5. Salivation is slight with both methyl cyclopropane and cyclopropane. Vomiting has not been observed after either anesthetic.

6. Post-anesthetic depression and loss of appetite appear to be more marked after methyl cyclopropane than after cyclopropane.

SUMMARY.

Methyl cyclopropane has been prepared in a condition of purity suitable for its evaluation as an inhalation anesthetic, by the reduction of 1,3-dibromobutane with zinc. The reduction of 1,3-dichloroisobutane under similar conditions gave almost entirely isobutylene instead of methyl cyclopropane.

Although the effective concentrations as well as the lethal concentrations of methyl cyclopropane and cyclopropane, and hence their margins of safety, are about the same, the methyl cyclopropane does not compare favorably with cyclopropane in a qualitative and quantitative comparison of their several side-effects.

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A PRACTICAL METHOD FOR TESTING NON-PHENOLIC DISINFECTANTS.*[1]

BY WILLIAM C. CLARK.

Jordan (1) defines a disinfectant as a substance that kills the microbes with which it comes into contact. This statement is a very broad and general definition and Reddish (2) clarifies this general statement by asking "What are disinfectants for and why are they used?" and answering "Disinfectants are germicides which are used on inanimate objects for the purpose of killing disease germs which cause epidemiologic diseases. Their primary purpose is to aid in preventing the spread of disease by killing the bacteria which cause them."

Varley (3) recognizes the limitations of the Phenol Coefficient Method and proposes a method for testing disinfectants which are chemically related to Phenol that closely simulates the conditions of use of these disinfectants. His method is adapted to testing odorous disinfectants which may be washed away after a relatively short time. A simpler method was desired for an odorless non-phenolic disinfectant that is recommended for disinfecting floors, woodwork and furniture in the sickroom. Because this disinfectant is colorless and odorless it is not removed from the woodwork by washing, but remains on the wood until the next regular washing of the woodwork.

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[1] A contribution from the Bacteriological Laboratory of James F. Ballard, Inc.

In devising this test the object was to have the conditions of the test as nearly identical as possible with the conditions of use in the sickroom.

Materials needed for this test are available in any bacteriological control laboratory and are as follows:

Three varnished sticks 8 x 4 x 100 mm. Three painted sticks of the same dimensions. These sticks are cut to the above dimensions and are dipped into the varnish or paint and then allowed to dry one week or longer. Strips of linoleum or other floor coverings having the same dimensions may also be used. One lot of tubed nutrient broth, each tube containing 10 cc. of nutrient broth made and sterilized as directed in Circular 198 of the United States Department of Agriculture (4).

One large pair of forceps such as is used in plugging tubes with absorbent cotton.

One culture of *Straphylococcus aureus* 22 to 26 hours old and meeting all the other requirements of Circular 198 of the United States Department of Agriculture.

Six sterile medication tubes 25 x 150 mm. plugged with absorbent cotton before sterilization.

The Test.—Mark one end of each stick by any suitable means and grasp the opposite end in the left hand. Sterilize the marked end of the stick by passing it through the flame. Take the forceps in the right hand, flame the tips and grasp the marked end of the stick with forceps and flame the entire stick and the lower end of the forceps. Allow the stick to cool a few seconds, place the stick into the culture and stir it about 15 seconds. Remove the stick from the culture and place it on a recently flamed and cooled wire gauze. The entire operation should be performed using the usual aseptic precautions. Repeat the above operation with the five remaining sticks. Allow the six sticks to remain on the wire gauze exposed to the air just as is the woodwork in a sickroom.

After 72 hours have elapsed completely immerse each of the inoculated sticks into the highest dilution of the disinfectant that is recommended for disinfecting woodwork in the sickroom. Keep each stick completely immersed in the diluted disinfectant for thirty seconds. Transfer each stick from the diluted disinfectant into a dry, sterile, medication tube. Flame the mouth of each medication tube and replace the cotton plugs. Invert each medication tube after the sticks are in the tubes and the cotton plugs have been replaced. The object in inverting the medication tube is to permit the cotton plug to absorb any excess liquid and to permit the stick to dry rapidly. Allow the medication tubes to remain inverted for one hour.

With sterile forceps remove a stick from a medication tube and place it into a tube of sterile broth marked number one. Stir the broth for thirty seconds and then transfer the stick to a tube of sterile broth marked number two. Stir thirty seconds and remove the stick. This operation is repeated with the five remaining sticks. Aseptic precautions must be strictly observed in this part of the test to prevent the possibility of contamination. Incubate all twelve of the tubes of broth for 48 to 72 hours. Examine the tubes for growth. The object of the second subculture is to render the disinfectant too dilute to inhibit the growth of *Straphylococcus aureus*.

If growth occurs in all of the second subculture tubes, the dilution of the disinfectant used in this test has failed to kill the test organism.

If growth occurs only in one or two of the second subculture tubes, contamination may have occurred. Make an agar slant streak, incubate 24–48 hours and examine. If the organism is not *Staphylococcus aureus*, contamination has occurred. If the organism proves to be *Staphylococcus aureus*, the test must be repeated, as this organism may or may not be a contamination.

If no growth occurs in any of the second subculture tubes, the disinfectant has killed the test organism under conditions which are as nearly identical as possible to the conditions of use in disinfecting the woodwork of a sickroom. If there are any reasons to suspect that inhibition and not killing has occurred, inoculate the six tubes that are marked number two and incubate 24 hours. Growth proves that no inhibition has occurred.

In the disinfection of woodwork, the concentration of the disinfectant begins to increase immediately as the water evaporates; this increase in concentration continues until practically all of the moisture has evaporated. As a matter of fact, the bacteria will remain in contact with the disinfectant until it has decomposed or evaporated. The time of exposure of the bacteria to the disinfectant will, of course, vary with each different disinfectant; therefore, a fifteen-minute exposure in no way approximates the time of exposure in this use of the disinfectant. As a rule the temperature of a sickroom is above 20° C. These three fundamental conditions—concentration of the disinfectant, time of exposure of the bacteria to the disinfectant and temperature at which the bacteria are exposed to action of the disinfectant—are conditions which must be observed in any test that purports to give a true indication of the value of a disinfectant.

Considering these wide variations in conditions, it is readily apparent that the Phenol Coefficient Determination is not well adapted to the testing of claims for a non-phenolic disinfectant for use on the woodwork in a sickroom. A disinfectant which can be diluted only as much as one part of disinfectant to three parts of water in order to kill *Staphylococcus aureus* within five minutes using the Phenol Coefficient Determination shows effective disinfection by killing the same culture when one part of disinfectant is added to ten parts of water by this method of testing!

Such a variation in the concentration of the disinfectant required to kill a standard strain of *Staphylococcus aureus* is to be expected under such wide variations in conditions as exist between the Phenol Coefficient Determination and the disinfection of woodwork in a sickroom. The end sought in the disinfection of woodwork is the death of the bacteria by the disinfectant; not necessarily the killing of an extremely large number of bacteria in a few minutes. It is of no material difference whether the bacteria are killed in five minutes or in several hours; disinfection has been accomplished.

There are several factors that give this test a reasonable margin of safety. The fact that *Staphylococcus aureus* is a more highly resistant organism than *E. typhi*, the official test organism used in the Phenol Coefficient Method, the fact that the disinfectant remains on the woodwork for a much longer period of time than the time of the test, and the presence of the organic matter in the test materials, offer a reasonable margin of safety for this test.

Naturally this test has very definite limitations and these limitations must be observed if this test is to be of value. This test is limited to the use of testing non-phenolic disinfectants which do not depend on a volatile constituent and which are odorless. The disinfectant must not be washed off the woodwork for several days.

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(4) Circular 198, United States Department of Agriculture, December 1931, page 1.