Effect of Aquaphor Ointment on Wound Healing

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The effect of Aquaphor and petrolatum ointments on the healing of open granulating wounds in rats was studied. Wounds were observed up to ten days after wounding. The rate of wound contraction and epithelization was measured. Aquaphor- and petrolatum-treated wounds were significantly smaller by day ten. The rate of epithelization was retarded in treated wounds observed six days after wounding, but returned to normal by day ten.

Research in wound healing has been concerned primarily with one of three clinical objectives. The first involves the search for agents that could improve the end result of the healing process. So far the search for such a material has been unsuccessful. The second concerns the search for treatments that could accelerate the early phases of the healing process under normal conditions. In our laboratory parathyroid hormone and Aquaphor ointment have been demonstrated to speed the onset of wound rupture strength of 7-day-old wounds. These are only two examples of agents that have been found to affect the early phases of the healing process. The third general area of research concerns the search for drugs that will offset the retarding effects of certain medicines that must be used in the treatment of patients with systemic disease, for example, the problem of delayed healing in patients subjected to prolonged cortisone therapy. Recent study has shown that topical vitamin A can reverse the inhibitory effect of cortisone on the healing of open wounds in rabbits and man.[1]

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The purpose of this study was to continue the search for agents that could influence the early phases of healing under normal conditions. As mentioned earlier, Aquaphor ointment was found to speed the onset of rupture strength in wound healing by primary intention.[2] After wounding, the wounds were coated with ointment three times per day. Seven days after wounding the animals were killed and the rupture strength of the wounds was determined using an Instron Tensiometer. The ointment-treated wounds showed a mean increase in rupture strength of 61%. In the present study the effect of Aquaphor ointment on the healing of an open granulating wound was studied.

Materials and Methods

Thirty-six Sprague-Dawley rats that weighed from 200 to 230 gm each were randomly distributed into three equal groups and caged individually. The dorsal hair was shaved and a 2 cm diameter circle was imprinted on the skin below the scapulae at the midline. With the use of surgical scissors, the circle of skin was removed; this left an open wound that extended down to the muscles of the back. All 36 rats were wounded in a similar manner. On the day after wounding the three groups were treated as follows: group 1: about 0.5 gm of Aquaphor ointment* applied to the wound surface three times per day; group 2: about 0.5 gm of petrolatum applied in the same manner as in group 1; group 3: wounds were not treated and served as nontreated controls.

On days two, four, six, and eight after

* Aquaphor absorbent ointment base, Duke Laboratories, South Norwalk, Conn.

wounding, photographs* were taken at a standard distance from the wound. These slides then were projected on a screen and the outline of the wound was traced onto tracing paper. The wound was cut out and the paper representing the wound surface area was weighed with a Metler balance.

On day six, half of the rats from each group were weighed and killed. Sections through the midportion of the wound were collected and fixed in buffered Formalin. Histologic sections were prepared using a routine paraffin embedding technique and the sections were stained with hematoxylin and eosin.

On day ten, the remaining rats were weighed and killed and histologic sections were prepared from the wounds.

The histologic slides were projected onto a screen and the wound was outlined on tracing paper. The amount of reepithelization also was indicated on the tracing paper. Then with the use of a map measurer the length of the cross section of the wound surface at its greatest width and the amount of reepithelization at both ends of the wound were measured. By dividing the total length of the wound into the length covered by "new" epithelium the percentage of reepithelization of the wound was estimated.

All data were tested for statistical differences using the Wilcoxin ranking method. Differences were considered significant only when the P value was 0.05 or less.

Results

Rats treated with Aquaphor and petrolatum did not gain weight as rapidly as the control rats during the first six days after wounding. However, by the end of the experiment, no significant difference was found among the mean group animal weights (Table 1).

Based on the findings of the projection-tracing technique used to estimate the surface area of the wounds from the photographs, we found that both the Aquaphor- and petrolatum-treated wounds were larger than the control wounds up to day four. But by day six the treated wounds were significantly smaller than the control wounds (Table 2).

Data from the histologic projection technique revealed that treated wounds were significantly smaller than control wounds on day ten (Table 3). Based on the technique used to estimate the percentage of wound covered by epithelium, we found that only about 5% of the treated wound was reepithelized compared with about 24% of the control wound on day six. By day ten about 30% of the surface of all wounds had been reepithelized (Table 4).

* Kodachrome, Eastman Kodak Co., Rochester, NY.

TABLE 1
ANIMAL WEIGHT IN GRAMS

	Initial Weight	Day 6	Day 10
Aquaphor	215	237	267
Petrolatum	216	234	273
Control	217	252	276

TABLE 3
WIDTH OF TEN-DAY WOUNDS MEASURED FROM HISTOLOGIC SECTIONS USING A PROJECTION TECHNIQUE

	Length in Centimeters	Wound Length Compared with Controls
Aquaphor	5.2	−16%
Petrolatum	4.8	−23%
Control	6.2	...

TABLE 4
PORTION OF WOUND COVERED BY EPITHELIUM AS DETERMINED FROM HISTOLOGIC SECTIONS USING A PROJECTION TECHNIQUE

	Day 6 (%)	Day 10 (%)
Aquaphor	2	37
Petrolatum	5	25
Control	24	33

TABLE 2
PERCENTAGE DIFFERENCE IN WOUND SURFACE AREA AS DETERMINED FROM PHOTOGRAPHS

	Day 2 (%)	Day 4 (%)	Day 6 (%)	Day 8 (%)
Aquaphor	+23	+ 7	−15	−33
Petrolatum	+19	+17	−18	−41
Control	...	...	...	...

Note: Control wounds were the standard for comparison.

Discussion

This study was concerned with the effects of Aquaphor ointment, an absorbent ointment base obtained from wool fat, on the healing of open wounds. Ten-day-old wounds treated with the ointment were significantly smaller than control wounds. Because of the possibility that the ointment contains an active biologic agent, petrollatum was included in the study to serve as an ointment control because it contains no active ingredients. Petrolatum-treated wounds also showed about the same magnitude of reduction in size ten days after wounding as the Aquaphor-treated wounds. Based on these findings, we concluded that the Aquaphor effect was not due to an active biologic agent but was in some way related to the ointment serving as a "protective coat" on the surface of the wound. The petrolatum also had a similar function.

The processes involved in wound contraction are not well understood at the present time. Therefore, it is difficult to speculate on how the treatment of open wounds with an ointment base would effect wound contraction. The ointment may reduce surface contamination, protect the wound surface from air contact, or aid in retention of a more moist wound scab.

It was interesting that during the earlier stages of healing, days two to six, the treated wounds were larger than control wounds and reepithelization was retarded. However, by day ten all wounds revealed about the same amounts of reepithelization and the treated wounds were significantly smaller than control wounds.

The results of this study were similar to those reported in another paper[2] about the effect of Aquaphor ointment on the rupture strength of wounds healing by primary intention. The ointment-treated wounds showed a significant increase in rupture strength. The explanation for the ointment effect on the healing of closed and open wounds is not clear at the present time.

Conclusions

An experiment was performed to test the effects of topical application of Aquaphor ointment on the healing of open granulating wounds. Rats were wounded and the rate of healing was evaluated on days two, four, six, eight, and ten after wounding. Wounds were evaluated by measuring the rate of wound closure and reepithelization using a projection-tracing technique from photographs and histologic sections of the wounds. Aquaphor-treated wounds were significantly smaller at ten days after wounding than control wounds. No significant effect was noted on the reepithelization of the Aquaphor-treated wounds at ten days, however, epithelization was retarded up to day six. The ointment appears to act as a physical covering agent.

References

1. HUNT, T.K.; EHRLICH, H.P.; GARCIA, J.A.; and DUNPHY, J.E.: Effect of Vitamin A on Reversing the Inhibitory Effect of Cortisone on Healing of Open Wounds in Animals and Man, *Ann Surg* **170:**633-641, 1969.
2. LITTLE, J.W.; BICKLEY, H.C.; BICKLEY, C.; and KING, D.R.: Effects of Phytohemagglutinin, Dimethylsulfoxide and Aquaphor Ointment on Wound Healing, unpublished manuscript.
3. LITTLE, J.W.; BICKLEY, H.G.; and HINES, G.: The Effect of Parathyroid Extract on Wound Tensile Strength, *J Dent Res* **45:** 848-852, 1966.