Short Report: Successful Treatment of Canine Cutaneous Leishmaniasis Using Radio-Frequency Induced Heat (RFH) Therapy

Anil A. Ahuja, Ram A. Bumb, Rajesh D. Mehta, Neha Prasad, Ram K. Tanwar, and Abhay R. Satoskar*

Department of Clinical Veterinary Medicine, RAJUVAS, Bikaner and Department of Skin and STD, S.P. Medical College, Bikaner, Rajasthan, India; Director Clinics, RAJUVAS, Bikaner, Rajasthan, India; Departments of Pathology and Microbiology, The Ohio State University, Columbus, Ohio

Abstract. Canine cutaneous leishmaniasis (CCL) is a significant veterinary problem. Infected dogs also serve as parasite reservoirs and contribute to human transmission of cutaneous leishmaniasis (CL). Current treatments for CCL are cumbersome and toxic because they are prolonged and involve multiple injections of antimonials. Radio-frequency induced heat (RFH) therapy has been found to be highly effective against CL in humans. Here, we examined the efficacy of topical RFH therapy in the treatment of CL in two pet dogs. We found that RFH therapy induced complete clinical cure and lesion healing within 45 days and both dogs have remained disease free for the last 16 months. This report is the first to demonstrate that a single topical application of RFH therapy is safe and effective in inducing long-term cure of CCL.

Leishmania are obligate intracellular parasites that infect both humans and animals. These parasites are transmitted by a sand fly vector and cause a wide range of diseases, such as cutaneous leishmaniasis (CL), mucocutaneous leishmaniasis (ML), and visceral leishmaniasis (VL). Over 12 million people currently suffer from leishmaniasis, and ~2 million are infected annually, making it a major global health problem and a World Health Organization (WHO) classified neglected tropical disease. In many *Leishmania*-endemic regions of the world, infected dogs serve as a significant reservoir for zoonotic transmission of VL and CL to humans.^{1–9} Many recent studies have also reported that dogs in the United States have tested positive for leishmaniasis,^{10–14} although no dog-to-human transmission has been documented.

Cutaneous leishmaniasis in dogs is a significant veterinary problem, but it is also well documented that infected dogs serve as parasite reservoirs and contribute significantly to human transmission of VL and CL in many parts of the world. Cutaneous leishmaniasis in dogs manifests as localized skin lesions that may either heal or become chronic, the latter leading to significant tissue destruction and disfigurement. Current treatments available for canine CL (CCL) are cumbersome, toxic, and sometimes associated with a relapse.^{15–20} Safe, inexpensive, and effective treatment that induces long-term cure is therefore needed for CCL.

Recently, topical radio-frequency induced heat (RFH) therapy has shown promise in the treatment of CL in immunocompetent individuals^{21–24} and immunocompromised patients infected with human immunodeficiency virus (HIV).²⁵ This treatment involves controlled and localized delivery of radio frequencies into lesions for 60 seconds under local anesthesia using a current field radio-frequency generator. The goal of this report was to determine whether RFH therapy could also be used for the treatment of CL in dogs. Our findings show that a single application of RFH therapy is safe and effective in inducing complete long-term clinical cure of CL in dogs, and could therefore represent a new treatment for CCL.

Two female dogs, a German Sheppard and Labrador retriever, with multiple ulcerative mucocutaneous lesions

on the snout, were brought to a small animal clinic at RAJUVAS, Bikaner, India (Figure 1A and B). Parasitological examination of the skin smear confirmed a diagnosis of CL, which is endemic in this region of India. The lesions were treated by a single application of RFH therapy for 30 seconds under local anesthesia (2% Lidocaine), as described previously.^{25,26} Topical antibiotic cream was applied daily over the

FIGURE 1. Cutaneous leishmaniasis in dogs manifesting as multiple nodules or ulcerating lesions on the snout. (A) German shepherd dog and (B) Labrador retriever.

*Address correspondence to Abhay R. Satoskar, Department of Pathology and Microbiology, The Ohio State University, Columbus, OH 43210. E-mail: abhay.satoskar@osumc.edu

treated lesions for 7 days to prevent secondary bacterial infection. Dogs were examined every 15 days to monitor lesion healing. Both dogs responded well to RFH therapy and resolved their lesions by Day 45 post treatment (Figure 2A and B). At follow-up 16 months later, both were disease free and showed no evidence of lesions.

The most effective treatment of CL in dogs involves injections of pentavalent antimonial, meglumine antimonite in combination with allopurinol, which is required to be administered for 6–12 months to prevent the relapses of the infection.^{15,18,27} Some studies have reported that a combination of meglumine antimonite and miltefosine is also effective in the treatment of CCL.^{28,29} We had previously found that once a week intralesional injection of 1% berberine sulphate for 4–6 weeks could be used to treat CL in domestic dogs, although its long-term efficacy was not determined.²⁰ Never-

theless, all these treatments are cumbersome and could induce significant side effects in some canines.

Non-pharmacological interventions such as cryotherapy and heat therapy have been used in the treatment of CL in humans but never tested in dogs.^{21–23,26,30–34} Cryotherapy using liquid nitrogen was found to be safe and promising against CL, but this treatment required multiple applications for a period ranging from 1 to 3 weeks.^{32,34–36} In addition, although cryotherapy was highly effective in the treatment of dryer and smaller (< 1 cm) lesions, it had to be administered in combination with intralesional antimonials for treating large lesions.^{31,37} The RFH therapy has also been found to be safe and effective in the treatment of CL in humans.^{21–23,38} Several short-term (4–5 months) follow-up studies have shown that RFH therapy is comparable or has even better efficacy than antimonials in the treatment of CL.^{22–24,39,40}

In this study, we found that a single application of RFH therapy under local anesthesia was safe and sufficient to induce complete clinical cure of CL within 45 days associated with the resolution of lesions with no or minimal scarring. Furthermore, both dogs remained disease free for 16 months after the treatment, indicating that this treatment was effective in inducing long-term cure of the infection. Together, these findings indicate that RFH therapy could be a new first-line treatment of CCL and may be a useful therapy to help limit the zoonotic spread of CL from dogs to man.

Received December 4, 2011. Accepted for publication February 27, 2012.

Financial Support: This study was funded by a grant from Thermosurgery Inc., to A.R.S.

Authors' addresses: Anil A. Ahuja and Ram K. Tanwar, RAJUVAS—Veterinary Medicine, Bikaner, India, E-mails: isacp2012@gmail.com and rktanward@rediffmail.com. Ram A. Bumb, PBM Hospital—Dermatology, Bikaner, India, E-mail: dr_bumb@rediffmail.com. Rajesh D. Mehta and Neha Prasad, SP Medical College—Ski and STD, Bikaner, India; and PBM Hospital—Dermatology, Bikaner, India, E-mails: rdm@hotmail.com and nehaprasd@hotmail.com.

REFERENCES

1. Abdeen ZA, Sawalha SS, Eisenberger CL, Khantar HM, Greenblatt CL, Yousef O, Schnur LF, Azmi K, Wargurg A, 2002. Epidemiology of visceral leishmaniasis in the Jenin District, West Bank: 1989–1998. *Am J Trop Med Hyg* 66: 329–333.
2. Abranches P, Silva-Pereira MC, Conceicao-Silva FM, Santos-Gomes GM, Janz JG, 1991. Canine leishmaniasis: pathological and ecological factors influencing transmission of infection. *J Parasitol* 77: 557–561.
3. Aguilar CM, Rangel EF, Garcia L, Fernandez E, Momen H, Garimaldi G Jr, Vargas Z, 1989. Zoonotic cutaneous leishmaniasis due to *Leishmania (Viannia) braziliensis* associated with domestic animals in Venezuela and Brazil. *Mem Inst Oswaldo Cruz* 84: 19–28.
4. Costa CH, Stewart JM, Gomes RB, Garcez LM, Ramos PK, Bozza M, Satoskar A, Dissanayake S, Santos RS, Silva MR, Shaw JJ, David JR, Maguire SH, 2002. Asymptomatic human carriers of *Leishmania chagasi*. *Am J Trop Med Hyg* 66: 334–347.
5. Desjeux P, Bryan JH, Martin-Saxton P, 1983. Leishmaniasis in The Gambia. 2. A study of possible vectors and animal reservoirs, with the first report of a case of canine leishmaniasis in The Gambia. *Trans R Soc Trop Med Hyg* 77: 143–148.
6. Falqueto A, Sessa PA, Varejao JB, Barros GC, Momen H, Grimaldi JG, 1991. Leishmaniasis due to *Leishmania braziliensis* in Espirito Santo State, Brazil. Further evidence on the role of dogs as a reservoir of infection for humans. *Mem Inst Oswaldo Cruz* 86: 499–500.

FIGURE 2. Complete healing of cutaneous lesions by Day 45 after RFH therapy. (A) German shepherd dog and (B) Labrador retriever.

7. Gavvani AS, Mohite H, Edrissian GH, Mohebbi M, Davies CR, 2002. Domestic dog ownership in Iran is a risk factor for human infection with *Leishmania infantum*. *Am J Trop Med Hyg* 67: 511–515.
8. Morsy TA, Hamadto HA, Rashed SM, el Fakahany AF, Abdalla KF, 1990. Animals as reservoir hosts for *Leishmania* in Qalyobia Governorate, Egypt. *J Egypt Soc Parasitol* 20: 779–788.
9. Padilla AM, Marco JD, Diosque P, Segura MA, Mora MC, Fernández MM, Malchiodi EL, Basombrio MA, 2002. Canine infection and the possible role of dogs in the transmission of American tegumentary leishmaniasis in Salta, Argentina. *Vet Parasitol* 110: 1–10.
10. Duprey ZH, Steurer FJ, Rooney JA, Kirchhoff LV, Jackson JE, Rowton ED, Schantz PM, 2006. Canine visceral leishmaniasis, United States and Canada, 2000–2003. *Emerg Infect Dis* 12: 440–446.
11. Gaskin AA, Schantz P, Jackson J, Birkenheuer A, Tomlinson L, Gramiccia M, Levy M, Steurer F, Kollmar E, Hegarty BC, Ahn A, Breitschwerdt EB, 2002. Visceral leishmaniasis in a New York foxhound kennel. *J Vet Intern Med* 16: 34–44.
12. Giger U, Oakley DA, Owens SD, Schantz P, 2002. *Leishmania donovani* transmission by packed RBC transfusion to anemic dogs in the United States. *Transfusion* 42: 381–383.
13. Grosjean NL, Vrabie RA, Murphy AJ, Mansfield LS, 2003. Seroprevalence of antibodies against *Leishmania* spp. among dogs in the United States. *J Am Vet Med Assoc* 222: 603–606.
14. Rosypal AC, Troy GC, Zajac AM, Duncan RB Jr, Waki K, Chang KP, Lindsay DS, 2003. Emergence of zoonotic canine leishmaniasis in the United States: isolation and immunohistochemical detection of *Leishmania infantum* from foxhounds from Virginia. *J Eukaryot Microbiol* 50 (Suppl): 691–693.
15. Torres M, Bardagi M, Roura X, Zanna G, Ravera I, Ferrer L, 2011. Long term follow-up of dogs diagnosed with leishmaniasis (clinical stage II) and treated with meglumine antimoniate and allopurinol. *Vet J* 188: 346–351.
16. Denerolle P, Bourdoiseau G, 1999. Combination allopurinol and antimony treatment versus antimony alone and allopurinol alone in the treatment of canine leishmaniasis (96 cases). *J Vet Intern Med* 13: 413–415.
17. Solano-Gallego L, Riera C, Roura X, Iniesta L, Gallego M, Valladares JE, Fisa R, Castillejo S, Alberola J, Ferrer L, Arboix M, Portús M, 2001. *Leishmania infantum*-specific IgG, IgG1 and IgG2 antibody responses in healthy and ill dogs from endemic areas. Evolution in the course of infection and after treatment. *Vet Parasitol* 96: 265–276.
18. Baneth G, Shaw SE, 2002. Chemotherapy of canine leishmaniasis. *Vet Parasitol* 106: 315–324.
19. Noli C, Auxilia ST, 2005. Treatment of canine Old World visceral leishmaniasis: a systematic review. *Vet Dermatol* 16: 213–232.
20. Ahuja A, Purohit SK, Yadav JS, Netra PR, 1993. Cutaneous leishmaniasis in domestic dogs. *Indian J Public Health* 37: 29–31.
21. Aronson NE, Wortmann GW, Byrne WR, Howard RS, Bernstein WB, Marovich MA, Polhemus ME, Yoon IK, Hummer KA, Gasser RA Jr, Oster CN, Benson PM, 2010. A randomized controlled trial of local heat therapy versus intravenous sodium stibogluconate for the treatment of cutaneous *Leishmania major* infection. *PLoS Negl Trop Dis* 4: e628.
22. Lobo IM, Soares MB, Correia TM, de Freitas LA, Oliveira MI, Nakatani M, Badaro R, David JR, 2006. Heat therapy for cutaneous leishmaniasis elicits a systemic cytokine response similar to that of antimonial (Glucantime) therapy. *Trans R Soc Trop Med Hyg* 100: 642–649.
23. Sadeghian G, Nilfroushzadeh MA, Iraj F, 2007. Efficacy of local heat therapy by radiofrequency in the treatment of cutaneous leishmaniasis, compared with intralesional injection of meglumine antimoniate. *Clin Exp Dermatol* 32: 371–374.
24. Velasco-Castrejon O, Walton BC, Rivas-Sanchez B, Garcia MF, Lazaro GJ, Hobart O, Roldan S, Floriani-Verdugo J, Munguia-Saldana A, Berzaluce R, 1997. Treatment of cutaneous leishmaniasis with localized current field (radio frequency) in Tabasco, Mexico. *Am J Trop Med Hyg* 57: 309–312.
25. Prasad N, Ghiya BC, Bumb RA, Kaushal H, Saboskar AA, Lezama-Davila CM, Salotra P, Satoskar AR, 2011. Heat, Oriental sore, and HIV. *Lancet* 377: 610.
26. Bumb RA, Satoskar AR, 2011. Radiofrequency-induced heat therapy as first-line treatment for cutaneous leishmaniasis. *Expert Rev Anti Infect Ther* 9: 623–625.
27. Ginel PJ, Lucena R, Lopez R, Molleda JM, 1998. Use of allopurinol for maintenance of remission in dogs with leishmaniasis. *J Small Anim Pract* 39: 271–274.
28. Manna L, Gravino AE, Picillo E, Decaro N, Buonavoglia C, 2008. *Leishmania* DNA quantification by real-time PCR in naturally infected dogs treated with miltefosine. *Ann N Y Acad Sci* 1149: 358–360.
29. Manna L, Vitale F, Reale S, Picillo E, Neglia G, Vescio F, Gravino AE, 2009. Study of efficacy of miltefosine and allopurinol in dogs with leishmaniasis. *Vet J* 182: 441–445.
30. Asilian A, Sadeghinia A, Faghihi G, Momeni A, Amini HA, 2003. The efficacy of treatment with intralesional meglumine antimoniate alone, compared with that of cryotherapy combined with the meglumine antimoniate or intralesional sodium stibogluconate, in the treatment of cutaneous leishmaniasis. *Ann Trop Med Parasitol* 97: 493–498.
31. Asilian A, Sadeghinia A, Faghihi G, Momeni A, 2004. Comparative study of the efficacy of combined cryotherapy and intralesional meglumine antimoniate (Glucantime) vs. cryotherapy and intralesional meglumine antimoniate (Glucantime) alone for the treatment of cutaneous leishmaniasis. *Int J Dermatol* 43: 281–283.
32. Gurei MS, Tatli N, Ozbilge H, Erel O, Seyrek A, Kocyigit A, Ulukanligil M, 2000. Efficacy of cryotherapy and intralesional pentostam in treatment of cutaneous leishmaniasis. *J Egypt Soc Parasitol* 30: 169–176.
33. Layegh P, Pezeshkpoor F, Soruri AH, Naviafar P, Moghiman T, 2009. Efficacy of cryotherapy versus intralesional meglumine antimoniate (glucantime) for treatment of cutaneous leishmaniasis in children. *Am J Trop Med Hyg* 80: 172–175.
34. Leibovici V, Aram H, 1986. Cryotherapy in acute cutaneous leishmaniasis. *Int J Dermatol* 25: 473–475.
35. el Darouti MA, al Rubaie SM, 1990. Cutaneous leishmaniasis. Treatment with combined cryotherapy and intralesional stibogluconate injection. *Int J Dermatol* 29: 56–59.
36. Mosleh IM, Geith E, Natsheh L, Schonian G, Abotteen N, Kharabsheh S, 2008. Efficacy of a weekly cryotherapy regimen to treat *Leishmania major* cutaneous leishmaniasis. *J Am Acad Dermatol* 58: 617–624.
37. Bassiouny A, El Meshad M, Talaat M, Kutty K, Metawaa B, 1982. Cryosurgery in cutaneous leishmaniasis. *Br J Dermatol* 107: 467–474.
38. Berman JD, Neva FA, 1981. Effect of temperature on multiplication of *Leishmania* amastigotes within human monocyte-derived macrophages *in vitro*. *Am J Trop Med Hyg* 30: 318–321.
39. Willard RJ, Jeffcoat AM, Benson PM, Walsh DS, 2005. Cutaneous leishmaniasis in soldiers from Fort Campbell, Kentucky returning from Operation Iraqi Freedom highlights diagnostic and therapeutic options. *J Am Acad Dermatol* 52: 977–987.
40. Navin TR, Arana BA, Arana FE, de Merida AM, Castillo AL, Pozuelos JL, 1990. Placebo-controlled clinical trial of meglumine antimonate (glucantime) vs. localized controlled heat in the treatment of cutaneous leishmaniasis in Guatemala. *Am J Trop Med Hyg* 42: 43–50.