

*Summary.* The results secured in these investigations show that the initial number of cells influences survival of microörganisms exposed to ultraviolet light. Dilute suspensions are destroyed more quickly than heavy ones. The order of decreasing resistance for the species used was: *Staphylococcus aureus*, *Escherichia coli* (Frank strain), *Escherichia coli* (American Type Collection No. 4348), and *Serratia marcescens*. For the mold-spores studied the order of decreasing resistance was: *Aspergillus glaucus*, *Aspergillus niger*, *Penicillium*, and *Mucor*.

Cardboard strips, which had been inoculated by immersion in a suspension of mold-spores, were not as thoroughly sterilized as were the suspensions themselves, when exposed to ultraviolet light in Petri dishes. On many of the strips it was possible to determine that the colonies originated from the under side of the strip more frequently than from the upper side. This was interpreted as indicating that indirect irradiation is much less effective than direct irradiation.

## 13034

**Bactericidal Effect of Ultraviolet Rays on Microorganisms on Restaurant Glass-Ware.**

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To study the bactericidal effect of irradiation with the Sterilamp on drinking glasses which had been used at soda fountains, it was necessary to select or devise a method of sampling the bacteriological contamination of the glasses. The multiple glass test proposed by the American Public Health Association<sup>1</sup> was used whenever a series of glasses from a soda fountain was examined. Other procedures were followed including the method used by Speck and Black.<sup>2</sup> In addition, a supplementary method was devised which permitted examination of the entire inner surface of the glass as well as the rims and proved to be simpler than that of Speck and Black. About 5 ml of a 25 ml water blank were poured into the drinking glass to be sampled and the entire inner surface of the glass was wetted and rubbed by means of a sterile cotton swab. The swab was then broken

<sup>1</sup> Am. Pub. Health Assn., Yearbook, 1936-1937, p. 48.

<sup>2</sup> Speck, M. L., and Black, L. A., *Food Research*, 1937, **2**, 559.

off into the rest of the 25 ml water blank and shaken vigorously until the cotton had become well disentangled. The cotton swab and the remainder of the water blank were then carefully transferred back to the drinking glass. The contents of the glass were mixed by a few rapid rotations and one ml portions were plated on agar.

During this investigation an opportunity was presented to make some tests with a Sterile-Ray Cabinet. A series of unirradiated dry drinking glasses, a series of irradiated dry drinking glasses, a series of unirradiated wet (but clean) drinking glasses and a series of irradiated (but clean) drinking glasses were examined bacteriologically by the American Public Health Association procedure and by the procedure of Speck and Black. The exposure time of 130 seconds was controlled by a timing device on the cabinet. The plate counts on nutrient agar were made after a 24-hour and a 60-hour incubation period at 37°C. The results appear in Table I. It will be noted

TABLE I.  
Exposure of Drinking Glasses for 130 Seconds in the Steril-Ray Cabinet.

|                                                                      | Estimates of colonies per glass after being incubated for: |       |               |       |
|----------------------------------------------------------------------|------------------------------------------------------------|-------|---------------|-------|
|                                                                      | 24 hr                                                      |       | 60 hr         |       |
|                                                                      |                                                            |       |               |       |
| A.P.H.A. proposed procedure used on:                                 |                                                            |       |               |       |
| 10 unexposed glasses                                                 | 3.5                                                        |       | 15.5          |       |
| 10 exposed glasses                                                   | 0.3                                                        |       | 9.3           |       |
| Estimates of colonies on rims from plate counts after incubation of: |                                                            |       |               |       |
|                                                                      | (Inside rim)                                               |       | (Outside rim) |       |
|                                                                      | 24 hr                                                      | 60 hr | 24 hr         | 60 hr |
| Procedure of Speck and Black (1937) used on:                         |                                                            |       |               |       |
| Dry unexposed glass                                                  | 25                                                         | 25    | 100           | 500   |
| " " "                                                                | 50                                                         | 125   | 0             | 75    |
| " " "                                                                | 0                                                          | 25    | 25            | 25    |
| " " "                                                                | 0                                                          | 75    | 0             | 0     |
| Dry exposed glass                                                    | 0                                                          | 0     | 0             | 0     |
| " " "                                                                | 0                                                          | 0     | 0             | 0     |
| " " "                                                                | 0                                                          | 25    | 0             | 0     |
| " " "                                                                | 0                                                          | 0     | 0             | 25    |
| Wet unexposed glass                                                  | 2,950                                                      | *     | 75            | 275   |
| " " "                                                                | 27,500                                                     | *     | 75            | 275   |
| " " "                                                                | 550                                                        | 1,325 | 175           | 450   |
| " " "                                                                | 1,150                                                      | 2,550 | 75            | 250   |
| Wet exposed glass                                                    | 0                                                          | 25    | 25            | 50    |
| " " "                                                                | 0                                                          | 0     | 0             | 25    |
| " " "                                                                | 50                                                         | 100   | 0             | 25    |
| " " "                                                                | 0                                                          | 50    | 150           | 350   |

\*Not possible to count colonies.

TABLE II.  
Effect of the Sterilamps on Drinking Glasses Rinsed with Suspensions of  
*Escherichia coli* (Frank Strain).

|         | Estimates of colonies on the inside surface of the glass after<br>being exposed for: (min.) |      |     |    |   |    |
|---------|---------------------------------------------------------------------------------------------|------|-----|----|---|----|
|         | 0                                                                                           | 1    | 2   | 3  | 4 | 5  |
| Glass A | 7500                                                                                        | 1300 | 0   | 0  | 0 | 0  |
| Glass B | (7500)*                                                                                     | 325  | 350 | 75 | 0 | 25 |

\*Count taken equal to the first because it was too difficult to estimate.

TABLE III.  
Effect of Sterilamps on Drinking Glasses Exposed in the Laboratory Cabinet.

| Estimates of colonies on the inside surface of the glass after<br>the exposure indicated: |                                 |                                 |
|-------------------------------------------------------------------------------------------|---------------------------------|---------------------------------|
| 5 unexposed glasses                                                                       | 5 glasses exposed<br>for 2 min. | 5 glasses exposed<br>for 4 min. |
| 250                                                                                       | 0                               | 75                              |
| 600                                                                                       | 0                               | 50                              |
| 200                                                                                       | 0                               | 50                              |
| 1000                                                                                      | 2120                            | 25                              |
| 550                                                                                       | 625                             | 25                              |
| Avg 520                                                                                   | Avg 549                         | Avg 45                          |

that, while the exposed and unexposed dry glasses show only small differences in counts, the opposite is true for wet (but clean) glasses.

In another series of experiments, sterile drinking glasses were rinsed with a suspension of approximately 120,000 bacteria per milliliter. The glasses were irradiated in pairs for definite time intervals in the laboratory cabinet. Table II shows clearly the marked lethal effect of the radiation on *Escherichia coli* (Frank strain).

In a still different type of experiment, 15 clean and dried drinking glasses were placed in individual sterile paper bags at the soda fountain and then transported to the laboratory for sampling and exposure. Five of the test glasses were sampled as untreated controls; 5 were exposed in the laboratory cabinet for 2 minutes and then sampled; the remaining 5 were exposed for 4 minutes before sampling. Sampling was done with 25 ml water blanks and single swabs as previously described in this article. The counts were made after 48 hours incubation at 37°C and 96 hours at room temperature. The results in Table III show a reduction of about 90% for a 4-minute exposure, but no reduction in the average count for a 2-minute exposure. It should be indicated, however, that in groups of glasses exposed for 2 minutes, three were apparently devoid of any living bacteria.

*Summary.* A sampling procedure, which was developed in order

to examine more completely the interior of a drinking glass, proved quite satisfactory and it was used extensively in this investigation.

The Steril-Ray Cabinet, employing 4 Sterilamps, was tested while in actual operation at a soda fountain. Dry clean glasses showed but slight bacteriological improvement as the result of being exposed in this cabinet. It should be noted that these were already of low bacterial content without being exposed. Wet clean glasses showed a very marked improvement as the result of being exposed. However complete sterilization did not result in every case.

The experimental evidence obtained in this investigation indicates that almost sterile surfaces are produced when clean drinking glasses are irradiated with the ultraviolet light produced by the Sterilamp.

### 13035

#### **Determination of Hydrogen Sulfide in Stomach Contents of Experimental Animals.**

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The problem of determining the sulfide content of solutions containing reducible or interfering substances presents difficulties. Since hydrogen sulfide is unstable and easily oxidized considerable trouble may be expected in recovering it from solution. Aeration with air, carbon dioxide, or hydrogen as well as distillation procedures have been employed<sup>1, 2, 4, 5</sup> but the completeness of the removal is open to question.

Recently in connection with another investigation it was necessary to determine the hydrogen sulfide concentration in the stomach contents. Due to the nature of the material it was evident that methods based on evolution procedures were the only approach to the problem.

Preliminary experiments with various evolution procedures on pure hydrogen sulfide solutions (1-10 mg per liter) consistently

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<sup>1</sup> Benade, W., *Mitt. Lab. preuss. geol. Landesanst.*, 1933, **19**, 3; Dept. Sci. Ind. Research, *Water Pollution Research*, Summary, current lit. **7**, 318.

<sup>2</sup> Buswell, *J. Am. Chem. Soc.*, 1925, **47**, 1381.

<sup>3</sup> Gibbs and Clardy, *Ind. and Eng. Chem. Anal. Ed.*, 1940, **12**.

<sup>4</sup> Marzahn, W., *Hyg. Rundschau*, 1919, **29**, 557; *Exp. Stat. Record*, **42**, 207.

<sup>5</sup> Rudolfs and Baumgartner, *Ind. and Eng. Chem.*, 1932, **24**, 1152.