Int J Adv Life Sci Res. Volume 9(4) 134-144
https://doi.org/10.31632/ijalsr.2026.v09i04.009

International Journal of Advancement in Life Sciences Research

Online ISSN: 2581-4877

Journal homepage http://ijalsr.org



Original Article

Integrating Ethnographic Knowledge and Rhizosphere Microbiology of *Peganum harmala* in Kyrgyzstan

Baktybek Isakov[1], Saykal Bobusheva[2], Hossein Zeinalzadeh-Tabrizi[3*]

[1] *Department of Sociology, Faculty of Humanities, Kyrgyz-Turkish Manas University, Bishkek, 720044, Kyrgyzstan*

[2] *Department of Plant Protection, Faculty of Agriculture, Kyrgyz-Turkish Manas University, Bishkek, 720044, Kyrgyzstan*

[3] *Department of Horticulture and Agronomy, Faculty of Agriculture, Kyrgyz-Turkish Manas University, Bishkek, 720044, Kyrgyzstan*

Corresponding Author's Email: h.zeynelzade@manas.edu.kg

Abstract

Background: *Peganum harmala* (locally known in Kyrgyzstan as adyrashman or ysyryk) is a medicinal plant with a long history of traditional use in Kyrgyzstan, with references to its applications preserved in the Manuscripts Fund of the National Academy of Sciences of the Kyrgyz Republic. However, little is known about the rhizosphere-associated bacterial communities of this species. This study examined *P. harmala* using an integrated microbiological and molecular approach to characterize its rhizosphere bacteria and investigate their microbial diversity. **Methods:** *P. harmala* plants were collected from the Kashka-Suu rural area of the Alamudun District, Chüy Region, Kyrgyzstan. Rhizosphere-associated bacteria were isolated using standard microbiological techniques and characterized by morphological and biochemical analyses. Microbial diversity was investigated through 16S rRNA gene amplification, sequencing, and taxonomic identification. **Results:** The analysis revealed the presence of bacterial genera including *Pseudomonas*, *Stenotrophomonas*, and *Paenarthrobacter*, highlighting the diversity of microorganisms associated with the plant root environment. These findings provide new insights into the bacterial communities associated with *P. harmala* and establish a foundation for future investigations into plant–microbe interactions and the ecological roles of root-associated microorganisms. **Conclusion:** This study represents the first characterization of the rhizosphere bacterial community associated with *Peganum harmala* in Kyrgyzstan using 16S rRNA gene-based identification. The findings expand current knowledge of the microbial diversity associated with this medicinal plant and provide a basis for future studies exploring the ecological, agricultural, and biotechnological significance of its root-associated microorganisms.

Keywords: *Genus Pseudomonas; Kyrgyzstan; Peganum harmala (Adyrashman); Rhizosphere Bacteria*

Introduction

Peganum harmala, frequently referred to in Kyrgyzstan as *adyrashman* or *ysyryk*, stands as a globally recognized medicinal herb with profound cultural, therapeutic, and ceremonial importance spanning Central Asia, the Middle East, and the Mediterranean. For generations, local communities across these territories have turned to *P. harmala* as a versatile healing resource, employing it to alleviate pain, manage respiratory ailments, treat gastrointestinal issues, and address diverse inflammatory conditions, alongside its traditional use in ritual cleansing and protective customs (Asgarpanah & Ramezanloo, 2012; Mamedov *et al*., 2018). Its role in traditional medicine is particularly profound in Central Asia, where it gained renewed attention during the COVID-19 pandemic as a potent antiviral

Received on : 16th February 2026; Revised version received on :31st July 2026; Accepted: 7th August 2026

herbal remedy utilized by local populations (Penkala-Gawęcka, 2023). Ethnographic research consistently indicates that *P. harmala* remains a cornerstone of traditional pharmacopeias, and historical archives in Kyrgyzstan verify its application since the nomadic era, specifically regarding its use as a fumigant, analgesic, and antiseptic agent.

This medicinal versatility is echoed in Traditional Chinese Medicine, where the plant is extensively utilized and its therapeutic properties are rigorously documented (Li, 2024). Currently, *P. harmala* is prescribed for muscle weakness, joint disorders, headaches, cough, and menstrual irregularities, highlighting its sustained relevance across diverse medical systems. The phytochemical composition of the plant is remarkably complex, featuring beta-carboline and quinazoline alkaloids, as well as flavonoids, coumarins, and essential oils. Because *P. harmala* possesses a degree of inherent toxicity, researchers have focused on isolating its alkaloids to harness their specific pharmacological benefits for treating cancer, inflammation, and depression (Gökkaya *et al*., 2023). Recent chemical and biological analyses have scrutinized peganine and deoxypeganine—the plant's primary alkaloids—noting that peganine levels peak during the branching stage of growth, marking it as a promising candidate for future pharmaceutical development (Khudoynazarova *et al*., 2025). Furthermore, harmaline and harmine have been successfully isolated from the seeds, providing high-purity compounds for biochemical and pharmacological research (El Fissi *et al*., 2025).

Experimental data from pharmacological trials reinforce these observations, with seed extracts showing analgesic efficacy on par with substances like aspirin and morphine (Koçak & Şahin, 2009). Nevertheless, the safety margin for therapeutic use is quite narrow; studies on animal models involving seed decoctions demonstrated that while testosterone levels were unaffected, higher concentrations led to a rise in sperm morphology abnormalities (Alghoul *et al*., 2025). Beyond analgesia, purified fractions show antimicrobial effects against *Staphylococcus aureus* and *Escherichia coli* (Borde *et al*., 2013). More recent molecular studies provide mechanistic explanations for these neurotropic actions, demonstrating strong binding affinities between harmine derivatives and several neural receptor systems, including serotonin 5-HT2C, dopamine D2, and monoamine oxidase A and B (Doskaliyev *et al*., 2021).

While the field of phytochemistry is largely established, the ecological and microbiological elements governing *P. harmala* are becoming a significant new frontier of inquiry. Recent investigations into the stems of *P. harmala* have isolated six endophytic bacteria that demonstrate strong antifungal activity, phosphate solubilization, and nutrient secretion, suggesting their potential as bio-fertilizers in agriculture (Babar *et al*., 2025). Moreover, since *P. harmala* is a xerophytic plant, it has been identified as a significant host for drought-resistant endophytic bacteria, which may be a key factor in its resilience in arid environments (Mukhtorova *et al*., 2024).

This study addresses the gap in current literature by moving beyond the plant acting in isolation and viewing *P. harmala* as a complex plant–microbe system. By merging microbiological assessments of rhizosphere bacterial structures with 16S rRNA sequencing and historical-ethnographic data from the From the Manuscripts Fund of the National Academy of Sciences of the Kyrgyz Republic, this work presents a unique perspective on the microbial diversity associated with a medicinal plant. The isolation and identification of bacterial genera such as *Pseudomonas*, *Stenotrophomonas*, and *Paenarthrobacter* provide valuable insights into the composition and diversity of root-associated microbial communities. By applying microbiological approaches, this study contributes to a better understanding of plant-associated bacteria and their ecological relationships with medicinal plants, providing a foundation for future investigations into their roles in plant–microbe interactions.

Materials and Methods

Plant Sample Collection

Peganum harmala samples were collected in accordance with standardized methodologies established for rhizosphere-associated microbial community studies (Bulgarelli *et al.,* 2013; Compant *et al.,* 2010; Lundberg *et al.,* 2012) during May 2025 from the Chui region of Kyrgyzstan. The species

was identified by botanist M. Ganybaeva, Ph.D. (Candidate of Biological Sciences) using the taxonomic keys described in Flora of the Kirghiz SSR, Vol. 7, and was further verified in the Plantarium database (https://www.plantarium.ru). A voucher specimen KG-PH-001 has been deposited in the Botany Laboratory of the National Academy of Sciences of the Kyrgyz Republic.

Bacterial Isolation Procedures

Following field collection, plant samples were immediately placed in sterile collection bags and transported to the laboratory within 24 hours to ensure microbial viability for subsequent isolation procedures. The sampling site was at an altitude of 1,296 meters above sea level. Plants were carefully excavated, and loosely adherent bulk soil was removed through gentle agitation. Root systems were subsequently sectioned longitudinally using sterile instruments (Doolotkeldieva & Bobusheva, 2016). Root segments measuring 2–3 cm in length were aseptically excised and transferred into sterile 50 mL conical tubes containing 25 mL of sterile phosphate-buffered saline (PBS, pH 7.2).

Sample suspensions underwent mechanical agitation via vortex mixing for 10 minutes to facilitate the detachment of tightly adherent rhizobacteria (Lundberg *et al.,* 2012). The resultant bacterial suspensions were subjected to serial decimal dilutions up to 10^{-6}, and 100 µL aliquots were plated onto Luria-Bertani (LB) agar medium using the spread plate technique. Inoculated plates were incubated at 28°C for 48–72 hours under aerobic conditions. Individual colonies exhibiting distinct morphological characteristics were selected and purified through successive streak plate isolation until pure cultures were established.

The Phenotypic Characterization of Bacteria

The phenotypic characterization of bacterial isolates was carried out following the biochemical approaches outlined in Bergey's Manual of Systematic Bacteriology (Krieg *et al.,* 2010). Preliminary identification of plant growth-promoting rhizobacteria (PGPR) was based on morphological traits. After 24 h of incubation on peptone agar at 30°C, purified colonies were examined for colony morphology, including pigmentation, form, size, surface characteristics, opacity, and texture. Cellular features, such as shape, dimensions, and division patterns, were further assessed using light microscopy.

DNA Extraction and PCR Analysis

For genetic characterization, genomic DNA was extracted from bacterial log phase liquid cultures grown overnight at 27°C in LB medium using the DNeasy Blood and Tissue kit (Qiagen) according to the standard protocol as provided by the manufacturer. The 16S rRNA gene was amplified using universal bacterial primers 27F (5′-AGAGTTTGATCMTGGCTCAG-3′) and 1492R (5′-TACGGYTACCTTGTTACGACTT-3′), as originally described by Weisburg *et al*. (1991). PCR reactions were carried out in 25 µL volumes containing 12.5 µL of 2× PCR Master Mix (Thermo Fisher Scientific), 1 µL of each primer (10 µM), 1 µL template DNA (~50 ng), and nuclease-free water to volume. The thermal cycling program consisted of an initial denaturation at 95°C for 3 min; 30 cycles of denaturation at 95°C for 30 s, annealing at 55°C for 30 s, and extension at 72°C for 90 s; followed by a final extension at 72°C for 10 min. PCR products were analyzed by electrophoresis on 1% agarose gels stained with ethidium bromide and visualized under UV illumination. Sanger sequencing was performed by Starseq (Mainz, Germany) using primers 16S rRNA 27F (5′-AGAGTTTGATCMTGGCTCAG-3′) and 1492R (5′-TACGGYTACCTTGTTACGACTT-3′) and raw sequence data were combined into a single consensus sequence for each bacterial isolate using the MEGA program version 6 (Tamura *et al*. 2013).

Results

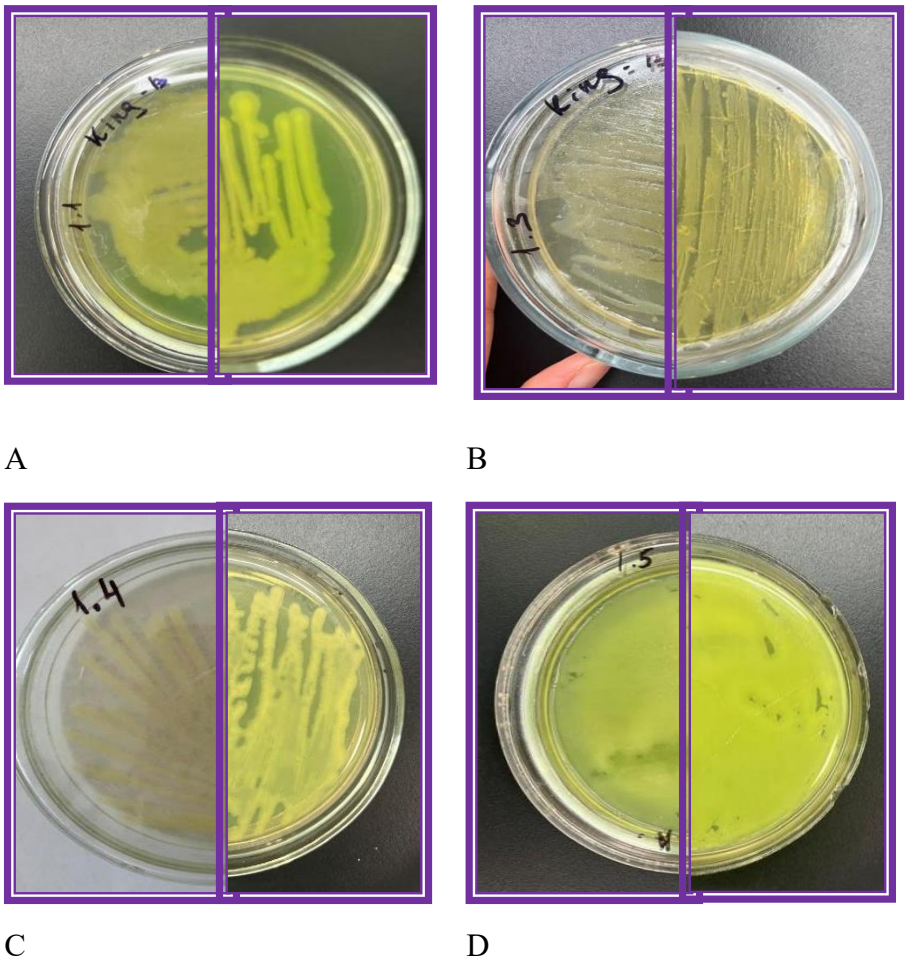
Peganum harmala plants collected from the Chui region of Kyrgyzstan exhibited well-developed root systems suitable for rhizosphere bacterial isolation (Figure 1). In our investigation, a total of 10 bacterial strains demonstrated robust growth on LB medium and on the King's B agar surface (Figure

2). Microorganisms associated with medicinal plants represent valuable sources to produce significant bioactive metabolites.



a b

Figure 1: *a: General View of the Field Showing the Growth of Peganum harmala; b: Individual Plant of Peganum harmala with Rhizosphere*



A B

C D

Figure 2: *Colony Morphologies on King's B Agar of Rhizosphere-Associated Bacteria Isolated from Peganum harmala: (A) Pseudomonas cedrina (P1.1); (B) Stenotrophomonas sp. (P1.3); (C) Paenarthrobacter aurescens (P1.4); (D) Pseudomonas salomonii (P1.5).*

Phylogenetic Identification Through 16S rRNA Gene Sequence Analysis

16S rRNA gene sequence analysis is a widely used molecular approach for the preliminary taxonomic characterization of bacterial isolates from various environmental sources (Clarridge, 2004). In the present study, this approach was applied to characterize plant growth-promoting rhizobacteria (PGPR) isolated from the rhizosphere of *Peganum harmala*.

The bacterial isolates were subjected to PCR amplification of the 16S rRNA gene, and successful amplification was confirmed by agarose gel electrophoresis, which showed clear PCR products for the isolates analyzed (Figure 3). The obtained sequences were compared with available reference sequences to determine their closest taxonomic affiliations.

Based on sequence similarity analysis, the isolates showed the highest similarity to members of the genera *Pseudomonas*, *Stenotrophomonas*, and *Paenarthrobacter*. The most closely related reference strains included *Pseudomonas cedrina, Pseudomonas koreensis, Stenotrophomonas sp., Paenarthrobacter aureus, Pseudomonas salomonii, Pseudomonas sp., and Pseudomonas fluorescens*.

Since 16S rRNA gene sequence analysis may have limited resolution for discrimination between closely related bacterial species, the obtained results are considered preliminary taxonomic assignments. Further genomic analysis and comprehensive phylogenetic reconstruction would be required for definitive species-level identification.

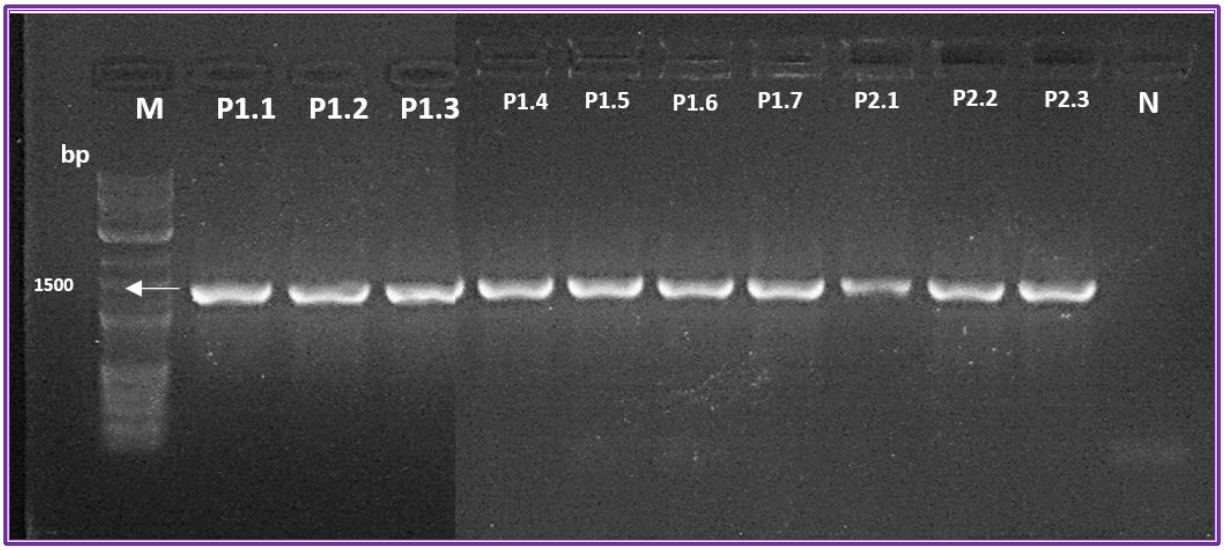


Figure 3: *Agarose Gel Electrophoresis Confirming PCR Amplification of 16S rRNA Gene Fragments from Bacterial Isolates*

Discussion

Following standardized protocols, the isolated bacterial strains underwent a thorough morphological and biochemical assessment. Key physical traits, such as cellular structure, colony topography, pigmentation, and peripheral margins, were documented in accordance with the frameworks established by Krieg *et al*. (2010). Gram differentiation was performed using the methodology of Cappuccino and Sherman (2014) and further validated through the 3% KOH alkaline test (Halebian *et al*., 1981; Dida *et al*., 2018).

Among the identified species, *P. fluorescens* stands out for its extensive use as a biological control agent, consistently proving effective against damping-off diseases in various cereals and legumes (Weller, 2007; Santoyo *et al*., 2012). Related species, including *P. synxantha*, *P. salomonii*, and *P. cedrina*, share many morphological and physiological characteristics with *P. fluorescens*, although they have not yet been as widely researched. Notably, *P. cedrina* demonstrates a unique ability to

adapt to cold environments, emphasizing the remarkable ecological flexibility of this genus (Bossis *et al*., 2000). Beyond *Pseudomonas*, the *P. harmala* rhizosphere was found to harbor *Stenotrophomonas* and *Paenarthrobacter*. *Stenotrophomonas* spp. are Gram-negative, non-fermentative rods recognized for their broad-spectrum antagonistic capabilities. While they may not be traditional antibiotic producers, they synthesize extracellular enzymes like proteases and chitinases, alongside volatile organic compounds that suppress competing microbes (Ryan *et al*., 2008). Their ecological contribution is largely tied to competitive exclusion and colonization resistance, which indirectly bolsters the plant's natural defenses. However, it is worth noting that certain strains, such as *S. maltophilia*, are considered opportunistic pathogens, exhibiting multidrug resistance through mechanisms like efflux pumps and biofilm development (Brooke, 2012).

Stenotrophomonas spp., much like the bacteria mentioned above, actively suppress the proliferation of harmful fungi and bacteria within the soil and the plant's immediate vicinity. This inhibitory effect stems from their capacity to generate enzymes such as proteases and chitinases. Stenotrophomonas spp. also releases Volatile Organic Compounds (VOCs) with antioxidant properties through the plant's scent, which serves to further impede microbial growth.

Paenarthrobacter nitroguajacolicus (formerly known as *Arthrobacter nitroguajacolicus*) is a Gram-positive actinobacterium characterized by its diverse metabolic potential, specifically its ability to break down nitroaromatic pollutants like 4-nitroguaiacol (Kotoučková *et al*., 2004). Certain PGPR strains produce secondary metabolites that simultaneously protect plants from soil-borne pathogens and enhance their growth under saline stress (Kumar *et al*., 2023). This dual functionality underscores its potential for both environmental bioremediation and sustainable agricultural practices.

Pseudomonas fluorescens and *Paenarthrobacter nitroguajacolicus* function as beneficial soil-dwelling organisms that curb the spread of pathogenic fungi and harmful bacteria around the plant's root zone. These characteristics - specifically their ability to neutralize fungal and bacterial threats - offer a scientific explanation for why *Peganum harmala* (adyrashman) has been esteemed as a powerful medicinal plant by local communities for centuries. Reinforcing this perspective, the Manuscript Fund of the National Academy of Sciences of the Kyrgyz Republic maintains archival records from Soviet researchers (1946–1990), which include numerous oral accounts describing adyrashman as an effective treatment for various ailments related to swelling and inflammation.

This perennial herb thrives in diverse habitats, including sun-drenched slopes, saline valleys, residential yards, and wasteland areas. It is characterized by highly branched stems reaching up to 25 cm, narrow foliage, and white or yellowish blossoms, all supported by an extensive vertical root system. Locally, it is frequently referred to as *ysyryk*. The plant typically emerges in May, enters its flowering stage in July, and produces fruit by August. Its fruit takes the form of a globular capsule containing seeds that mature into a deep brown hue. Notably, livestock generally refrain from grazing on the plant due to its chemical properties.

According to archival records, the Kyrgyz nomads—whose primary means of transport was the horse—faced frequent problems of joint swelling due to the harsh mountainous terrain, where horses were constantly ridden, loaded with heavy burdens, and driven over long distances. This specific condition, a swelling of the knees known as *Buba* disease, was traditionally managed using *adyrashman*. Specifically, the fruits, leaves, and stems of the plant were crushed, ground, and boiled in water. The horse's swollen leg was exposed to the steam of this boiling mixture for about half an hour, then wrapped warmly, after which the swelling subsided (Tügelov, 1946: 17).

Many inflammatory conditions and swellings are associated with complex biological processes, including microbial interactions. The traditional use of adyrashman highlights its importance as a medicinal plant; however, the contribution of its associated microbial communities to these properties requires further investigation.

Collectively, the bacterial isolates from the rhizosphere of *P. harmala* demonstrate diverse antagonistic mechanisms, with *Pseudomonas* spp. serving as the primary drivers of pathogen

suppression, while *Stenotrophomonas* and *Paenarthrobacter* contribute complementary functions through enzymatic degradation, volatile-mediated inhibition, and stress-adaptive interactions. These microbial consortia not only enhance the resilience of *P. harmala* but also represent valuable reservoirs of novel antimicrobial compounds with applications in agriculture, environmental biotechnology, and natural product discovery.

Our results indicated that *Pseudomonas* genus was the dominant group in this study based on 16S rRNA gene sequence analysis. Five isolates were classified within the Proteobacteria class, predominantly consisting of *Pseudomonas*. The presence of *Pseudomonas* as a common soil inhabitant of plant, biocontrol agent and antimicrobial compound producer is already reported (O'Sullivan & O'Gara, 1992).

The subsequent taxonomic identification based on partial 16S rRNA gene sequences revealed a diverse array of bacterial groups spanning different genera. In the present study, it has been seen that medicinal plants harbored a large fraction of potential bacteria similar as reported in previous studies (Aravind *et al*., 2010). Secondary metabolite production is the phenomenon used by bacteria to promote plant growth and defend against different soil pathogens (Dobbelaere *et al*., 2003). In previous studies on medicinal plants, it has been seen that each plant harbors a specific kind of bacterial population depending upon their alkaloids and secondary metabolites produced by associated bacteria (Qi *et al*., 2012).

Regarding the ethnographic context, local communities are naturally unaware of the specific bacterial isolates inhabiting the roots and rhizosphere of *Peganum harmala*, or the antagonistic properties these microbes grant the plant. Instead, they have historically valued it as a holistic medicinal herb used to treat a vast range of conditions. Oral accounts collected by Soviet researchers in Kyrgyzstan between 1946 and 1990—now preserved in the Manuscripts Fund of the National Academy of Sciences of the Kyrgyz Republic—include records dating back to pre-1930s nomadic Kyrgyz life. These archives provide extensive inventories of folk medicine, in which the use of *P. harmala* is a recurring and significant theme.

One illustrative example concerns horses, the primary means of transport and livelihood for Kyrgyz nomads. Due to the mountainous terrain—where horses constantly bore riders, carried loads, and travelled across long distances—the animals often developed swollen and painful knees. This condition, known among nomads as *Buba* disease, was consistently treated with *P. harmala*. Specifically, its fruits, leaves, and stems were crushed, finely chopped, and boiled in water. The affected horse's swollen legs were then exposed to the steam of this decoction for about half an hour, after which the legs were wrapped warmly (Tügelov, 1946: 17). This treatment was traditionally believed to alleviate the swelling effectively.

It is also possible that the swelling observed in horses' knees resulted not only from physical strain but also from exposure to cold following perspiration. In any case, it remains clear that in the traditional therapeutic practices of Kyrgyz nomads, *Peganum harmala* was consistently employed as the principal remedy for such inflammatory conditions (Miftakhov, 1946).

From a microbiological perspective, this traditional therapeutic practice can be reinterpreted by considering the bacterial isolates present in the rhizosphere of *Peganum harmala* and the secondary metabolites released by these microorganisms. It is highly plausible that the steam from decoctions prepared with the plant carried such bioactive compounds, thereby contributing to the observed therapeutic effects.

According to the same archival ethnographic records, livestock such as sheep, cattle, and horses occasionally developed ulcers located above the tail, characterized by a distinct pale appearance. The Kyrgyz referred to this condition as *ködööyara* ("pale sore") and traditionally treated it with *Peganum harmala*. In this practice, the fruits, leaves, and stems of the plant were crushed into a fine mass and directly applied to the affected area (Tügelov, 1946: 18). In reality, *Peganum harmala* is

neither consumed by humans nor by livestock; however, ethnographic narratives suggest that it is regarded as a non-poisonous plant. One oral account describes the following:

“At one time, a thousand soldiers fell ill and halted in one place. Among them, a sick soldier picked a single seed of *Peganum harmala* growing nearby, placed it in his mouth, and chewed it. Observing this, another soldier took the dried stem of the plant, rolled it, and smoked it like tobacco. A third soldier broke off a stalk and used it to clean his teeth. In this way, many of the soldiers used parts of the plant, and all eventually recovered from their illness. For this reason, a saying spread among the people that *adyrashman* is a cure for a thousand diseases” (Abdieva 2019).

While the aforementioned account may be categorized as a legend, it remains plausible that during periods of warfare—when access to formal medical treatment was non-existent—soldiers may have resorted to chewing *Peganum harmala* seeds or using its stalks for dental hygiene, despite the plant’s inherent bitterness and pungent odor. Such practices could indeed have contributed to recovery from infectious diseases, given the plant’s association with rhizosphere-derived compounds possessing both antibacterial and anti-fungal activity.

Further references within archival ethnographic materials shed light on additional medicinal applications. One such record notes: “During the nomadic era, Kyrgyz communities occasionally grazed their livestock along roadsides where *Peganum harmala* grew in abundance. Although the animals never consumed the plant, it was believed that exposure to its potent aroma could shield livestock from certain ailments” (Askarbek & Tulobayev, 2021). This observation can reasonably be linked to the properties of *Stenotrophomonas* bacteria identified in the *P. harmala* rhizosphere. *Stenotrophomonas* are Gram-negative, non-fermentative rods recognized for their broad-spectrum antagonistic effects against competing microorganisms. While they may not produce traditional antibiotics, they secrete enzymes such as proteases and chitinases, alongside Volatile Organic Compounds (VOCs), which collectively inhibit the growth of pathogenic microbes.

The ethnographic observation regarding the “aroma of *adyrashman*” appears to align closely with the biological activity of these VOCs. It is highly probable that the volatile compounds emitted through the plant’s metabolism functioned within the immediate environment to reduce the microbial load, thereby exerting a natural sanitizing effect on the animals as they graze.

Furthermore, the volatile organic compounds (VOCs) associated with *Stenotrophomonas* bacteria may be linked not only to Kyrgyz ethnographic accounts but also to broader cultural traditions across Central Asia. In particular, this connection resonates with the deeply rooted ritual of burning *Peganum harmala* during the celebration of Nowruz. Historically and into the present day, Central Asian communities mark Nowruz in late March, symbolizing the end of winter and the rebirth of nature. During this festival, it is customary for households to place dried juniper and *Peganum harmala* in a vessel to smoulder, creating dense smoke. As the ritual is performed, the smoke is carried throughout the home and courtyard with the belief that it cleanses the space of "bad odors, the evil eye, and ill wishes.

While the element of spiritual belief is central to this ritual, when viewed from a microbiological perspective, the practice of burning *Peganum harmala* may have fulfilled a genuine biological function. The VOCs released during the smouldering of the dried plant act to inhibit the growth of harmful airborne microorganisms, effectively sanitizing the domestic environment. This practice of "purification" through fumigation was not exclusive to Nowruz; it remains a common feature of everyday life in rural Kyrgyzstan today, bridging the gap between ancient tradition and microbial defense.

The present study offers a comprehensive, interdisciplinary exploration of *Peganum harmala* and its associated rhizosphere bacterial communities by merging rigorous microbiological analysis with historical and ethnographic data.

Limitations

First, this study was restricted to isolates from a single geographic site in the Chui region using 16S rRNA gene sequencing, which may not fully capture the complete functional diversity or spatial variability of the *Peganum harmala* rhizosphere microbiome. Second, while our interdisciplinary approach links microbial profiles to ethnographic practices, the direct pharmacological mechanisms and specific bioactive compounds responsible for these traditional therapeutic effects remain to be empirically validated.

Future Scope

Future research should expand sampling across broader ecological zones and utilize multi-omics platforms—such as metagenomics and metabolomics - to fully elucidate the functional traits of these rhizobacteria. Additionally, targeted bioassays are needed to isolate active secondary metabolites, thereby rigorously validating traditional fumigation and therapeutic applications for potential agricultural and pharmaceutical use.

Conclusion

In conclusion, the present study demonstrates the phylogenetic diversity of potential bacteria from medicinal plants. It appears that medicinal plants can be an important source of antagonistic bacteria that confers many advantages to the host plant. Further investigations are required to study the associated rhizospheric bacteria population of the host plant.

Conflict of Interest

The authors declare no conflicts of interest in this work.

Acknowledgement

The authors independently conducted the study without external assistance.

References

Abdieva, A. M. (2019). XIX kylymdynayagındajana - XX kylymdynbashynda Fergana öröönününtüshtük-batyshchölkömündöjashagankyrgyzdardynfloranydarykatarypaydalanyshy [Flora used by Kyrgyz people living in the southwest desert region of Fergana valley in the late 19th and early 20th centuries]. *Izvestia Universities of Kyrgyzstan, (11)*, 92–96.

Alghoul, N., Sasi, S., Awayn, N., Prastiya, R., & Salem, K. (2025). Assessment of Sperm Function and Embryo Morphology Following Exposure to Peganum Harmala Seeds Decoction in Mice. *AlQalam Journal of Medical and Applied Sciences*, *8*(2), 1000-1004. https://doi.org/10.54361/ajmas.258271

Aravind, R., Eapen, S. J., Kumar, A., Dinu, A., & Ramana, K. V. (2010). Screening of endophytic bacteria and evaluation of selected isolates for suppression of burrowing nematode (Radopholus similis Thorne) using three varieties of black pepper (Piper nigrum L.). *Crop Protection*, *29*(4), 318-324. https://doi.org/10.1016/j.cropro.2009.12.005

Asgarpanah, J., &Ramezanloo, F. (2012). Chemistry, pharmacology and medicinal properties of Peganum harmala L. *African Journal of Pharmacy and Pharmacology*, *6*(22), 1573-1580. https://doi.org/10.5897/AJPP11.876

Askarbek, G., & Tulobayev, A. (2021). Mal ylaŋdarynaldynaluudagy Kyrgyz elininsalttuuykmalary [Traditional Kyrgyz remedies for snakebites]. *Manas Journal of Agriculture Veterinary and Life Sciences, 11*(2), 138–144. https://doi.org/10.53518/mjavl.884684

Babar, Z. Z., Younis, T., & Shinwari, Z K. (2025). Isolation and Characterization of Plant Growth Promoting Endophytic Bacteria FromPeganum Harmala L. *Biological and Clinical Sciences Research Journal, 6*(8), 75–81. https://doi.org/10.54112/bcsrj.v6i8.1975

Borde, V., Hivrale, V., &Kachole, M. (2013). Identification and purification and antimicrobial activity of alkaloid from Peganumharmale (L), a medicinal Plant. *Journal of Biology, Agriculture and Healthcare*, *3*(19), 105–111. https://www.iiste.org/Journals/index.php/JBAH/article/view/9798

Bossis, E., Lemanceau, P. P., Latour, X., & Gardan, L. (2000). The taxonomy of Pseudomonas fluorescens and Pseudomonas putida: current status and need for revision. *Agronomie*, *20*(1), 51-63. https://doi.org/10.1051/agro:2000112

Brooke, J. S. (2012). Stenotrophomonas maltophilia: an emerging global opportunistic pathogen. *Clinical Microbiology Reviews*, *25*(1), https://doi.org/10.1128/cmr.00019-11

Bulgarelli, D., Schlaeppi, K., Spaepen, S., Van Themaat, E. V. L., & Schulze-Lefert, P. (2013). Structure and functions of the bacterial microbiota of plants. *Annual Review of Plant Biology*, *64*, 807-838. https://doi.org/10.1146/annurev-arplant-050312-120106

Cappuccino, J. G., & Sherman, N. (2014). *Microbiology: A Laboratory Manual*. Pearson Higher. 10th Ed. https://www.scribd.com/doc/272782862/Proced-DilSeriada-1

Clarridge J. E., 3rd (2004). Impact of 16S rRNA gene sequence analysis for identification of bacteria on clinical microbiology and infectious diseases. *Clinical Microbiology Reviews*, *17*(4), 840-862. https://doi.org/10.1128/cmr.17.4.840-862.2004

Compant, S., Clément, C., &Sessitsch, A. (2010). Plant growth-promoting bacteria in the rhizo-and endosphere of plants: their role, colonization, mechanisms involved and prospects for utilization. *Soil Biology and Biochemistry*, *42*(5), 669-678. https://doi.org/10.1016/j.soilbio.2009.11.024

Dida, G., Beshir, H. M., & Muleta, D. (2018). Isolation and characterization of starch degrading rhizobacteria. *Open AccessJournal of Microbiology & Biotechnology, 3*, 2576-7771. https://doi.org/10.23880/oajmb-16000129

Dobbelaere, S., Vanderleyden, J., & Okon, Y. (2003). Plant growth-promoting effects of diazotrophs in the rhizosphere. *Critical Reviews in Plant Sciences*, *22*(2), 107-149. https://doi.org/10.1080/713610853

Doolotkeldieva, T., & Bobusheva, S. (2016). Fire blight disease caused by Erwinia amylovora on Rosaceae plants in Kyrgyzstan and biological agents to control this disease. *Advances in Microbiology*, *6*(11), 831-851. https://doi.org/10.4236/aim.2016.611080

Doskaliyev, A., Seidakhmetova, R., Tutai, D. S., Goldaeva, K., Surov, V. K., & Adekenov, S. M. (2021). Alkaloids of Peganum harmala L. and their Pharmacological Activity. *Open Access Macedonian Journal of Medical Sciences,9(A)*, 766-775. https://doi.org/10.3889/oamjms.2021.6654

El Fissi, H., Ameziane, H., Bouzid, F., Achqra, I., Msanda, F., & Alif, N. (2025). Isolation, digital analysis, and semi-quantification of harmaline and harmine from Peganum harmala L. *Euro-Mediterranean Journal for Environmental Integration*, *10*(3), 1805-1810. https://doi.org/10.1007/s41207-024-00624-x

Gökkaya, İ., Renda, G., Subaş, T., & Özgen, U. (2023). Phytochemical, pharmacological, and toxicological studies on Peganum harmala L.: An overview of the last decade. *Clinical and Experimental Health Sciences*, *13*(3), 664-678. https://doi.org/10.33808/clinexphealthsci.1125345

Halebian, S., Harris, B., Finegold, S. M., & Rolfe, R. D. (1981). Rapid method that aids in distinguishing Gram-positive from Gram-negative anaerobic bacteria. *Journal of Clinical Microbiology*, *13*(3), 444-448. https://doi.org/10.1128/jcm.13.3.444-448.1981

Khudoynazarova, G., Kodirova, Z., Ganiev, B., Jalilov, S., & Mavluda, R. (2025). Chemical Composition and In Silico Study of the Biological Activities of Alkaloids from Peganum harmala. In *E3S Web of Conferences* (Vol. 661, p. 05003). EDP Sciences. https://doi.org/10.1051/e3sconf/202566105003

Koçak, Y., & Şahin, A. (2009). *Peganum harmala* L. (üzerlik) tohumekstresininanaljezikaktivitesiveakuttoksisitesininfarelerüzerindebelirlenmesi [Analgesic activity and acute toxicity determination of *Peganum harmala* L. (üzerlik) seed extract in mice]. *YüzüncüYılÜniversitesiVeterinerFakültesi Dergisi, 20*(1), 27–30. https://dergipark.org.tr/tr/pub/yyuvfd/article/166269

Kotoučková, L., Schumann, P., Durnova, E., Spröer, C., Sedláček, I., Neča, J., ... & Němec, M. (2004). Arthrobacter nitroguajacolicus sp. nov., a novel 4-nitroguaiacol-degrading actinobacterium. *International journal of Systematic and Evolutionary Microbiology*, *54*(3), 773-777. https://doi.org/10.1099/ijs.0.02923-0

Krieg, N. R., Ludwig, W., Whitman, W. B., Hedlund, B. P., Paster, B. J., Staley, J. T., Brown, D. R., Parte, A., & Ward, A. C. (Eds.). (2010). *Bergey's manual of systematic bacteriology*: *The Bacteroidetes, Spirochaetes, Tenericutes (Mollicutes), Acidobacteria, Fibrobacteres, Fusobacteria, Dictyoglomi, Gemmatimonadetes, Lentisphaerae, Verrucomicrobia, Chlamydiae, and Planctomycetes*. Springer. (2nd ed., Vol. 4). Springer.

Kumar, V., Raghuvanshi, N., Pandey, A. K., Kumar, A., Thoday-Kennedy, E., & Kant, S. (2023). Role of halotolerant plant growth-promoting rhizobacteria in mitigating salinity stress: Recent advances and possibilities. *Agriculture*, *13*(1), 168. https://doi.org/10.3390/agriculture13010168

Li, L. N. (2024). Peganum harmala L.: A review of botany, traditional use, phytochemistry, pharmacology, quality marker, and toxicity. *Combinatorial Chemistry & High Throughput Screening*, *27*(6), 797-822. https://doi.org/10.2174/1386207326666230622164243

Lundberg, D. S., Lebeis, S. L., Paredes, S. H., Yourstone, S., Gehring, J., Malfatti, S., ... & Dangl, J. L. (2012). Defining the core Arabidopsis thaliana root microbiome. *Nature*, *488*(7409), 86-90. https://doi.org/10.1038/nature11237

Mamedov, N. A., Pasdaran, A., &Mamadalieva, N. Z. M. (2018). Pharmacological studies of Syrian rue (*Peganum harmala L., Zygophyllaceae). International Journal of Secondary Metabolite*, *5*(1), 1-6. https://doi.org/10.21448/ijsm.335539

Miftakhov, K. (1946). *Kyrgyz el meditsinasy* [Kyrgyz folk medicine]. National Academy of Sciences of the Kyrgyz Republic, Manuscripts Fund, 305/526, Folder 1, pp. 1–2.

Mukhtorova, S., Alikulov, B., Yuldosheva, M., Maxammadieva, D., Khidirova, U., Kabulova, F., & Ismailov, Z. (2024). Diversity of endophytic bacteria isolated from Peganum harmala distributed in arid re-gions in Uzbekistan. *Regulatory Mechanisms in Biosystems*, *15*(2), 286-291. https://doi.org/10.15421/022441

O'sullivan, D. J., & O'Gara, F. (1992). Traits of fluorescent Pseudomonas spp. involved in suppression of plant root pathogens. *Microbiological Reviews*, *56*(4), 662-676. https://doi.org/10.1128/mr.56.4.662-676.1992

Penkala-Gawęcka, D. (2023). Poganekrutowaty (Peganum harmala L.) w tradycjachludówAzjiŚrodkowejijegopopularność w dobie pandemii COVID-19 – perspektywaantropologiimedycznej *MedycynaNowożytna,* [Peganum routed (Peganum harmala L.) in the traditions of the peoples of Central Asia and its popularity in the era of the COVID-19 pandemic – a perspective of medical anthropology, *Modern Medicine Studies on Medical Culture*]. *29*(1), 135-164. https://doi.org/10.4467/12311960MN.23.007.18448

Qi, Q., Chu, A. Y., Kang, J. H., Jensen, M. K., Curhan, G. C., Pasquale, L. R., ... & Qi, L. (2012). Sugar-sweetened beverages and genetic risk of obesity. *New England Journal of Medicine*, *367*(15), 1387-1396. https://doi.org/10.1056/NEJMoa1203039

Ryan, R. P., Germaine, K., Franks, A., Ryan, D. J., & Dowling, D. N. (2008). Bacterial endophytes: recent developments and applications. *FEMS Microbiology Letters*, *278*(1), 1-9.https://doi.org/10.1111/j.1574-6968.2007.00918.x

Santoyo, G., Orozco-Mosqueda, M. D. C., &Govindappa, M. (2012). Mechanisms of biocontrol and plant growth-promoting activity in soil bacterial species of Bacillus and Pseudomonas: A review. *Biocontrol Science and Technology*, *22*(8), 855-872.https://doi.org/10.1080/09583157.2012.694413

Tamura, K., Stecher, G., Peterson, D., Filipski, A., & Kumar, S. (2013). MEGA6: molecular evolutionary genetics analysis version 6.0. *Molecular Biology and Evolution*, *30*(12), 2725-2729. https://doi.org/10.1093/molbev/mst197

Tügelov, N. (1946, September 16). [Interview: Age 72, Choro lineage, resident of ZhanïTalap kolkhoz, Ak-Talaa District, Tien Shan Region] (S. Tabyshaliev, A. Chorobaev, & Umurzakov, Interviewers). Notebook 3, Materials of the Tien Shan Expedition (p. 17). (archival interview.)

Weisburg, W. G., Barns, S. M., Pelletier, D. A., & Lane, D. J. (1991). 16S ribosomal DNA amplification for phylogenetic study. *Journal of Bacteriology*, *173*(2), 697-703. https://doi.org/10.1128/jb.173.2.697-703.1991

Weller, D. M. (2007). Pseudomonas biocontrol agents of soilborne pathogens: Looking back over 30 years. *Phytopathology*, *97*(2), 250-256. https://doi.org/10.1094/PHYTO-97-2-0250