Research for his helpful criticism in the preparation of this paper.

SUMMARY

Mycelial fragments of *Aspergillus niger* strain 1617, prepared by blending mycelial pellets, were found to be usable as a test organism in the quantitative evaluation of mycostatic (or mycoidal) activity of antifungal agents. Using these fragmented mycelia and spores, a comparative and quantitative method for determining fungitoxicity against fungal mycelia and spores was presented.

The sensitivity to various agents and heating of mycelia was compared with that of spores, and the general concept of fungitoxicity was discussed. The loss of germicidal and heat resistance of the spores in the early phase of germination was demonstrated.

REFERENCES


Studies on Disinfection of Clinical Thermometers

I. Oral Thermometers from a General Hospital

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The literature contains many different test procedures for the determination of the efficacy of disinfectants for the clinical thermometer. None of these methods is representative of the actual conditions encountered in the field. The purpose of our investigation was to develop a test method that reflects actual field conditions and to evaluate the efficacy of some common disinfectants by this method.

In the past, the investigators of thermometer disinfectants have used artificial conditions in their test procedures. For example, Gershenfeld et al. (1951) contaminated thermometers with a broth culture of microorganisms containing citrated plasma. Ritter (1956) contaminated glass rods with mucus inoculated with microorganisms. Ecker and Smith (1937) smeared thermometers with sputum taken from patients with Type 1 lobar pneumonia, and Frobisher et al. (1953) also used sputum, but from patients with active pulmonary tuberculosis. It is obvious that broth cultures of microorganisms with organic matter do not duplicate practical field conditions. Sputum serves as a source of microorganisms in a natural environment; however, it is unlikely that sputum would be found on oral thermometers after use. In our study we decided to use thermometers from mouths of patients suffering with a variety of infections in order to obtain on the thermometer a representative spectrum of microorganisms in their natural environment.

MATERIALS AND METHODS

**Thermometers.** Becton, Dickinson 1 oral thermometers, from which the mercury was removed by breaking the upper end and centrifuging, were used for the test. The broken end was sealed and fire polished before using.

**Disinfectants.** All disinfectant solutions were prepared on a volume per volume basis in sterile distilled water. The following disinfectants were tested:

- Ethyl alcohol
- Synthetic phenolic (p.c. 10)2 ortho-hydroxydiphenyl and para-tertiary-amyl phenol solubilized by potassium ricinoleate.
- Synthetic phenolic (p.c. 5) ortho-hydroxydiphenyl solubilized by potassium ricinoleate.
- Commercial iodophor, iodine solubilized by nonionic detergent; 1.6 per cent available iodine.

1 Becton, Dickinson and Company, Rutherford, New Jersey.

2 p.c. = Phenol coefficient.
Iodophor, iodine solubilized by nonionic detergent; 2 per cent available iodine.

Benzalkonium chloride

Tincture of benzalkonium chloride, 0.1 per cent anhydrous benzalkonium chloride in 50 per cent ethyl alcohol.

Formaldehyde

Method. Thermometers were cultured in Difco fluid thioglycollate medium. Thermometers that had been immersed in the quaternary ammonium compound were cultured in Difco fluid thioglycollate broth containing 0.5 per cent Tamol X \(^8\) (Goetchius, 1949) in order to obviate bacteriostasis.

Method. About 50 thermometers were placed in a covered enameled tray and sterilized each time before use at 165 \(^\circ\)C for 90 min. The thermometers were taken to the wards of a general hospital and were placed in the mouths of surgical and medical patients by the nursing staff. They were collected in a second sterile tray and returned to the laboratory for disinfection studies. The above routine was continued once weekly for about 8 months.

The thermometers were disinfected 3 to 4 hr after removal from the patients' mouths. It was observed that all thermometers were dry at this time. Each was placed in a sterile capped tube containing sufficient disinfectant (about 20 to 25 ml) for complete immersion. After 15 min at 20 \(^\circ\)C, each thermometer was rinsed by agitating for 10 sec in a tube containing 30 ml of sterile distilled water. Each was then transferred to a tube containing 10 ml of broth. All tubes were incubated for 7 days at 37 \(^\circ\)C. At the end of the incubation period, each tube was observed for the presence of growth. Gram stains were made from all tubes exhibiting growth to determine the type of microorganism present. A few tubes were found to contain spore-bearing bacilli and these were eliminated from the test results since none of the disinfectants is sporicidal under the test conditions.

Two types of controls were included. The first series of controls was made to ascertain the presence of viable microorganisms on all thermometers and to determine the types of microorganisms found. For these tests, sterile distilled water was substituted for the disinfectant in the above procedure. The second series of controls was made to determine the probability of air contamination during transfer of the thermometers since this operation was carried out in a large laboratory. In this instance, sterile thermometers and sterile distilled water were used and the procedure as described above was followed.

Method to demonstrate elimination of bacteriostasis in the culture tubes. Following the above procedure, five thermometers from each of the disinfectant solutions were placed in one tube containing 10 ml of the appropriate culture broth. On inoculation with 0.01 ml of fresh saliva, growth was normal in 24 hr.

Results and Discussion

The results of all tests and findings are summarized in Table 1. It is evident from the transfer of 100 sterile thermometers that 2 per cent became contaminated during transfer due to air-borne microorganisms. Consequently, in the interpretation of the results, allowance should be made for the possibility of air contamination. In drawing conclusions as to the reliability of a disinfectant, we have considered the number of positive tests and the types of microorganism found as contaminants. The presence of streptococci indicated that the disinfectant was not reliable. It is estimated that air contamination is probably of the order of 0 to 4 per cent. The results in table 1 have been interpreted as follows:

Ethyl alcohol: 70 per cent ethyl alcohol is a reliable disinfectant for oral thermometers, whereas 50 per cent ethyl alcohol is not.

Phenolics: both synthetic phenolics were found to be reliable disinfectants for oral thermometers.

Iodine: iodine solubilized with nonionic detergents was found to be an unreliable disinfectant for oral thermometers at concentrations as high as 200 ppm available iodine.

TABLE 1

Results of treatment of thermometers with various disinfectants at 20 \(^\circ\)C

<table>
<thead>
<tr>
<th>Test</th>
<th>No. Tested</th>
<th>No. Positive</th>
<th>Type of Microorganism Found</th>
<th>Per Cent Negative</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Ethyl alcohol</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>50%</td>
<td>50</td>
<td>7</td>
<td>1 1 1 5</td>
<td>86</td>
</tr>
<tr>
<td>70%</td>
<td>100</td>
<td>3</td>
<td>1 1 1</td>
<td>97</td>
</tr>
<tr>
<td><strong>Phenolics</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2% Synthetic pheno-</td>
<td>100</td>
<td>1</td>
<td>1</td>
<td>99</td>
</tr>
<tr>
<td>nol (p.e. 10)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3% Synthetic pheno-</td>
<td>100</td>
<td>3</td>
<td>1 1 1</td>
<td>97</td>
</tr>
<tr>
<td>nol (p.e. 5)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Iodine</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0.5% Commercial i-</td>
<td>50</td>
<td>18</td>
<td>12 3 2 2 1</td>
<td>64</td>
</tr>
<tr>
<td>odophor (75 ppm</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>available iodine)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2% Iodophor (200</td>
<td>100</td>
<td>6</td>
<td>2 3 3 1</td>
<td>94</td>
</tr>
<tr>
<td>ppm available i-</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>odine)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>**Benzalkonium chlori-</td>
<td>100</td>
<td>2</td>
<td>1 1 1</td>
<td>98</td>
</tr>
<tr>
<td>de** (p.e. 10)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Benzalkonium chloride: an aqueous solution of 0.1 per cent benzalkonium chloride was found to be an unreliable disinfectant for oral thermometers. The addition of alcohol improved the performance of the compound. The results with 0.1 per cent tincture benzalkonium chloride indicate that this preparation may be a reliable disinfectant for oral thermometers. However, it should be pointed out that the survivors found from this test were gram-negative bacilli. The quaternary ammonium compounds have their least activity against this type of microorganism.

Formaldehyde: an aqueous solution of 10 per cent formaldehyde was found to be a reliable disinfectant for oral thermometers.

Our results differ in several instances with those of previous investigators. For example, Ecker and Smith (1937) found that 70 per cent ethyl alcohol did not disinfect thermometers in 5, 10, and 30 min. Frobisher et al. (1953), after evaluating 70 per cent ethyl alcohol and 10 per cent formaldehyde, did not recommend either preparation as reliable disinfectants. We have found both to be reliable disinfectants in this study. However, it should be pointed out that the latter authors used contact periods of 10 min vs 15 min in our study; and they also were evaluating disinfectants against Mycobacterium tuberculosis. All the preparations found to be reliable disinfectants in this study also exhibit tuberculocidal action. Since this organism was not a consideration in this investigation, a study is in progress on the disinfection of thermometers used by patients with active pulmonary tuberculosis and will be reported later.

ACKNOWLEDGMENTS

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SUMMARY

A test procedure is described that is representative of conditions encountered in the field in the disinfection of oral thermometers. The efficacy of several common disinfectants was evaluated according to this procedure. It was found that 70 per cent ethyl alcohol, 2 per cent synthetic phenolic (p.c. 10), 3 per cent synthetic phenolic (p.c. 5), 10 per cent formaldehyde, and a tincture of 0.1 per cent benzalkonium chloride were reliable disinfectants for thermometers at 20 °C in 15 min. The results indicate 50 per cent ethyl alcohol aqueous, 0.1 per cent benzalkonium chloride, and 2 per cent of an iodophor (200 ppm available iodine) failed to disinfect.

REFERENCES

ECKER, E. E. AND SMITH, R. 1937 Disinfecting clinical thermometers. Modern Hospital, 48, 86.