Effects of topical application of taurolidine on second intention healing of experimentally induced wounds in rats

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Objective—To determine the macroscopic effects of topical application of taurolidine on second intention healing of experimentally induced wounds in rats.

Animals—32 adult Sprague-Dawley female rats.

Procedures—In each rat, 2 skin wounds were created in the lumbar area. Groups of 7 rats were assigned to have 1 wound treated topically with hydroxycellulose gel (HDCG), 2% taurolidine in HDCG (T-HDCG), 2% taurolidine–sodium citrate solution, or bacitracin-neomycin-polymyxin B ointment; the other wound was not treated. Four control rats (8 untreated wounds) were used. Wounds were monitored for contraction, epithelialization, and complete healing at 4, 8, and 14 days after wound creation. The number of days to complete healing was also recorded for each wound.

Results—Compared with other treatments or untreated wounds, wounds treated with T-HDCG had decreased total healing at day 8 and decreased epithelialization and decreased total healing at day 14. Wounds treated with T-HDCG required approximately 3 days longer to completely heal than all other treated and untreated wounds. Application of bacitracin-neomycin-polymyxin B ointment did not enhance wound healing. Mean time to complete healing of untreated wounds in all treatment and control groups was 10.00 to 10.14 days.

Conclusions and Clinical Relevance—In rats, topical application of T-HDCG to wounds had a negative effect on second intention healing by delaying the epithelialization process. In mammals, generally, wounds treated topically with taurolidine may need to be treated and monitored for a longer period than other wounds treated with other common wound-healing compounds or untreated wounds. (*Am J Vet Res* 2008;69:1210–1216)

Taurolidine (bis [1,1-dioxoperhydro-1,2,4-thiadiazinyl-4]-methane) is a synthetic derivative of the amino acid taurine. It consists of 2 aromatic rings that are connected with a CH_2 group.[1] Taurolidine has a broad-spectrum antimicrobial activity against gram-positive and gram-negative bacteria (both aerobic and anaerobic organisms) and activity against fungi.[2,3] Chemically reactive hydroxymethyl groups react with cell wall constituents of microbial pathogens that have become damaged. Cells lose their ability to undergo mitosis and lose their pathogenic and invasive features.[1,2] Emergence of bacterial resistance to taurolidine is not presently evident.[2] The hydroxymethyl groups of taurolidine can also react with the lipopolysaccharides of the endotoxins and polypeptides of exotoxins produced by bacteria, thereby diminishing their lethal effects.[1,4] In clinical settings, taurolidine has been administered via lavage after abdominal exploratory surgery in humans to reduce development of postoperative infections and adhesions and to treat peritonitis.[3] Because of its broad-spectrum antimicrobial activity, it is included in solutions used for flushing central venous catheters.[2,5]

Abbreviations

BNPB	Bacitracin-neomycin-polymyxin B
CI	Confidence interval
HDCG	Hydroxycellulose gel
T-HDCG	2% taurolidine in hydroxycellulose gel
T-SCS	2% taurolidine–sodium citrate solution

Additionally, taurolidine has been found to have antitumor activity by selectively inducing apoptosis.[3] Its activity against many tumors in vitro, against experimentally induced tumors in vivo, and against naturally occurring tumors has been identified.[1,3,6–11] Taurolidine is nontoxic to humans and other animals at therapeutic doses.[2] Because of its many attributes, several clinical applications may be found for this agent. Currently, the applications for its use that are being investigated often involve surgery. Potential applications include its use for treatment of septic peritonitis, infected open wounds, and osteomyelitis or as a local adjuvant to excision of tumors. However, it is unknown what effects taurolidine has on wound healing. If taurolidine is to be used in conjunction with surgery or to treat wounds, it seems important to know the effects taurolidine has on wound healing. The objective of the study of this report was to determine the macroscopic effects of topical ap-

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plication of taurolidine on second intention healing of experimentally induced wounds in rats.

Materials and Methods

Animals—Thirty-two adult Sprague-Dawley female rats weighing 250 to 300 g were included in the study. All procedures were approved by the Institutional Animal Care and Use Committee of the Oregon State University. The rats were premedicated with 0.015 mg of buprenorphine SC and administered carprofen (1.5 mg, SC); by use of an induction tank and facemask, anesthesia was induced and maintained at 1.25% to 2% isoflurane mixed with 100% oxygen. Five milliliters of physiologic saline (0.9% NaCl) solution was administered SC in the ventral abdominal region (so that it would not interfere with the surgical field). Physiologic saline (0.9% NaCl) solution was administered SC to maintain adequate hydration during surgery.

Surgical procedure—The surgical procedure was based on that used in a study[12] of cutaneous wound healing in cats. Each rat was placed in sternal recumbency. The dorsal right and left lateral aspects of the body were shaved with a clipper[a] from the level of the scapulae to the level of the pelvis. The surgical field was prepared with 4% chlorhexidine and physiologic saline solution. An 8-mm-diameter dermal biopsy punch[b] was used to create a full-thickness circular defect in the skin. A defect was created 1.5 cm lateral to the dorsal midline on each side at the level of the 13th (caudal-most) rib. Bleeding was stopped by applying gentle pressure on the wounds with sterile gauze squares until hemostasis was achieved. The day that the wounds were created was designated as day 0.

Experimental procedures—Rats were randomly allocated to 1 of 4 treatment groups (n = 7 rats each) or a control group (4 rats). For the rats in the treatment groups, 1 wound was randomly chosen to receive treatment and the other was not treated. Treatments were as follows: HDCG[c] alone, T-HDCG,[c] T-SCS (pH, 6.8),[d] or BNPB ointment.[e] Just enough gel, ointment, or solution was applied to cover the selected wound. The investigators remained unaware of the treatment groups that received HDCG alone or T-HDCG; the investigators were not blinded to treatment with BNPB ointment or T-SCS treatment. The tubes were labeled as A and B by the manufacturer, and the investigators did not know which contained the HDCG with and without taurolidine until the data were analyzed. Topical application of the appropriate treatment was performed the day the wounds were created, repeated daily for 4 consecutive days after surgery, and then performed every other day for as long as 12 days after surgery; treatment was discontinued earlier if the wound was completely healed. In all rats, the wound on the opposite side did not receive any topical treatment.

Postoperative procedures—For rats in the treatment and control groups, wounds were covered with a triple-layer bandage. The contact layer consisted of a nonadherent, semiocclusive pad[f] placed directly on the wound surface. The secondary layer was open-weave cotton roll gauze applied in several layers. The tertiary layer consisted of porous adhesive tape.[g] The secondary and tertiary layers were wrapped around 1 forelimb to prevent slipping of the bandage. Buprenorphine (0.05 mg/kg) and carprofen (5 mg/kg) were each administered SC once 12 hours after surgery.

Rats had their bandage changed daily for the first 5 days and then every other day until 19 days after surgery. If it appeared that the wound was almost completely healed, the bandage was changed daily to accurately record the number of days to complete healing. When changing a bandage for each rat, anesthesia was induced and maintained with 1% to 2% isoflurane mixed with 100% oxygen and delivered via a facemask. Anesthesia was required to prevent trauma to the wound sites or hampering of healing that might result from resistance to the bandage change and other procedures. The wounds were gently irrigated with physiologic saline solution to allow direct observation of the tissues without exudate covering the wound. Nineteen days after surgery, all rats were euthanatized with an overdose of pentobarbital injected intraperitoneally. No postmortem examination, including histologic examination of tissues, was performed.

Evaluation of wounds—Wound evaluation was similar to that used in a previous study[12] of cutaneous wound healing in cats. Visual inspection of wound sites was performed at the time of bandage changes. Information recorded from visual inspection of the wounds included amount and character of any wound fluid in the bandage, bandage state, and any appearance of wound-site infection or other abnormalities. The interval (time in days) to complete coverage of the wound by epithelium, wherein granulation could no longer be detected, was also noted; this was considered to be the time of complete healing of the wound. If a scab was present, it remained undisturbed until day 8 after wound creation. Thereafter, the scab was moistened with physiologic saline solution and was atraumatically peeled off with a 25-gauge hypodermic needle to allow visual inspection of the underlying tissues.

Planimetry was performed immediately after wounds were created on day 0 and on days 4, 8, and 14 during bandage change by the same 2 investigators (JMS and BS). By use of a 5X magnifying lamp, the perimeter of the wound was traced onto a sterile piece of clear acetate film with a fine-point indelible marking pen.[h] The acetate film was laid on the wound surface, smoothed flat, and kept immobile while the tracing was made by the examiner. Care was taken so that the pressure placed on the wound did not distort the wound edges during tracing. A tracing was made at the border between apparently normal skin and the wound, and the outlined area was considered the total wound area (**Figure 1**). Next, a tracing was made at the margin of the leading edge of the advancing epithelium. The area between the 2 trace lines was considered the area of epithelialization. The area within the margin of the advancing epithelium was considered the area of open or unhealed wound.

The areas outlined on the acetate films were digitized by use of a computer and digital scanning software.[i] The percentages of epithelialization, wound contraction, and total wound healing were calculated for

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each wound at each time point according to the following formulas:

$$\text{Percentage of epithelialization on day}_n = (\text{area of epithelium/total wound area}) \times 100$$

where day_n is the day of evaluation.

$$\text{Percentage of wound contraction on day}_n = (100 - [\text{total wound area on day}_n/\text{original wound area on day 0}]) \times 100$$

$$\text{Percentage of total wound healing on day}_n = (100 - [\text{open wound area on day}_n/\text{original wound area on day 0}]) \times 100$$

Data analysis—Number of wounds per group required to achieve statistical significance was derived by use of a formula, as follows:

$$\text{No. of wounds/group} = 4(zs)^2/E^2$$

where z is a multiplier representing the 97.5th percentile from a *t* distribution, s is the estimated sample SD, and E is the desired width of a 95% CI. If z = 2, s = 10, and E = 14, the number of wounds per group was calculated to be 8. We chose to have 7 rats in each treatment group and 4 in the control group; because each rat in the control group had 2 untreated wounds, this provided 8 wounds in the control group.

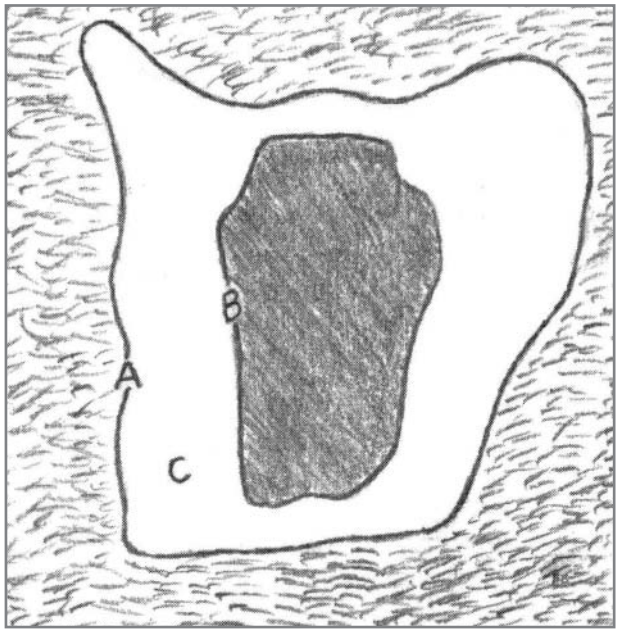

Figure 1—Illustration of the tracings of the wounds in the skin of rats made on an overlaid acetate film at various time points during a 14-day period. The tracings were used to calculate the percentage epithelialization, percentage wound contraction, and percentage total wound healing. A = Outer margin between apparently normal skin and wound epithelium. The area circumscribed by margin A is the total wound area. B = Margin between the wound epithelium and the open wound area. The area circumscribed by margin B is the open wound area. C = Area of wound epithelium (A minus B). (Adapted from Bohling MW, Henderson RA, Swaim SF, et al. Cutaneous wound healing in the cat: a macroscopic description and comparison with cutaneous wound healing in the dog. *Vet Surg* 2004;33:579–587. Reprinted with permission.)

Because each rat had 2 wounds (1 treated and 1 untreated), we analyzed within-rat differences for each of the measurements, namely the percentages of epithelialization, contraction, and total healing, and time to complete healing. At days 4, 8, and 14 after surgery (ie, when planimetry was performed), differences between percentages of epithelialization, contraction, or healing in treated and untreated wounds were assessed. In this way, the responses that were used for the statistical analysis were within-rat differences for each of these measurements. Positive percentage differences (values for treated wounds minus values for untreated wounds) in any of the active treatment groups suggested that that particular treatment was better than no treatment. Differences in these percentage differences among the treatment groups suggested that efficacies of the treatments themselves differed. The data for each of the 3 days were analyzed separately. Because the percentage wound contraction and percentage total wound healing were calculated from the original wound measurement (at day 0), the data at each time point (days 4, 8, and 14) represented healing from day 0 and not necessarily from the previous time point; therefore, for example, the percentage contraction and percentage total wound healing calculated at day 8 were not the amount of change since day 4 but were the amount of change from day 0.

In each analysis, we first assessed the correlation between responses (ie, percentage wound contraction, percentage healing, and percentage epithelialization) as well as the distributional nature of those responses (ie, normal or skewed distribution). Depending on these evaluations, different analytic approaches were used (these included 1-way ANOVA, paired *t* tests, and permutation tests). Percentages of contraction, epithelialization, and total healing and time to complete healing of the untreated wounds in the treatment groups and the control group were also compared; this allowed assessment of whether treatments had systemic effects on wound healing. Statistical analysis was performed by use of computer software,[j] and significance was set at a value of $P < 0.05$.

Results

In all rats, wounds produced minimal amounts of serous fluid. None of the wounds or rats developed any signs of infection throughout the study period. One rat in the control group died 4 days after wound creation; a full necropsy (including histologic examination of tissue specimens) was performed, and no cause of death could be determined. All other rats survived throughout the study period. For statistical analyses, data obtained from the 3 surviving rats in the control group were used. Mean percentages of epithelialization, contraction, and total healing of wounds in the treatment and control groups at the 3 time points at which planimetry was performed were calculated (**Table 1**).

Analysis of percentages of epithelialization, wound contraction, and total healing—On day 4, there was no epithelialization of any of the treated or untreated wounds in any of the rats. Because there was no wound epithelialization, percentage total wound

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Table 1—Mean ± SE percentages of epithelialization, contraction, and total healing of experimentally created wounds in rats at days 4, 8, and 14 after wound creation (day 0); wounds received no treatment (control group) or topical treatment with HDCG, T-HDCG, T-SCS, or BNPB ointment after creation daily for 4 days, and then every other day for up to 12 days. Two wounds were created in each rat. In the control group, neither wound was treated (of the 4 rats in this group, 1 died 4 days after surgery, and those data were not included in the analyses); in the treatment groups (n = 7 each), only 1 wound received treatment.

		Day 4		Day 8		Day 14	
Variable	**Group**	**Treated wounds**	**Untreated wounds**	**Treated wounds**	**Untreated wounds**	**Treated wounds**	**Untreated wounds**
Epithelialization (%)	HDCG	0	0	68.81 ± 2.85	72.15 ± 3.00	100	100
	T-HDCG	0	0	50.48 ± 2.80	67.44 ± 2.56	85.73 ± 7.18	100
	T-SCS	0	0	67.76 ± 4.44	62.62 ± 4.76	100	100
	BNPB	0	0	59.67 ± 2.43	76.38 ± 4.55	100	100
	Control	NA	0	NA	58.85 ± 5.76	NA	100
Contraction (%)	HDCG	45.99 ± 5.10	49.01 ± 7.34	39.52 ± 7.07	35.52 ± 12.10	86.45 ± 2.54	89.73 ± 1.93
	T-HDCG	27.25 ± 6.92	43.67 ± 5.85	–0.83 ± 10.44	38.05 ± 3.55	82.13 ± 3.24	88.89 ± 2.40
	T-SCS	43.70 ± 9.30	28.80 ± 7.74	38.49 ± 9.77	29.07 ± 7.80	89.36 ± 1.94	89.30 ± 1.92
	BNPB	52.32 ± 5.92	56.34 ± 4.11	42.39 ± 6.13	50.57 ± 8.71	87.75 ± 1.18	84.07 ± 2.29
	Control	NA	46.62 ± 2.57	NA	49.64 ± 3.52	NA	90.72 ± 3.14
Total wound healing (%)	HDCG	45.99 ± 5.10	49.01 ± 7.34	80.63 ± 2.74	81.57 ± 4.40	100	100
	T-HDCG	27.25 ± 6.92	43.67 ± 5.85	50.18 ± 5.89	79.82 ± 2.15	96.22 ± 1.95	100
	T-SCS	43.70 ± 9.30	28.80 ± 7.74	78.90 ± 4.83	73.85 ± 4.11	100	100
	BNPB	52.32 ± 5.92	56.34 ± 4.11	76.23 ± 3.43	87.48 ± 3.83	100	100
	Control	NA	46.62 ± 2.57	NA	79.15 ± .04	NA	100

NA = Not applicable.

Table 2—Mean ± SE differences in percentages of epithelialization, contraction, and total healing between treated and untreated wounds (1 of each in each rat) in 7 rats in each of the 4 treatment groups on day 8 after wound creation (day 0).

	Treatment group			
Variable	**HDCG**	**T-HDCG**	**T-SCS**	**BNPB**
Epithelialization (%)	–3.34 ± 4.27	–16.95 ± 4.39	5.14 ± 7.98	–16.69 ± 4.39
Contraction (%)	4.00 ± 12.35	–38.88 ± 8.92	9.42 ± 12.64	–8.18 ± 10.92
Total wound healing (%)	–0.94 ± 4.32	–29.65 ± 5.28*	5.06 ± 8.01	–11.25 ± 5.75

*Value significant at the 0.05 familywise level (used to account for the multiple comparisons or simultaneous inferences being made).[13]

healing was therefore equivalent to percentage wound contraction at this time point. Differences in mean ± SE percentage total healing (and thus differences in mean percentage contraction as well) between the treated and untreated wounds in rats in the HDCG, T-HDCG, T-SCS, and BNPB ointment treatment groups were –3.01 ± 8.14%, –16.42 ± 9.82%, 14.90 ± 11.26%, and –4.03 ± 6.95%, respectively.

None of these mean differences provided evidence of differences between treated and untreated wounds in any of the 4 treatment groups ($P > 0.1$ for all paired t tests). A 1-way ANOVA provided no evidence of differences in the mean wound healing differences across the 4 treatment groups ($P = 0.2$). Therefore, at day 4 after wound creation, there was no evidence that any of the treatments were better or worse than any other in terms of wound contraction or healing. Also, there were no differences in percentages of epithelialization, wound contraction, and total healing of the untreated wounds among the 4 treatment groups and the control group (ANOVA; $P = 0.06$).

On day 8, both the difference in percentage contraction and the difference in percentage epithelialization were strongly (positively) correlated with the difference in percentage healing (sample correlations, $r = 0.77$ and $r = 0.88$, respectively). Mean percentage differences of epithelialization, contraction, and total healing between the treated and untreated wounds for the 4 treatment groups were calculated (**Table 2**).[13] The only significant ($P = 0.001$) mean percentage difference identified was total wound healing in the T-HDCG treatment group.

On examination of the differences among the treatment groups, only ANOVA results for the difference in percentage healing were reported because the other 2 responses were highly correlated with healing. A 1-way ANOVA provided strong evidence of a difference among the treatment groups ($P = 0.003$). Specifically, the mean difference in percentage healing in the HDCG treatment group was estimated to be 28.7% higher than that in the T-HDCG treatment group (95% CI derived by use of Tukey method for multiple comparisons, 4.3% to 53.1%). In addition, the mean percentage healing difference in the T-SCS treatment group was estimated to be 34.7% higher than the mean percentage healing difference in the T-HDCG treatment group (a significant difference by use of the Tukey method; 95% CI, 10.3% to 59.1%). No other pairwise comparisons between treatment groups indicated significant differences. Therefore, there was evidence that the T-HDCG treat-

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ment did not compare favorably with either the HDCG or T-SCS treatments for wound healing (ANOVA; P = 0.003). There was also evidence that the T-HDCG treatment was less effective in wound healing than no treatment, as applied to the control group (paired t test; P = 0.001). As for the untreated wounds, there were no differences in percentages of epithelialization, wound contraction, and total healing among the 4 treatment groups and the control group (ANOVA; P = 0.15).

On day 14, all treated and untreated wounds were healed in all treatment and control groups except for certain wounds treated with T-HDCG. In the T-HDCG group, 3 treated wounds still had exposed granulation tissue on day 14. Therefore, the mean percentage differences of epithelialization and total wound healing were 0 for all groups except the T-HDCG treatment group. The mean ± SE percentage differences of epithelialization and total wound healing for the T-HDCG treatment group were –14.27 ± 7.18% and –3.78 ± 1.95%, respectively. The mean percentage contraction differences for the HDCG, T-HDCG, T-SCS, and BNPB ointment treatment groups were –3.29 ± 3.51%, –6.77 ± 3.97%, 0.07 ± 3.35%, and 3.69 ± 2.63%, respectively. By use of a permutation test, we determined that the chance that the 3 wounds with exposed granulation tissue in the T-HDCG treatment group would all be on the treated side was small ($P < 0.001$), suggesting that the T-HDCG treatment did detrimentally affect wound healing at day 14. A similar evaluation suggested that the T-HDCG treatment had a significant negative impact on epithelialization (permutation test; $P < 0.001$). Also, as determined by use of a permutation test, it was unlikely ($P < 0.001$) that all of the nonzero differences in healing and epithelialization would appear in the T-HDCG treatment group only. This indicated that the T-HDCG treatment was significantly different from the other treatments. All treatment groups had nonzero differences in percentage contraction at day 14, and standard ANOVA tools were used for analysis of that response. There were no significant (ANOVA; P = 0.25) differences in the differences in percentage contraction among the 4 treatment groups. There were also no significant (paired t tests; all $P > 0.10$) differences in percentage contraction of the untreated and treated wounds in all treatment groups. As well, there were no differences (ANOVA; P = 0.25) in percentage wound contraction, epithelialization, and total healing of the untreated wounds among the 4 treatment groups and the control group.

Number of days to complete healing—The mean number of days to complete healing in the HDCG treatment group for treated and untreated wounds was 10.14 (range, 10 to 11) and 10 (all wounds were healed at 10 days), respectively. The mean number of days to complete healing in the T-HDCG treatment group for treated and untreated wounds was 13.43 (range, 10 to 16) and 10 (all wounds were healed at 10 days), respectively. The mean number of days to complete healing in the T-SCS treatment group for treated and untreated wounds was 10 (all wounds were healed at 10 days) and 10.14 (range, 10 to 11), respectively. The mean number of days to complete healing in the BNPB ointment treatment group for treated and untreated wounds was 10.29 (range, 10 to 11) and 10.14 (range, 10 to 11), respectively. The number of days to complete healing in the control group was 10 days for all wounds. Treated wounds in the T-HDCG group required significantly (P = 0.01) more days to complete healing.

Discussion

The objective of the present study was to macroscopically evaluate the effect of topical application of taurolidine on second intention healing of experimentally induced wounds in rats. Wound healing has been described by a classification system that involves stages of inflammation, debridement, repair, and maturation.[14] Epithelialization occurs during the repair phase. In open wounds, epithelialization occurs only after a granulation bed is formed.[14,15] Typically, a period of 4 to 5 days will elapse before epithelial cells begin to migrate over granulation tissue.[14] A full-thickness open wound may or may not have a scab associated with the healing process. A scab is the desiccated wound surface that includes clotted blood, fibrin, collagen, and trapped debris.[14] Nontoxic topically applied agents can enhance epithelialization because of their moisture contents.[14] In the present study, this benefit was not evident because none of the topical treatments significantly enhanced wound healing, compared with healing of untreated wounds in the rats included in the treatment or control groups. Application of T-HDCG, however, did delay wound healing. The difference came from a delay in the epithelialization process. The 2 possible explanations are that taurolidine inhibited the proliferation of epithelial cells or inhibited the formation of granulation tissue; the latter would delay migration of the epithelial cells because that process requires granulation tissue as a supporting surface. In 1 study,[3] taurolidine inhibited the proliferation of murine fibroblasts in vitro; the concentration required to inhibit the proliferation of 50% of the cells (ie, IC_{50}) for those cells was 11.9 ± 1.8μM. Interestingly, in 1 experiment in that study,[3] in which murine fibroblasts were incubated with taurolidine for 3 days, the fibroblasts resumed proliferation within 1 week after taurolidine removal. It is possible that taurolidine inhibited proliferation, at least temporarily, of fibroblasts in the rats of the present study, which would have delayed the generation of the granulation tissue and hence interfered with the ability of epithelial cells to migrate. However, subjectively, there was no difference in the appearance of open wounds in any group over time in terms of granulation tissue coverage. Granulation of the wounds appeared to progress at the same rate, but it can be quite difficult to grossly compare the healing of open wounds.[16] Wounds treated with taurolidine contracted at the same rate as other wounds in our study. Given that fibroblasts are responsible for wound contraction,[17] this suggests that fibroblast function was not affected by taurolidine. Microscopic evaluation of wound healing with and without taurolidine treatment is needed to investigate this speculation more definitively, but for now, we conclude that taurolidine delayed wound healing via inhibition of epithelialization.

Application of the T-SCS did not delay wound healing, unlike the effect of T-HDCG treatment. In both preparations, 2% taurolidine was used. The most likely

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explanation for the delayed healing associated with 1 preparation and not the other is related to the physical properties of the respective vehicle. The T-HDCG was a gel formulation. Pharmaceutically, hydroxycellulose is used as a thickener, stabilizer, and dispersing agent in creams, jellies, ointments, and lotions.[18] It is nontoxic when used externally and is often used as a vehicle for drugs.[19,20] The findings of the present study did confirm that HDCG does not negatively affect wound healing. The T-SCS preparation was an aqueous solution. When applied to a wound, the solution remained as a drop on the surface. It is suspected the T-SCS was mostly absorbed through the nonadhesive pad, which dramatically decreased the amount of taurolidine that penetrated into the wound. The gel preparation was easily applied and remained in the wound bed. We speculate that the taurolidine in the gel formulation was absorbed less into the nonadhesive pad, which therefore allowed the taurolidine to penetrate the wound.

The authors were able to find only 1 report of a previous study[21] in which the effects of taurolidine on healing were evaluated. In that study, taurolidine was prepared as a peritoneal lavage solution and its use increased fibroblastic activity, tissue hydroxyproline concentration, and bursting pressure of primary colonic anastomoses in rats. However, in that study,[21] secondary peritonitis was induced in the rats before the anastomosis was performed and the concentration of taurolidine was 0.5%.[21] Nevertheless, the findings of that experiment are seemingly in contradiction to those of the present study, which indicated that taurolidine inhibited wound healing. There are 2 likely reasons why taurolidine improved wound healing of the colon tissues in the other study. First, the antimicrobial properties of taurolidine most likely resulted in a healthier local environment within the peritoneal cavity of treated rats, thereby promoting wound healing.[21] In our study, however, there were no signs of wound infection in any rat; thus, the antimicrobial benefit of taurolidine treatment could not have influenced outcome. Second, the concentration of taurolidine was only 0.5% in the previous study. It is possible that at a lower concentration than that used in our study, taurolidine would not delay wound healing yet still maintain its beneficial biological (eg, antimicrobial and tumoricidal) properties. As an analogous example, chlorhexidine at concentrations of 1.25 to 5μM does not impair fibroblast proliferation and maintains its bactericidal activity; however, at higher concentrations, chlorhexidine has toxic effects on fibroblasts.[22]

Topical application of BNPB ointment was investigated in our study because it is reported to enhance second intention wound healing.[16] Compared with untreated wounds, treatment with BNPB ointment increases the rate of reepithelialization by 25%, and this beneficial effect is not attributable to its antimicrobial action.[16] In the event that taurolidine would improve wound healing, our study was designed to compare taurolidine treatment with another topical treatment known to improve wound healing. However, data obtained in our study did not indicate that application of BNPB ointment improved wound healing. There are several possible explanations for those conflicting results. In the study in which treatment of wounds with BNPB ointment resulted in improved epithelialization, a brand-name preparation[k] was used, whereas a generic product was used in our study. This difference may account for the different results. For example, the petrolatum used as the carrier in the brand-name preparation has a lower melting point than the United States Pharmacopeia petrolatum, and differences in healing properties between the 2 have been noted.[16] Two other important differences are that the study to evaluate the brand-name preparation was carried out in pigs and the investigators created partial-thickness wounds.[16] Healing of partial-thickness wounds differs from that of full-thickness wounds,[14] and there is considerable heterogeneity in wound healing among species.[12]

Among the treatment and control groups of the present study, there was no difference in any of the variables measured for wound healing of the untreated wounds. These results indicated that there was no systemic effect of these topically applied treatments on wound healing and justified our study design involving comparison of treated and untreated wounds in the same individual rats.

For our study, we elected to experimentally create full-thickness wounds in rats. By use of a dermal biopsy punch instrument, repeatability and consistency among wounds were ensured. Also, by virtue of their dorsal location, the wounds were away from the ground surface, thereby unlikely to be rubbed on the ground during movement of the rats, which could have affected wound healing.

In experimentally created wounds in rats, topical application of T-HDCG delayed second intention healing. Future studies including microscopic evaluations of the wounds are necessary to better understand the mechanisms by which taurolidine can exert this effect and to determine the effect of taurolidine concentration on wound healing. If a surgeon elects to use this product for other proven effects, such as antibacterial properties, it is important to expect and therefore prepare for delayed wound healing at the site of drug application. Also, on the basis of the findings of our study, there was no apparent advantage in wound healing for any of the agents evaluated, when compared with healing of wounds without topical treatment in a control group of rats.

a. Model BTF, Andis, Sturtevant, Wis.
b. Miltex, York, Pa.
c. Provided by Vet Norfolk Products, Skokie, Ill.
d. TCS, Access Technologies, Skokie, Ill.
e. G&W Laboratories, South Plainfield, NJ.
f. Telfa pad, Kendall, Chicopee, Mass.
g. Elastikon, Johnson & Johnson, Skillman, NJ.
h. Sharpie (ultra-fine point), Sanford LP, Oak Brook, Ill.
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