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SHORT REPORT OPEN ACCESS

Effectiveness of Carbon Dioxide Cryotherapy for the Treatment of Localized Cutaneous Leishmaniasis in Ethiopia

Feleke Tilahun Zewdu[1,2,3] | Saba Maria Lambert[4] | Michael Marks[4,5] | Yematawork Kebede Aragaw[1] | Derese Bekele Daba[1,6] | Kassahun Alemu[2] | Endalamaw Gadisa[1] | Stephen L. Walker[4,5]

[1]Malaria and Neglected Tropical Diseases Research Division, Armauer Hansen Research Institute, Addis Ababa, Ethiopia | [2]Department of Epidemiology and Biostatistics, Institute of Public Health, College of Medicine and Health Sciences, University of Gondar, Gondar, Ethiopia | [3]Dermatology Department, Boru Meda General Hospital, Dessie, Amhara, Ethiopia | [4]Department of Clinical Research, Faculty of Infectious and Tropical Diseases, London School of Hygiene & Tropical Medicine, UK Hospital, London, UK | [5]Hospital for Tropical Diseases, University College London Hospitals, NHS Foundation Trust, London, UK | [6]Department of Public Health, College of Medicine and Referral Hospital, Ambo University, Ambo, Ethiopia

Correspondence: Feleke Tilahun Zewdu (momflk@gmail.com)

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ABSTRACT

Background: Cutaneous leishmaniasis (CL) is a public health problem in Ethiopia. Diagnosis is often delayed, and treatment options are limited. Liquid nitrogen cryotherapy is a recommended treatment but not widely available. Carbon dioxide (CO_2) cryotherapy is used for the prevention of cervical cancer and is widely available in Ethiopia and might be a suitable therapy for treating localized CL.

Objectives: The aim of this short report is to assess the effectiveness of carbon dioxide cryotherapy for the treatment of CL in CL treatment centre, Ethiopia.

Methods: We performed a prospective study assessing the effectiveness of CO_2 cryotherapy for the treatment of localized CL between September 2022 and June 2023 at an established CL treatment centre.

Results: Seventeen individuals with 24 CL lesions were enrolled. Twelve (70.6%) were confirmed using a skin slit smear and five by histopathology (29.4%). Nine (52.9%) individuals received a single session of CO_2 cryotherapy, five received two (29.4%) and three (17.65%) received three sessions of cryotherapy. At Day 90, 16 participants were assessed and 14 (82.4%) had healed.

Conclusions: CO_2 cryotherapy shows promise as a potential treatment strategy for CL. Formal evaluations are required.

1 | Introduction

Cutaneous leishmaniasis (CL) is a vector-borne skin neglected tropical disease (NTD) caused by an intracellular parasite of the genus *Leishmania* [1]. In Ethiopia, CL is predominantly due to *Leishmania aethiopica* and is a significant public health problem with an estimated 50,000 new cases annually [2]. Many of those affected live in rural areas making access to treatment challenging and expensive [1, 2]. CL is associated with reduced health-related quality of life [2].

The choice of treatment for CL is influenced by the clinical phenotype, location, number of lesions [3–5], and the preference of the affected individual. In localized CL physical

Endalamaw Gadisa and Stephen L. Walker have equal contribution.

Summary

What is already known about this topic?

- CL is a common parasitic skin disease in Ethiopia.
- Liquid nitrogen cryotherapy is recommended but not widely available.
- Seeking care for CL is expensive for affected individuals

What does this study add?

- Carbon dioxide cryotherapy appears to be effective for treating localized CL.
- Carbon dioxide cryotherapy appears to give similar results to liquid nitrogen.
- Carbon dioxide cryotherapy offers an innovative approach to the decentralized management of CL.

and pharmacological therapies are recognized treatment modalities. Cryotherapy may be used as monotherapy or as an adjunct to pharmacological agents such as sodium stibogluconate (SSG) administered intralesionally or systemically. Liquid nitrogen cryotherapy, applied weekly, using a "cotton applicator" to provide a 10–30-s freeze, with a thawing interval of 20 s, was reported to cure 93% (83/89) of confirmed CL lesions in southern Ethiopia [6].

Cryotherapy is included as a treatment option in Ethiopia's Guideline for Diagnosis, Treatment, and Prevention of Leishmaniasis [3]. Nearly 33.4% of Ethiopians live in CL endemic areas [7]. Ethiopia aims to improve access to CL diagnosis and treatment, with a target to increase the number of treatment centres to 30 and diagnostic centres to 170, by 2030 [8]. At present, liquid nitrogen and other treatments for CL are only available in the 25 treatment centres based in hospitals, mostly in urban areas [8], and the supply of liquid nitrogen is not always assured.

Carbon dioxide (CO_2) cryotherapy is used for the treatment of cervical intraepithelial neoplasia in Ethiopia [9] with 1400 cryotherapy machines distributed to health facilities in 770 districts. CO_2 cryotherapy has been reported to have a high cure rate for CL, in a prospective case series in Yemen [10]. One hundred and eighty-three individuals, with 1–9 lesions received CO_2 cryotherapy. A session of cryotherapy consisted of a slush of CO_2, proportionate to the size of the lesion, applied directly to the lesion with slight pressure for 60 s followed by a thaw interval (unspecified duration), and then a further 60-s application. One session was sufficient to cure the lesions in 163 individuals (89%). CO_2 cryotherapy is a potentially useful treatment for CL, but there have been no studies of its effectiveness in Ethiopia.

In January 2023, a multi-stakeholder meeting with health officials of the Ethiopian Federal Ministry of Health and the Amhara Regional Health Bureau was convened in Addis Ababa to discuss approaches to decentralization of CL care. The outcome of the meeting was that CO_2 cryotherapy for CL was an acceptable avenue for research. We therefore aimed to conduct a pilot study of CO_2 cryotherapy to determine its efficacy in the treatment of localized CL in Ethiopia and to inform work focused on decentralizing care for people with CL.

2 | Methods

We performed a prospective open-label study to gather preliminary data on the effectiveness of CO_2 cryotherapy for the treatment of localized CL between September 2022 and June 2023 at Boru Meda Hospital. CL was confirmed by clinical examination and subsequent slit-skin smear and/or biopsy. Treatment naïve individuals over the age of 4 years with parasitologically confirmed localized CL were eligible if the lesion was less than 5 cm (largest diameter); if there were fewer than four lesions; and if no lesion was involving or within 2 cm of a mucosal surface, on the eyelid or a joint. Lesions were measured using a disposable tape measure. The operational definition of localized CL was a parasitological confirmed case of leishmaniasis, with no mucosal involvement, characterized by ten or fewer cutaneous papules and/or nodules and/or plaques with or without ulceration involving one body site [11]. Convenience sampling was used for recruitment.

CO_2 cryotherapy was delivered using the Medgyn Cryotherapy System MGC-200. The system uses compressed CO_2 which is released on the depression of a trigger leading to the cooling of a metallic tip which is then applied to the lesion. Three different tip sizes were available (1, 3, and 5 cm diameter). The following CO_2 cryotherapy regimen was employed: an appropriate size of cryo-tip was cooled until ice appeared on the tip (for a maximum of 15 s, using a timer); the cooled tip was applied to the CL lesion for 40 s, followed by a thawing period of 20 s, and a second application of 40 s. Participants were evaluated every 2 weeks and CO_2 cryotherapy was repeated if deemed necessary by the clinician. An individual could receive a maximum of six treatment sessions over a period of 12 weeks. Participants were reviewed every 2 weeks until Day 90. All individuals were assessed at Day 90 after the first administration of CO_2 cryotherapy to determine the outcome which was a dichotomous global physician assessment of the CL lesion as cured or not cured using clinical parameters of reduction in size, flattening, re-epithelialization, and resolution of erythema [12]. The type of posttreatment pigmentary changes at the site of the CL lesion was recorded as no pigmentary change, hypopigmented, or hyperpigmented. The outcome for individuals with more than one CL lesion at enrolment was based on the largest lesion. Individuals who did not respond to CO_2 cryotherapy were offered treatment with intra-lesional SSG. Adverse effects of cryotherapy were recorded.

3 | Results

Of the 17 included in the study, 12 (70.6%) had a single CL lesion and 5 (29.4%) had more than one (range 2–3). The median age was 25 years (IQR 14–39), and 9 (53%) were female. Twelve (70.6%) cases were confirmed using skin slit smear and 5 (29.4%) by histopathology (Table 1). Seven participants (41.2%) had a facial lesion and 11 participants (64.7%) had a

TABLE 1 | Summary table for the study participants with localized cutaneous leishmaniasis and their outcomes following carbon dioxide cryotherapy, 2024.

Sex	Age (inyears)	Number of lesions	Size of the most significant lesion (in cm)	Site of the largest lesion	Duration of the largest lesion (in months)	Number of cryotherapy sessions	Day 90 outcome (cured, not cured)	Posttreatment pigmentary changes (Y/N)
Female	28	1	5	Forearm	12	2	Cured	**Y**
Female	22	1	2	Nose	2	2	Cured	N
Male	4	1	4	Nose	8	1	Cured	**Y**
Male	25	1	1	Forearm	4	1	Cured	**Y**
Female	33	3	3	Forearm	7	2	Cured	**Y**
Male	19	1	1.5	Neck	4	1	Cured	**Y**
Female (A)	7	1	1	Chin	5	1	Cured	N
Male	56	1	3	Leg	8	1	Cured	N
Female (B)	63	2	5	Forehead	18	2	Cured	N
Female	5	2	4	Forearm	12	1	Cured	**Y**
Male	9	3	2	Cheek	13	1	Cured	N
Male (C)	37	1	2	Forearm	3	1	Cured	**Y**
Female	21	1	4	Leg	6	2	Cured	**Y**
Female	28	1	5	Forearm	6	3	Cured	**Y**
Female	41	2	3	Nose	4	2	Not Cured	Not applicable
Male	4	1	4	Nose	9	3	Not Cured	Not applicable
Male	19	1	2	Neck	3	1	Lost to follow up	Unknown

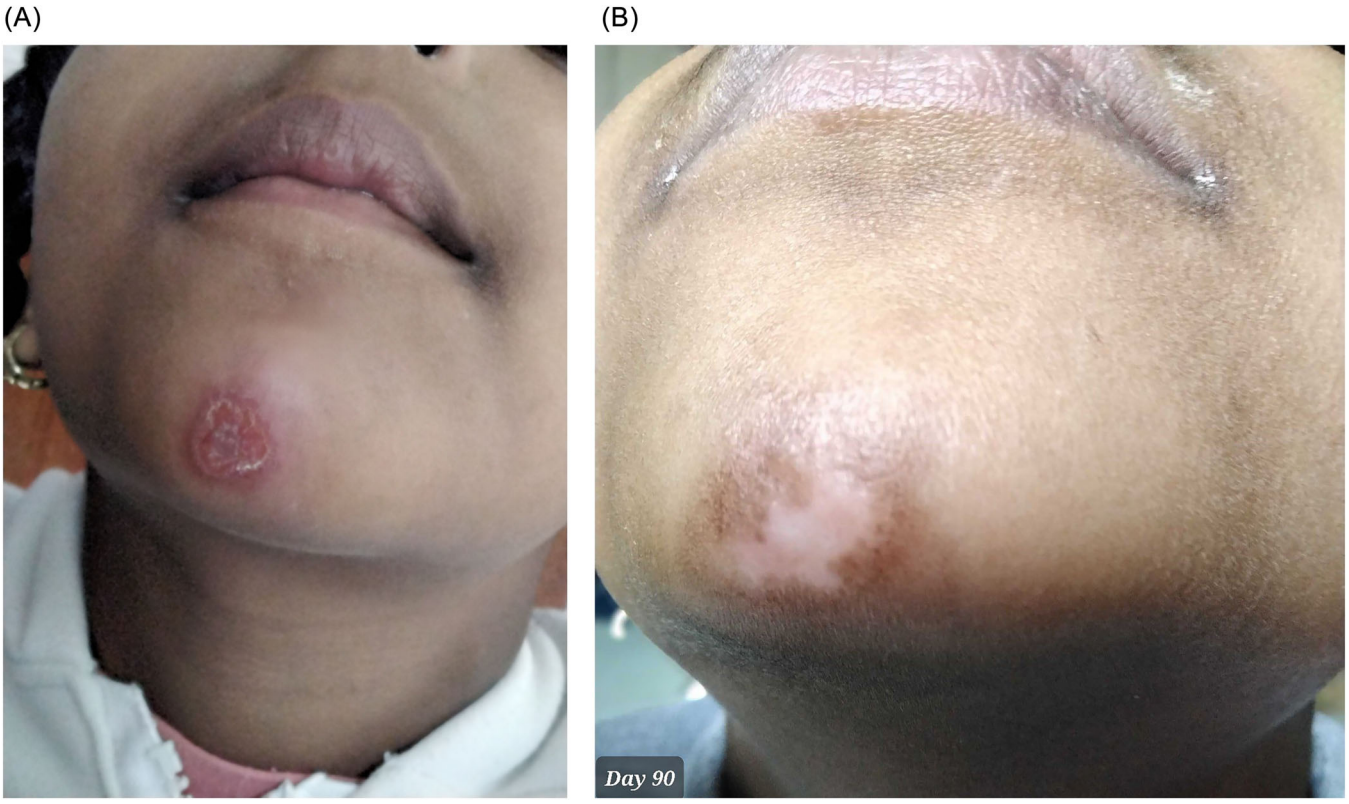

FIGURE 1 | (A) A solitary erythematous ulcerated plaque on the chin at enrolment. (B) A hypopigmented scar on the chin at Day 90 following treatment with CO_2 cryotherapy.

lesion larger than 2 cm. Ten individuals (58.8%) had lesions that had been present for less than 6 months.

Nine (52.9%) individuals received a single session of cryotherapy, five (29.4%) received two sessions, and three (17.65%) received three sessions of cryotherapy. No adverse effects were reported other than dyspigmentation. At Day 90, outcome data were available for 16 participants (Table 1). One participant was lost to follow-up after the first treatment session. Of these, 14 (87.5%) were cured. Two participants did not respond to CO_2 cryotherapy (after 2 and 3 sessions, respectively) and were subsequently treated with intra-lesional SSG. Of the 14 who responded to CO_2 cryotherapy, 6 had hyperpigmentation and 3 had hypopigmentation. Five did not have any dyspigmentation.

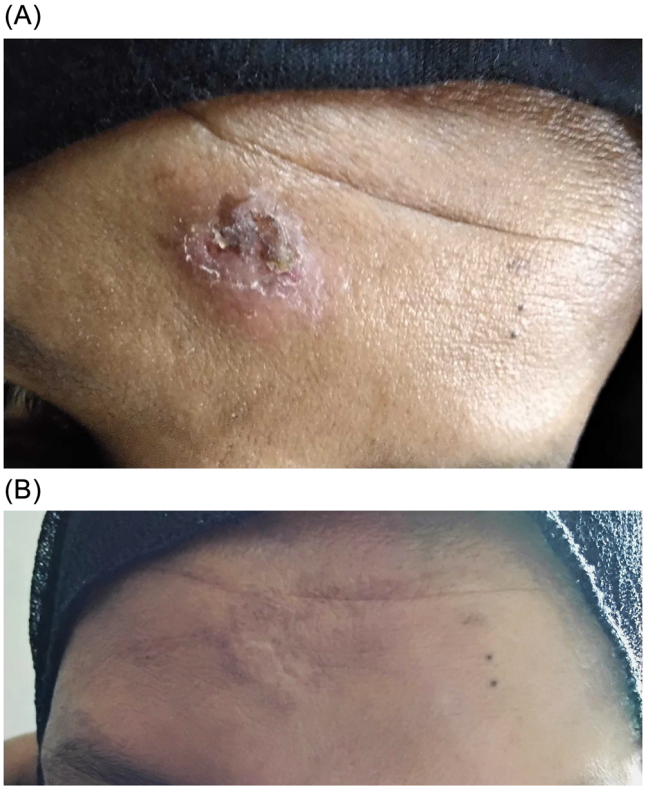

FIGURE 2 | (A) An ulcerated plaque lesion on the forehead at enrolment. (B) Hyperpigmentation on the forehead at Day 90 following treatment with CO_2 cryotherapy.

4 | Discussion

Our small case series suggests that CO_2 cryotherapy may be an effective intervention with comparable cure rates to those reported with liquid nitrogen in Ethiopia [13] and CO_2 cryotherapy in Yemen [10].

Cryotherapy works by rapidly freezing and damaging target cells and tissues through direct physical effects and triggering an inflammatory response, destroying abnormal tissue [14]. The adverse effects associated with cryotherapy include dyspigmentation, which occurred in 68.8% of individuals in our study who responded to CO_2 cryotherapy assessed at Day 90. This is less than dyspigmentation reported at 3 months in 93.9% of individuals who received miltefosine or cryotherapy in a community-based study in southern Ethiopia, with most (86%) still exhibiting dyspigmentation at 6 months [13].

Lesion-directed therapy early in the course of CL, when lesions are still small, is preferable as they are associated with fewer adverse effects. Currently, in Ethiopia, lesion-directed therapies such as intra-lesional SSG and liquid nitrogen cryotherapy have to be administered repeatedly by trained healthcare workers at one of the 25 CL treatment centres. These facilities are at a considerable distance from the rural areas where CL prevalence is highest, presenting major financial challenges for affected individuals wishing to access care [15]. The decentralization of lesion-directed treatment to the communities affected by CL has been

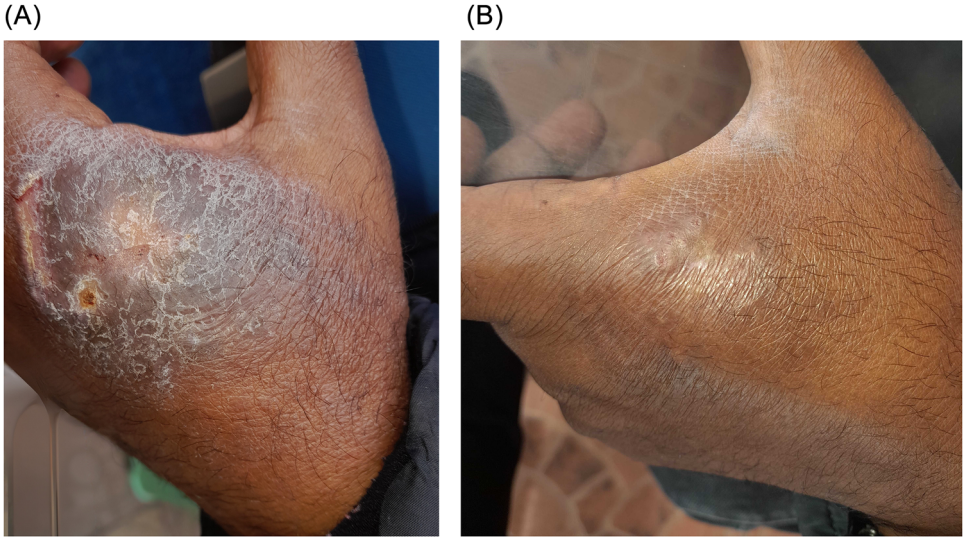

FIGURE 3 | (A) A hyperpigmented, ulcerated plaque on the dorsum of the hand at enrolment. (B) A hypopigmented scar on the dorsum of the hand following treatment with CO_2 cryotherapy.

conducted in research studies but has not been operationally implemented in Ethiopia. The lower temperature of liquid nitrogen, compared to CO_2, has made this the preferred cryogen for cutaneous lesions in dermatology [14]. In Ethiopia, the procurement, safe storage, and training required to use liquid nitrogen cryotherapy appropriately is challenging outside treatment facilities located in large urban centres. The widespread availability of CO_2 cryotherapy for the treatment of cervical dysplasia in more easily accessible health facilities makes this an attractive candidate for the management of localized CL. CO_2 cryotherapy has the potential to deliver synergistic health benefits, increase the cost-effectiveness of the provision of healthcare infrastructure, and improve universal health coverage. Repurposing drugs for better control of NTDs is a well-recognized approach [16] and CO_2 cryotherapy may be repurposed to improve access to treatment for localized CL in Ethiopia. This will require robust evidence of efficacy, tolerability, and acceptability from well-designed, pragmatic randomized controlled trials and a strategy to ensure cryotherapy for CL is feasible and can be implemented sustainably in primary health centres. The dermatology and skin NTD community will need to work with a wide range of colleagues with responsibilities for a variety of health policies including cancer prevention (Figures 1–3).

Our small case series has limitations: the number of participants was small, the outcome data were short-term, and no measure of acceptability was obtained. A single clinician conducted the outcome assessment. Participants were not questioned systematically about adverse effects.

Author Contributions

Conceptualization: Stephen L. Walker, Feleke Tilahun, Endalamaw Gadisa, Saba Lambert and Michael Marks. **Formal analysis:** Feleke Tilahun. **Methodology:** Feleke Tilahun, Kassahun Alemu, Endalamaw Gadisa, Michael Marks, Stephen L. Walker and Saba Lambert. **Investigation:** Feleke Tilahun and Yematawork Kebede. **Supervision:** Yematawork Kebede, Endalamaw Gadisa, Michael Marks and Stephen L Walker. **Writing – original draft:** Feleke Tilahun. **Writing – review and editing:** Endalamaw Gadisa, Saba Lambert, Yematawork Kebede, Derese Bekele, Stephen L. Walker and Michael Marks.

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Ethics Statement

This study was conducted by the ethical standards of the Institutional Review Board/Ethics Committee and received approval number (APHI/Dessie/4/947/2024). All participants provided written informed consent/assent for the data to be collected and the photos taken and to be used for teaching and publication purposes too before data collection, ensuring the confidentiality of their responses throughout the research process. No identifiable participant information will be disclosed in the study results. The parents/guardians of minor patients have given written informed consent for their child's participation in the study, as well as for the use of their child's deidentified, anonymized, aggregated data, and case details (including photographs) for publication. Adult patients have given written informed consent for participation in the study and the use of their deidentified, anonymized, aggregated data and their case details (including photographs) for publication. An ethical approval letter was taken from the Amhara public health institution with the approval number APHI/Dessie/4/947/2024.

Conflicts of Interest

The authors declare no conflicts of interest.

Data Availability Statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

References

1. S. Burza, S. L. Croft, and M. Boelaert, "Leishmaniasis," *Lancet* 392, no. 10151 (2018): 951–970, https://doi.org/10.1016/S0140-6736(18)31204-2.

2. S. Henten, van, W. Adriaensen, H. Fikre, et al., "Cutaneous Leishmaniasis Ddue to *Leishmania aethiopica*," *EClinicalMedicine* 6 (January 2018): 69–81, https://doi.org/10.1016/j.eclinm.2018.12.009.

3. Ethiopian Ministry of Health Gfd, "Treatment and Prevention of Leishmaniasis," 2nd ed. (2013), https://www.afrikadia.org/wp-content/uploads/2018/08/VL_Guidelines_Ethiopia_2013.pdf.

4. N. Aronson, B. L. Herwaldt, M. Libman, et al., "Diagnosis and Treatment of Leishmaniasis: Clinical Practice Guidelines by the Infectious Diseases Society of America (IDSA) and the American Society of Tropical Medicine and Hygiene (ASTMH)," *Clinical Infectious Diseases* 63, no. 12 (December 2016): e202–e264, https://doi.org/10.1093/cid/ciw670.

5. J. Blum, P. Buffet, L. Visser, et al., "Leishman Recommendations for Treatment of Cutaneous and Mucosal Leishmaniasis in Travelers, 2014," *Journal of Travel Medicine* 21, no. 2 (2014): 116–129, https://doi.org/10.1111/jtm.12089.

6. E. Negera, E. Gadisa, J. Hussein, et al., "Treatment Response of Cutaneous Leishmaniasis due to *Leishmania aethiopica* to Cryotherapy and Generic Sodium Stibogluconate From Patients in Silti, Ethiopia," *Transactions of the Royal Society of Tropical Medicine and Hygiene* 106, no. 8 (2012): 496–503, https://doi.org/10.1016/j.trstmh.2012.02.006.

7. Ethiopian Statistical Agency, 2024. http://www.statsethiopia.gov.et/wp-content/uploads/2024/07/Projected_Population-2024.pdf.

8. Ministry of Health, the Third National Neglected Tropical Diseases Strategic Plan 2021-2025 (2013/14–2017/18 e.c.), November 2021, P49.

9. Federal Democratic Republic of Ethiopia, Ministry of Health, *Guideline for Cervical Cancer Prevention and Control in Ethiopia*, 2015.

10. Y. Al-Qubati, MD, E. J. Janniger, and R. A. Schwartz, "Cutaneous Leishmaniasis: Cryosurgery Using Carbon Dioxide Slush in a Resource-Poor Country," *International Journal of Dermatology* 51 (2012): 1217–1220.

11. A. B. Mohammed, F. S. Mohammed, F. T. Zewdu, et al., "Protocol for a Prospective Observational Cohort Study of Cutaneous Leishmaniasis in Ethiopia," *NIHR Open Research* 3 (2023): 49, https://doi.org/10.3310/nihropenres.13432.1.

12. P. Olliaro, M. Grogl, M. Boni, et al., "Harmonized Clinical Trial Methodologies for Localized Cutaneous Leishmaniasis and Potential for Extensive Network With Capacities for Clinical Evaluation," *PLoS Neglected Tropical Diseases* 12, no. 1 (2018): e0006141, https://doi.org/10.1371/journal.pntd.0006141.

13. S. Henten, van, M. Pareyn, D. Tadesse, et al., "Community-Based Treatment of Cutaneous Leishmaniasis Using Cryotherapy and Miltefosine in Southwest Ethiopia: The Way Forward?," *Frontiers in Medicine* 10 (October 2023): 1196063, https://doi.org/10.3389/fmed.2023.1196063.

14. G. B. Colver and R. P. R. Dawber, "Cryosurgery, the Principles, and Simple Practice," *Clinical and Experimental Dermatology* 14, no. 1 (January 1989): 1–6, https://doi.org/10.1111/j.1365-2230.1989.tb00873.x.

15. Y. Hailemichael, J. Novignon, L. Owusu, et al., "The Role of Economic Factors in Shaping and Constituting the Household Burden of Neglected Tropical Diseases of the Skin: Qualitative Findings From Ghana and Ethiopia," *Social Science & Medicine* 356 (July 2024): 117094, https://doi.org/10.1016/j.socscimed.2024.117094.

16. P. J. Hotez, B. Pecoul, S. Rijal, et al., "Eliminating the Neglected Tropical Diseases: Translational Science and New Technologies," *PLoS Neglected Tropical Diseases* 10, no. 3 (March 2016): e0003895, https://doi.org/10.1371/journal.pntd.0003895.