Understanding the Genomics and Metabolism of Rare Actinobacteria as Potential Sources of New A Antibiotics

Ali Arkan Majhool[1], Abbas Abduladeem Abbas[2], Noor M. Faisal[3]

[1] Faculty of Dentistry, University of Kerbala, Kerbala, Iraq.

[2] Faculty of Dentistry, University of Kerbala, Kerbala, Iraq.

[3] Faculty of Dentistry, University of Kerbala, Kerbala, Iraq.

E-mail: ali.arkan@uokerbala.edu.iq (Corresponding Author)

Received: 19 / 1 /2026
Accepted: 10 / 3 /2026
Published: 31 / 3 /2026

DOI:
10.65682/kjnhs.v2.i1.102-112

Abstract

Introduction: There is an urgent requirement to develop novel antimicrobial agents that can counter the rising problem of antibiotic-resistant disease-causing microorganisms. The actinobacteria are a prolific source of natural products, and most of the known antibiotics are produced by these mycelial bacteria. Studies of rare actinobacteria can enhance the chances of new compounds with new chemical structures.

Methods: This review relates to intensive isolation and screening procedures of rare actinobacteria of understudied environments. The natural habitats like sandy, beach, desert and forest soils were viewed as potential harvest for collecting the less-represented genera in the past. The importance was also put on the assessments of ecological conditions, such as microbial interactions, which might trigger the expression of antibiotics.

Results: Natural discovery libraries and the probability of natural bioactive discovery are enhanced as a result of the screening of rare actinobacteria in unexploited natural habitats. It has been indicated that some antibiotics are solely expressed in some conditions of environmental or microbial interaction and there is a necessity to conduct research in other ecological niches such as soil, dry and marine ecosystems.

Conclusion: The rare actinobacteria can also be used as a good source of novel antimicrobial drugs. Increasing bio-discovery initiatives by focusing on isolating, screening, and ecologically discovering new antimicrobial agents can empower scientific capacity to discover new, desperately required antimicrobial agents.

Keywords: Soil Microbiology, Actinobacteria, Antibiotic resistance, Natural product.

1. Introduction

Multidrug-resistant (MDR) organisms have emerged and spread, and there are few treatment choices for infections, resulting in significant death rates (Catalano et al., 2022). It resulted in an urgent need to identify new highly effective and productive antimicrobial medicines; yet, despite scientific progress, there has been a weakness in the discovery of novel antibiotics during the last 30 years (Cook & Wright, 2022). Actinobacteria are a kind of Gram-positive bacteria that are unicellular and filamentous, forming a branching network and producing spores. They have been identified as sources of smells in drinking water, which spread widely throughout many settings (Majhool et al., 2025). Many species of actinobacteria have been isolated from natural caves (Fattah et al., 2024), clinical material (Alenazi et al., 2023), ocean sediments (Savitha et al., 2022), and plant roots (Oloumi et al., 2023). The unique habitat is not only in soils where rare actinobacteria species can be found but also in the fact that a range of possible actinobacteria and their unique metabolites have been discovered (Al-Shaibani et al., 2021). This paper set out to examine actinobacteria and their secondary metabolites in soils, including the soils at beaches, seas, and deserts, and their possible medical applications. Actinobacteria is untapped future source of natural products discovery as it has been identified through literature review that actinobacteria cultured in harsh conditions can perform diverse biological behaviors of compounds in medical and therapeutic products.

2. Taxonomy of Actinobacteria

Actinobacteria are classified into 6 classes, 23 orders, and 53 families in the current edition of Bergey's Manual of Systematic Bacteriology, which is based on comparative 16S rRNA gene sequence data (Bergey, 1994). Although Waksman isolated a variety of bacteria, several colonies have been discovered. They discovered that certain bacteria differed in appearance from their typical colonies (Rasmussen, 2022). These bacterial colonies penetrated deeply into the agar medium, were thick, pyramid-shaped, leathery, and often coated in surface fuzz that was different from the surface growth. They categorized this bacterium under a distinct class called actinobacteria because they were aware that such bacteria existed. Actinobacteria are defined by having a high DNA content of guanine and cytosine (69–73 mol%), as well as lengthy substrates that branch with aerial mycelia. Additionally, Figure 1 displays the taxonomy of actinobacteria (Rasmussen, 2022).

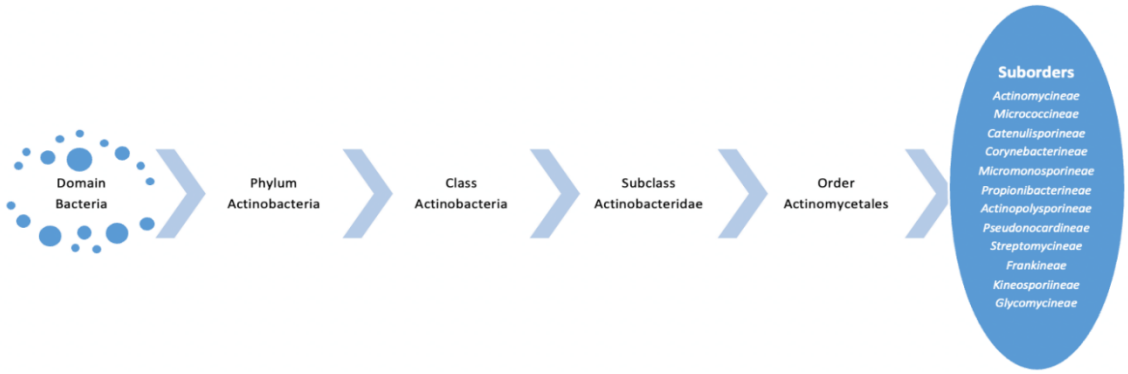

Figure 1. The current systemic classification of actinobacteria (Hartemink, 2021).

3. Occurrence and Distribution of Actinobacteria

Most of the actinobacteria live in most types of water, soil, and colonizing plants (Narsing Rao & Li, 2022). Actinobacteria are widely distributed in natural ecosystems around the world (Alenazi et al., 2023). Actinobacteria are found in a wide range of aquatic environments, including beach sands, marine sediments, rivers, streams, lake silt, and river sediments. *Micromonospora, Rhodococcus*, and *Streptomycetes* are among the most common actinobacteria in these environments (Ribeiro et al., 2020). Al- Shaibani in 2021, discovered actinobacteria species that produce beneficial secondary metabolites against malignant tumors and Plasmodium in marine environments (Al-Shaibani et al., 2021). Other studies have also confirmed the being of *Micromonospora* in rivers, river

sediments, and streams, and are considered to be an important part of the aquatic microflora (Zhang et al., 2020).

4. Identification Techniques of Actinobacteria

4.1 Morphological Techniques

The gold standard for identifying actinobacteria is culture. Its identification may be inferred from its shape on agar (Ribeiro et al., 2020).Therefore, physical traits, such as the presence or absence of spores at the substrate mycelium or the formation of zoospores in specialized spore cysts or sporangia, are frequently utilized to characterize its taxa. However, Rotich in 2018, found that actinobacteria on culture medium have a powdery look, colors ranging from white to grey to pinkish to yellow, and are typically difficult to remove due to their toughness. Depending on the media, some actinomycete strains can also generate melanin colors that vary from light-yellow brown to strong brown or distinguishing green. Some strains have the potential to create a distinctive color on the back of the colony, while others produce soluble pigments that range from yellow, orange, red, blue, and green in addition to melanin (Majhool et al., 2021).

4.2 Molecular Approach

The modern molecular techniques, including the 16S rRNA sequences, have been applied to characterize actinobacteria, and this technique has been beneficial in the taxonomy of actinobacteria (Idris, 2022). Characterization of the lineages is the most effective way to accomplish this because it is easier to discover the relationship between two lineages because these nucleic acids are either genes or products of genes (Majhool et al., 2025). Initial studies on nucleic acid hybridization even resulted in the establishment of the organism classification and identification in molecular biology (Idris, 2022).

4.2.1 RNA Analysis

Bonetti in 2020 argues that ribonucleic acid (RNA) is one of the basic elements of taxonomic analysis of organism (Bonetti et al., 2020). This molecule plays a critical role in protein synthesis and is present in all forms of bacteria and therefore it is an effective predictor in establishing the relationship between microorganisms. 16S RNA possesses highly conserved as well as more or less diverse sequences. This feature coupled with the horizontal movement of RNA among organisms makes it very practical in the study of evolutionary ties among different species (Majhool et al., 2021). Moreover, conserved RNA regions are also significant in that they are used as primer binding sites in polymerase chain reaction (PCR) and optimal hybridization targets of cloning RNA genes. The research on the RNA sequencing methods should therefore help in the development of the identification processes (Dorado et al., 2021). Oligonucleotide signatures are unique nucleotide sequences identified in 16S rRNA genes across all phylogenetic groups (Idris, 2022). Oligonucleotides are sequences that exist in some or all members of a specific phylogenetic group and may occur during the manufacture of primers that are unique to a genus or species According to Schloss in 2016, the characteristics of the 16SrRNA genes make it a useful phylogenetic tool and also make it useful for laboratory bacterial detection and identification (Schloss et al., 2016). These features include the fact that its presence in any bacteria hence is a general target to identification bacteria, the function of 16S rRNA has remained constant implying that random sequence mutations are a major accurate measure of interval (evolution) and lastly the 16S rRNA gene is long enough (approximately 1,500 bp) to presence statistically relevant sequence data, also, more importantly, the molecule consists of around 50 functional regions (Aßhauer et al., 2015).

4.2.2 Phylogenetic Studies

Actinobacteria and other bacteria are increasingly being studied using phylogenetic analyses based on 16S ribosomal DNA sequences (Bonetti et al., 2020). The 1542 bp 16S rRNA gene is an ideal tool for phylogenetic relationship analysis since it is highly conserved among microorganisms, particularly actinobacteria (Idris, 2022). DNA extraction is the first step in the analysis of 16S rDNA (Majhool et al., 2021). Following that, the gene encoding for 16S rRNA is amplified using a polymerase chain reaction. Following purification, the amplified products are sequenced using a DNA

sequences, which establishes the orientation of the bases throughout the samples' length (Schloss et al., 2016). The produced 16S rRNA sequences are aligned and compared to sequences in the GenBank database on the National Center for Biotechnology Information website using phylogenetic analysis techniques. These simple sequence setting comparisons will provide an assessment of the species' degree of relatedness. However, because the 16S rRNA gene is substantially conserved to provide a meaningful analysis at the species and subspecies levels, this study only permits identification up to the genus level (Rotich, 2018).Therefore, analysis of the 16S rRNA gene gives a time-saving option to the standard systems of classification and also allows for the evaluation of a broader range of variety than that received by physiological systems (Majhool et al., 2025).

4.2.3 Whole Genome Sequencing

Whole-genome sequencing refers to the technique of determining an organism's full DNA sequence. This procedure entails sequencing all of the organism's chromosomal DNA as well as the DNA contained in its mitochondria and, in the case of plants, chloroplasts (Watson et al., 2022). In the 1970s and 1980s, DNA sequencing techniques like Sanger and Maxam-Gilbert sequencing were done manually. However, the change to quicker, automated sequencing methods in the 1990s enabled more efficient sequencing of whole genomes (Watson et al., 2022). The complete genome of *Haemophilus influenzae* was sequenced for the first time in 1995. Following that, the genomes of several bacteria and archaea will be shown. *H. influenzae's* genome contains 1,830,140 base pairs of DNAS (Gandham et al., 2023). Amoebas have a genome of roughly 700 billion nucleotide pairs spread over many chromosomes (Gandham et al., 2023). Human germ cells contain approximately 3.2 billion base pairs; however, the total genome size is substantially bigger than that of bacteria. The first eukaryotic genome, S*accharomyces cerevisiae* was sequenced in 1996. This unicellular eukaryote was the first to undergo whole-genome sequencing (WGS), and it has over 12 million nucleotide pairs. In 1998, the nematode worm Caenorhabditis elegans was the first multicellular eukaryote to have its genome sequenced. The full DNA sequence of human chromosome 22 was reported in 1999 (Berberich & Hegele, 2023). In 2000, *Arabidopsis thaliana* became the first plant whose genome was sequenced. By 2002, the genome of the laboratory mouse, Mus musculus had also been completed. Illumina launched its Personal Full Genome Sequencing Service in 2009, offering 30× sequencing at a cost of $48,000 for per genome. According to various reports from 2015, the cost of whole genome sequencing (WGS) had reduced to roughly $1,000 (Balloux et al., 2018). However, as of 2017, significant portions of the human genome had still to be fully sequenced (Berberich & Hegele, 2023). The advent of whole genome sequencing has the potential to open up new avenues for forecasting bacterial pathogenicity. Currently, there are several high-throughput sequencing technologies available, and about 56,168 bacterial genomes have been read, 6,997 of which are Actinobacteria. Amongst these, only 486 actinobacterial genomes have been completely sequenced and annotated till date with the majority of them representing organisms that are a source of commercially important drugs or are contagious and infectious to humans and animals (Gandham et al., 2023).

5. Role of Actinobacteria in Soil

Soil is an acceptable medium for growing a variety of microorganisms, including bacteria and fungi. A single gram of soil can contain approximately one million microbial cells (Fattah et al., 2024). Actinobacteria perform critical functions in organic matter cycling and are an important complement to soil biological buffering. They help to regulate soil ecosystems naturally by performing nitrogen fixation and degrading high molecular weight natural substances like hydrocarbons found in contaminated soils (Ribeiro et al., 2020). The topic of actinobacteria is currently under the close study in the effort of understanding their place in the soil environment and their internalization with the co-existing microorganisms, especially those that produce the antibacterial compounds (Cook & Wright, 2022).

Bacteria, fungi, algae, and actinobacteria are known as separate classes of microorganisms in the subsurface environment (Zhang et al., 2020). These microbial communities facilitate the process of breakdown of senescent vegetation thus providing the necessary nitrogenous and organic food to support optimal plant growth (Ribeiro et al., 2020). The quality of the soil can be described as the

ability of soil system to play its ecological roles within the environmental constraints in an effective way, thus sustaining the biodiversity and ecosystem sustainability. Different soil characteristics, including chemical, physical and biological, are critical factors of this quality. Therefore, the physicochemical milieu of the soil is influenced by these qualities, thus affecting micro-communities in abundance, taxonomic richness and functional potential (Zhang et al., 2020). The extent of microbial heterogeneity, biomass, and metabolic activity all give information on the determination of soil health. Empirical studies suggest that soil quality is evaluated by a combination of physical, chemical, and biological indicators as shown in Figure 2.

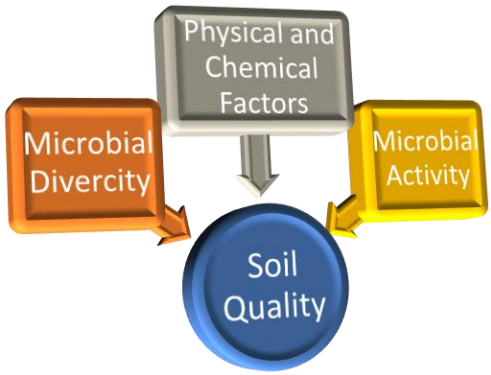

Figure 2. Parameters affecting on quality of soil (Zhang et al., 2020).

Actinobacteria play a crucial role in maintaining soil health, particularly in antibiotic production. They contribute to the breakdown and regeneration of various elements through the action of different types of microorganisms. According to one study, actinobacteria also play a role in chemical decomposition and recycling. They are also vital for organic matter mineralization, nitrogen fixation, and the restoration of physical and environmental health. The majority of Actinobacteria are free-living and found throughout nature, thriving in both terrestrial and aquatic habitats. They are highly tolerant of harsh environmental conditions (Soung & Han, 2022).

6. Identification and Profiling of Bioactive Compounds

New technology is critical to advancing the discovery of natural substances. Several screening techniques have been developed to efficiently isolate various beneficial chemicals from microbes (Majhool et al., 2021). As a result, systematic programs and creative methods for studying novel medical substances must be developed (Chanthasena et al., 2022). These techniques entail conducting investigations to identify and address known secondary metabolites within the research framework, a technique known as dereplication (Daigham & Mahfouz, 2020). Repeat compound detection is a screening paradigm that takes advantage of available libraries of secondary metabolites to prospectively identify new entities, so that redundant analyses of previously studied samples are avoided (Majhool et al., 2025). A set of spectroscopic modalities is essential to the tentative identification of the natural product constituents, and may frequently allow the initial recognition of individual compounds directly within the unrefined feedstocks. The most commonly used analytical stacks used to discover repeat-compounds include gas chromatography-mass spectrometry (GC-MS), high-performance liquid chromatography (HPLC) (Daigham & Mahfouz, 2020), liquid chromatography-mass spectrometry (LC-MS), nuclear magnetic resonance (NMR) spectroscopy, and infrared spectroscopy (IR); all of them have been demonstrated to be effective in detecting the presence of repeat-compounds (Chanthasena et al., 2022).

6.1 Thin Layer Chromatography (TLC)

TLC is a widespread and effective method of separation, analysis and detection of a large variety of compounds. It is anticipated to be applied to about 60 percent of lab tests. Hence, it is important to know its mechanism of action and technique. Saleem et al. (2023) refer to a wide range

of natural chemicals such as alkaloids, aromatic amines, flavonoids, flavanones, polyphenols, proteins, alcohols, acids, amino acids, glycoses, peptides, amides, antibiotics, vitamins, pesticides, and bile acid that can be isolated using TLC (Saleem et al., 2023). Several steroids such as hormones, estrogens, sterols, cholesterol, bile salts and progesterone can also be isolated using it. Moreover, this system identifies synthetic performance-enhancing substances like oxandrolone, mifepristone, nandrolone, and drostanolone, and are unlawful in sports like the Olympic Games (Daigham & Mahfouz, 2020). TLC is a cheap, quick, and straightforward isolation, method of a large diversity of compounds and semi-quantitative determination of these compounds using optical techniques. TLC, as a technique, has only recently gained the attention of researchers, complementing high-performance liquid chromatography (HPLC) and gas chromatography-mass spectrometry (GC-MS), because it is able to separate complex extracts, and generate quantitative data (Majhool et al., 2025). The improvement of plate quality, novel stationary phases, and novel methods of reagent detection have also been advanced in TLC. Besides that, density scanning and sample preparation have been visited (Al-Shaibani et al., 2021). The key benefits of thin-layer chromatography (TLC) are that it is cheap and has a large separation ability. The technique needs development chamber, chromatographic plates, guide materials, development reagents and certain solvents. The fact that it is very sensitive relative to other techniques, including gas chromatography/mass spectrometry (GC/MS) or high-performance liquid chromatography (HPLC), and that it requires a larger sample size are its primary drawbacks (Saleem et al., 2023).

6.2 High Performance Liquid Chromatography (HPLC)

High-performance liquid chromatography (HPLC) is a complex chemical analysis technique that is used to separate and identify compounds in complex mixtures (Clark, 2025). In an HPLC configuration, a sample mixture that is constrained by a comparatively low pressure of solvent flow is then passed through a chromatographic column that has a stationary phase absorber. The reaction of each analyte in the mixture with the selected reagents is unique and hence, each molecular entity has its own unique eluent profile. Proper diagnosis results are achieved through separation of the compounds separated and as they pass through the column at specific retention times. HPLC is widely used in the drug industry and in the quantitative analysis of biologically obtained products. Furthermore, it is rooted in the scientific activity of the separation of substances out of complex mixtures or the desired purification. High-performance liquid chromatography has also been used in the present research environment to explain the levels of vitamin D in serum (Chen et al., 2024). A typical HPLC system consists of a degassing system, an inlet system, a detector and pumping systems. The flow rate is very sensitive and its careful control is important to the proper functioning of the column. The output of the detector is proportional to the abundance of the analyte, thus making possible to perform a quantitative analysis after the separation of the species (Chen et al., 2024). A digital microprocessor is used to operate the system, and the data is represented through easy-to-use software interfaces. Also, there is usually a thermostatted furnace to maintain a constant temperature state during the chromatographic process (Faddetta et al., 2023). HPLC finds extensive use in an array of analytical methods, such as UV/VIS spectroscopy, photodiode array detection and mass spectrometry. Thin-layer chromatography (TLC) and sample separation by HPLC are considered compliant techniques of extracting the compound in different raw materials. Nevertheless, HPLC is usually regarded as a method that is more superior to TLC in regard to resolution during a separation process. Moreover, the closed-system mode of HPLC allows operating the flow of a mobile phase to be refined (Clark, 2025).

6.3 GC-MS (Gas Chromatography-Mass Spectrometry)

Gas chromatography-mass spectrometry (GC-MS) is a complex method of analysis that allows to identify various components of a sample in one sample through a combination of the separation properties of gas chromatography and the mass-spectrometric identification efficiency (Cunha et al., 2024). The technique has been widely used in a wide field of investigation and research, which includes fire forensics, environmental surveillance, pharmaceutical surveillance, and the

determination of complex organic compounds (Chen et al., 2024). The first GC-MS device appeared in 1959 and the further integration of cheap and small-sized computer platforms has radically streamlined the workflow arrangement which has significantly shortened the time taken to achieve conclusive analytical outcomes (Cunha et al., 2024). In 1968, Michael gave model quadrupole GC/MS systems to Stanford and Purdue universities (Michael, 1968). A mass spectrometer normally operates in one of two modes: entire scan or selective ion monitoring (SIM). The basic GC-MS instrument may do both procedures. The primary goal of using this technique is to quantify a material's components by comparing the relative concentrations of the atomic masses in the resulting spectrum. There are two types of analyses available: comparative and original. Comparative analysis often entails matching the given spectrum to a database of spectra to see if its characteristics are present in any of the samples kept in the archive (Chen et al., 2024).

7. Elucidation of The Structure

The identification of the structure of naturally occurring compounds may be both expensive and time-consuming, as it has been shown in recent literature (Stuart et al., 2020). However, there are numerous different innovative techniques in identification of compounds in semi-pure cultures. The major benefit of such systems is that they use lower amounts of pure chemicals (Lane et al., 2015). This allows known compounds and unknown molecules in extracts to be quickly distinguished by the researcher and removes the time-consuming, labor-intensive process of separating known compounds and allows novel compounds or specific structural motifs to be selectively isolated. This process uses a combination of analysis methods, such as HPLC DAD, HPLC MS, HPLC MS/MS, LC NMR, HPLC DAD Ms, HPLC NMR and HPLC MS- NMR (Stuart et al., 2020). Additionally, the combination of the liquid chromatography approach with the tandem mass spectrometry (HPLCUS/MS) has led to the evolution of the state-of-the-art methodologies in the analysis of microbial culture extract and plant-based samples, thus, aiding in the acquisition of the most unbiased and selective information on familiar structures with a remarkable sensitivity and selectivity (Lane et al., 2015).

8. Future Prospect of The Actinobacteria

Actinobacteria phylum produces a highly varied range of secondary metabolites, many of which have strong therapeutic potential. Actinomycetes still represent one of the main reserves of clinically useful antibiotics, with many having synthetically intractable structures, which are not readily accessible by traditional industrial chemistry (Sharma et al., 2018). In 2010, Goodfellow and Fiedler described the identification of actinobacterial strains with a wide range of structural motifs and biological activities, such as antibacterial, anticancer, antifungal, antiparasitic and antiviral activities (Goodfellow & Fiedler, 2010). Currently, actinobacterial diversity in terrestrial settings has been under systematic survey of less than one-third of the total terrestrial habitat (Earth). Actinobacteria have only been discovered to be the source of about 3 -percent of the known antibiotic agents, with 97 remaining to be studied. Such a gap highlights the urgency of the development of innovative technologies that will support the systematic screening and isolation of natural products that can be obtained out of actinobacteria (Sharma et al., 2018). The future opportunities of new pharmacophores are signaled by the chemical heterogeneity of naturally occurring bioactive molecules, especially those of the rare actinobacterial taxa. However, definitive identification of strong actinobacterial strains requires a profound knowledge of their taxonomic diversity, and ecological niche, without which it is impossible to inform the guided purification approaches (Prole et al., 2025). With the imperatives of biomedicine and biotechnology exploding to meet new demands in finding solutions to challenges of antibiotic resistance, emerging infectious diseases, and environmental pollution, the search after renewable microbial resources that may produce long-term, ecologically friendly solutions is becoming more urgent. Actinobacteria and their secondary metabolites have a broad-spectrum antimicrobial activity against a broad range of pathogens, including multidrug-resistant ones, like MRSA, *Shigella dysenteriae, Klebsiella spp.* and *Escherichia coli, Pseudomonas aeruginosa*, and *vancomycin-resistant Enterococcus* (Prole et al., 2025).

9. Conclusion

The continuously increasing problem of antibiotic resistance has shown the inefficiency of the current methods of antibiotic creation, therefore, underlining the urgent necessity to find new treatment prospects. The actinobacteria, including those occurring in problematic and non-understandable soil ecologies, are the potential source of new antibacterial agents with new molecular structures and action mechanisms. The present study discusses how these microorganisms excel in their biotic and ecological stability and metabolic diversity in order to put them at the forefront to find the next generation of natural products. Even after decades of study, the actinobacterial diversity has been studied only to a small fraction, and their metabolic capabilities have not well studied. To achieve the full potential of these odd actinobacteria in therapy, sampling of largely unexplored environments needs to be directed, and interdisciplinary methods are needed. Nowadays, the study of rare actinomycetes is not only scientifically necessary but also one of the basic components of the long-term research of antibiotics in the era of altering antimicrobial resistance and the emergence of novel pathogenic threats.

10. Acknowledgements

Our sincere appreciation to the University of Kerbala for their help and for providing the facilities in this study.

11. REFERENCES

Alenazi, A. M., Anwar, Y., Abo-Aba, S. E. M., & Bataweel, N. M. (2023). A review on Actinomycetes distribution, isolation, and their medical applications. *Nov. Res. Microbiol. J*, *7*, 1918–1931.

Al-Shaibani, M. M., Radin Mohamed, R. M. S., Sidik, N. M., Enshasy, H. A. El, Al-Gheethi, A., Noman, E., Al-Mekhlafi, N. A., & Zin, N. M. (2021). Biodiversity of secondary metabolites compounds isolated from phylum Actinobacteria and its therapeutic applications. *Molecules*, *26*(15), 4504.

Aßhauer, K. P., Wemheuer, B., Daniel, R., & Meinicke, P. (2015). Tax4Fun: predicting functional profiles from metagenomic 16S rRNA data. *Bioinformatics*, *31*(17), 2882–2884.

Balloux, F., Brynildsrud, O. B., Van Dorp, L., Shaw, L. P., Chen, H., Harris, K. A., Wang, H., & Eldholm, V. (2018). From theory to practice: translating whole-genome sequencing (WGS) into the clinic. *Trends in Microbiology*, *26*(12), 1035–1048.

Berberich, A. J., & Hegele, R. A. (2023). Genetic testing in dyslipidaemia: An approach based on clinical experience. *Best Practice & Research Clinical Endocrinology & Metabolism*, *37*(3), 101720.

Bergey, D. H. (1994). *Bergey's manual of determinative bacteriology*. Lippincott Williams & Wilkins.

Bonetti, A., Agostini, F., Suzuki, A. M., Hashimoto, K., Pascarella, G., Gimenez, J., Roos, L., Nash, A. J., Ghilotti, M., & Cameron, C. J. F. (2020). RADICL-seq identifies general and cell type–specific principles of genome-wide RNA-chromatin interactions. *Nature Communications*, *11*(1), 1018.

Catalano, A., Iacopetta, D., Ceramella, J., Scumaci, D., Giuzio, F., Saturnino, C., Aquaro, S., Rosano, C., & Sinicropi, M. S. (2022). Multidrug resistance (MDR): A widespread phenomenon in pharmacological therapies. *Molecules*, *27*(3), 616.

Chanthasena, P., Hua, Y., Rosyidah, A., Pathom-Aree, W., Limphirat, W., & Nantapong, N. (2022). Isolation and identification of bioactive compounds from Streptomyces

actinomycinicus PJ85 and their in vitro antimicrobial activities against methicillin-resistant Staphylococcus aureus. *Antibiotics*, *11*(12), 1797.

Chen, J.-J., Cheng, M.-J., Tseng, M., Wu, M.-D., & Weng, J.-R. (2024). Natural products from the culture broth of an actinobacteria Streptomyces sp. M5205 and evaluation of their antifungal activity. *Phytochemistry Letters*, *59*, 59–63.

Clark, G. M. (2025). *Screening of South African marine filamentous actinobacteria for antitubercular compounds*.

Cook, M. A., & Wright, G. D. (2022). The past, present, and future of antibiotics. *Science Translational Medicine*, *14*(657), eabo7793.

Cunha, M. B., Jorge, A. F., Nunes, M. J., Sousa, J. R., Lança, M. J., Gomes da Silva, M., & Gaudêncio, S. P. (2024). GC/MS Fatty Acid Profile of Marine-Derived Actinomycetes from Extreme Environments: Chemotaxonomic Insights and Biotechnological Potential. *Marine Drugs*, *23*(1), 1.

Daigham, G. E., & Mahfouz, A. Y. (2020). Isolation, characterization, and screening of actinomycetes producing bioactive compounds from Egyptian soil. *Egyptian Pharmaceutical Journal*, *19*(4), 381–390.

Dorado, G., Gálvez, S., Rosales, T. E., Vásquez, V. F., & Hernández, P. (2021). Analyzing modern biomolecules: the revolution of nucleic-acid sequencing–review. *Biomolecules*, *11*(8), 1111.

Faddetta, T., Polito, G., Abbate, L., Alibrandi, P., Zerbo, M., Caldiero, C., Reina, C., Puccio, G., Vaccaro, E., & Abenavoli, M. R. (2023). Bioactive metabolite survey of actinobacteria showing plant growth promoting traits to develop novel biofertilizers. *Metabolites*, *13*(3), 374.

Fattah, A., Bakeer, S. R., El-Gendy, W., Alkhalifah, A. O., Hozzein, D. H. M., Alenazi, W. N., Anwar, A. M., Abo-Aba, Y., Bataweel, S. E., Amaresan, N. M., Kumar, N., Naik, K., Bapatla, J. H., & Mishra, K. G. (2024). Antimicrobial and Anticancer Potential of Actinobacteria newly-isolated from Caves. In *The International Journal of Environmental Sciences* (Vol. 32, Number 1).

Gandham, P., Vadla, N., Saji, A., Vadlamudi, S., Ruperao, P., Selvanayagam, S., Saxena, R. K., Rathore, A., Gopalakrishnan, S., & Thakur, V. (2023). *Whole genome sequencing of actinobacterial strain Amycolatopsis sp.(BCA-696) identifies the underlying plant growth-promoting genes*.

Goodfellow, M., & Fiedler, H.-P. (2010). A guide to successful bioprospecting: informed by actinobacterial systematics. *Antonie Van Leeuwenhoek*, *98*(2), 119–142.

Hartemink, A. E. (2021). *Soil Science Americana: Chronicles and Progressions 1860─ 1960*. Springer Nature.

Idris, H. (2022). *Chemical compounds and antimicrobial activities of actinomycetes isolates from BRIS soil*.

Lane, A. L., Mandelare, P. E., & Ban, Y. H. (2015). New developments in NMR methodologies with special roles in natural product discovery. In *Applications of NMR Spectroscopy: Volume 3* (pp. 79–117). Bentham Science Publishers.

Majhool, A. A., Idris, H. B., Abdullah, M. D. D., Zin, N. M., & Gany, S. A. (2025). BRIS soil actinomycetes and their antimicrobial activity. *Microbiology Australia*, *46*(3), 158–162.

Majhool, A. A., Idris, H., Hakimi, W. M. N., & Abdullah, M. D. D. (2021). Chemical Compounds and Antimicrobial Activities of Actinomycetes Isolates from BRIS Soil. *Research Journal of Pharmacy and Technology*, *14*(9), 4783–4788.

Michael, W. R. (1968). Metabolism of linear alkylate sulfonate and alkyl benzene sulfonate in albino rats. *Toxicology and Applied Pharmacology*, *12*(3), 473–485.

Narsing Rao, M. P., & Li, W.-J. (2022). Diversity of actinobacteria in various habitats. In *Actinobacteria: Microbiology to Synthetic Biology* (pp. 37–58). Springer.

Oloumi, H., Khaleghi, M., & Dalvand, A. (2023). Isolation and identification of endophytic actinobacteria from Iris persica and Echium amoenum plants and investigation of their effects on germination and growth of wheat plant. *Food Science & Nutrition*, *11*(9), 5296–5303.

Prole, J. R., Allenby, N., Manning, D. A. C., & Goodfellow, M. (2025). Identification of four novel Streptomyces isolated from machair grassland soil using a culture-based bioprospecting strategy: Streptomyces caledonius sp. nov., Streptomyces machairae sp. nov., Streptomyces pratisoli sp. nov. and Streptomyces achmelvichensis sp. nov. *International Journal of Systematic and Evolutionary Microbiology*, *75*(4), 006736.

Rasmussen, N. (2022). René Dubos, the autochthonous flora, and the discovery of the microbiome. *Journal of the History of Biology*, *55*(3), 537–558.

Ribeiro, I., Girão, M., Alexandrino, D. A. M., Ribeiro, T., Santos, C., Pereira, F., Mucha, A. P., Urbatzka, R., Leão, P. N., & Carvalho, M. F. (2020). Diversity and bioactive potential of actinobacteria isolated from a coastal marine sediment in northern Portugal. *Microorganisms*, *8*(11), 1691.

Rotich, M. C. (2018). *BIO-PROSPECTING FOR BROAD SPECTRUM ANTIBIOTIC PRODUCING ACTINOMYCETES ISOLATED FROM VIRGIN SOILS IN KERICHO COUNTY*.

Saleem, M., Hassan, A., Li, F., Lu, Q., Ponomareva, L. V, Parkin, S., Sun, C., Thorson, J. S., Shaaban, K. A., & Sajid, I. (2023). Bioprospecting of desert actinobacteria with special emphases on griseoviridin, mitomycin C and a new bacterial metabolite producing Streptomyces sp. PU-KB10–4. *BMC Microbiology*, *23*(1), 69.

Savitha, T., Khalifa, A., & Sankaranarayanan, A. (2022). Isolation of Actinobacteria from soil and marine sediment samples. In *Methods in Actinobacteriology* (pp. 1–5). Springer.

Schloss, P. D., Jenior, M. L., Koumpouras, C. C., Westcott, S. L., & Highlander, S. K. (2016). Sequencing 16S rRNA gene fragments using the PacBio SMRT DNA sequencing system. *PeerJ*, *4*, e1869.

Sharma, P., Dutta, J., & Thakur, D. (2018). Future prospects of actinobacteria in health and industry. In *New and Future Developments in Microbial Biotechnology and Bioengineering* (pp. 305–324). Elsevier.

Soung, S. H., & Han, S.-I. (2022). Isolation and characterization of odorous compounds (geosmin and 2-MIB) producing Actinobacteria in North Han River Watershed. *The Microbiological Society of Korea*, *58*(1), 23–32.

Stuart, K. A., Welsh, K., Walker, M. C., & Edrada-Ebel, R. (2020). Metabolomic tools used in marine natural product drug discovery. *Expert Opinion on Drug Discovery*, *15*(4), 499–522.

Watson, A. K., Kepplinger, B., Bakhiet, S. M., Mhmoud, N. A., Chapman, J., Allenby, N. E. E., Mickiewicz, K., Goodfellow, M., Fahal, A. H., & Errington, J. (2022). Systematic whole-genome sequencing reveals an unexpected diversity among actinomycetoma pathogens and provides insights into their antibacterial susceptibilities. *PLOS Neglected Tropical Diseases*, *16*(7), e0010128.

Zhang, L., Zhao, F., Li, X., & Lu, W. (2020). Contribution of influent rivers affected by different types of pollution to the changes of benthic microbial community structure in a large lake. *Ecotoxicology and Environmental Safety*, *198*, 110657.