# Comparison of Hyperbaric Oxygen and Dapsone Therapy for Loxosceles Envenomation

*Gregory D. Hobbs, MD, Alena R. Anderson, MD, Thomas J. Greene, MD, Donald M. Yealy, MD*

## ABSTRACT

**Objective:** To determine whether hyperbaric $O_2$ (HBO), dapsone, or HBO plus dapsone affects lesion size in a swine model of Loxosceles envenomation.

**Methods:** In a randomized controlled animal laboratory experiment, 32 piglets were assigned to 1 of 4 equal groups. Each piglet received 15 μL of purified venom intradermally on day zero. Group 1 received no treatment; group 2 received HBO at 2 atm for 2 hours on days 1–3; group 3 received 50 mg of dapsone orally on days 1–3; and group 4 received dapsone 50 mg orally and HBO at 2 atm for 2 hours on days 1–3. On days 1–7, 14, and 21, an investigator blinded to the treatment groups measured necrosis and induration. Mean necrosis and induration rates were compared using analysis of variance for repeated measures.

**Results:** Comparing groups on any day, no significant difference was noted in necrosis, induration, reduction in necrosis from day 1, or rate of change in lesion size from days 1–7. A difference was seen in the reduction of induration between all 3 treatment groups and the control group on days 7 and 14 only. The sample size permitted a power of 0.8 to detect a 12-mm mean change in lesion size.

**Conclusion:** Compared with the control, neither dapsone, HBO, nor the combination of dapsone and HBO reduced necrosis from Loxosceles envenomation on days 3–21. An increase was seen in the rate of reduction in induration between all 3 treatment groups and the control group on days 7–21. However, the magnitude of this effect was clinically insignificant. In this animal model, treatment with either dapsone or HBO or a combination offers little clinical benefit in Loxosceles envenomation.

**Key words:** *Loxosceles reclusa*, hyperbaric oxygen, dapsone, brown recluse spider, envenomation.

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Two species of the brown recluse spider, Loxosceles, are found in the United States. Both *L. reclusa* and *L. deserta*, found in the Southeast and the Southwest, respectively, produce necrotic skin lesions in humans. These spiders are reclusive and nocturnally active, hiding in closets, garages, or woodpiles. The spider bites when it is provoked, as when trapped or crushed against the skin.[1] The bite is usually painless. The spider often is crushed or escapes, making positive identification difficult.[2] Erythema and pain typically develop at the bite site within 2–6 hours. A small blister appears, which progresses to form a bluish-colored macule. The lesion then may progress to form a necrotic ulcer that can be up to 30 cm in diameter and take several months to heal. However, <10% of lesions progress to form these large ulcers, with most spontaneously regressing. Suggested treatments for Loxosceles envenomation include dapsone[3–5] and hyperbaric $O_2$ (HBO) therapy.[6]

No controlled human study has been published that evaluates these treatments in proven Loxosceles envenomation, although case series and retrospective data exist.[4,6] In the absence of a control group, reduction in lesion size may be secondary to either treatment effect or spontaneous healing. Most of the reported human series are based on presumed brown recluse envenomation, because the spider is rarely captured or identified. Because envenomation often produces insignificant lesions, many patients do not seek medical evaluation.[2] Another confounding variable in human trials is that in the United States, 6 species of spiders (including *L. reclusa*)[7] can produce necrotic lesions.

All published trials involving animal models of Loxosceles envenomation have used rabbits or rodents. The

*From the Scott & White Clinic and Memorial Hospital; Scott, Sherwood and Brindley Foundation; Texas A&M University Health Science Center, College of Medicine, Temple, TX, Department of Emergency Medicine (**GDH, ARA, TJG**); and the University of Pittsburgh School of Medicine, Pittsburgh, PA, Department of Emergency Medicine (**DMY**).*

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*Address for correspondence and reprints: Gregory D. Hobbs, MD, Scott & White Clinic, 2401 South 31st Street, Temple, TX 76508. Fax: 817-724-1044.*

**TABLE 1** Mean Necrosis/Induration for Swine*

| | Mean Necrosis (mm) (±SD) | | | | Mean Induration (mm) (±SD) | | | |
|---|---|---|---|---|---|---|---|---|
| | Control | Dapsone | HBO | Dapsone + HBO | Control | Dapsone | HBO | Dapsone + HBO |
| Day 3 | 11.1 (4.4) | 11.1 (6.5) | 9.9 (6.4) | 13.3 (5.3) | 30.1 (7.7) | 24.1 (8.1) | 25.0 (11.3) | 27.9 (6.4) |
| Day 7 | 11.1 (6.2) | 9.1 (6.2) | 5.8 (5.2) | 8.7 (7.4) | 31.4 (14.4) | 19.5 (12.1) | 17.0 (11.8) | 20.2 (12.3) |
| Day 14 | 7.1 (5.3) | 3.4 (2.9) | 2.7 (2.7) | 5.7 (4.6) | 18.9 (12.8) | 8.4 (9.6) | 7.1 (6.1) | 12.3 (10.7) |
| Day 21 | 4.7 (4.6) | 2.4 (1.9) | 1.5 (1.7) | 3.4 (4.6) | 8.9 (7.2) | 3.4 (2.8) | 1.8 (1.9) | 7.2 (9.2) |
| Peak | 13.0 | 16.9 | 11.4 | 17.2 | 40.7 | 32.9 | 29.6 | 35.7 |

*Time zero was at envenomation. HBO = hyperbaric $O_2$.

external validity of these data, particularly with respect to humans, is uncertain because of the skin dissimilarities between these species.[8,9] Swine skin is structurally more similar to human skin than that of these other species. We previously reported our swine model of Loxosceles envenomation.[10] The null hypothesis for the current study was that the use of either dapsone, HBO, or a combination of these therapies would offer no benefit in clinical wound healing after Loxosceles envenomation, using a swine model.

## METHODS

### Study Design

A randomized, controlled trial was performed comparing the effects of HBO, dapsone, and dapsone plus HBO on lesion size in a previously developed porcine model of Loxosceles envenomation. Necrosis was the primary outcome and induration was the secondary outcome. An investigator blinded to animal treatment group made all wound evaluations.

### Animal Subjects

Light-skinned Yorkshire piglets were obtained from a local breeder at 6 weeks of age. They were numbered with ear tattooing and randomized to 1 of 4 groups. They were housed until they reached 13 kg prior to beginning the experimental protocol. Previous experience with this model showed a high mortality rate with smaller piglets. The animals were housed in groups of 6 in open-air pens with cement flooring, which were cleaned twice daily. The study protocol was approved by the animal use committee.

### Experimental Protocol

*Loxosceles deserta* venom is available commercially in purified form[11] and produces lesions similar to those seen with *L. reclusa* envenomation.[3,12,13] Thirty-two weanling piglets were randomized to 1 of 4 equal groups. An area on the right flank then was prepped by clipping the hair and cleaning with povidone-iodine. Each animal was injected intradermally in the flank with 15 μL of venom on day 0. The venom was pooled and the same investigator performed all injections to reduce variation. Since all envenomations were not performed on the same day, the venom was frozen to maintain potency. The animals were not sedated for venom injection or during HBO treatment. No prophylaxis was given to prevent barotrauma.

Group 1 served as the control group and received no intervention. The other 3 groups started treatment 24 hours after envenomation. Group 2 received HBO in an animal research chamber at 2 atm for 2 hours daily for 3 consecutive days. Group 3 received 50 mg of oral dapsone, crushed and mixed in peanut butter, daily for 3 doses. Group 4 received both dapsone and HBO by the treatment regimens described above. No attempt was made to keep the animals from rubbing against the lesions, and no local wound care was provided.

### Measurements

An investigator blinded to the treatment groups measured necrosis and induration on days 1–7, 14, and 21. Necrosis was measured with a ruler using the greatest diameter of the necrotic area. Induration was defined as the area of firmness and swelling around the central necrotic lesion, and was measured with a ruler at the point of greatest diameter. At the end of the study, the piglets were sacrificed. No animal was followed past 21 days.

### Data Analysis

Necrosis and induration rates over this 21-day period after envenomation were compared using analysis of variance (ANOVA) for repeated measures. The comparison α error was set at 0.05. Comparisons were made between all treatment groups, and all treatment groups were compared with the control.

## RESULTS

All the animals developed typical necrotic lesions with surrounding induration and erythema. Table 1 shows the mean induration and necrosis for each group on days 3, 7, and 14 and the peak means for each group. Figure 1

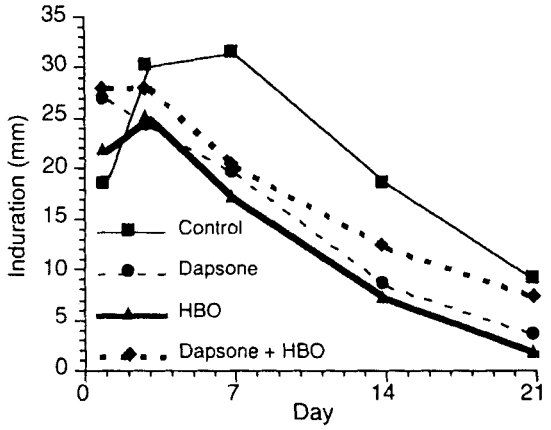


**FIGURE 1.** Mean induration for each of the groups. Time zero was at envenomation. HBO = hyperbaric $O_2$.

depicts each group's mean induration, while Figure 2 shows mean necrosis over the 21-day period. Comparison of the 4 groups on any day showed no significant difference in mean necrosis rates ($p = 0.13–0.17$), mean induration rates ($p = 0.07–0.38$), mean reductions in necrosis from day 1 ($p = 0.15–0.65$), or mean rates of change in lesion size from day 1 through day 7 ($p = 0.15$). A difference was noted in mean reduction of induration between all 3 treatment groups (but not among these groups themselves) and the control group on days 7 and 14 only ($p = 0.02$ and 0.01, respectively). The sample size permitted a power of 0.8 to detect a 12-mm mean change in lesion size, based on post-hoc calculations.

## DISCUSSION

The venom of the brown recluse spider is cytotoxic.[14] Purification of the venom yields 8 subcomponents. The primary dermonecrotic factor is a phospholipase, sphingomyelinase D.[15] The venom causes polymorphonuclear neutrophil (PMN) chemotaxis, platelet aggregation, and necrosis.[1,16] Inactivation of the complement system also occurs, with the venom attaching to the red blood cell membrane, causing hemolysis. A rare systemic reaction may occur, with several deaths reported, although none has occurred in the United States.[17] Systemic symptoms include massive hemolysis with disseminated intravascular coagulation, generalized petechial rash, thrombocytopenia, and hemolytic anemia.

Treatment of the cutaneous lesion involves local wound care, elevation, immobilization, observation, and close follow-up for management of secondary cellulitis and other complications. Dapsone has been thought to reduce skin necrosis by inhibiting PMN chemotaxis, thus limiting the inflammatory reaction. Although a number of case reports support this conclusion, there is no randomized or controlled human study demonstrating its usefulness. The recommended dose of dapsone is 50–200 mg/day.[3–5] We chose the lower dose because of the small size of the piglets in our study. Dapsone also has serious side effects such as hypersensitivity, hemolytic anemia in patients with G6PD deficiency, toxic hepatitis, and cholestatic jaundice.[18,19]

Hyperbaric $O_2$ also has been suggested as a potential therapy.[6,20] One proposed mechanism for the dermonecrotic action of Loxosceles venom is via rapid coagulation and occlusion within small capillaries.[21] HBO causes neovascularization in necrotic tissue, and also inactivates enzymes that contain sulfhydryl groups such as phospholipids in vitro.[20] However, HBO has its own negative features such as claustrophobia, barotrauma, and expense.

Mack,[22] using white rabbits as the animal model, showed that HBO had a significant effect in wound healing in histologic samples obtained 21 days after Loxosceles envenomation. However, Phillips et al.,[23] using the same model, showed no significant difference, either histologically or cutaneously, among animals treated with dapsone, HBO, or cyproheptadine. Our results support the latter study, with no cutaneous healing benefit of HBO or dapsone. We did not evaluate histologic changes.

The lack of a suitable animal model has limited Loxosceles research.[24] In developing an animal model, we sought to simulate actual acute human envenomation as closely as possible. While no other species has skin structures identical to that of humans, we believe that swine provide the best model currently available for research. The skins of both humans and swine have a sparse hair coat, a thick epidermis, a dermis that has a well-differentiated papillary body, and a large content of elastic tissue.[8] Both also share similar epidermal histologic characteristics and cell proliferation kinetics.[9] Even with these similarities, swine skin is distinct. Notable differences from human skin are poor vascularization

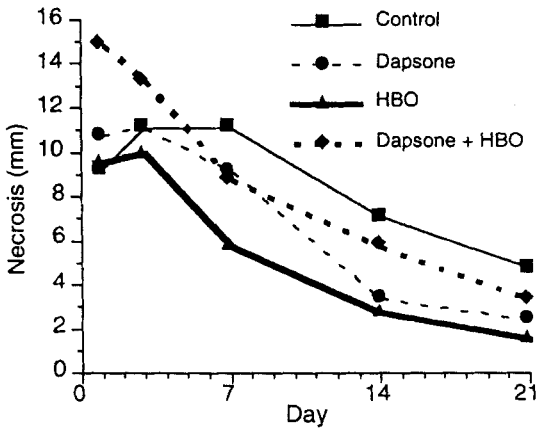


**FIGURE 2.** Mean necrosis for each of the groups. Time zero was at envenomation. Repeated-measures analysis of variance revealed no significant difference among the groups. HBO = hyperbaric $O_2$.

and slightly greater thickness of the dermis. Rabbit skin and rodent skin, in contrast, have a thick coat of fur, a thin epidermis and dermis, and a sparse subcutaneous layer with little elastic tissue. Given the difference between species, the applicability of any animal data to humans is unclear. However, the fact that both HBO and dapsone are of little benefit in 2 different animal models strengthens the validity of our results.[25] Our sample size provided a power of 0.8 to detect a 12-mm reduction in lesion size. This was equivalent to an all-or-nothing response given the size of the lesions obtained. Although our study lacked sufficient power to detect smaller changes in lesion size, we believe that smaller changes in lesion size are clinically unimportant.

We identified a difference in induration on days 7 and 14 when comparing all the treatment groups with the control group, but no difference was seen among the treatment groups. There is a strong correlation between induration and necrosis ($r = 0.78$). The clinical importance of induration is unknown in this setting. There may be increased discomfort associated with an increase in induration, but this is not possible to establish in a porcine model.

## LIMITATIONS AND FUTURE QUESTIONS

Our study has several limitations. We did not investigate histologic differences between the treatment groups and the controls. We measured the lesions using the greatest diameter since the lesions were primarily circular. Calculating the lesion area might have been more precise. Because we did not follow any animals past 21 days, the time course for wound healing in pigs is unknown, and may be different from that of humans. Our sample size and mean lesion size permitted detection of only an "all-or-nothing" response. A larger sample size, or the development of a model that produces consistently larger lesions, might detect a smaller change in lesion size. Dapsone and HBO treatment regimens were arbitrarily chosen based on common clinical practice. Future studies could examine more aggressive regimens.

## CONCLUSION

Using a previously reported swine model, we found no improvement in the reduction of lesion size with dapsone, HBO, or a combination of dapsone and HBO when compared with the control. Based on these results, as well as those reported in other recent studies, we recommend only routine wound care for Loxosceles envenomation.

## REFERENCES


1. Wilson DC, King LE Jr. Spiders and spider bites. Dermatol Clin. 1990; 8:277–86.
2. Berger RS. The unremarkable brown recluse spider bite. JAMA. 1973; 225:1109–11.
3. Rees R, Campbell D, Rieger E, King LE Jr. The diagnosis and treatment of brown recluse spider bites. Ann Emerg Med. 1987; 16:945–9.
4. King LE Jr, Rees RS. Dapsone treatment of a brown recluse bite. JAMA. 1983; 250:648.
5. Rees RS, Altenbern DP, Lynch JB, King LE Jr. Brown recluse spider bites: a comparison of early surgical excision versus dapsone and delayed surgical excision. Ann Surg. 1985; 202:659–63.
6. Svendsen FJ. Treatment of clinically diagnosed brown recluse spider bites with hyperbaric oxygen: a clinical observation. J Ark Med Soc. 1964; 68:768–73.
7. Anderson PC. Necrotizing spider bites. Am Fam Physician. 1982; 26:198–203.
8. Eaglestein WH, Mertz PM. Effects of topical medications on the rate of repair of superficial wounds. In: Dineen P (ed). The Surgical Wound. Philadelphia: Lea & Febiger, 1981, pp. 150–1.
9. Bustad LK, McClellan RO (eds). Swine in Biomedical Research. Seattle: Frayn Printing Co., 1966.
10. Hobbs GD, Yealy DM. Development of an acute cutaneous swine model for Loxosceles envenomation. Vet Hum Toxicol. 1994; 36: 298–300.
11. Phillips K. Spider man. Discover. 1991; 6:48–53.
12. Russell FE, Waldron WG, Madon MB. Bites by the brown spiders *Loxosceles unicolor* and *Loxosceles arizonica* in California and Arizona. Toxicon. 1969; 7:109–17.
13. Duffey PH, Limbacher HP. Brown spider bites in Arizona. Ariz Med. 1971; 28:89–95.
14. Smith CW, Micks DW. A comparative study of the venom and other components of three species of Loxosceles. Am J Trop Med Hyg. 1968; 17:651–6.
15. Rekow MA, Civello DJ, Geren CR. Enzymatic and hemolytic properties of brown recluse spider (*Loxosceles reclusa*) toxin and extracts of venom apparatus, cephalothorax and abdomen. Toxicon. 1983; 21:441–4.
16. Kurpiewski G, Forrester LJ, Barrett JT, Campbell BJ. Platelet aggregation and sphingomyelinase D activity of a purified toxin from the venom of *Loxosceles reclusa*. Biochem Biophys Acta. 1981; 678:467–76.
17. Wasserman GS, Anderson PC. Loxoscelism and necrotic arachnidism. J Toxicol Clin Toxicol. 1983–84; 21:451–72.
18. Hobbs GD, Harrell RE Jr. Brown recluse spider bites: a common cause of necrotic arachnidism. Am J Emerg Med. 1989; 7:309–12.
19. Wille RC, Morrow JD. Case report: dapsone hypersensitivity syndrome associated with treatment of the bite of a brown recluse spider. Am J Med Sci. 1988; 296:270–1.
20. Maynor ML, Abt JL, Osborne PD. Brown recluse spider bites: beneficial effects of hyperbaric oxygen. J Hyperbar Med. 1992; 7:89–102.
21. Berger RS, Adelstein EH, Anderson PC. Intravascular coagulation—the cause of necrotic arachnidism. J Invest Dermatol. 1973; 61:142–50.
22. Mack RB. The bite of the spider woman: *Loxosceles reclusa* (the brown recluse). N C Med J. 1992; 53:200–3.
23. Phillips S, Kohn M, Baker D, Vander Lest R, Gomez H, McKinney P. Therapy of brown spider envenomation: a controlled trial of hyperbaric oxygen, dapsone and cyproheptadine. Ann Emerg Med. 1995; 25:363–8.
24. Futrell JM. Loxoscelism. Am J Med Sci. 1992; 304:261–7.
25. Yealy DM. How much "significance" is significant: the transition from animal models to human trial in resuscitation research. Ann Emerg Med. 1993; 22:11–6.