

## CICATRIZATION OF WOUNDS.

### IX. INFLUENCE ON THE HEALING OF WOUNDS OF VARIATIONS IN THE OSMOTIC TENSION OF THE DRESSING.

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In previous articles it has been shown that the curve representing the process of cicatrization of an aseptic wound is geometric,<sup>1, 2</sup> and can be calculated by the formula of du Noüy. The comparison of the calculated and observed curves alters the study of the effect upon cicatrization of a substance applied to the surface of the wound. If the daily decrease of the area of a wound is known, and if the wound is maintained in a condition of surgical asepsis, the modifications in the rate of the healing process can be attributed to the special action of the substance applied, and the extent of this action can be accurately measured.

Up to the present it has not been known to what degree the rate of cicatrization of a wound can be affected by the dressing. Surgeons have studied the influence of so called healing substances when applied to wounds, but no precise conclusion has resulted from these observations. No method existed of measuring exactly the surface of a wound and of calculating with any degree of accuracy the rate of the healing process. On the other hand, the bacteriological condition of the wounds experimented upon was never taken into consideration. Such modifications of the rate of healing as were noticed might be equally attributable to the action of the substance employed upon the bacteria of the wound as to the tissues themselves.

<sup>1</sup> Carrel, A., and Hartmann, A., *J. Exp. Med.*, 1916, xxiv, 429.

<sup>2</sup> du Noüy, P. L., *J. Exp. Med.*, 1916, xxiv, 451.

The following experiments were undertaken to ascertain whether modifications in the osmotic tension of the dressing exert an influence on the rate of repair.

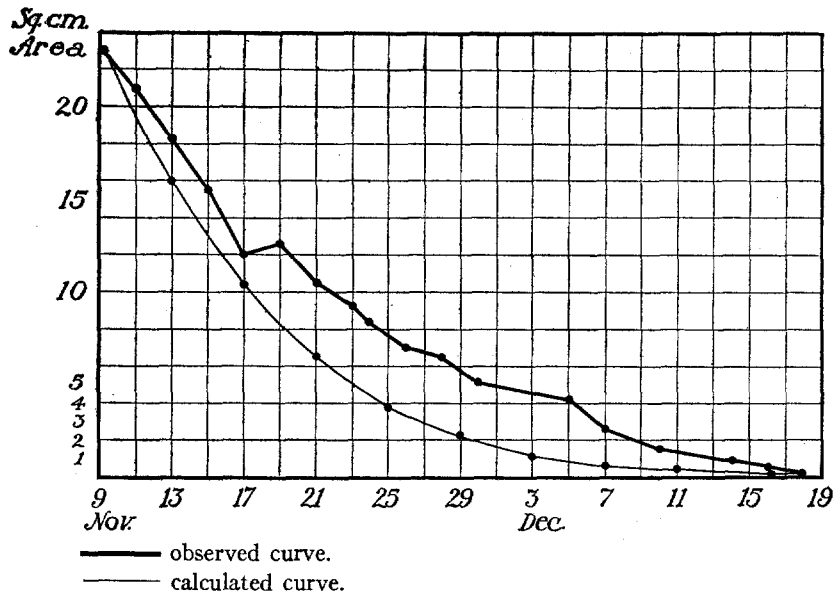
#### EXPERIMENTAL.

Surface wounds already covered with granulating tissue were selected. The normal rate of cicatrization of the wound was first obtained by sterilization with Dakin's hypochlorite solution or Daufresne's chloramine paste. The experiment was started as soon as the regular progress of the observed curve, as compared with the curve calculated according to du Noüy's formula, was established. The measurements of the wound and the plotting of the curve were made according to the technique previously described. Distilled water or hypertonic solution was applied to the surface of the wound by means of small perforated rubber tubes bound at the extremities and enveloped by a small cylindrical pad of Turkish toweling. The length of the pad varied from 4 to 8 cm., according to the size of the wound. Four threads placed transversely across the pad extended over the wound and were attached to the skin by means of small adhesive patches in such a manner as to keep the flushing tube in a fixed position over the granulations. The sterile liquid was contained in a flask placed about 1 meter above the patient's mattress, and reached the flushing tube by means of a Murphy drop tube. As a rule, in these experiments about 125 gm. of fluid flowed out per hour. Another technique was also used to bring into contact the surface of the wound and the distilled water or hypertonic solution. Agar cakes containing distilled water or hypertonic sodium chloride solution were applied to the surface of the wound. These constantly gave up their fluid contents, and were left at the surface of the wound, according to the nature of the experiment, from 6 to 12 hours and even 24 hours per day. In order to prevent immediate reinfection, the wound was flushed from four to six times in 24 hours with Dakin's solution. Another method consisted in applying for several hours every day a dressing composed of sodium stearate and chloramine paste. Chloramine paste, 10 parts per 1,000 was generally used, because reinfection takes place under 4 parts per 1,000. The wound

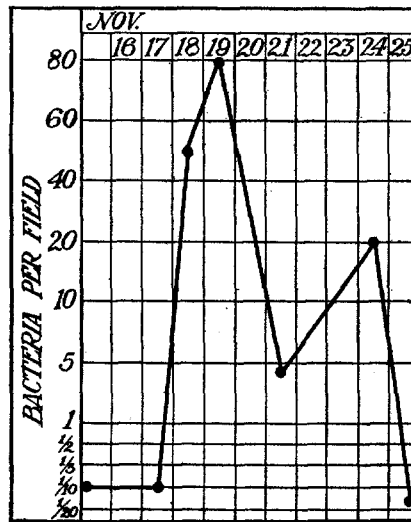
was examined bacteriologically every day. As soon as reinfection occurred the experiment was interrupted and the wound thoroughly sterilized. Every 4 days the surface of the wound was measured and the curve was plotted and compared with the calculated curve. The patients were kept in bed during the experiment and were under the supervision of a nurse day and night.

*Influence of Distilled Water on the Rate of Healing of a Sterile Wound.*

In the following experiments the wounds were flushed with distilled water from 2 to 4 hours every day.



Cicatrization curve.



Bacteriological curve.

TEXT-FIG. 1. Experiment 1. Case 646.

*Experiment 1 (Text-fig. 1).*—Case 646, age 25 years.

Nov. 17, 1916. Sterile wound in the calf, 12 sq. cm. in area. The wound is surgically aseptic. Flushing with distilled water for 2 hours. Then flushing with 30 per cent hypertonic solution for 2 hours. The dressing was kept in place until the following morning.

Nov. 18. The appearance of the wound has not changed. Surface area 12.1 sq. cm. 30 to 50 bacteria per field. Same dressing. Flushing with distilled water for 2 hours. Flushing with a hypertonic solution for 2 hours.

Nov. 19. The appearance of the wound has remained the same. Surface area 12.6 sq. cm. 50 to 100 bacteria per field. Dressing with chloramine paste, 10 parts per 1,000.

Nov. 20. Same dressing. Surface area 11.1 sq. cm.

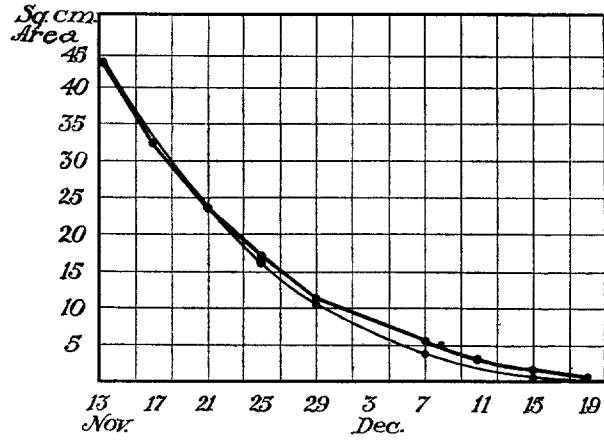
Nov. 21. Chloramine dressing, 4 parts per 1,000. Surface area 10.5 sq. cm. 4 bacteria per field.

Nov. 22. Flushing with distilled water for 4 hours. Chloramine dressing, 4 parts per 1,000 for 20 hours.

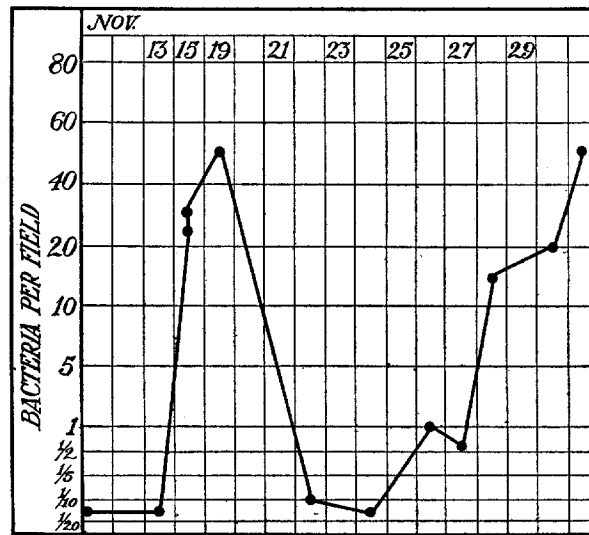
Nov. 23. Granulations smaller. Surface area 9.2 sq. cm. Flushing with distilled water for 4 hours. Chloramine dressing, 4 parts per 1,000.

Nov. 24. Same appearance. Surface area 8.4 sq. cm. 18 to 20 bacteria per field. Numerous cocci. Flushing with distilled water for 8 hours. Chloramine dressing, 10 parts per 1,000.

Nov. 25. 1 bacterium in 15 fields.



Cicatrizization curve.



Bacteriological curve.

TEXT-FIG. 2. Experiment 2. Case 694.

*Experiment 2 (Text-fig. 2).*—Case 694, age 20 years. Sterile wound 33.5 sq. cm. in area.

Nov. 17, 1916. Flushing with distilled water for 2 hours, followed by 2 hours' flushing with 30 per cent hypertonic sodium chloride solution. The dressing was not renewed until the following morning.

Nov. 18. The wound appears the same. Surface area 30.4 sq. cm. 20 to 30 bacteria per field. Flushing with distilled water for 2 hours and hypertonic solution for 2 hours.

Nov. 19. Same appearance. Owing to the infection, the rate of cicatrization has diminished. The surface area is 29.6 sq. cm. 55 bacteria per field. Chloramine dressing, 10 parts per 1,000.

Nov. 20. Same appearance. The curve has caught up with the normal curve. Surface area 26 sq. cm. 20 bacteria per field. Chloramine paste, 10 parts per 1,000.

Nov. 21. Same appearance. Surface area 23.6 sq. cm. 4 bacteria per field. Chloramine dressing, 4 parts per 1,000.

Nov. 22. Chloramine dressing, 4 parts per 1,000. 1 bacterium per 8 or 10 fields.

Nov. 23. Same treatment. Surface area 17.3 sq. cm. The wound is sterile and the experiment with distilled water alone can be resumed.

Nov. 24. Flushing with distilled water for 8 hours. Dressing with chloramine paste, 4 parts per 1,000. 1 bacterium in 10 to 15 fields.

Nov. 25. Flushing with distilled water for 8 hours.

Nov. 26. Flushing with distilled water for 24 hours. In order to keep the wound sterile, an injection of Dakin's solution is made every 6 hours. Surface area 14.6 sq. cm.; calculated area 14 sq. cm. 1 bacterium per field.

Nov. 27. Flushing with distilled water for 24 hours. 1 bacterium in 3 fields.

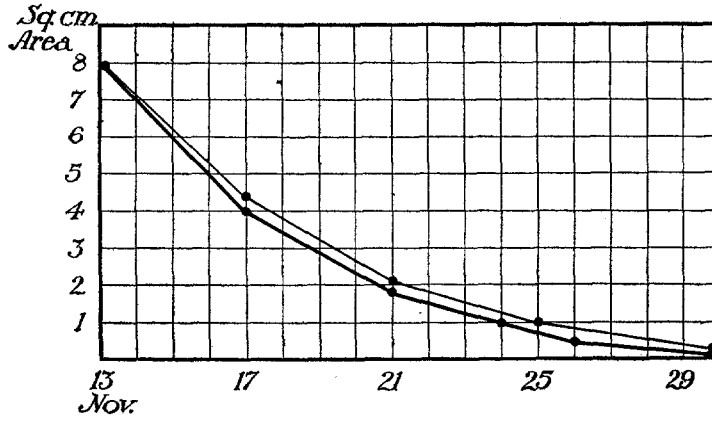
Nov. 28. Flushing with distilled water for 24 hours. Observed area 12 sq. cm. No apparent change. Calculated area 11.8 sq. cm. 10 to 15 bacteria per field.

Nov. 29. Flushing with distilled water for 24 hours.

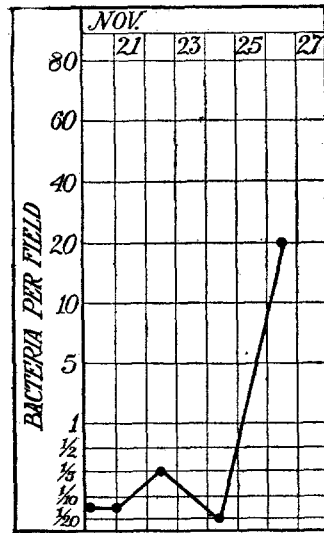
Nov. 30. Retardation of the healing process. Observed area 11 sq. cm.; calculated area 9 sq. cm. 15 to 20 bacteria per field. Flushing with distilled water for 24 hours.

Dec. 1. Experiment interrupted on account of infection. Application of compresses soaked in Dakin's solution; renewed six times in 24 hours.

Dec. 2. Same dressing. Innumerable small bacilli.



Cicatrization curve.



Bacteriological curve.

TEXT-FIG. 3. Experiment 3. Case 694.



*Experiment 3 (Text-fig. 3).*—Case 694, age 20 years.

Nov. 21, 1916. Sterile wound of the wrist, measuring 1.8 sq. cm. Since Nov. 13 the process of repair has been regular. Flushing with distilled water for 2 hours, followed as in the previous experiment by application of chloramine paste, 4 parts per 1,000, in order to prevent infection.

Nov. 22. Flushing with distilled water for 4 hours, followed by chloramine dressing. Observed surface 1.5 sq. cm.; calculated surface 1.8 sq. cm. 1 bacterium in 4 fields.

Nov. 23. Flushing with distilled water for 6 hours. Dressing with chloramine paste.

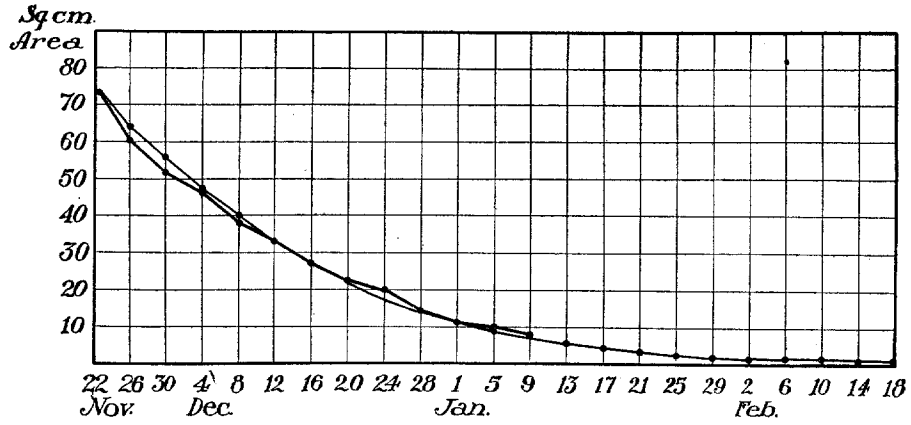
Nov. 24. Flushing with distilled water for 8 hours. Dressing with chloramine paste. 1 bacterium in 15 to 20 fields.

Nov. 25. Scale formed on the wound. Dry dressing.

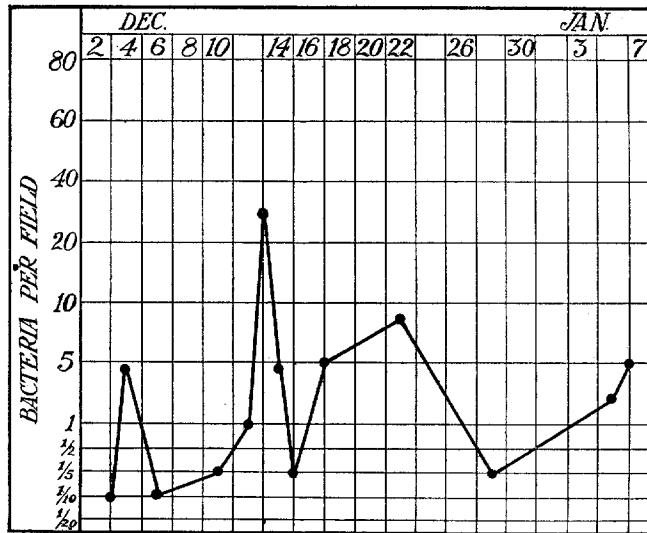
Nov. 26. Scale is removed. Area of the wound is 0.37 sq. cm.; calculated surface 0.7 sq. cm. 15 to 20 bacteria per field. Dressing with chloramine paste.

*Influence of Hypertonic Sodium Chloride Solution on the Rate of Healing of a Sterile Wound.*

In the following experiments the wounds were flushed with hypertonic sodium chloride solution.



Cicatrization curve.



Bacteriological curve.

TEXT-FIG. 4. Experiment 4. Case 639.

*Experiment 4 (Text-fig. 4).*—Case 639, age 36 years.

Dec. 1, 1916. The wound is practically sterile. 1 bacterium in 10 to 12 fields.

Dec. 2. Flushing with 40 per cent sodium chloride solution for 6 hours. Four injections with Dakin's solution. Surface area of wound 48.5 sq. cm.

Dec. 4. Flushing with 40 per cent sodium chloride solution for 12 hours. Four injections of Dakin's solution. Observed surface 46.5 sq. cm.; calculated surface 47.3 sq. cm. 3 bacteria per field.

Dec. 5. Same treatment.

Dec. 6. Same treatment. 1 bacterium in 8 to 10 fields.

Dec. 7. Same treatment.

Dec. 8. Same treatment. Six injections of Dakin's solution, instead of four. Area 38.5 sq. cm.; calculated area 39.7 sq. cm.

Dec. 9. Same treatment.

Dec. 10. Flushing with 40 per cent sodium chloride solution for 24 hours. Six injections of Dakin's solution. 1 bacterium in 8 fields.

Dec. 11. Same treatment.

Dec. 12. Same treatment. Surface of wound 33 sq. cm.; calculated surface 33 sq. cm. 1 bacterium per field. The curve as observed has finally overtaken the calculated curve and they now coincide exactly.

Dec. 13. Same treatment. 20 to 30 bacteria per field. The experiment is interrupted on account of infection.

Dec. 14. Sterilization of the wound by twelve flushings with Ringer solution.

Dec. 15. Same treatment. 1 bacterium in 6 fields.

Dec. 16. The experiment is resumed. A cake of agar-agar made of 40 per cent sodium chloride solution is applied to the wound during 12 hours. During the night the wound is sterilized by six flushings with Dakin's solution. Observed area 27 sq. cm.; calculated area 27.1 sq. cm. Sterile. The cake is applied for 9 hours.

Dec. 17. Same treatment. 4 bacteria per field.

Dec. 18. Same treatment. Surface observed 24.3 sq. cm.; calculated surface 24.2 sq. cm.

Dec. 19. A few bacteria.

Dec. 24. The wound is sterile. Agar cakes containing 50 per cent sodium chloride solution are applied during the day, and chloramine paste during the night.

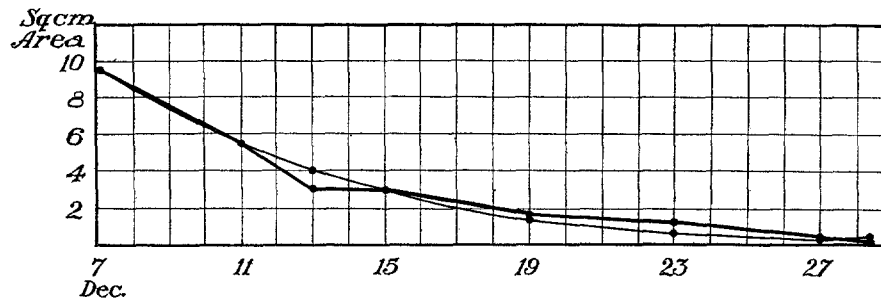
Dec. 28. The surface of the wound measures 14.2 sq. cm.; calculated surface 14.25 sq. cm. Same treatment.

Dec. 29. Same treatment.

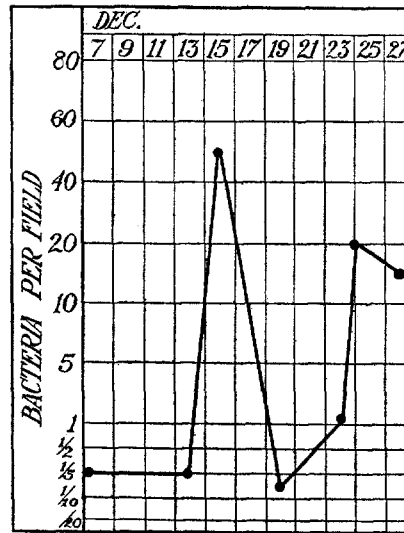
Dec. 30. Flushing with 80 per cent sodium chloride solution for 24 hours. Four flushings with Dakin's hypochlorite solution.

Jan. 1, 1917. Same treatment. Granulations congested. Small hemorrhages. Observed area 11 sq. cm.; calculated area 11.3 sq. cm.

Jan. 5. Same treatment with 80 per cent solution from Jan. 2 to 4. Observed area 10 sq. cm.; calculated area 9 sq. cm. 4 bacteria per field. The experiment is discontinued.



Cicatrization curve.



Bacteriological curve.

TEXT-FIG. 5. Experiment 5. Case 715.

*Experiment 5 (Text-fig. 5).*—Case 715, age 23 years; Arabian. Leg wound.

Dec. 7, 1916. Surface of wound 9.5 sq. cm. Dressing with Dakin's solution.

Dec. 11. Surface of wound 5.5 sq. cm.; calculated area 5.5 sq. cm. Flushing with 50 per cent sodium chloride solution for 24 hours, interrupted with two injections of Dakin's solution. Same treatment until Dec. 15.

Dec. 15. 30 to 50 bacteria per field. Four flushings with Dakin's solution in 24 hours. Surface observed 2.85 sq. cm.; calculated area 2.9 sq. cm.

Dec. 18. Flushing with hypertonic solution is substituted by the application of a cake of agar-agar containing 40 per cent sodium chloride. During the night three injections with Dakin's solution are made.

Dec. 19. Surface observed 1.5 sq. cm.; surface calculated 1.35 sq. cm. The injections are replaced by applications of chloramine paste, 10 parts per 1,000, the agar cakes being kept in contact with the wound for 12 hours every day.

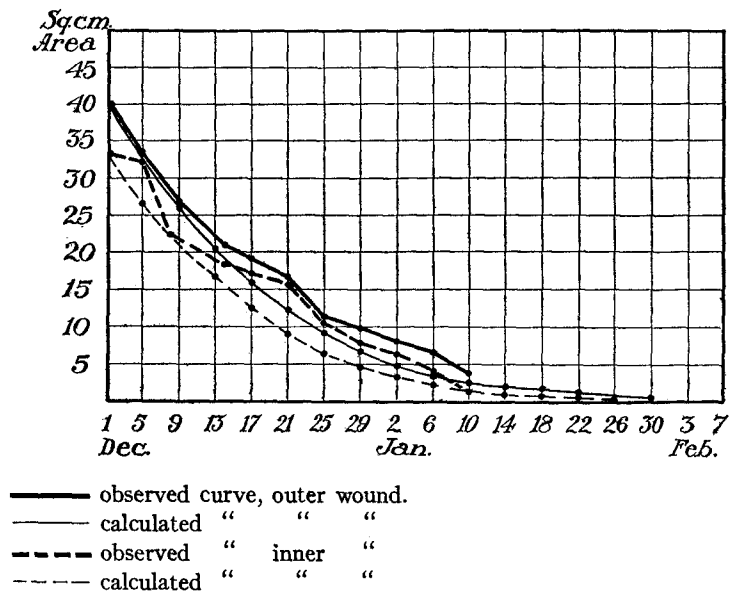
Dec. 23. Same treatment until Dec. 23. Observed area 1.1 sq. cm.; calculated area 0.6 sq. cm. The difference may be attributed to a slight infection. 2 bacteria per field.

Dec. 24. The 40 per cent agar cakes are replaced by a 50 per cent cake. The application of chloramine paste is continued during the night.

Dec. 27. Same treatment until Dec. 27, on which date it is calculated that healing will be effected. The wound, which is too small to be measured (less than 0.1 sq. cm.), is infected (10 to 15 bacteria per field), and this explains the retardation. Dressing with chloramine paste.

Dec. 29. The wound is completely healed.

*Comparison of Distilled Water and of Hypertonic Sodium Chloride Solution on the Same Patient.*



Cicatrization curve.

TEXT-FIG. 6. Experiment 6. Case 721.

*Experiment 6 (Text-fig. 6).*—Case 721. The patient has two wounds in the thigh, of about equal dimension, a small distance apart. The inner wound is flushed out with distilled water; the other with 40 per cent sodium chloride solution.

Dec. 3, 1916. Flushing of inner wound with distilled water for 12 hours. Four injections of Dakin's solution. Flushing of outer wound for 12 hours with 40 per cent sodium chloride. Four injections of Dakin's solution.

Dec. 4. 30 to 50 bacteria per field. Sterilization with Dakin's solution.

Dec. 8. Sterilization with Dakin's solution.

Dec. 12. Flushing for 24 hours per day under the same conditions described above, with six injections of Dakin's solution, continued until Dec. 16.

Dec. 17 and 18. Reinfection. Sterilization.

Dec. 20. Application to the external wound of an agar-agar cake, in the proportion of 40 per cent sodium chloride, and to the inner wound of a cake compounded with distilled water. 7 p.m. Dressing with chloramine paste. Same treatment until Dec. 27.

#### DISCUSSION AND SUMMARY.

In the study of the action of non-antiseptic substances on the rate of cicatrization, the chief obstacle encountered is the facility with which wounds become reinfected under an aseptic dressing. At the beginning of Experiment 1 the wound was sterile. It was subjected to flushing with distilled water for 2 hours, then to flushing with 30 per cent sodium chloride solution for another 2 hours. During that time no special precaution was taken to sterilize the wound and the dressing was left intact until the following morning. It was then found that the wound contained from 30 to 50 bacteria per field. The following day, after the wound had been subjected to the same treatment, the number of bacteria had increased to 50 and 100 per field, and as an immediate consequence the surface of the wound increased from 12 to 12.6 sq. cm. in 2 days. The wound was then dressed antiseptically and was found to be sterile 3 days later. Reinfection again took place the following day in spite of antiseptic dressing with chloramine paste 4 parts per 1,000, which was applied for 20 hours. In Experiment 2 similar results were observed. After 2 days of flushing with distilled water, the number of bacteria had increased to 50 per field. The wound was thereupon sterilized, but new reinfection ensued a few days later. Another wound on the same patient became reinfected under the same conditions after 1

day of sterile dressing. In none of the patients could the wounds be kept in a sterile condition throughout the whole experiment. It was impossible to maintain the sterility of a wound under aseptic dressing. Dakin's solution was therefore injected every 4 hours, or less often, according to the degree of infection, or chloramine paste was applied during the night. If there were 3 or 4 bacteria per field, the experiment was discontinued in order that the wound might be sterilized again. The cicatrization and bacteriological curves of Experiment 4 show that by the application of chloramine paste a wound may be maintained in an appropriately bacteriological condition for carrying out an experiment. Nevertheless, in spite of the antiseptic precautions taken, it was necessary to interrupt this experiment on two occasions, on December 13 to 15 and on December 18 to 22, in order that a complete sterilization of the wound might be effected. When the sterilization was performed as soon as the bacteria were discovered, little retardation occurred in the process of cicatrization. Moreover, the reinfection from the skin was often due to fine bacilli which have but mild retarding action on the rate of healing. The use of at least six flushings in 2 hours with Dakin's solution or of 12 hours' dressing with chloramine paste 10 parts per 1,000, was necessary to keep the wound in a condition of surgical asepsis.

The action of distilled water was studied in Experiments 1, 2, and 3. In Experiment 1 the wound was subjected to flushing with distilled water first for 2 hours, then 4 hours, and later for 8 hours per day. The wound was maintained in a condition of mild infection. No marked modification, either acceleration or retardation, was noted in the rate of repair during the period that the treatment was applied. From November 21 to 25 the wound was almost clean and the observed curve remained parallel to the calculated curve, showing that distilled water did not retard the rate of healing. In Experiment 2 the wound was subjected to uninterrupted flushing with distilled water, first for 2 and 8 hours, then for 24 hours. It was continued from November 24 to 30; *viz.*, for 112 hours out of 120, without the occurrence of any marked modification of the course of healing. The bacteriological curve showed that from November 22 to 27 inclusive the wound was kept aseptic. The slight retardation which occurred afterwards

was probably brought about by the infection. In Experiment 3 the wound was subjected to flushing with distilled water, first for 2, then for 4, 6, and 8 hours, a total of 20 hours in 4 days. From November 21 to 24 the wound remained surgically aseptic. No modification in the rate of healing occurred.

The action of the hypertonic sodium chloride solution was studied in a similar way. In Experiment 4 the wound was flushed at first with 40 per cent sodium chloride solution, from December 4 to 9 for 12 hours a day, and from December 10 to 13 for 24 hours a day, making a total of 144 hours out of 240 hours. At the end of this time the surface area of the wound coincided exactly with the calculated area. Owing to reinfection the experiment was suspended. From December 24 to 29 the wound was kept in contact with 50 per cent sodium chloride solution for 54 hours, and after December 30 flushing with 80 per cent solution for 24 hours a day was resorted to. The total amount of time involved in the above treatments was 174 hours with 40 per cent solution, 72 hours with 50 per cent solution, and 120 hours with 80 per cent solution. On January 1, the surface measured 11 sq. cm. and the calculated surface was 11.3 sq. cm. On January 5 the surface observed was 10 sq. cm. and the calculated surface was 9 sq. cm. It should be noticed that on January 5 the bacteria numbered 4 per field, which might account for the difference. In Experiment 5 the wound was flushed for 24 hours every day with 50 per cent sodium chloride solution from December 11 to 18, a total of 192 hours. From December 18 to 24 the wound was dressed with agar-agar cakes containing 40 per cent sodium chloride. The concentration was raised to 50 per cent from December 24 to 27. The cicatrization curve indicates only a slight retardation of the repair which can be attributed to infection when both cicatrization and infection curves are compared. The temporary acceleration on the 13th may have been due to the influence of the dressing, but as it did not occur again an experimental error is probably the cause of the change observed in the curve. In Experiment 6 two practically identical wounds at a distance of but a few centimeters from each other were located on the right thigh of Patient 721. The areas of the wounds were respectively 40 and 33 sq. cm. One of the wounds was flushed with distilled



water only. The other was subjected to the action of 40 per cent sodium chloride solution. From December 20 to 25 both wounds were in a condition of surgical asepsis. However, the cicatrization curves show that in spite of the difference of treatment the rate of healing was not modified.

The rate of healing of the wounds did not therefore apparently undergo any measurable modification under the influence of distilled water or hypertonic salt solution. It is well known that the osmotic changes of the medium have a marked influence on tissues deprived of circulation. But it seems that a tissue with normal circulation is protected by it against the changes of the osmotic pressure occurring at its surface. The above experiments show that apparently the conditions of the tissues of a wound are not modified by the changes of the osmotic pressure of the dressing. The beneficial effects of hypertonic sodium chloride solution on the sterilization of wounds and on the rate of healing recently described by various surgeons are possibly an illusion due to lack of precise technique.

#### CONCLUSIONS.

1. The flushing of an aseptic granulating wound with hypertonic sodium chloride solution or distilled water brings about an immediate reinfection.
2. Distilled water and hypertonic sodium chloride solution do not modify to a measurable extent the rate of healing of an aseptic wound.