THE PHENOL COEFFICIENT METHOD OF TESTING DISINFECTANTS.*

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Ten years ago, S. Rideal and J. T. A. Walker (Journ. Roy. San. Inst., London, 1903, 424), devised a method for the bacteriological standardization of disinfectants, known as the Rideal-Walker Method. It is extensively used in England and the British colonies. Later, a modification of it was proposed in the London Lancet (Vol. 177, Nos. 4498, 4999 and 4500), known as the Lancet Method, and this was believed to be a distinct advance over the Rideal-Walker method. Still later a third modification was evolved by J. F. Anderson and T. B. McClintic, of the Hygienic Laboratory of the Public Health Service (Bulletin No. 82, April, 1912), known as the Hygienic Laboratory Method. This method has some of the features of the Rideal-Walker method as well as the Lancet Method, but also, important modifications. It is now being used by the Federal and state authorities in connection with the purchase of disinfectants, and has been officially adopted by some state boards of health.

The original method and its modifications consist in an attempt to measure the phenol-coefficient, or relative killing-power of disinfectants upon certain bacteria, under standard conditions, compared with phenol. Briefly stated, a coefficient is "a number or known quantity prefixed in algebra as a multiplier to a variable or an unknown quantity." The phenol or carbolic coefficient of a disinfectant is determined "by dividing the figure indicating the degree of dilution of the disinfectant that kills an organism in a given time, by that expressing the degree of dilution of the phenol or carbolic acid that kills the same organism, in the same time, under exactly similar conditions."

In determining the Rideal-Walker coefficient, the technical procedure is substantially as follows:

"Phenol solutions of known strength are used; cultures are grown in a standard medium, transplants being made every 24 hours; the loops used for all inoculations are of a standard size (about 4 mm. in diameter). Usually four dilutions of suitable strengths of the disinfectant to be used are made. Phenol controls of a suitable strength are also prepared. Five cc. of each of these dilutions are placed in sterile test tubes, to which are added at intervals of one-half minute a 24-hour broth culture of B. typhosus in the proportion of 1 drop of culture to each cubic centimeter of disinfectant used.

"At the end of two and a half minutes a loopful of each of the mixtures is inoculated into a test tube containing 5 cc. of standard broth, an interval of half a minute being thus allowed between taking the samples from the different dilutions. This is repeated at 5, 7½, 10, 12½ and 15 minutes. The broth tubes, after being incubated at 37° C. for 48 hours, are examined for growth.

"The results of the examination are then noted, and if suitable, comparative strengths of the disinfectant and phenol have been selected, the phenol coefficient is determined as above stated."

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Although the toxic power of phenol is taken as the unit of comparison, it is influenced, to a certain extent, by conditions, or in other words, it is not a constant unit.

The conditions that influence results are: "Organisms to be acted upon; number of micro-organisms and amount of organic matter to be added; strength and number of dilutions; time during which the distinfectant is allowed to act, and temperature." (Tr. B. P. C. vide Journ. A. Ph. A., 1912, 637).

By this method of testing disinfectants, no arbitrary standard of phenol toxicity upon bacteria is assumed, but the killing-power of a disinfectant upon bacteria is compared with the killing-power of phenol upon the same bacteria, under the same conditions, so that if the bacteria are of a "weak strain" or a "strong strain," the results of the test are comparative, because they have been made under exactly analogous conditions.

If, however, a "weak strain" of bacteria be used as a standard for a series of tests, and a "strong strain" for another series, the results of the test as to the coefficients will be somewhat different. But such variations are equalized by dividing the figure representing the percentage strength of the weakest killing solution of the phenol, by the figure representing the percentage strength of the weakest killing solution of the disinfectant tested, both at 2½ and at 15, or 30 minutes. The mean resulting figure is assumed to be the true coefficient.

The nature of the test-organism has a great deal to do with the results obtained. With different species of organisms the coefficients obtained may vary as much as 300 percent. Walker and Rideal and Anderson and McClintic use the typhoid bacillus, the London Lancet the colon bacillus, and the Department of Health of the State of Maryland (which has a law directing that the labels of disinfectants shall give their coefficient value) specifies the use of either the typhoid or colon bacillus; though it is but fair to state that Charles Caspari, Jr., State Food and Drug Commissioner of Maryland, advises the writer that "in view of the fact that investigations during the past year have shown the great desirability of conforming the test to one specific bacillus, I think that a change in our regulations should and will be made very shortly."

The method originally devised, did not specify the use of organic matter in making the test, but it has been modified so that the test may be made with or without organic matter. Various forms of organic matter have been tried; peptone and gelatin are recommended by Anderson and McClintic.

The test *without* organic matter yields higher results than *with* organic matter. Thus, the phenol coefficient of Phenol Liquid (U. S. P. 1890), *without* organic matter is 177, and *with* organic matter is 176; of Crude Carbolic Acid (Navy Department) is 2.75 and 2.63, of Cresol is 2.90 and 1.75, and of Compound Solution of Cresol is 3.00 and 1.87, respectively.

The phenol coefficient method of testing disinfectants apparently marks a distinct step forward in methods of testing disinfectants, but while it has important possibilities, it has its limitations.

Woodward and Kingsett state that: "While the phenol coefficient method may serve to determine the relative germicidal value of similarly prepared preparations of a coal tar nature, it is not applicable for ascertaining the real or relative

values of other disinfectants of a different chemical nature." (Woodward and Kingsett, Trans. B. P. C., vide Journ. A. Ph. A. 1912, 637.)

During the past year, a number of bills have been introduced into different State legislatures, not only for standardizing disinfectants by the Anderson and McClintic method, but also for standardizing by the same method, deodorants, antiseptics and germicides; and also providing that the labels of such must be marked with the phenol coefficient in every case, and failure to do so shall be considered a misdemeanor subject to fine for each offense. In every case, fortunately, the bills have been killed or vetoed, and the reasons are obvious. It was assumed that these four classes of products could all be standardized by the phenol coefficient method, and such is not the case.

A disinfectant is a substance that destroys the cause of infection, such as phenol and compounds of a similar type.

A deodorant is a substance that destroys the odors or effects of bacterial action. It is not necessarily a disinfectant. Thus, charcoal is a deodorant, but it does not destroy bacteria, and has no coefficient value. Sulphur is a deodorant, but it is not a disinfectant until burned and converted into sulphurous acid gas.

Formaldehyde is both a deodorant and a disinfectant, while corrosive sublimate is a powerful germicide but not a deodorant.

A germicide is a substance that kills germs. It is synonymous in meaning with disinfectant.

An antiseptic is a substance that inhibits or prevents the growth and development of bacteria, but it does not kill bacteria, and can have, therefore, no coefficient value. Saturated solutions of salt or sugar will preserve meat or vegetable substances from decomposition and decay, that is they are antiseptic in action, but they are not germicidal.

The class of antiseptics embraces a long list of substances which are of material importance in practical medicine and surgery. There are many conditions of the human body in which it is desirable to restrain or prevent bacterial action, and yet in which the use of germicides and disinfectants are contradicted by reason of their corrosive action. The list of antiseptics embraces such commonly used substances as Boric Acid, Iodoform, Bismuth Subiodide, Naphthalene, Salol, Menthol, Thymol, Guaiacol, Acetanilide, etc.

Many antiseptics are insoluble in water and cannot be tested against organisms until made soluble, and even then if such a test could be made, it would be valueless, because it would not represent the body-conditions under which such antiseptics act. Iodoform is a striking example. Iodoform is of recognized value in the treatment of wounds. It is insoluble in water. Hehn and Rosving (Chem. News, 55), state that "sterilized iodoform jelly, when inculcated with microorganisms, was found to be full of them, all growing freely on the third day." Bouillat (Zeitsch f praktisch Chem. 25, 300), finds that 10 percent of iodoform does not check putrefactive change in pancreas. But it is an unquestioned clinical fact that iodoform applied to a body-wound prevents putrefaction and promotes granulation and cicatrization, and this is probably because the wound-secretions decompose the iodoform into iodine products that cause sterility. And what is true of iodoform, as an antiseptic, in the treatment of wounds, is probably true of other insoluble antiseptics.

Boric acid is a most widely used antiseptic for the treatment of the eye conditions, and yet its solution (1 to 100) does not kill typhoid bacilli even after 15 minutes.

Hydrogen Peroxide is one of the most largely used antiseptics, and yet its germicidial powers are so weak, compared with phenol, that the determination of its coefficient is admittedly impracticable. (Bulletin No. 82, Hygienic Laboratory, 1912, 65).

The Hygienic Laboratory Method of standardizing disinfectants, with and without organic matter, has been adopted by the Council of Pharmacy and Chemistry of the American Medical Association, and it is very probable that the use of this method will become general in the United States, and displace other methods. It is not a perfect method and is not claimed to be, but its use within certain limitations (that is, applied only to disinfectants of the coal tar group), will do much to standardize a very variable group of commercial products.

THE EFFECT OF PARAFORMALDEHYDE, PHENOL AND CREOSOTE ON THE DIGESTIVE ACTION OF PEPSIN, PANCREATIN AND DIASTASE.*

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Paraform, the crystallized, polymeric form of formaldehyde, has for a considerable number of years been to a slight extent employed in doses of from 5 to 15 grains as an intestinal antiseptic. Owing to the readiness with which formaldehyde is liberated from paraform, and the well-known disturbing action of this vigorous gas on digestive processes and its tendency to harden tissues and render protein substances insoluble, the use of paraform in these heroic doses has been limited.

Our interest was recently directed to a more careful study of effect of paraform on the digestive ferments by a suggestion from Dr. Walter J. Freeman of Philadelphia, who wished to employ this drug in small doses in the form of lozenges or pastilles. Some tablets were prepared, each containing Paraform ¼ grain, Sodium Bicarbonate 2½ grains, Talcum 1 grain, Sugar q. s. 20 grains, Oil of Peppermint q. s. to flavor, and tests were then made to determine what interfering effect, if any, these would have on the artificial digestion of proteids and starch.

As we wished to have something with which to compare the paraform, we also ran tests in which the effects of phenol and of creosote were studied. In order to make the experiments comparable as nearly as possible to its use in the case of patients under normal food conditions, we considered the average amount of egg albumin taken at one time as a "food dose" to be that contained in two eggs, or about 30 grams. In the tests with starch, 22.5 to 24 grams of dried starch were taken as normal food dose. The amounts of paraform, phenol

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