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## ABSTRACT OF DISCUSSION.

Lyman F. Kebler referred to some investigations, not yet completed, on surgical dressings. D. F. Jones suggested the use of the Denver Steam Cooker as a sterilizer for small quantities of dressings, and J. Leon Lascoff stated that it was a very serviceable part of the drug store equipment.

# DISINFECTION SOLUTION (SPECIAL APPLICATION).\*

## BY L. E. SAYRE AND F. A. PATTY.

During the fearful period of the influenza epidemic the almost terror-stricken inhabitants of nearly every community were asking the authorities connected with boards of health for protection from every possible source of infection. It is needless to say that these authorities, while doing their utmost, were groping in the dark to locate the source or sources of contagion. They centered their efforts upon processes of purification and disinfection in every imaginable place where a possibility of a spread of the disease was suspected. One of the places under suspicion was the soda fountain. It was reported that in not a few places there was carelessness displayed in washing soda water glasses. In Kansas, a ruling was made that glasses should be washed in water at or near the boiling point. Many of the enterprising pharmacists installed sterilization schemes such as running hot water; others used live steam; and still others installed expensive apparatus for complete sterilization. Of course, such methods could not be used in the majority of small places where soda trade did not warrant the expenditure. The result was that in many of the small towns, stores discontinued their fountain business.

Dr. S. J. Crumbine, secretary of the Board of Health, and F. E. Rowland, assistant chief of inspection, asked for the preparation of a solution, if possible, that would be a harmless but reliable disinfectant for washing soda glasses—a solution that would stand a fairly severe test and one that any ordinary druggist could make himself. The off-hand remark to these officials was that a sodium hypochlorite solution would most likely meet the requirements they had in mind, but they urged that they would be glad to have any statement verified and substituted by experimental data.

Laboratory experiments to cover the situation were accordingly designed and carried out. This was accomplished by first procuring cultures of three organisms, one a culture of *micrococcus aureus*, an organism which produces boils and abscesses and is found abundantly in the mouth and throat. It is relatively easy to kill. Another, a culture of typhoid bacillus, the bacteria causing typhoid fever; and last, a culture of *streptococcus pyogenes*, which causes sore throat and various skin eruptions. This organism is very resistant and difficult to kill with ordinary disinfectants.

Broth cultures of these organisms were prepared and incubated 24 hours at body heat to obtain a luxuriant growth, then poured into glasses, previously sterilized in an autoclave. The glasses were drained, one at a time, dipped into a disinfectant bath and removed immediately. Each glass was next rinsed with sterile broth and a portion of the broth streaked on agar and incubated at body

<sup>\*</sup> Scientific Section, A. Ph. A., City of Washington meeting, 1920.

temperature to ascertain whether or not the organism had been killed by the disinfectant bath.

The time element in this way is reduced to a minimum, but so would it be in actual practice. Many solutions can be prepared, which will be good disinfectants if allowed to remain in contact for some time and at a temperature somewhat above room temperature  $(72^{\circ} \text{ F.})$ .

Tabulation of experiments with various solutions, together with results obtained, are herewith subjoined. It may be of interest to note here that results obtained from some solutions were somewhat surprising, as will be indicated.

The calcium hypochlorite solution employed was made by triturating bleaching powder with water and straining the mixture. The sodium hypochlorite solution used is best made as directed in making Solution No. 1.

The formula for the disinfecting bath is given below, for convenience, in both the Apothecaries and Metric Systems.

### FORMULA IN APOTHECARIES SYSTEM.

Solution No. 1.—Triturate 12 ounces (one can) bleaching powder to a smooth paste in 20 fluidounces of water and wash the mixture into a large flask or bottle (about one gallon) with 14 fluidounces of water. Add 8.4 ounces pure sodium carbonate dissolved in 17 fluidounces of hot water. Shake the contents thoroughly; if it becomes gelatinous, warm gently. Transfer the whole to a wet muslin strainer in a large funnel and return the first portion which comes through until the liquid is clear. Drain and wash with small successive portions of water sufficient amount to make 50 fluidounces.

Use four fluidounces to each three gallons (1.3 fluidounces to a gallon) of the disinfectant bath.

Solution No. 2.—Measure exactly 3 fluidounces C. P. hydrochloric acid (sp. gr., 1.17) into a container graduated for 32 fluidounces, and add enough water to make 32 fluidounces.

Use 1 fluidounce Solution No. 2 to each gallon of the disinfectant bath in its final preparation for use.

#### FORMULA IN METRIC SYSTEM.

Solution No. 1.—Triturate 200 grammes bleaching powder to a smooth paste, using water, and wash into 2-liter flask with 200 Cc. water. Add 140 grammes pure sodium carbonate dissolved in 250 Cc. hot water. Shake thoroughly; if contents become gelatinous, warm. Transfer to a wetted muslin strainer in a funnel and return first portion of filtrate until it comes through clear. Drain and wash with small successive portions of water to make 1000 grammes of the solution.

Use 10 Cc. of solution of No. 1 to make 1 liter disinfectant bath.

Solution No. 2.—Measure exactly 69 Cc. C. P. hydrochloric acid (sp. gr., 1.17) into a liter flask and make up to a liter with distilled water.

Use 10 Cc. to each liter disinfectant bath in final preparation for use.

*Precautions.*—All weights and measurements must be made *exact* for maximum results. Distilled water must be used in making solutions 1 and 2 and preferably but not necessarily in making the final bath. The bleaching powder must have at least 30% available chlorine present. The most favorable temperature for the bath is between  $80-95^{\circ}$  F.  $(27-35^{\circ}$  C.).

The method of application is simply to dip the previously washed glass into the bath, allowing all parts to come in contact with the liquid, and sterilization is complete. The bath is harmless to the hands.

It must be understood that no disinfectant, no matter how efficient, can so readily disinfect a dirty glass. The particles of dirt (organic matter) envelop the organisms and form a protective coating. To illustrate the truth of this statement, glasses infected by a broth culture of *streptococcus pyogenes*, were the organisms found to be alive. Therefore, after use at a fountain, glasses must not be permitted to become dry but should be, preferably, rinsed in tap water before going into the bath. This precaution would also help to maintain the efficiency for a longer period than otherwise would be possible. Care must be taken not to increase alkalinity by careless addition of wash water. Glasses are to be rinsed with clean water after the disinfecting bath.

## TABULATION.

|  |                        | Killed.        |        |                           |
|--|------------------------|----------------|--------|---------------------------|
| Disinfectant bath.                                     | Organism.              | Fresh<br>bath, | 24 hr. | Growth<br>in-<br>hibited. |
| Exp. 1.—Boiling water                                  | Micrococcus aureus     |                |        |                           |
|  | Bacillus typhosus      |                |        | +                         |
|  | Streptococcus pyogenes | -+-            |        | +                         |
| Exp. 2.—Calcium hypochlorite solution                  | Micrococcus aureus     | +              |        | +                         |
| 0.04% available chlorine made alkaline                 | Bacillus typhosus      |                |        | +-                        |
| with sodium carbonate                                  | Streptococcus pyogenes | _              | —      |                           |
| Exp. 3.—Calcium hypochlorite solution                  | Micrococcus aureus     |                |        |                           |
| 0.04% available chlorine made acid                     | Bacillus typhosus      |                |        |                           |
| with salicylic   | Streptococcus pyogenes |                |        |                           |
| Exp. 4.—Calcium hypochlorite solution                  | Micrococcus aureus     | +-             | -+-    | +-                        |
| 0.04% available chlorine                               | Bacillus typhosus      |                |        | ÷                         |
|  | Streptococcus pyogenes |                |        | ·                         |
| Exp. 5.—Calcium hypochlorite solution                  | Micrococcus aureus     | +              | +-     | +                         |
| with 5 Gm. sodium carbonate per liter.                 | Bacillus typhosus      | +              | +      | +-                        |
| 0.04% available chlorine                               | Streptococcus pyogenes | _              | _      |                           |
| Exp. 6.—Calcium hypochlorite solution                  | Micrococcus aureus     | +-             |        | +-                        |
| 0.06% available chlorine                               | Bacillus typhosus      | +-             |        | +-                        |
|  | Streptococcus pyogenes | -+-            |        | +                         |
| Exp. 7.—Calcium hypochlorite solution                  | Micrococcus aureus     | -+-            | +      | +                         |
| 0.06% available chlorine with 5 Gm.                    | Bacillus typhosus      | +-             | +      | -+-                       |
| sodium carbonate per liter                             | Sireplococcus pyogenes |                |        | +                         |
| Exp. 8.—Experiment 7 repeated with same results        | 100                    |                |        |                           |
| Exp. 9.—Calcium hypochlorite solution                  | Micrococcus aureus     | -+-            |        | -+-                       |
| 0.06% available chlorine made acid                     | Bacillus typhosus      | -+-            | _      | -+-                       |
| with hydrochloric acid                                 | Streplococcus pyogenes | +              |        | -+-                       |
| Exp. 10.—Calcium hypochlorite solution                 | Micrococcus aureus     |                |        | +-                        |
| 0.06% available chlorine with 2 Gm.                    | Bacillus typhosus      | +              |        | +                         |
| sodium salicylate per liter                            | Streptococcus pyogenes | · · · ·        |        |                           |
| Exp. 11.—Calcium hypochlorite solution                 | Micrococcus aureus     | _              |        | +                         |
| 0.06% available chlorine with 10 Gm.                   | Bacillus typhosus      | +              | +      | +                         |
| sodium carbonate and 2 Gm. sodium                      | Streptococcus pyogenes |                |        |                           |
| salicylate per liter                                   |                        |                |        |                           |
| Exp. 12Calcium hypochlorite solution                   | Micrococcus aureus     | +-             | +-     | +-                        |
| 0.06% available chlorine with 1 Gm.                    | Bacillus typhosus      | +              | +      | +                         |
| mercuric chloride per liter                            | Streptococcus pyogenes | +              | +      | +                         |
| Exp. 13.—Sodium hypochlorite solution                  | Micrococcus aureus     |                |        | +                         |
| 0.04% available chlorine                               | Bacillus typhosus      |                |        | +                         |
|  | Streptococcus pyogenes | —              |        |                           |
| Exp. 14.—Sodium hypochlorite solution                  | Micrococcus aureus     |                |        | +                         |
| $0.4^{e_{10}}_{10}$ available chlorine nearly neutral- | Bacillus typhosus      | +              |        | +                         |
| ized with hydrochloric acid                            | Streptococcus pyogenes |                |        |                           |

Killed.

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|  |                                      | mineu.         |                        |                           |
|--|--------------------------------------|----------------|------------------------|---------------------------|
| Disinfectant bath.   | Organism,                            | Fresh<br>bath. | Bath<br>24 hr.<br>old. | Growth<br>in-<br>hibited. |
| Exp. 15.—Sodium hypochlorite solution                            | Micrococcus aureus                   |                |                        | _                         |
| 0.05% available chlorine   | Bacillus typhosus                    | <u> </u>       |                        | +                         |
|  | Streptococcus pyogenes               |                |                        |                           |
| Exp. 16.—Sodium hypochlorite solution<br>0.1% available chlorine | Results same as for<br>Experiment 15 |                |                        |                           |
| Exp. 17.—Sodium hypochlorite solution                            | Micrococcus aureus                   | +              | +                      | +                         |
| $0.05\%$ available chlorine + 50 Cc. $\frac{N}{10}$              | Bacillus typhosus                    | +              | +                      | +                         |
| HCl per liter  | Streptococcus pyogenes               |                | —                      | +                         |
| Exp. 18.—Sodium hypochlorite solution                            | Micrococcus aureus                   | +              | +                      | +                         |
| 0.05% available chlorine + 80 Cc.                                | Bacillus typhosus                    | +              | +                      | +                         |
| $\frac{N}{10}$ HCl per liter                                     | Streptococcus pyogenes               | +              | +                      | +                         |
| Exp. 19.—Sodium hypochlorite solution                            | Micrococcus aureus                   | +              |                        | +                         |
| 0.05% available chlorine + 120 Cc.                               | Bacillus typhosus                    | +              | +                      | +                         |
| $\frac{N}{10}$ HCl per liter                                     | Streptococcus pyogenes               | +              | <u> </u>               | +                         |
|  |                                      |                |                        |                           |

Solution No. 1 was made up by four different formulas using less sodium carbonate, and neutral baths of 0.05% available chlorine made from them, but all proved unsatisfactory.

Bath 18 was tried again with the same good results, and was found to be germicidal to *micrococcus aureus* and typhoid bacillus even after standing seven days. A liter was then tried on 150 glasses in a series and found to be efficient for that number. Although this bath under good conditions is germicidal more than 24 hours, it is strongly recommended that it be prepared fresh each day.

It may be noted in the experimental data that the hot water is not at all efficient in the time allowed. By comparing the results, it can be seen that a 0.06% available chlorine solution is of little more value than a 0.04% solution. A strong alkaline solution appears to be as good the second day as the first and to be as powerful as a neutral solution. The sodium carbonate tends to hold the chlorine in solution. A solution made acid with hydrochloric acid is strongly germicidal when freshly prepared, but of no value after standing 24 hours, because the chlorine is liberated and soon escapes from the solution. Sodium salicylate, germicidal in itself, appears to add no power to the hypochlorite solutions.

The sodium hypochlorite solution proved to be of no value even in a strong (0.1) available chlorine solution. This was a revelation very disappointing, to say the least. The idea of reducing the alkalinity suggested itself. Therefore, varying amounts of  $\frac{N}{10}$  hydrochloric acid, sufficient to neutralize the solution, were added to Baths 17, 18 and 19. The results were somewhat striking in that it was found that the maximum results were obtained, as will be seen, in the mean between 17 and 19, namely, 18.

Baths 12 and 18 both proved highly satisfactory as far as germicidal power is concerned, but Bath 12 is undesirable and impracticable because of the highly poisonous character of mercuric chloride even in a 1: 1000 solution. Bath 18, on the other hand, is ideal in that it is non-poisonous, safe to handle, easily made, and inexpensive—one can of bleaching powder and one-half pound of sodium carbonate being sufficient for about 40 gallons of bath.

### CONCLUSIONS.

1.—It will be seen that 19 different solutions of various compositions were experimented with. The solutions containing available chlorine were the only ones that were available for making a disinfectant bath. Of the various sodium hypochlorite baths tested, it was found that the amount of free chlorine was not the only factor that determined efficiency. For example, a bath containing 0.1% of available chlorine was no more efficient than one containing 0.05%.

2.—The efficiency of the hypochlorite bath seems to depend not only upon the available chlorine but also upon the degree of alkalinity of the solution. The most efficient solution seems to be that containing 0.05% of available chlorine and a degree of alkalinity represented by almost the neutral point. In order to determine the exact amount of alkalinity, an aliquot portion of the solution neutralized with its equivalent of HCl (the amount designated in Formula 18) was evaporated to dryness; the residue re-dissolved in distilled water and the resulting solution titrated. It was found that one Cc. of the original solution thus prepared corresponded to 5.0 Cc. of  $\frac{N}{10}$  HCl. Since 10 Cc. are used in a liter of bath the alkalinity of this would make a liter of the bath correspond to 50 Cc.  $\frac{N}{10}$  HCl.

3.—This solution of the above alkalinity can be arrived at by using the proportions indicated in Formula 18, using the solutions and mixing same as prescribed in said formula.

4.—Since one of the microörganisms resisted the boiling water bath and only two were inhibited in growth, therefore, it would seem that any treatment with boiling water, much less warm water, would be absolutely unreliable unless care was taken that the utensils were permitted to remain in such a bath for some minutes. It is also suggested that the wiping of the cleansed tumbler with a fabric would be objectionable for the reason that repeated use of same would tend toward a risk of contamination.

# COMMERCIAL DISINFECTANT LIQUIDS.

Commercial proprietary disinfectants for the above purpose are on the market. We have examined two of these which have quite a wide distribution. These we will designate as A and B.

A, we found to be quite efficient and equal to our own solution.

B, although apparently surcharged with chlorine gas and claimed to be a bacillus destroyer, was wholly ineffective for making a disinfectant bath as above indicated.

Mention is made of these merely to show that laboratory tests should be made of such proprietary solutions and these preparations should be required to state on label the efficiency of the article in such terms as can be understood and the claimed efficiency tested by proper authorities.