Received: 22 FEB 2025 | Accepted: 22 SEPT 2025 | Published Online: 27 SEPT 2025
Recoletos Multidisciplinary Research Journal Vol. 13 no. 2 (December 2025)
DOI: https://doi.org/10.32871/rmrj2513.02.01

Original Article

Biochemical Characterization and Antimicrobial Potential of Soil-Derived *Bacillus* Isolates

Daniel Tunde Abbey*, Dim Emmanuel Ifeanyi, and Leonard Ebube Onyibo

Department of Microbiology and Biotechnology, Federal University Dutse, Dutse, Nigeria

*Correspondence: danielabbey020@gmail.com

Abstract

Background: *There is an increasing need for the exploration of alternative sources of antimicrobial agents as a result of the increasing prevalence of multi-drug resistant (MDR) pathogens. Bacillus species from diverse soil samples were isolated within Dutse, Jigawa State, Nigeria.*

Methods: *Six different Bacillus isolates were identified based on cultural, morphological, and biochemical characteristics: P. polymyxa, B. subtilis, B. cereus, B. pumilus, B. megaterium, and B. licheniformis. Antibacterial activities of these isolates were assessed using agar well diffusion against clinically isolated pathogens: Staphylococcus aureus, Escherichia coli, Klebsiella pneumoniae, and Pseudomonas aeruginosa.*

Results: *Results showed that B. licheniformis showed the broadest spectrum with zones of inhibition of (23. 0 ± 0.8 mm) against S. aureus and (14. 0 ± 0.6 mm) against P. aeruginosa. B. subtilis likewise displayed extremely potent inhibitory action against S. aureus (19. 0 ± 1.0 mm) and P. aeruginosa (21. 0 ± 0.7 mm). On the other hand, B. cereus and P. polymyxa displayed moderate activity while B. megaterium and B. pumilus showed little to no zone of inhibition against some of the pathogens.*

Conclusion: *These findings highlight soil-borne Bacillus species as promising candidates for novel antimicrobial drug discovery with potential contributions to addressing the global challenge of antibiotic resistance.*

Keywords

Antimicrobial resistance (AMR), Bacillus spp, soil microbiota, antibiotic discovery, novel antimicrobial agents, sustainable development goal 3, multidrug-resistant pathogens (MDR), secondary metabolites, bioprospecting, pathogenic bacteria

INTRODUCTION

One very real worldwide public health concern is antimicrobial resistance (AMR). Pathogens, including *Streptococcus pneumoniae, Klebsiella pneumoniae, Escherichia coli, Staphylococcus aureus,* and *Pseudomonas aeruginosa*, were responsible for an alarming 27.3 deaths per 100,000 people (Antimicrobial Resistance Collaborators, 2022). A 2022 study by the Antimicrobial Resistance Collaborators reports that there were 4.95 million deaths associated with AMR in 2019 alone, with West Africa bearing one of the highest mortality rates. Because of their presence all around in the natural world, Bacillus species are everywhere. Thriving in all kinds of places, ranging from air, water, and soil, and in harsh conditions such as hot springs, hydrothermal

vents, and polar regions (Abriouel et al., 2011). Commonly found in soil is an excellent variety of antibiotic-producing Bacillus species. Notable examples include *Bacillus subtilis, B. cereus, B. licheniformis, B. thuringiensis, Paenibacillus polymyxa, B. pumilus, B. megaterium, B. circulans,* and *B. brevis* (Alyousif 2022; Sitotaw et al., 2022).

Different identification techniques are proposed because members of the *Bacillus* genus display many morphological, physiological, and genetic resemblances (Kuta et al., 2009; Okpani et al., 2019). A great value can be seen in accurately identifying novel antibiotic-producing *Bacillus* strains, which is especially crucial at these times, as many pathogenic microorganisms are developing resistance to conventional antibiotics (Al-Turk et al., 2020). It is known that *Bacillus* species play a role in the production of a wide range of very effective antibiotics, which they use in nature as a competitive advantage against other microorganisms (Islam et al., 2022). Also of note is that metabolites produced by *Bacillus* species have many applications in medical, pharmaceutical, agricultural, and industrial settings (Su et al., 2020). Some antibiotics discovered to be generated by members of the *Bacillus* genus include polymyxin, bacitracin, colistin, difficidin, circulin, fengycin, iturin, and gramicidin. Moreover, these substances have shown significant efficacy against Gram-positive and Gram-negative bacteria (Alqahtani et al., 2023; Singh et al., 2012).

The discovery of new antibiotics worldwide is a growing problem, and it can be viewed as a response to the development of antimicrobial resistance in recent years. In the past, *Bacillus* species have been identified as good candidates for antibiotic production. However, the soil microbiota of Dutse Metropolis in the semi-arid part of North-west Nigeria has not been looked into very much. Given its atypical climate and soil makeup, it may host unknown *Bacillus* strains with antibiotic-producing properties, which presents a significant research gap that needs to be filled. This study presents findings that aim to fill that gap by isolating and characterizing the *Bacillus* species from the soil of Dutse to present Bacillus strains with the potential for antibiotic production. This study aims to contribute significant data on the soil microbiota within Dutse Metropolis in a way that supports global efforts in tackling antimicrobial resistance to improve public health (Su et al., 2020).

METHODS

Study Area

The research was carried out in the Microbiology laboratory of Federal University Dutse, Jigawa State. Dutse, the capital city of Jigawa State, lies in the northwestern region of Nigeria at 11.7460°N and 9.3409°E. Ecologically, Dutse is characterized by semi-arid conditions, low annual rainfall, and relatively high temperatures.

Sample Collection

Seven soil samples were collected from different areas within Dutse metropolis, especially areas with different soil and ecological characteristics, such as a car park, a cattle ranching site, a garden, a poultry shed, and a marshy area. 3g each of soil samples was collected at a depth of 8–10 cm from seven different locations, totaling 21g of soil samples collected altogether. The samples were packaged in labelled sterile polyethene bags containing the date, location, and time of collection. The soil samples were collected at Yalwawa, Mechanic Village, Bokoto, Federal University Dutse botanical garden, Hakimi, Rafin Sanyi, and Jigawa State Polytechnic campus.

Sample Preparation

All the glassware used in this research was sterilized in an oven. Other materials were sterilized using 70% ethanol. All media used in this research, which include Nutrient agar, Tryptic Soy agar, and Mueller-Hinton agar (MHA), were prepared following the manufacturer's instructions and sterilized at 121^{0}C at 15 lbs. pressure for 15 mins. The soil samples were weighed to determine actual weight after heavy sediments were isolated. The pH levels of the soil samples were determined based on the method carried out by Kuta et al. (2009). Heat treatment of the collected soil samples was carried out at 80°C for 10 minutes based on the method carried out by Thapa et al. (2021) to select for spore-forming Bacillus species. 1g each of collected soil samples from designated locations L1 to L7 was dissolved in 9ml of normal saline to make a homogenate stock solution. The

prepared stock homogenate was serially diluted in sterile test tubes labelled from 10^{-1} to10^{-6} dilution. The exact process was carried out for the samples obtained from the designated locations L1 to L7. 1ml of stock homogenate was added to 9ml of distilled water. From each dilution, 0.1ml was inoculated via the spread plate method on Tryptic Soy agar and incubated at 37^0c for 24 hours. Upon successful overnight incubation, colonies were identified based on morphology for further purification. The isolates were purified by repeated streaking on fresh agar plates (Abdulkadir & Umaru, 2012).

Identification of Bacillus species

The characterization of the colony isolates was achieved by initial morphological examination of the colonies in the plates for colony appearance, shape, size, colour, margin, elevation, surface, and texture of colonies. Microscopic examination of the isolates was conducted to determine cell shape, cell arrangement, endospore formation, spore shape, size, and Gram staining. Other distinguishing features were also determined, which include motility as some Bacillus species are motile due to the presence of flagella (Kearns & Losick, 2003), as well as hemolysis on blood agar, as some Bacillus species may exhibit beta hemolysis, alpha hemolysis, or gamma hemolysis (Sonenshein et al., 1993). Biochemical tests were also carried out to identify the various Bacillus species further. Organisms were identified by comparing morphological and biochemical characteristics with profiles from Bergey's Manual of Determinative Bacteriology (Holt et al., 1994).

Confirmation of Clinical Isolates

Clinically isolated pathogens (*Escherichia coli, Streptococcus pyogenes, Staphylococcus aureus, Klebsiella pneumoniae*) were obtained from the University Health Center, Federal University Dutse. The clinical isolates were subjected to biochemical tests for confirmation.

Screening for Antibiotics Production

The agar well diffusion method was used to evaluate the antibacterial activity of the *Bacillus* isolates. The test pathogens *Escherichia coli, Staphylococcus aureus, Streptococcus pyogenes, and Klebsiella pneumoniae* were prepared in sterile saline or nutrient broth, clinical bacterial suspensions. The turbidity of each suspension was made to stand for a 0.5 McFarland standard, which is at about 1.5 x 10^8CFU/ml. Mueller Hinton agar plates were swabbed to produce a uniform growth of the standardized bacteria. With a sterile cork borer, 9mm wells were aseptically made into the agar at equal distances. Cell-free supernatants from 24-hour-old. Bacillus culture broths, which were spun at 4000 rpm for 10 min, were obtained. Each well was filled with 100 µL of the Bacillus supernatant. 100 µL of sterile normal saline was added to separate wells, which served as the negative control to verify the absence of any antimicrobial action from the solvent. Ciprofloxacin at a concentration of 10 µg/mL was added into separate wells and was used as the positive control to which the antibacterial action can be compared. The plates were left to pre diffuse at room temperature for 30 minutes before incubation at 37^0 °C for 18–24 hours. All tests were run in triplicate for reproducibility. After incubation, the zones of inhibition around the wells were measured in millimeters using a transparent sterile ruler across the diameter zones. Results were as mean ± standard deviation. Statistical analysis using one-way ANOVA was conducted to determine the significance of differences observed, with $p < 0.05$ considered statistically significant.

RESULTS

Morphological description of the six isolates (B1–B6) as presented in table 1 revealed different colony morphologies that were all Gram-positive rods consistent with *Bacillus* spp. The isolates differed in colony morphologies from rough, irregular, and lobate (B1, B2, B3, B5) to smooth, round, and transparent colonies (B4, B6). These traits are typical of *Bacillus* species, which are capable of surviving under diverse environments, and the described traits, as seen against Bergey's Manual of Determinative Bacteriology, agree with their provisional classification at the family level.

Table 1. *Morphological Characterization of Isolates*

Isolate ID	Colony Morphology	Gram Staining	Shape	Suspected Organism
B_1	off-white, irregular, rough, convex, translucent, lobate colony	+	Rod	*Bacillus* spp
B_2	Yellowish green, large, irregular rough, entire flat colony	+	Rod	*Bacillus* spp
B_3	White, rough, opaque, irregular, abundantly growing colony	+	Rod	*Bacillus* spp
B_4	White, circular, flat, smooth, translucent colony	+	Rod	*Bacillus* spp
B_5	White, circular irregular, rough, translucent, lobate colony	+	Rod	*Bacillus* spp
B_6	White, large, smooth, round, convex, entire, creamy colony	+	Rod	*Bacillus* spp

**Consistent with standard definitions in Bergey's Manual of Determinative Bacteriology (Holt et al., 1994), suspected genus-level identifications of isolates were based on observed morphological properties like as Gram reaction, colony appearance, and rod-shaped cell morphology.*

Biochemical differentiation discriminated the six *Bacillus* isolates into different species (Table 2). B1 possessed a profile corresponding to Bacillus subtilis, and B2 to *Bacillus cereus*. B3 was *Bacillus licheniformis*, and B4 *Bacillus pumilus*. B5 possessed characteristics corresponding to *Paenibacillus polymyxa*, and B6 to *Bacillus megaterium*. These were based on their hydrolytic characteristics (gelatin, casein, starch), citrate utilization, reduction of nitrate, motility, and lecithinase, corresponding to reference values from Bergey's Manual.

Table 2. *Biochemical Test of Bacillus species isolates*

Test	B_1	B_2	B_3	B_4	B_5	B_6
GH	+	+	+	+	+	+
CH	+	+	+	+	+	+
SH	+	+	+	–	–	+
CI	+	+	+	+	–	+
Motility	+	–	+	+	+	+
NR	+	+	+	–	+	–
LC	–	+	–	–	+	–
Suspected Bacillus spp	*Bacillus subtilis*	*Bacillus cereus*	*Bacillus licheniformis*	*Bacillus pumilus*	*Paenibacillus polymyxa*	*Bacillus megaterium*

**Comparing biochemical test results of the isolates to reference standards stated in Bergey's Manual of Determinative Bacteriology (Holt et al., 1994), presumptive species identification was accomplished.*
Key: GH= Gelatin Hydrolysis, CH = Casein Hydrolysis, SH = Starch Hydrolysis, CI = Citrate Utilization, NR = Nitrate Reduction, LC = Lecithinase, (+)= Positive, (-)= Negative, B_1 – B_6 = Bacillus spp isolates

Test organisms were confirmed by Gram staining, cell morphology, and biochemical tests (Table 3). CI1 was confirmed to be *Escherichia coli* as Gram-negative rods with positive indole and casein but negative oxidase. CI2 was *Pseudomonas aeruginosa*, Gram-negative rod with positive VP, casein, and oxidase. CI3 was *Staphylococcus aureus*, Gram-positive cluster cocci that yielded positive indole, catalase, coagulase, methyl red, and VP tests. CI4 was *Klebsiella pneumoniae*, a Gram-negative rod that gave positive oxidase but negative for all other biochemistry parameters.

Table 3. *Biochemical Confirmation of Clinical Isolates Used for Antibiotic Screening*

Isolates Code	Gram Staining	Shape	IN	CA	CO	MR	VP	OX	Confirmed Bacteria
Cl_1	–	Rod	+	+	–	+	–	–	*Escherichia coli*
Cl_2	–	Rod	–	+	–	+	+	+	*Pseudomonas aeruginosa*
Cl_3	+	Clustered cocci	+	+	+	+	+	–	*Staphylococcus aureus*
Cl_4	–	Rod	–	–	–	–	+	–	*Klebsiella pneumonia*

Key: Cl_1 – Cl_6 = Clinical Isolates, (+)= Positive, (-)= Negative

Antibiotic screening of *Bacillus* isolates against pathogenic clinical isolates as presented in table 4 showed differential inhibitory potential. *Bacillus licheniformis* possessed the widest and strongest activity, with zones of inhibition of 23 ± 0.8 mm against *S. aureus*, 19 ± 0.5 mm against *E. coli*, 21 ± 0.9 mm against *K. pneumoniae*, and 14 ± 0.6 mm against *P. aeruginosa*, which in some cases was comparable with that of ciprofloxacin. *Bacillus subtilis* also possessed strong inhibition, particularly against *P. aeruginosa* (21 ± 0.7 mm). *B. cereus* showed moderate activity against all the pathogens, while *P. polymyxa, B. pumilus,* and *B. megaterium* showed comparatively weak or selective inhibitory activities. The negative control (normal saline) inhibited nothing, validating the antimicrobial activity of the *Bacillus* isolates.

Table 4. *Mean Zone of Inhibition (mm ± SD) of Antibiotics Producing Bacillus species isolates against Pathogenic Microorganisms*

Antibiotics Producing Bacillus spp	***Clinical Isolates (zone of inhibition in mm)***			
	Staphylococcus aureus	***Pseudomonas aeruginosa***	***Escherichia coli***	***Klebsiella pneumonia***
B. cereus	11 ± 0.6	17 ± 0.5	13 ± 0.7	8 ± 0.3
B. licheniformis	23 ± 0.8	14 ± 0.6	19 ± 0.5	21 ± 0.9
B. subtilis	19 ± 1.0	21 ± 0.7	14 ± 0.6	16 ± 0.4
P. polymyxa	ND	12 ± 0.4	9 ± 0.3	11 ± 0.6
B. pumilus	12 ± 0.5	6 ± 0.2	ND	8 ± 0.3
B. megaterium	ND	6 ± 0.4	ND	9 ± 0.2
Ciprofloxacin (Positive control)	21 ± 0.4	18 ± 0.3	8 ± 0.7	18 ± 0.4
Normal Saline (Negative control)	0	0	0	0

Key: (ND)= Not Determined, SD = Standard Deviation from triplicate measurements

DISCUSSIONS

Six Bacillus isolates: *B. subtilis, B. cereus, B. licheniformis, B. pumilus, P. polymyxa, and B. megaterium,* were chosen based on their morphological and biochemical characteristics, which are consistent with those described in Bergey's Manual of Determinative Bacteriology (Holt et al., 1994) (Table 1). All isolates exhibited gelatin, casein, and starch hydrolysis; this denotes the generation of extracellular enzymes, a typical characteristic of the *Bacillus* genus. In addition, *B. subtilis* and *B. licheniformis* had great citrate utilization and motility, which reflects their metabolic versatility and ecological adaptability to soil environments (Table 2). Such traits help identify *Bacillus* species as they present the phenotypic diversity and enzymatic capabilities. The findings of this study also support what was put forth by Torome et al. (2015), which is that substrate-associated *Bacillus* strains have similar biochemical profiles and can produce bioactive secondary metabolites.

This study takes it a step further to look at the antibacterial activity of these isolates against clinically isolated pathogens using the agar well diffusion method (Table 3).

B. licheniformis seemed to have the strongest and broadest range of antibacterial activity, with a zone of inhibition spanning from (14.0 ± 0.6 mm) against *P. aeruginosa* to (23.0 ± 0.8 mm) against *S. aureus* (Table 4). Likewise, *B. subtilis* exhibited potent inhibitory activity against *P. aeruginosa* (21.0 ± 0.7 mm) and *S. aureus* (19.0 ± 1.0 mm). *B. cereus* showed a moderate inhibitory effect against *P. aeruginosa* (17.0 ± 0.5 mm) and *E. coli* (13.0 ± 0.7 mm). In contrast, *P. polymyxa, B. pumilus,* and *B. megaterium* had variable or little effect; no zone of inhibition was seen, indicating a lower or strain-specific antibacterial capacity. A comparison with the positive control was conducted using ciprofloxacin (10 µg/mL) which showed zones of inhibitions of (21.0 ± 0.4 mm) against *S. aureus*, (18.0 ± 0.3 mm) against *P. aeruginosa*, (8.0 ± 0.7 mm) against *E. coli*, and (18.0 ± 0.4 mm) against *K. pneumoniae* which in turn underlines the very strong bioactivity of *B. licheniformis* and *B. subtilis* whose results in some cases were equal to or better than the positive control. Also, no zones of inhibition were seen from our negative control, which was normal saline, which confirmed the assay's reliability and ruled out any solvent-related effects.

These results are in agreement with past studies of Torome et al. (2015), who reported that the antimicrobial effects of *B. licheniformis* and *B. subtilis* are the result of the production of lipopeptides and polyketides, which are known for their wide range of bioactive properties. In a similar study conducted in 2021, Thapa et al. (2021) reported very high efficacy of soil found *Bacillus* strains against *S. aureus* and *E. coli*, which was also reported in this study. Also of note is the moderate sensitivity of *P. aeruginosa* to *B. subtilis* and *B. licheniformis*, which is a particular point of interest as this pathogen is known to possess intrinsic resistance to many antibiotics. The findings of this study are also consistent with the study conducted by Yahya et al. (2021), who also reported that *Bacillus* species produced compounds that target *P. aeruginosa*. A variation in the inhibition patterns was seen against *E.coli* and *K.pneumoniae*, which may be attributed to the structural differences in the outer membrane and resistance mechanisms like efflux pumps and beta-lactamase enzymes. While this study reports findings of some *Bacillus* strains with broad-spectrum activity, others may produce more specific or strain-related antimicrobial compounds.

The large-scale antibacterial activity from *B. licheniformis* and *B. subtilis* toward multi-drug resistant pathogens positions them as promising candidates for future antibiotic discovery. Also, the findings of this study reinforce that soil microbiota plays a key role as a host to novel antimicrobial agents. A point put forth by Thapa et al. (2021) as well. However, at the same time, it is put forward that the use of traditional biochemical-based methods for species identification presents limitations. Many *Bacilli* species present similar phenotypic traits, and environmental factors like pH, temperature, and nutrient availability also play a role in how they express themselves biochemically, which in turn may cause misclassification. Hence, the isolates are referred to as "suspected *Bacillus* species" to acknowledge the preliminary nature of their identification.

Molecular techniques, including 16S rRNA gene sequencing, polymerase chain reaction (PCR), or matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS), should be included in the subsequent research for exact species-level resolution and to prevent possible misidentification. These tools will enhance the taxonomic findings since they are more targeted. Additional research should center on isolating and structurally characterizing the bioactive substances causing the observed inhibition. This helps to create a direct connection between the phenotypic inhibition zones and the underlying chemical processes, therefore influencing the evaluation of therapeutic benefit and the development of new and enhanced antibacterial compounds. This will help establish a direct link between the phenotypic inhibition zones and the underlying chemical mechanisms, which play a role in assessing therapeutic value and in designing new and improved antimicrobial agents.

CONCLUSION

Six *Bacillus* isolates: *B. subtilis, B. licheniformis, B. cereus, B. pumilus, B. megaterium*, and *P. polymyxa*. were found, and their antibiotic-producing ability against pathogenic bacteria was assessed. Utilizing the agar well diffusion method, *B. licheniformis* and *B. subtilis* demonstrated the most excellent antibacterial effect, particularly against *Staphylococcus aureus* and *Pseudomonas aeruginosa*, pointing to their potential as producers of bioactive substances. These findings present the critical role of soil microflora as a natural reservoir of antimicrobial agents and further report on the role of *Bacillus* species in fighting multi-drug resistant pathogens. This study contributes to continuous research for alternative sources of antimicrobial agents. Also, it brings out the value of *Bacillus* species, which play a role in developing new antibiotics.

Author Contributions

D. T. Abbey: Conceptualization, Methodology, Investigation, Supervision, Writing – original draft; **D. E. Ifeanyi:** Formal analysis, Writing – review & editing; **L. E. Onyibo:** Validation, Data curation

Funding

This research received no external funding.

Ethical Approval

Not applicable.

Competing interest

The authors declare no conflicts of interest.

Data Availability

Data will be made available by the corresponding author on request.

Declaration of Artificial Intelligence Use

In this work, the author utilized artificial intelligence (AI) tools, specifically ChatGPT, to assist with grammar correction and language refinement. The author reviewed and edited the output carefully and assumes full responsibility for the final manuscript.

Acknowledgement

Our sincere gratitude to the Microbiology and Biotechnology Department, Federal University Dutse.

REFERENCES

Abdulkadir, M., & Umaru, W. (2012). Screening and isolation of the soil bacteria for ability to produce antibiotics. European Journal of Applied Sciences, 4(5), 211–215. https://www.semanticscholar.org/paper/Screening-and-Isolation-of-the-Soil-Bacteria-for-to-Abdulkadir-Umaru/ef87bbce207b3dbbaac861113a3cce25943c4cd9

Abriouel, H., Franz, C. M. A. P., Omar, N. B., & Gálvez, A. (2011). Diversity and applications of Bacillus bacteriocins. FEMS Microbiology Reviews, 35(1), 201–232. https://doi.org/10.1111/j.1574-6976.2010.00244.x

Al-Turk, A., Odat, N., & Massadeh, M. I. (2020). Isolation and molecular characterization of antibiotic producing Bacillus Licheniformis strains isolated from soil. Journal of Pure and Applied Microbiology, 14(4), 2363–2370. https://doi.org/10.22207/jpam.14.4.14

Alqahtani, O., Stapleton, P., & Gibbons, S. (2023). Production of antibacterial compounds using Bacillus spp. isolated from thermal springs in Saudi Arabia. Saudi Pharmaceutical Journal, 31(7), 1237–1243. https://doi.org/10.1016/j.jsps.2023.05.015

Alyousif, N. A. (2022). Distribution, occurrence and molecular characterization of Bacillus related species isolated from different soil in Basrah Province, Iraq. Biodiversitas Journal of Biological Diversity, 23(2), 679–686. https://doi.org/10.13057/biodiv/d230209

Antimicrobial Resistance Collaborators. (2022). Global burden of bacterial antimicrobial resistance in 2019: A systematic analysis. The Lancet, 399(10325), 629–655. https://doi.org/10.1016/S0140-6736(21)02724-0

Holt, J. G., Krieg, N. R., Sneath, P. H. A., Staley, J. T., & Williams, S. T. (1994). Bergey's manual of determinative bacteriology (9th ed.). Williams & Wilkins.

Islam, T., Rabbee, M. F., Choi, J., & Baek, K.-H. (2022). Biosynthesis, molecular regulation, and application of Bacilysin produced by Bacillus species. Metabolites, 12(5), 397. https://doi.org/10.3390/metabo12050397

Kearns, D. B., & Losick, R. (2003). Swarming motility in undomesticated Bacillus subtilis. Molecular Microbiology, 49(3), 581–590. https://doi.org/10.1046/j.1365-2958.2003.03584.x

Kuta, F. A., Nimzing, L., & Orka'a, P. Y. (2009). Screening of Bacillus species with potentials of antibiotics production. Applied Medical Informatics, 24(1-2), 42–46. https://ami.info.umfcluj.ro/index.php/AMI/article/view/79

Okpani, O. A., Chijioke, E. S., Kelechi, A. A., Kalu, N. K., & Peter, N. N. (2019). Evaluation of Bacillus species with antimicrobial producing potentials from dump sites in Abakaliki. World Applied Sciences Journal, 37(6), 446–452. https://www.idosi.org/wasj/wasj37(6)19/1.pdf

Singh, A. P., Singh, R. B., & Mishra, S. (2012). Studies on isolation and characterization of antibiotic-producing microorganisms from industrial waste soil sample. The Open Nutraceuticals Journal, 5, 169–173. https://benthamopenarchives.com/contents/pdf/TONUTRAJ/TONUTRAJ-5-169.pdf

Sitotaw, B., Ayalew, F., Girma, A., Mekonnen, A. B., Bin Jardan, Y. A., Nafidi, H.-A., & Bourhia, M. (2022). Isolation and identification of promising antibiotic-producing bacteria. Open Chemistry, 20(1), 1283–1291. https://doi.org/10.1515/chem-2022-0233

Sonenshein, A. L., Hoch, J. A., & Losick, R. (Eds.). (1993). Bacillus subtilis and other gram-positive bacteria: Biochemistry, physiology, and molecular genetics. American Society for Microbiology Press. https://doi.org/10.1128/9781555818388

Su, Y., Liu, C., Fang, H., & Zhang, D. (2020). Bacillus subtilis: A universal cell factory for industry, agriculture, biomaterials and medicine. Microbial Cell Factories, 19. https://doi.org/10.1186/s12934-020-01436-8

Thapa, A., Budhathoki, A., Sapkota, A., Sainju, M., & Shrestha, P. (2021). Isolation, identification and screening of Bacillus species with antimicrobial activity from different soil samples of Kathmandu Valley. Nepal Journal of Biotechnology, 9(2), 1–6. https://doi.org/10.54796/njb.v9i2.41892

Torome, T. K., Matasyoh, L. G., Orinda, G., & Gakuya, F. (2015). Isolation and characterization of antibiotic producing Bacillus species in Lake Bogoria, Kenya. African Journal of Microbiology Research, 9(14), 1037–1043. https://doi.org/10.5897/AJMR2015.7441

Yahya, G., Ebada, A., Khalaf, E. M., Mansour, B., Nouh, N. A., Mosbah, R. A., Saber, S., Moustafa, M., Negm, S., El-Sokkary, M. M. A., & El-Baz, A. M. (2021). Soil-Associated Bacillus species: A reservoir of bioactive compounds with potential therapeutic activity against human pathogens. Microorganisms, 9(6), 1131. https://doi.org/10.3390/microorganisms9061131

How to cite this article:
Abbey, D. T., Ifeanyi, D. E., & Onyibo, L. E. (2025). Biochemical Characterization and Antimicrobial Potential of Soil-Derived *Bacillus* Isolates. Recoletos Multidisciplinary Research Journal 13(2), 1-8. https://doi.org/10.32871/rmrj2513.02.01