ance of the time of contact between the host and organism prior to treatment in the development of immunity has been pointed out by Harrison(17) and Curnen and MacLeod (18). Although the production of serum precipitins and mouse protective antibodies in pneumococcal infections in man is not affected by the exhibition of sulfonamide or penicillin, there is some depression of antibody formation if animals are treated early(19). With delay in therapy or the use of a very large infecting inoculum immunity develops to a satisfactory level(19).

Although only a very small number of patients developed secondary streptococcal complications, there was no correlation between the appearance of clinical or bacteriologic relapse and the development of bacteriostatic antibody. In one case only was there an indication that a suppurative sequela may have influenced the degree of antibacterial immunity.

The inhibition of the development of bacteriostatic antibody in patients ill with streptococcal infections by penicillin therapy probably is of greater clinical importance than a similar depression of anti-streptolysin O, anti-streptokinase and anti-hyaluronidase. The antibacterial antibody protects against reinvasion by a homologous serological type; the other antibodies probably play little or no role in the defense against invasion by the organism. Certain schedules of penicillin treatment, therefore, actually depress the development of effective immunity and permit reinfection by the same serologic type of *Streptococcus* which caused the initial disease.

Conclusions. (1) The administration of penicillin to patients with streptococcal pharyngitis depresses the formation of type-specific antibacterial antibody. (2) There is a relation between the dose and route of administration of penicillin and the degree to which antibacterial activity develops. (3) Suppression of antibody formation by penicillin is probably due to rapid removal of the antigenic focus. (4) Failure to develop type-specific antibacterial antibody for the *Streptococcus* following penicillin therapy increases the risk of subsequent infection by homologous serological types.

17. Harrison, P. E., *J. Inf. Dis.*, 1946, v79, 101.
18. Curnen, E. C., and MacLeod, C. M., *J. Exp. Med.*, 1942, v75.
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Microwave Diathermy Treatment of Frostbite. (19008)

ROBERT B. LEWIS. (Introduced by Herman I. Chinn.)
From the Department of Pathology, USAF School of Aviation Medicine, Randolph Air Force Base, Randolph Field, Texas.

Several investigators(1-5) have demonstrated the beneficial effect of rapid thawing with warm water on frostbitten tissue with respect to necrosis.

The purpose of this investigation was to determine the effect of immediate rewarming of frostbitten tissue with microwave diathermy on the incidence and extent of necrosis.

Materials and methods. The experiments were performed on 78 male albino rabbits of more than 2,500 g body weight, of which 39 were treated with diathermy and the remainder served as controls. Two degrees of cold

1. Arev, T. Y., *Sovet. vrach. Zhur.*, 1939, v43, 391; Translated by E. Hope.
2. Fuhrman, F. A., and Crismon, J. M., *J. Clin. Invest.*, 1947, v26, 476.
3. Lempke, R. E., and Shumacker, H. B., Jr., *Yale J. Biol. and Med.*, 1949, v21, 321.
4. Finneran, J. C., and Shumacker, H. B., Jr., *Surg., Gynec. and Obst.*, 1950, v90, 430.
5. Pichotka, J., and Lewis, R. B., *U. S. Armed Forces Med. J.*, in press.

TABLE I. Results with Diathermy Treatment of Local Cold Injuries.

No. of animals	Exposure temp (°C)	Treatment	Survival time	Muscle necrosis (%)	Skin necrosis (cm^2)
12		Diathermy		1.3 ± .5	0
—	−12°	—	4 wk	—	—
10		Controls		5 ± 1.3	0
25		Diathermy		45.7 ± 4.4	4.6 ± 1.5
—	−15°	—	8-9 days	—	—
24		Controls		61.9 ± 4	12.3 ± 2.1

exposure were used, —12°C for 30 minutes for the determination of muscle necrosis and —15°C for 30 minutes for the determination of muscle and skin necrosis. The exact procedure is given elsewhere(6). Muscle necrosis was determined in per cent and skin necrosis in square centimeters as described previously (7). After exposure to cold, the frozen legs were immediately treated with microwave diathermy* (wave length 12.2 cm in the 2,400 to 2,500 megacycle band) in the following manner: The corner reflector-type director was employed so that the 6-cm electrode was 1½ inches above the skin and parallel to the long axis of the leg which was laid flat on a table with the external surface up. At this distance 6 to 8 square inches receive 50% or more of the heat output, according to the manufacturer, which is more than the lateral surface area of the leg from ankle to knee. All exposures were for 20 minutes with a power setting of 40%. The tendency for the microwave energy to concentrate in the small areas of the body being treated resulted in burning of the ankle with the first trials. To avoid this mishap, the shape of the leg was transformed approximately to that of a cylinder by wrapping a single sheet of muscle from a rabbit's abdominal wall around the distal half of the leg and tying it in place. Except for rewarming all animals received similar care. The legs were covered with 3% sulfamylon ointment and sterile dressings applied which were changed daily. The animals exposed to —12°C were sacrificed after 4 weeks and those exposed to —15°C after 8 to 9 days.

The average results are given with the standard deviation of the mean.

For the statistical analysis, a nonparametric test of significance was used which gives a statistic which is distributed approximately as Student's "t"(8).

Results. Twenty-four rabbits had one hind leg exposed to an alcohol bath at —12°C for 30 minutes for the determination of the effect of diathermy on the extent of muscle necrosis. Twelve were treated with short waves as described above, and 12 were allowed to thaw at room temperature in air. Two control animals died before the extent of necrosis was apparent. Two control and 6 diathermy-treated animals escaped muscle necrosis but were included in the calculations. The results are given in Table I. The lesser muscle necrosis obtained in the diathermy-treated animals is statistically significant at the 1% level ($P < .01$ with 20 degrees of freedom).

For the determination of the extent of skin and muscle necrosis, another series of 54 rabbits was exposed to —15°C for 30 minutes. Twenty-seven were immediately rewarmed with diathermy, and 27 were thawed at room temperature in air. Two treated and 3 control animals died before the extent of necrosis was apparent. The extent of muscle and skin necrosis is given in the table. The difference in the extent of skin necrosis in the treated and untreated animals is significant ($P = .01$ with 47 degrees of freedom). Five control and 12 diathermy-treated animals did not develop skin necrosis, but were included in the computation.

The extent of muscle necrosis also showed

6. Pichotka, J., and Lewis, R. B., PROC. SOC. EXP. BIOL. AND MED., 1949, v72, 130.

7. Pichotka, J., Lewis, R. B., and Luft, U. C., *Texas Rep. Biol. and Med.*, 1951, v9, 601.

8. Pitman, E. J., *Suppl. J. Roy. Stat. Soc.*, 1937, v4, 119.

* Raytheon Manufacturing Co., Model CMD-5.

a significant decrease in the diathermy-treated rabbits. ($P = .01$ with 47 degrees of freedom.) All animals in the treated and control groups showed muscle necrosis.

Discussion. Rapid rewarming of frostbitten rabbit legs with diathermy resulted in less extensive necrosis of skin and muscle in comparison to slow thawing in air at room temperature.

The manner in which rapid rewarming of frostbitten tissue is effective in reducing the incidence and extent of necrosis is controversial. We agree with Crismon and Fuhrman (9) that the reduction in the extent of necrosis obtained by rapid rewarming must be due to some action other than vasodilatation. These investigators pointed out that the results from rapid rewarming of frozen rabbit ears were superior to those obtained by stellate ganglion block. The poor results obtained by Finnerean and Shumacker(4) by denervating frostbitten rabbit ears and their reported good results from rapid rewarming suggest the same conclusion. However, Lempke and Shumacker(3) hypothesized that the beneficial results in frozen mice tails obtained by rapid rewarming plus the vasodilatory effect of either tetra-ethyl-ammonium chloride† or sympathectomy are due to the prevention of tissue ischemia and subsequent thrombosis.

It is difficult to interpret these beneficial results attributed to vasodilatation in the light of the favorable results reported by Crismon and Fuhrman(10) from the local application of plaster casts or pressure dressings to frostbitten rabbit feet.

The shortening of the exposure time to cold by the rapid rewarming may be a factor, although Fuhrman and Crismon(2) doubt this explanation since rewarming in water at 36°C did not produce beneficial results in contrast to rewarming at 42°C.

That some metabolic tissue process must be favorably influenced by the local application of heat to the frozen tissue seems to be the best explanation for the beneficial results.

Conclusions. Rapid rewarming of frostbitten rabbit legs by means of short wave diathermy after two standard cold injuries resulted in a decrease in the extent of muscle and skin necrosis in comparison to control frost-bitten animals thawed at room temperature in air.

† Etamon Chloride, Parke, Davis & Co.

9. Crismon, J. M., and Fuhrman, F. A., *J. Clin. Invest.*, 1947, v26, 468.

10. Crismon, J. M., and Fuhrman, F. A., *J. Clin. Invest.*, 1947, v26, 486.

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Inhibiting Effect of Inositol on Serum Cholesterol and Phospho-Lipids Following Cholesterol Feeding in Rabbits.*† (19009)

LOUIS B. DOTTI, WILLIAM C. FELCH, AND STEPHANIE J. ILKA.

From the Department of Laboratories and the Department of Medicine, St. Luke's Hospital, New York City.

The role that inositol may play in cholesterol metabolism and in atherosclerosis has not been determined. Herrmann(1) found that inositol lowered the cholesterol in serum, aorta, heart and liver of old hens and(2) in the serum of hypercholesteremic patients. We(3) have found a similar effect in hypercholesteremic diabetic patients, as have Leinwand and Moore(4). On the other hand, Lupton *et al.*(5) found no effect of inositol on

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1. Herrmann, G. R., PROC. SOC. EXP. BIOL. AND MED., 1946, v63, 436.

2. Herrmann, G. R., *Exp. Med. and Surg.*, 1947, v5, 149.

3. Felch, W. C., and Dotti, L. B., PROC. SOC. EXP. BIOL. AND MED., 1949, v72, 376.