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Microbial diversity in cutaneous leishmaniasis lesions and potential implications for disease progression and treatment outcomes

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Abstract

Objective Beyond the parasitic infection in Cutaneous leishmaniasis (CL), secondary bacterial colonization can influence disease chronicity, delay healing, and reduce treatment efficacy. This study investigated the bacterial diversity in CL lesions, its association with lesion duration, and its potential impact on treatment outcomes among Sri Lankan patients.

Results Fifteen bacterial species were identified, including both Gram-positive and Gram-negative organisms. *Staphylococcus aureus* was associated with the longest lesion duration (up to 12 months) and extended treatment (15 cycles of intralesional sodium stibogluconate and cryotherapy). In contrast, species such as *Kocuria palustris* and *Acinetobacter baylyi* were linked to shorter treatment durations. Multivariate analysis revealed that lesion type significantly influenced treatment duration ($P < 0.05$), while larger lesion size and diabetes showed marginal associations with prolonged therapy. The presence of opportunistic and antibiotic-resistant species, particularly *S. aureus*, suggests a potential contributory role of bacterial co-infections in CL progression and highlights the need to consider their presence in treatment planning. Integrating microbial profiling into clinical protocols may enhance treatment efficacy and inform personalized care strategies. However, the limited sample size and convenience-based recruitment may affect the generalizability of these findings, and the potential influence of bacterial colonization on treatment response warrants further investigation in larger cohorts.

Keywords Cutaneous leishmaniasis, Secondary infection, Bacterial colonization, Treatment duration

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Background

Leishmaniasis is a vector-borne protozoan disease caused by *Leishmania* spp. and transmitted through the bite of infected phlebotomine sand flies (Diptera; Psychodidae). In Sri Lanka, the disease is primarily attributed to *Leishmania donovani* zymodeme MON-37, predominantly manifesting as cutaneous leishmaniasis (CL) [1]. The CL exhibits diverse clinical presentations, including papules, nodules, plaques, and ulcers, with variations in lesion morphology influenced by host immune response and parasite virulence [2]. Although localized cutaneous leishmaniasis (LCL) is the most common form, chronic ulcerative skin lesions, often persisting for months or even years without treatment, contribute to significant morbidity and psychosocial impact [3].

In Sri Lanka, CL is primarily treated using local infiltration of sodium stibogluconate (SSG) or cryotherapy with liquid nitrogen. For over two decades, intra-lesional SSG (IL-SSG) has been the first-line treatment for CL, with an estimated average of 10 injections administered at weekly intervals to achieve complete lesion healing [4]. However, an increasing number of CL cases exhibit treatment failure to IL-SSG, raising concerns about emerging drug resistance and the need for alternative therapeutic strategies [5].

Chronicity of ulcerative lesions, continuous environmental exposure, and inadequate hygiene, particularly in lesions occurring on the lower limbs, contribute to secondary bacterial colonization, increasing disease complexity [6]. Polymicrobial colonization within CL lesions has been associated with delayed wound healing, lesion chronicity, and reduced therapeutic efficacy [7, 8]. Secondary bacterial infections involving *Streptococcus pyogenes*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Morganella morganii*, and *Enterococcus durans* have been linked to prolonged lesion chronicity and lower cure rates following systemic SSG treatment [9].

Despite the growing recognition of the role of bacterial colonization in disease progression and therapeutic outcomes, limited research has been conducted on the microbiome associated with CL lesions in Sri Lanka. Understanding the microbial diversity and distribution within CL lesions and their influence on treatment response is critical for developing improved therapeutic strategies. Integrating microbiome profiling techniques, such as metagenomics and 16 S rRNA sequencing, could provide novel insights into host-microbe interactions, facilitating the identification of microbial biomarkers predictive of treatment outcomes. This knowledge may inform personalized therapeutic interventions, ultimately enhancing treatment efficacy and reducing the disease burden associated with CL.

This study aims to investigate the diversity of bacteria isolated from cutaneous leishmaniasis lesions in patients in Sri Lanka. It seeks to analyze the variation in bacterial diversity with disease progression and clinical presentation of CL lesions. Additionally, the study will assess the relationship between bacterial colonization and treatment outcomes following intra-lesional sodium stibogluconate (IL-SSG) therapy. Furthermore, it will explore the potential role of bacterial colonization in influencing lesion chronicity and delayed wound healing in CL patients Therefore, this study is expected to provide an understanding of the microbiome associated with CL lesions and its potential impact on disease progression and therapeutic outcomes.

Method

Study design and setting

A convenient sample comprising individuals who came to the dermatology clinic at the Kurunegala Teaching Hospital during October to November 2024 were considered form this study. The patients clinically diagnosed with ulcerative CL lesions and confirmed through either by slit skin smear or histopathological methods were invited for the study. Exclusion criteria included individuals with systemic infections, immunocompromised conditions, or recent antibiotic therapy.

Patient data collection

Demographic data, including age, gender, occupation, and environmental exposure, were recorded using a structured questionnaire (Supplementary material 1). Clinical data on lesion characteristics (size, location, type), duration, and associated symptoms were documented by trained medical personnel. Treatment history, including the use of IL-SSG, cryotherapy and others were collected from the patient records.

Sample collection and bacterial culturing

Samples of cutaneous lesions were collected under sterile condition using cotton swabs. The lesion swabs were steaked on Blood Agar and MacConkey Agar media to screen fastidious and Gram-negative bacteria, respectively. These plates were incubated aerobically at 37 °C for 24 to 48 h. Distinct bacterial colonies were observed, recorded, and subcultured to obtain pure isolates. Preliminary bacterial identification was carried out using several techniques: Gram staining was employed to classify bacteria as Gram-positive or Gram-negative, the catalase test was performed to differentiate catalase-positive *Staphylococcus* species from catalase-negative *Streptococcus* species, and the oxidase test was conducted to identify oxidase-positive bacteria, such as *P. aeruginosa*. Additionally, biochemical tests, including the coagulase test, were used for further differentiation of *S. aureus*.

To minimize the risk of contamination, all procedures were conducted under aseptic conditions using sterile

equipment and materials. Negative controls (blank swabs and uninoculated media) were included during culturing to monitor for contamination during sample processing.

Molecular identification of bacteria

Genomic DNA was extracted from pure bacterial isolates using the QIAamp DNA Mini Kit (Qiagen GmbH, Hilden, Germany). The 16 S rRNA gene was amplified using universal primers 27 F (5′-AGAGTTTGATCCT GGCTCAG-3′) and 1492R (5′-TACGGCTACCTTGTT ACGACTT-3′). The PCR reaction mixture consisted of PCR buffer (1×), $MgCl_2$ (2.5 mM), dNTPs (0.2 mM), Taq polymerase (0.3 U), 200 ng of DNA template, and sterile water as a negative control. Thermal cycling conditions included an initial denaturation at 94 °C for 10 min, followed by 35 cycles of denaturation at 95 °C for 30 s, annealing at 57.5 °C for 40 s, and extension at 72 °C for 30 s. A final extension was carried out at 72 °C for 8 min. The PCR products were analyzed by agarose gel electrophoresis (1% w/v), stained with ethidium bromide, and visualized under a UV transilluminator.

Table 1 Socio-demographic characteristics of patients recruited for the study

Factor	Response Category	No. of respondents ($n=25$)	Percentage (%)
Gender	Male	12	48.0
	Female	13	52.0
Age (years)	< 15	1	4.0
	15–30	5	20.0
	31–45	6	24.0
	46–60	9	36.0
	> 60	4	16.0
Duration at residence (years)	< 1	3	12.0
	> 1	22	88.0
Occupation	Nurse	1	4.0
	Teacher	1	4.0
	Farmer	4	16.0
	Driver	1	4.0
	Laborer	3	12.0
	Student	2	8.0
	Unemployed	9	36.0
	Service worker	4	16.0
Travel history to other districts	Yes	7	28.0
	No	18	72.0
Travel history to overseas	Yes	1	4.0
	No	24	96.0
Pets or farm within premises	Yes	17	68.0
	No	8	32.0
Scrub jungle in visible distance	Yes	15	60.0
	No	10	40.0
Engage in outdoor activities	Yes	7	28.0
	No	18	72.0

Phylogenetic analysis of bacteria isolates

Sanger sequencing of the purified 16 S rRNA amplicons was performed. The obtained sequences were analyzed using the Basic Local Alignment Search Tool (BLAST) algorithm against the NCBI GenBank database to determine bacterial species. The bacterial 16 S rRNA gene sequences were aligned using ClustalW in MEGA (Molecular Evolutionary Genetics Analysis) software version 11. Phylogenetic relationships were inferred using the Neighbor-Joining (NJ) method with the Kimura 2-parameter model. The robustness of the tree topology was assessed using 1,000 bootstrap replicates to determine the reliability of the branching patterns.

Statistical analysis

Statistical analyses were conducted using SPSS version 26.0. Descriptive statistics, including frequencies and percentages, were used to summarize demographic and clinical characteristics. A multivariate linear regression model was developed to explore factors influencing treatment duration. Treatment duration (in months) was set as the dependent variable. Independent variables were selected based on clinical relevance and findings from prior literature, including lesion size, lesion site, lesion type, and diabetic status. Prior to model construction, bivariate associations were examined to assess variable inclusion feasibility. Continuous variables were tested for normality and linearity. Categorical variables were dummy-coded. A stepwise forward selection method was applied to identify significant predictors, while checking for multicollinearity using variance inflation factors (VIF < 5). Model assumptions (linearity, homoscedasticity, normality of residuals) were verified through residual plots. Statistical significance was determined at a threshold of $P < 0.05$.

Results

Socio- economic information of the CL patients

The study included a total of 25 respondents, with a nearly equal gender distribution (48% male, 52% female). Most respondents (88%) had resided in their current location for more than one year (Table 1). Among occupational categories, unemployed individuals constituted the largest group (36%), followed by farmers and service workers (16% each). Travel history to other districts was reported by 28% of respondents. With regard to environmental exposure, 68% of respondents had pets or farm animals within visible distance, while 60% had scrub jungles in proximity to their residences. The presence of scrub jungles did not significantly deviate from an expected equal distribution ($\chi^2 = 1.00$; $P = 0.317$). Furthermore, only 28% of participants engaged in outdoor activities.

Clinico-epidemiological characteristics of the CL patients

The analysis of lesion characteristics among respondents revealed that the majority (84%) had a single lesion, while 16% had two. The lower limb was the most commonly affected site (48%), followed by the upper limb (36%), face (24%), and back (8%). Regarding lesion type, ulcerating nodules were the most prevalent (44%), followed by nodules (36%), papules (20%), and complete ulcers (16%). More than half of the lesions (56%) measured between 2 and 3 cm, in size. The majority of lesions (96%) had persisted for 2–7 months (Table 2).

Table 2 Clinico- epidemiological characterizes of CL patients recruited for the study

Feature	Category	No. of respondents	Percentage (%)
Number of lesions	1	21	84.0
	2	4	16.0
Site of the lesion	Upper limb	9	36.0
	Lower limb	12	48.0
	Face	6	24.0
	Back	2	8.0
Lesion type	Papule	5	20.0
	Nodule	9	36.0
	Ulcerating Nodule	11	44.0
	Complete Ulcer	4	16.0
Lesion size (cm)	< 1	9	36.0
	2–3	14	56.0
	> 3	9	36.0
Duration (months)	< 2	2	8.0
	2–7	24	96.0
	7–12	3	12.0
Pain in the lesion	Yes	3	10.7
	No	26	89.3
Itching in the lesion	Yes	5	17.9
	No	82.1	
Regional lymph node enlargement	Yes	4	16.0
	No	21	84.0
History of similar skin lesion	Yes	5	20.0
	No	20	80.0
History of similar skin lesion in others	None	17	68.0
	Family	1	4.0
	Neighbour	3	12.0
	Co-workers	4	16.0
Previous treatment	None	11	44.0
	SSG	1	4.0
	Cryotherapy and SSG	10	40.0
	Metronidazole	3	12.0
Treatment duration (months)	< 1	1	4.0
	1–5	10	40.0
	> 5	3	12.0
Previously diagnosed with Diabetes	Yes	10	40.0
	No	15	60.0
Previously undergone any surgeries	Yes	8	32.0
	No	17	68.0
Use of any immunosuppressive drugs	Yes	11	44.0
	No	14	56.0

Symptoms associated with the lesions varied, with only 10.7% reporting pain and 17.9% experiencing itching. Regional lymph node enlargement was noted in 16% of cases. Regarding treatment, 44% of respondents had received no prior treatment, while 40% had undergone cryotherapy combined with SSG. The treatment duration varied, with most patients (40%) undergoing treatment for 1–5 months, while 12% required more than 5 months. Among the respondents, 40% had been diagnosed with diabetes, and 32% had previously undergone surgeries. The use of immunosuppressive drugs was reported in 44% of cases, indicating a possible contributing factor to lesion development.

Factors affecting treatment duration in cutaneous leishmaniasis

Multivariate analysis demonstrated that larger lesion size and diabetic status were associated with prolonged treatment durations, although these associations were marginally non-significant (Table 3). Lesion type significantly influenced treatment outcomes, with ulcerating nodules and complete ulcers requiring longer durations compared to papular lesions ($P < 0.05$). Lesion site, however, did not significantly affect treatment time. These findings suggest that lesion severity and host factors such as diabetes are important predictors of therapeutic response in cutaneous leishmaniasis.

Lesion bacteria with disease duration and treatment

The duration of lesions varied among bacterial species isolated from cutaneous leishmaniasis patients. *Staphylococcus warneri* and *M. osloensis* were associated with prolonged lesion duration (8 months), whereas *S. haemolyticus* was found in a patient with the shortest lesion duration (1 month). *Staphylococcus aureus*, a known pathogenic species, was detected in patients having lesion duration of 12 months and 3 months (Fig. 1).

Among the bacterial isolates, only *S aureus*, *B. thuringiensis*, *K. palustris*, *Pseudomonas* sp., *(A) baylyi*, *M. lylae*, and *(B) toyonensis* were found in patients who had received treatment. *Staphylococcus aureus* was detected in a patient who underwent the longest treatment duration (12 months with 15 cycles of SSG and cryotherapy). Other bacterial species, such as *K. palustris* and *(A) baylyi*, were found in patients who had undergone shorter treatment durations (2–3 months), indicating a possible variation in bacterial persistence with treatment. Patients

Table 3 Multivariate linear regression analysis of predictors of treatment duration in CL patients (β coefficients indicate the estimated change in treatment duration (in months) for each predictor variable relative to the reference group)

Predictor	Coefficient (β)	*P*-value
Intercept	1.8	0.02
Lesion Size (cm)	0.45	0.06
Diabetic Status (Yes vs. No)	+ 1.1	0.07
Lesion Site (Ref: Face)		
• Lower limb	+ 0.8	0.12
• Upper limb	+ 0.5	0.19
• Back	+ 0.2	0.35
Lesion Type (Ref: Papule)		
• Nodule	+ 0.6	0.22
• Ulcerating Nodule	+ 1.5	0.04*
• Complete Ulcer	+ 1.7	0.03*

*$P < 0.05$ considered statistically significant

who received combination therapy (SSG and cryotherapy) had bacterial isolates belonging to both Gram-positive and Gram-negative groups. Gram-positive bacilli such as *(B) thuringiensis* and *B. toyonensis* were found in treated individuals, while Gram-negative bacilli, including *Pseudomonas* sp. and *A. baylyi*, were also present in individuals undergoing treatment.

Phylogenetic analysis of bacteria isolated from the lesions

The phylogenetic tree illustrates the evolutionary relationships among bacterial species isolated in the study based on 16 S rRNA gene sequences. The tree shows distinct clustering of bacterial isolates, reflecting their genetic similarity (Fig. 2). Notably, *B. cereus*, *Bacillus sp.*, and *B. paranthracis* form a well-supported clade, indicating a close evolutionary relationship within the *Bacillus* genus. Similarly, *S. aureus*, *S. arlettae*, and *S. haemolyticus* cluster together, suggesting their genetic relatedness within the *Staphylococcus* genus.

The presence of high bootstrap values, such as 99, indicates strong support for certain branches, while lower values suggest weaker statistical confidence in specific separations. The tree also highlights the diversity of bacterial genera, including *Staphylococcus*, *Bacillus*, *Micrococcus*, *Acinetobacter*, *Kocuria*, and *Moraxella*, some of which are known to be associated with wound infections and lesion persistence. The close grouping of *A. pittii* isolates suggests a common evolutionary origin, while the separation of *M. osloensis* into different clusters may indicate genetic variability within the species.

Discussion

In recent years, research has increasingly highlighted that the microbiome of cutaneous leishmaniasis (CL) lesions plays a much more active role in disease progression and treatment response than previously understood [10]. Rather than being passive cohabitants, bacterial communities within these lesions may contribute to chronicity, inflammation, immune modulation, and even treatment failure.

This study indicated that the bacterial communities associated with lesions were far from uniform. Instead, they consisted of a complex mixture of pathogenic, opportunistic, and commensal species, including *Staphylococcus*, *Bacillus*, *Acinetobacter*, *Moraxella*, *Micrococcus*,

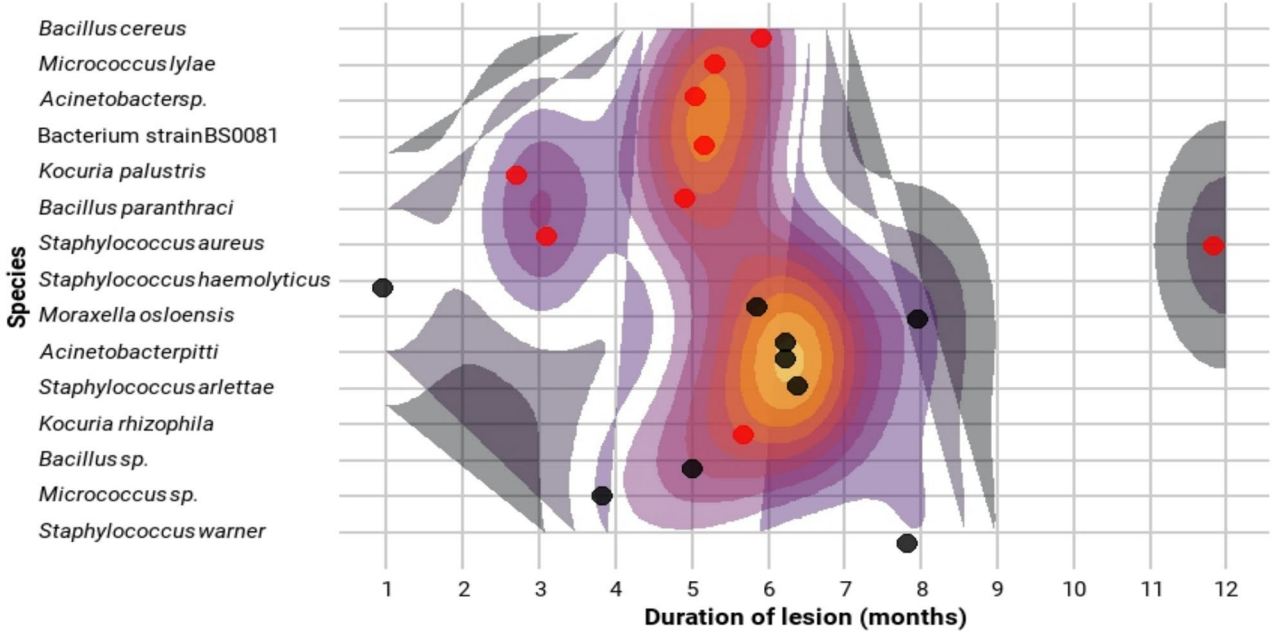

Fig. 1 Association of bacterial species with lesion duration and treatment received (Bubble size corresponds to treatment duration)

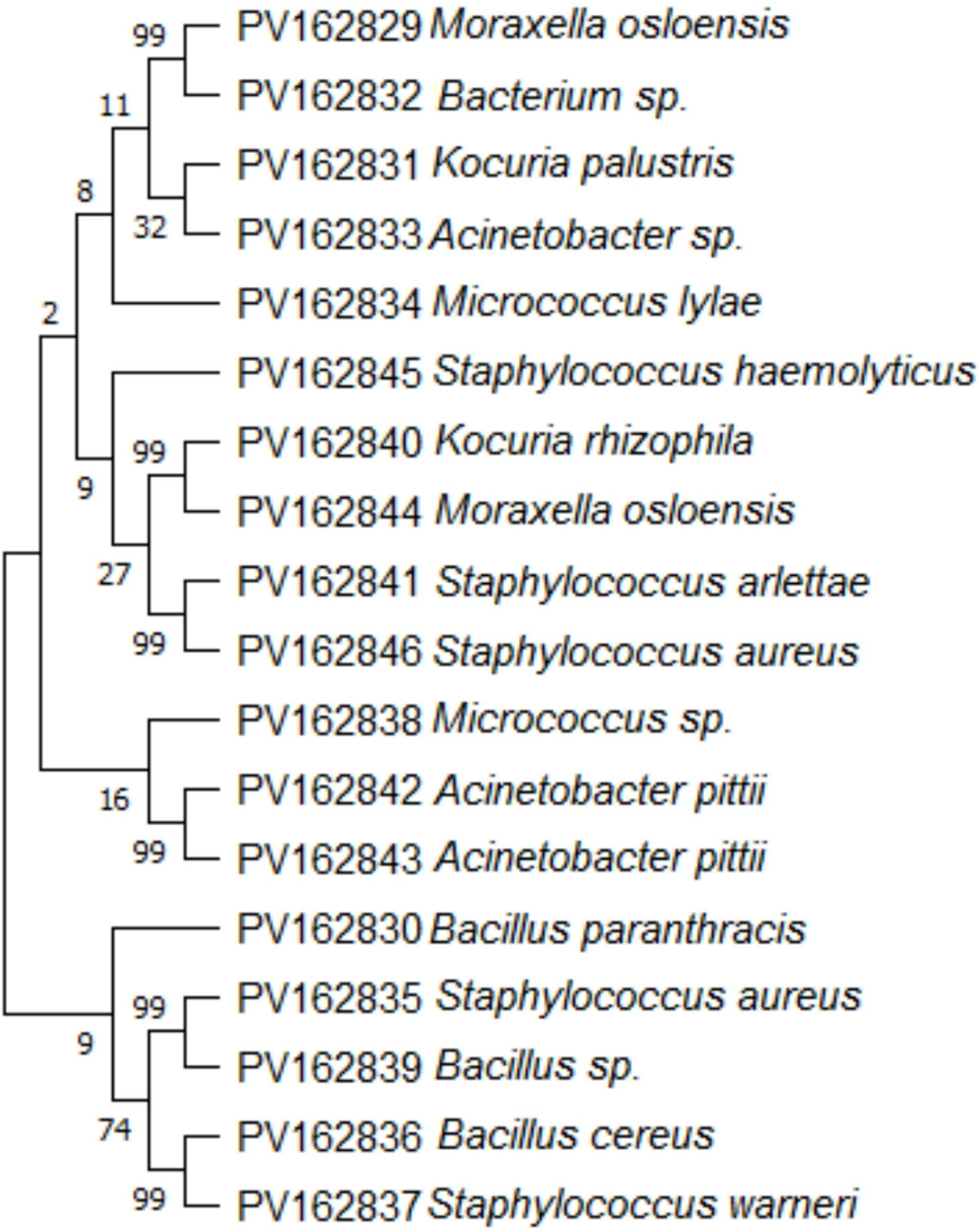

Fig. 2 Phylogenetic relationships among bacterial species isolated from CL lesions based on 16 S rRNA gene sequences. The tree was constructed using the Neighbor-Joining method with the Kimura 2-parameter model in MEGA 11. Bootstrap values (based on 1,000 replicates) are shown at branch points. GenBank accession numbers are indicated for each isolate. The scale bar represents the number of nucleotide substitutions per site

and *Kocuria*. This diversity mirrors what has been seen in other parts of the world and reinforces the idea that secondary bacterial colonization is a common and potentially impactful feature of CL [11].

Phylogenetic analyses, represented in linear tree, identified the presence of pathogenic, opportunistic, and commensal bacteria within CL lesions. Among these, *S. aureus* was the most frequently isolated species, consistent with its well-documented role in secondary bacterial infections and wound complications in CL patients [12, 13]. Click or tap here to enter text. Additionally, *S. arlettae* and *S. warneri*, known for their opportunistic pathogenicity and antibiotic resistance, were detected [14, 15]. The identification of *B. cereus* and *B. paranthracis* suggests a role in wound colonization, inflammation, and prolonged healing [16].

Certain environmental and opportunistic species, including *A. pitti* and *M. osloensis*, were also prevalent. These bacteria, known for their resilience in harsh conditions, may contribute to persistent infections and delayed healing [17]. Moreover, the presence of *M. lylae* and *Kocuria* spp., typically part of the normal skin flora, suggests that under immunocompromised conditions, these bacteria may act as opportunistic pathogens [18, 19].

The composition of the bacterial community appears to be influenced by the stage of CL lesions. Early-stage lesions predominantly harbored commensal and environmental bacteria, whereas chronic or ulcerative lesions exhibited an increased abundance of opportunistic and pathogenic species [20]. The clustering of *S. aureus* and *Bacillus* spp. in the phylogenetic analysis suggests a possible role in prolonged infections and delayed healing [21]. Additionally, the persistence of *Acinetobacter* spp. across multiple branches indicates that this bacterium may be involved throughout different disease stages [22]. Nevertheless, due to the limited sample size per bacterial species, especially for less frequently detected organisms, these associations with treatment duration and lesion chronicity should be viewed as exploratory and not definitive.

A critical factor contributing to therapeutic failure is bacterial biofilm formation within lesions, which can impede drug penetration and delay healing [20]. The identification of biofilm-forming bacteria such as *Acinetobacter* sp. and *M. osloensis* highlights the need for further studies to evaluate their impact on CL treatment outcomes. Understanding bacterial contributions to lesion chronicity and inflammation may help refine treatment strategies, potentially incorporating adjunct therapies targeting bacterial co-infections.

This study has several limitations. The use of convenience sampling from a single hospital may not fully represent the broader CL-affected population. The relatively small sample size limits the comprehensive assessment of bacterial diversity. Additionally, the cross-sectional design provides only a single time-point analysis, restricting insights into bacterial dynamics over time. Functional analyses, such as metagenomics or transcriptomics, were not conducted, limiting understanding of bacterial interactions and their impact on lesion progression and treatment response.

While this study identified 15 distinct bacterial species across patient lesions, it is important to interpret these findings with caution. Given the open nature of cutaneous lesions and the potential for skin surface flora to be captured during swabbing, contamination cannot be entirely ruled out. Although strict aseptic techniques were followed and controls were used during culturing, the possibility of environmental or skin microbiota being introduced during sample collection or processing remains a limitation. Future studies should consider incorporating metagenomic profiling with proper decontamination pipelines to more confidently differentiate true colonizers from contaminants.

Despite limitations, this study emphasizes the need to integrate bacterial profiling into CL management strategies. By combining clinical data with molecular identification and phylogenetic analysis, this study offers a foundation for understanding how specific bacterial species may influence treatment outcomes. Patients with poor responses to standard therapies exhibited a higher prevalence of antibiotic-resistant bacteria, suggesting a contributory role in delayed healing. While earlier studies have noted bacterial colonization, this study uniquely integrates lesion duration, treatment modality, and host comorbidities into the microbiological framework, providing a more holistic view of disease complexity However, given the limited number of patients and absence of longitudinal follow-up, conclusions about bacterial colonization as a primary or major cause of treatment failure should be interpreted cautiously. These findings should be considered as hypothesis-generating, supporting the need for confirmatory studies with broader sampling.

The findings further support the development of adjunct therapies, including topical antibiotics, probiotics, or bacteriophage therapy, to mitigate secondary infections and enhance treatment success [23, 24]. Future research should focus on functional metagenomic and transcriptomic analyses to elucidate the role of microbial communities in CL lesion pathophysiology and biofilm formation potential. Additionally, systematic antibiotic susceptibility profiling of isolates is warranted to inform targeted antimicrobial interventions. These advanced approaches could offer deeper insights into microbial-host-parasite interactions and guide optimized, evidence-based treatment protocols for CL.

Limitations

- Limited generalizability due to the small sample size and use of convenience sampling from a single hospital setting.
- Cross-sectional design restricts the ability to assess temporal changes or establish causal relationships between bacterial colonization and treatment outcomes.
- Potential for contamination from skin flora or the environment cannot be entirely excluded, and the absence of functional metagenomic or antibiotic susceptibility profiling limits deeper insight into microbial behavior and resistance patterns.

Conclusion

This study highlights the diverse bacterial communities associated with cutaneous leishmaniasis (CL) lesions, revealing a complex interplay between commensal, opportunistic, and pathogenic bacteria. The consistent presence of *S. aureus*, *Bacillus spp.*, and *Acinetobacter spp.* suggests their potential role in secondary infections, prolonged lesion inflammation, and delayed healing. However, the small sample size and use of convenience sampling from a single hospital limit the external validity and broader applicability of the findings. Future studies with larger, randomized, and geographically diverse samples are necessary to confirm these associations and support the development of targeted therapeutic strategies. To strengthen clinical translation, future studies should integrate functional metagenomic analyses and antibiotic susceptibility testing. These tools would help identify resistance mechanisms, characterize microbial functions, and guide adjunct antimicrobial therapies, especially for patients with persistent or treatment-resistant lesions.

Abbreviations

CL	Cutaneous leishmaniasis
SSG	Sodium stibogluconate
IL-SSG	Intra-lesional SSG
BLAST	Basic Local Alignment Search Tool
MEGA	Molecular Evolutionary Genetics Analysis
NJ	Neighbor-Joining

Supplementary Information

The online version contains supplementary material available at https://doi.org/10.1186/s13104-025-07420-y.

Supplementary Material 1: The questionnaire used to receive patient information

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Author contributions

NG: Conceptualization, designing the research, sample collection data analysis, writing the manuscript, TS, SE: sample collection and culturing of bacteria, WR: Molecular analysis and editing the manuscript. HG, BS: Providing sample collection and critical review. All authors read and approved the manuscript.

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Data availability

The datasets generated during the current study are available in the the GenBank database of the National Center for Biotechnology Information (NCBI) repository (Accession numbers, PV162829, PV162830, PV162831, PV162832, PV162833, PV162834, PV162835, PV162836, PV162837, PV162838, PV162839, PV162840, PV162841, PV162842, PV162843, PV162844, PV162845, PV162846.)

Declarations

Ethics approval and consent to participate

Ethical clearance for the study was obtained from the Ethics Review Committee (ERC) of the Faculty of Medicine, University of Kelaniya, Sri Lanka (P/103/08/2023). Informed Consent to participate in this study was obtained from all participants. For the individuals under the age of 16, informed consent to participate was obtained from their parents or legal guardians. This study was conducted in accordance with the principles of the Declaration of Helsinki.

Consent for publication

Not applicable.

Competing interests

The authors declare no competing interests.

Clinical trial

Not applicable.

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References

1. Karunaweera ND, Pratlong F, Siriwardane HVYD, Ihalamulla RL, Dedet JP. Sri Lankan cutaneous leishmaniasis is caused by *Leishmania donovani* zymodeme MON-37. Trans R Soc Trop Med Hyg. 2003;380–1.
2. Reithinger R, Dujardin JC, Louzir H, Pirmez C, Alexander B, Brooker S. Cutaneous leishmaniasis. Lancet Infect Dis. 2007;7(9):581–96.
3. Thakur L, Singh KK, Shanker V, Negi A, Jain A, Matlashewski G, Jain M. Atypical leishmaniasis: A global perspective with emphasis on the Indian Subcontinent. PLoS Negl Trop Dis. 2018;12(9):e0006659.
4. Tilahun Zewdu F, Tessema AM, Zerga AA, van Henten S, Lambert SM. Effectiveness of intralesional sodium stibogluconate for the treatment of localized cutaneous leishmaniasis at Boru Meda general hospital, amhara, ethiopia: pragmatic trial. PLoS Negl Trop Dis. 2022;16(9):e0010578.
5. Silva H, Liyanage A, Deerasinghe T, Chandrasekara V, Chellappan K, Karunaweera ND. Treatment failure to sodium stibogluconate in cutaneous leishmaniasis: A challenge to infection control and disease elimination. PLoS ONE. 2021;16(10):e0259009.
6. Aimon Khan Y-S, Koh. The role of skin Microbiome in human health and diseases. J Bacteriol Virol. 2024;54(3):191–202.
7. Farias Amorim C, Lovins VM, Singh TP, Novais FO, Harris JC, Lago AS, Carvalho LP, Carvalho EM, Beiting DP, Scott P, Grice EA. The skin microbiome enhances disease through IL-1b and delays healing in cutaneous leishmaniasis patients. medRxiv [Preprint]. 2023 Feb 6:2023.02.02.23285247. https://doi.org/10.1101/2023.02.02.23285247. Update in: Sci Transl Med. 2023;15(718):eadh1469. doi: 10.1126/scitranslmed.adh1469.
8. Scales BS, Huffnagle GB. The Microbiome in wound repair and tissue fibrosis. J Pathol. 2013;229(2):323–31.
9. Isaac-Márquez AP, Lezama-Dávila CM. Detection of pathogenic bacteria in skin lesions of patients with chiclero's ulcer. Reluctant response to antimonial treatment. Mem Inst Oswaldo Cruz. 2003;98(8):1093–5.

10. Gashaw B, Yizengaw E, Yismaw G, Tebeje S, Tilahun F, Sebsibe B, et al. Multifaceted impact of cutaneous leishmaniasis: treatment challenges and implications for healthcare systems and society in Boru Meda hospital, North-Central Ethiopia. Ethiop J Health Dev. 2023;2(37):1–7.
11. Borbón TY, Scorza BM, Clay GM, Lima Nobre de Queiroz F, Sariol AJ, Bowen JL, Chen Y, Zhanbolat B, Parlet CP, Valadares DG, Cassel SL, Nauseef WM, Horswill AR, Sutterwala FS, Wilson ME. Coinfection with *Leishmania major* and *Staphylococcus aureus* enhances the pathologic responses to both microbes through a pathway involving IL-17A. PLoS Negl Trop Dis. 2019;13(5):e0007247.
12. Lowy FD. *Staphylococcus aureus* infections. N Engl J Med. 1998;339(8):520–32.
13. Tong SYC, Davis JS, Eichenberger E, Holland TL, Fowler VG. Staphylococcus aureus infections: epidemiology, pathophysiology, clinical manifestations, and management. Clin Microbiol Rev. 2015;28(3):603–61.
14. Kloos WE, Bannerman TL. Update on clinical significance of coagulase-negative Staphylococci. Clin Microbiol Rev. 1994;7(1):117–1140.
15. Ramírez-Granillo A, Bautista-Hernández LA, Bautista-De Lucío VM, Magaña-Guerrero FS, Domínguez-López A, Córdova-Alcántara IM, Pérez NO, Martínez-Rivera MD, Rodríguez-Tovar AV. Microbial warfare on three fronts: mixed biofilm of Aspergillus fumigatus and Staphylococcus aureus on primary cultures of human limbo-corneal fibroblasts. Front Cell Infect Microbiol. 2021;11:646054.
16. Ravaioli S, De Donno A, Bottau G, Campoccia D, Maso A, Dolzani P, Balaji P, Pegreffi F, Daglia M, Arciola CR. The opportunistic pathogen *Staphylococcus warneri*: virulence and antibiotic resistance, clinical features, association with orthopedic implants and other medical devices, and a glance at industrial applications. Antibiot (Basel). 2024;13(10):972.
17. Peleg AY, Seifert H, Paterson DL. *Acinetobacter baumannii*: emergence of a successful pathogen. Clin Microbiol Rev. 2008;21(3):538–82.
18. Becker K, Heilmann C, Peters G. Coagulase-negative Staphylococci. Clin Microbiol Rev. 2014;27(4):870–926.
19. Kaluarachchi TJ, Weerasekera MM, Wickremasinghe R, Yasawardene S, De Silva H, Yapa PD, Menike C, Ranasinghe S. Clinical challenges in cutaneous leishmaniasis: A cohort from Southern province, Sri Lanka. Int J Multidisciplinary Stud. 2024;11(2):01–21.
20. Høiby N, Bjarnsholt T, Givskov M, Molin S, Ciofu O. Antibiotic resistance of bacterial biofilms. Int J Antimicrob Agents. 2010;35(4):322–32.
21. Wolcott RD, Rhoads DD, Dowd SE. Biofilms and chronic wound inflammation. J Wound Care. 2008;17(8):333–41.
22. Howard A, O'Donoghue M, Feeney A, Sleator RD. *Acinetobacter baumannii*: an emerging opportunistic pathogen. Virulence. 2012;3(3):243–50.
23. O'Toole GA, Kolter R. Flagellar and twitching motility are necessary for Pseudomonas aeruginosa biofilm development. Mol Microbiol. 1998;30(2):295–304.
24. Guarner F, Malagelada JR. Gut flora in health and disease. Lancet. 2003;361(9356):512–9.

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