*Sphingobacterium psychroaquaticum*	*Paenochrobactrum glaciei*

Table 3: Biochemical Characterization of Bacterial Isolates

1	Catalase	+	+	+	+
2	Urease	-	-	-	-
3	Oxidase	+	+	+	+
4	Citrate	+	+	-	+
5	Methyl Red	-	-	-	-
6	Voges Proskauer	-	-	-	-
7	Indole Production	-	-	+	-
	Expected Organism	*Bacillus Subtilis*	*Bacillus Cereus*	*Sphingobacterium Psychroaquaticum*	*Paenochrobactrum glaciei*

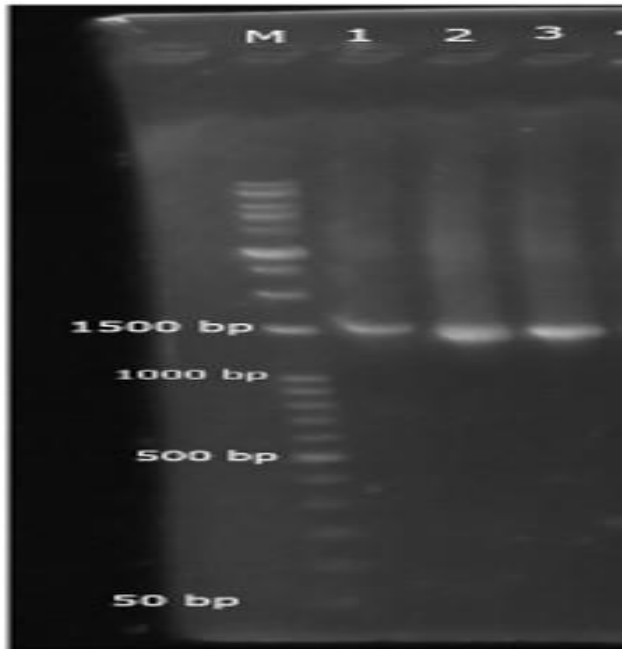

Figure 1: Genomic DNA from the bacterial isolates of salt tolerance bacteria strain. M (Ladder) Key: (Lane 1 *S psychroaquaticum* ; Lane 2 *P glaciei*; and Lane 3 *B subtilis*)

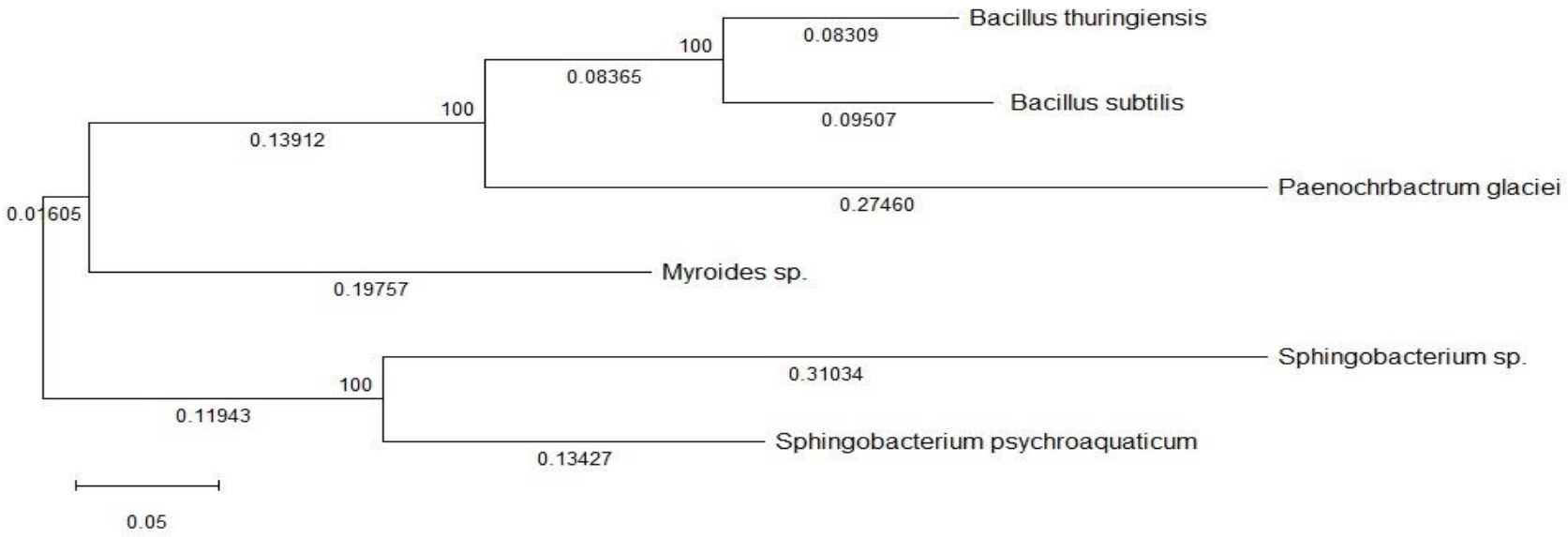

Figure 2: Phylogenetic Trees of Some of the Bacterial Isolates Identified

DISCUSSION

The high pH found in this study may be due to excess sodium accumulation on soil exchange sites, causing sodicity, which elevates soil pH because of low leaching of the excess sodium (Jibrin *et al.,* 2008). Conversely, high electrical conductivity (EC) may result from the accumulation of soluble salts from the irrigated water, which increases its ability to conduct electricity, and is more common in the arid and semi-arid regions of Nigeria. Intense evaporation due to high temperature, leaving salts behind at the surface, and possibly poor drainage, which raises the salt capillarity action (Maina *et al.,* 2012). The possible reason behind high nitrogen (N) and phosphorus (P) in the soil may result from the recent application of ammonium and phosphate fertilizer, or the water used for irrigation probably contains dissolved nitrate and phosphate. Although, high salinity can inhibit microbial activities that normally convert nitrogen into gaseous forms, this slows nitrogen loss and makes more mineral N accumulate in soil. Due to the high saline nature of the soil, crops struggle to absorb nutrients efficiently, leaving higher residual nitrate after application of fertilizer (Adamu and Dawaki, 2008). The possible reason to gram negative bacteria to withstand saline environment is their ability to actively synthesize or accumulate small organic molecules called compatible solutes in their cytoplasm. These solutes include compounds like glycine betaine, ectoine, sugars, and polyols. In addition, the bacteria have evolved specialized ion pumps to actively pump potassium ions into the cell while excluding sodium ions. Gram positive bacteria have another advantage over gram negative bacteria based on the accumulation of compatible solutes and the robust nature of their thick peptidoglycan cell wall. (Nina *et al.,* 2018). Both *Bacillus subtilis* and *Bacillus cereus* are typically catalase-positive, but they may show differences in their motility or other biochemical tests. *Sphingobacterium psychroaquaticum* and *Paenochrobactrum glaciei* have distinct enzymatic metabolism (Debarati et al., 2016; Xioa *et al.,* 2013, and Kampfer *et al.,* 2010). The result of the DNA PCR shown the movement of the DNA fragments with a size of 1500bp base on the molecular weight marker. This implies that the fragments of the DNA were smaller which enables them to move faster up to 1500bp ladder. Hence, this may be likely why it has a faint, thin band. Phylogenetic trees on bases of 16S rRNA the three bacterial isolates were constructed by neighbor joining method using mega 12 and the topology of the tree was evaluated by bootstrapping score over 1000replicates. The three bacterial isolates showed no relatedness or likely common ancestors as seen from their differences in their branch lengths and uncommon node.

CONCLUSION

The assessment of bacteria that may reduce the salinity stress is highly significant, coupled with the challenge of global food insecurity. Hence, this research was able to isolate and identify certain bacterial isolates that are found in saline soil, whose future exploration may alleviate crop salt stress.

REFERENCES

Adamu, G. K. & Dawaki, M. U. (2008). N P K level in multiple crop land in a small holder

Akram, M. S., Shahid, M., Tariq, M., Azeem, M., and Riaz, S. (2016). Deciphering Staphylococcus sciuri SAT-17 Mediated Anti-oxidative Defense Mechanisms and Growth Modulations in Salt Stressed Maize (*Zea mays* L.). *Frontiers in Microbiology*, *7*, 867.

Albdaiwi, R. N., Khyami-Horani, H., Ayad, J. Y., Alananbeh, K. M., and Al-Sayaydeh, R. (2019). Isolation and characterization of halotolerant plant growth promoting rhizobacteria from durum wheat (*Triticum turgidum* subsp. durum) cultivated in saline areas of the dead sea region. *Oxidat. Med. Cell. Long.* 10, 1–16.

Anderson, J.M. and Ingram J.S.I, (1998). Tropical Soil Biology and Fertility: A Handbook of Methods. 2nd Edition, CAB International, Wallingford, UK, pp (221).

Bhattacharya, D., Sarma, P.M., Krishnan, S., Mishra, S. and Lal, B. (2002): Evaluation of genetic diversity among *Pseudomonas citronellosis* strains isolated from oily sludge contaminated sites.*Journal of Applied Environmental Microbiology* 69(3); 14351-441

Chesebrouhg, M. (2003). *Medical Laboratory Manual.* Tropical Health Technology, Low Price Edition. Doddington, Cambridgegeshire, England pp 20-35.

Danbarati H, Pardita D, Arpita M, and Malini B (2016). Isolation and Characterization of Halophilic Bacteria from Sundarban Soil, *International. Journal of Life Science and Scientific Research,* 2(4); 442-450.

Dubey, R.C. (2002). *Practical Microbiology*. 1st Edition, S. Chand Publications, New Deldi, India,

Eno J. U., Ibia T. O., Ogunwale J. A., Ano A.O. and Esu I. E (2009) *Manual of Soil, Plant and Water Analyses*. Sibon Books Publishers, Lagos. pp (6 – 77).

FAO. (2020). Soil salinity: A hidden threat to food security. Food and Agriculture Organization of the United Nations.

farmland in Kano closed settled zone: *Biological and environmental sciences journal*

for the tropics, 5. BUK

Jibrin, J. M., Abubakar, S. Z., & Suleiman, A. (2008). Soil Fertility Status of the Kano River Irrigation Project Area in the Sudan Savanna of Nigeria. *Journal of Applied Sciences*, *8*(4), 692-696.

Kämpfer, P., Poppel, M. T., Wilharm, G., Glaeser, S. P., and Busse, H.-J. (2014). Paenochrobactrum pullorum sp. nov. isolated from a chicken. *International Journal of Systematic and Evolutionary Microbiology*, *64*(Pt 5), 1724–1728.

Kapadia, C., Patel, N., Rana, A., Vaidya, H., Alfarraj, S., Ansari, M. J., (2022). Evaluation of plant growth-promoting and salinity ameliorating potential of halophilic bacteria isolated from saline soil. *Frontiers in Plant Science.* 13, 946217.

Ke, J., Wang, B., and Yoshikuni, Y. (2020). Microbiome engineering: synthetic biology of plant-associated microbiomes in sustainable agriculture. *Trends in Biotechnology.* 39, 244–261.

Khumairah, F. H., Setiawati, M. R., Fitriatin, B. N., Simarmata, T., Alfaraj, S., Ansari, M. J., (2022). Halotolerant plant growth-promoting rhizobacteria isolated from saline soil improve nitrogen fixation and alleviate salt stress in rice plants. *Frontiers in Microbiology.* 13, 905210.

Kohler, J., Hernández, J. A., Caravaca, F., and Roldán, A. (2009). Induction of antioxidant enzymes is involved in the greater effectiveness of a PGPR versus AM fungi with respect to increasing the tolerance of lettuce to severe salt stress. *Journal of Environmental Experiment and Botony.* 65, 245–252. doi:

Kumar, P., and Sharma, P. K. (2020). Soil salinity and food security in India. *Front. Sustain. Food Syst.* 4, 533781

Kummerer, K. (2004): Resistance in the environment. *Journal of Antimicrobial Chemotherapy* 45; 311–320.

Maina, M. M., Amin, M. S. M., Aimrun, W., & Sani, I. (2012). Soil salinity assessment of Kadawa irrigation of the Kano River Irrigation Project (KRIP). *Journal of Food, Agriculture & Environment*, *10*(3&4), 1028-1034.

Nina Gunde-Cimerman, Ana Plemenitaš, Aharon Oren, (2018) Strategies of adaptation of microorganisms of the three domains of life to high salt concentrations, *FEMS Journal of Microbiology Reviews*, 42(3), 353–375,

Olsen, R. A., and Bakken, L. R. (1987). Viability of soil bacteria: optimization of plate-counting technique and comparison between total counts and plate counts within different size groups. *Journal of Microbial Ecology* 13(1); 59-74.

Olsen, S.R., Cole, C.V., Watanabe, F.S. and Dean, L.A. (1954) In: Klute, A. (Ed), *Methods of Soil Analysis: Physical Properties*, Part 1, 2nd Edn. Agron Monograph, No 9. Madison WI, pp (403-430).

Orhan, F., and Gulluce, M. (2015). Isolation and characterization of salt-tolerant bacterial strains in salt-affected soils of erzurum, Turkey. *Geomicrobiol. J.* 32 (6), 521–529

Reang, L., Bhatt, S., Tomar, R. S., Joshi, K., Padhiyar, S., Vyas, U. M., et al. (2022). Plant growth promoting characteristics of halophilic and halotolerant bacteria isolated from coastal regions of saurashtra Gujarat. *Sci. Rep.* 12 (1), 4699.

Singh, R. P., Jha, P. & Jha, P. N. (2015). The plant-growth-promoting bacterium Klebsiella sp. SBP-8 confers induced systemic tolerance in wheat (*Triticumaestivum*) under salt stress. *J. Plant Physiol.* 184, 57–67

Tamura, K., Stecher, G. and Kumar, S. (2021) MEGA11: Molecular evolutionary genetics analysis version 11. *Mol. Biol. Evol.* **38**(7), 3022– 3027.

Todar, K., Ubukata M. and Hamada, M. (2005): Microbiology of Human Perspective. McGraw Hill Publishers, London.

Udeani, C.K.T., Obroh, A.A., Okwuosa, N.C., Achukwu, U.P. and Azubike, N. (2009): Isolationand characterization of bacterial isolates. *African Journal of Biotechnology* 8(22); 6301-6303.

Xiao, T., He, X., Cheng, G., Kuang, H., Ma, X., Yusup, K., Hamdun, M., Gulsimay, A., Fang, C., and Rahman, E. (2013). Sphingobacterium hotanense sp. nov., isolated from soil of a Populus euphratica forest, and emended descriptions of Sphingobacterium daejeonense and Sphingobacterium shayense. *International Journal of Systematic and Evolutionary Microbiology*, *63*(Pt 3), 815–820.