bacteriostatic, but more actively bactericidal. Although Gram-negative bacteria are as a rule more resistant against the actinomycin than the Gram-positive forms, there is no sharp line of demarcation between the two groups. Marked differences in sensitivity exist between the bacteria within each group.

11769

Germicidal Efficiency of Some Medicinal Dyes Compared to a Group of Non-Dye Disinfectants.

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The phenol coefficient test is employed universally for rating disinfectants. The test was designed originally for the evaluation of germicides which were closely related to phenol in structure. However, the test is of little value for determining the efficiency of compounds which are unlike phenol. Also, the test gives no information concerning the degree of inactivation of a germicide in the presence of organic matter, nor its toxicity towards living tissue.

In a series of papers published over a number of years[1] a new method was proposed and employed for the evaluation of germicidal substances intended for clinical application. The germicides were tested for their effect on living embryonic chick heart tissue cultivated *in vitro* as well as for their ability to kill bacteria. All tests were performed at 37°C and in the presence of a standard amount of organic matter. A number known as the toxicity index was determined which was defined as the ratio of the highest dilution of germicide required to kill the tissue in 10 minutes to the highest dilution required to kill the test organism in the same period of time under identical conditions. The smaller the toxicity index the more nearly perfect the chemotherapeutic agent. Theoretically, an index less than one means that the germicide is more toxic to bacteria than to the embryonic tissue while an index greater than one means that the germicide is more toxic to the embryonic tissue than to the bacteria.

Some of the dyes, especially those belonging to the triphenylmethane group, are used clinically and strong claims have been made

[1] Salle, A. J., Shechmeister, I. L., and McOmie, W. A., *J. Bact.*, 1939, **37**, 639.

for their efficiency. At one time they were employed to a considerable extent but they have been largely replaced by other germicidal agents.

Some of the characteristics of dyes are: (1) They are strongly bacteriostatic. This property, more than their germicidal action, probably explains the favorable results obtained from their use; (2) they are markedly inactivated in the presence of organic matter. This property is more pronounced in the case of the dyes than in the non-dye compounds; (3) they are relatively unstable in solution and especially in the presence of light; (4) the triphenylmethane dyes and probably others are more effective against Gram-positive than Gram-negative bacteria.

A number of medicinal dyes were rated by this method[1] and the toxicity indices determined. The organisms used were *Staphylococcus aureus* and *Eberthella typhosa*. The results are given in Table I. It is seen that with the exception of brilliant green the dyes tested were unable to kill *E. typhosa* in 10 minutes at 37°C, in the presence of a standard amount of organic matter. The efficiency of brilliant green is very poor, the index showing that it is 161 times more toxic to tissue than to *E. typhosa* under the conditions of the test. However, the dyes rated better against *Staphylococcus aureus* (Gram positive) than against *E. typhosa* (Gram negative). All of the triphenylmethane dyes killed *Staph. aureus* and the toxicity indices were much lower in every case. It may be seen that there is no relationship between toxicity indices and phenol coefficients.

TABLE I.
Toxicity of Dyes to Chick Heart Tissue and Bacteria.

Compound	Killing dilution: Tissue = A	Killing dilution: Bacteria = B	Toxicity Index = A/B	Phenol Coefficient
	Eberthella typhosa.			
Brilliant green	33,600	208	161	1.1
Crystal violet	60,800	will not kill	....	....
Methyl " B	25,000	" " "	....	....
" " 6B	25,000	" " "	....	....
" "	25,000	" " "	....	....
Methylene blue	7,000	" " "	....	....
Proflavine	3,200	" " "	....	....
Acriflavine base	3,000	" " "	....	....
" HCl	2,700	" " "	....	....
	Staphylococcus aureus.			
Brilliant green	33,600	5,500	6.0	50.0
Crystal violet	60,800	2,800	21.8	25.5
Methyl " B	25,000	1,200	20.8	10.9
" " 6B	25,000	1,150	21.4	10.5
" "	25,000	1,150	21.4	10.5
Methylene blue	7,000	will not kill	....	....
Proflavine	3,200	" " "	....	....
Acriflavine base	3,000	" " "	....	....
" HCl	2,700	" " "	....	....

TABLE II.
Toxicity of Germicides to Chick Heart Tissue and Bacteria.

Compound	*Eberthella typhosa* Killing dilution: Tissue A	Bacteria B	Toxicity Index A/B	Phenol Coefficient	Compound	*Staphylococcus aureus* Killing dilution: Tissue A	Bacteria B	Toxicity Index A/B	Phenol Coefficient
Halogens:					Halogens:				
Azochloramid	7,800	101,000	0.08	543	Iodine	650	3,520	0.2	32
Iodine	650	3,370	0.20	18	Azochloramid	7,800	26,000	0.3	236
Chloramine	900	2,000	0.45	11	Chloramine	900	2,000	0.45	18
Silver compounds:					Phenols:				
Silver lactate	100	690	0.15	3.7	Hexylresorcinol	1,450	1,620	0.9	15
" citrate	610	4,500	0.14	24	P-hydroxy phenyl-n-amyl sulfide	2,800	2,700	1.0	25
" nitrate	140	1,250	0.11	6.7	O-n-hexylphenol	3,125	2,900	1.1	26
" protein strong	25	175	0.14	1.0	P-hydroxy-di-phenyl sulfide	3,125	2,500	1.3	23
Silver protein mild	30	70	0.43	0.4	Phenol	224	110	2.0	—
Phenols:					Silver compounds:				
Hexylresorcinol	1,450	1,770	0.8	9.5	Silver lactate	100	110	0.9	1.0
P-hydroxy phenyl-n-amyl sulfide	2,800	2,700	1.0	14.5	" citrate	610	600	1.0	5.5
O-n-hexylphenol	3,125	2,800	1.1	15.0	" nitrate	140	80	1.8	0.7
P-hydroxy diphenyl sulfide	3,125	3,125	1.0	17.0	" protein strong	25	15	1.7	0.14
Phenol	224	186	1.2	—	Silver protein mild	30	12*	2.5+	0.11—
Mercurials:					Mercurials:				
Metaphen	2,070	5,660	0.4	30.5	Metaphen	2,070	1,370	1.5	13
Mercurochrome	90	140	0.6	0.8	Mercurochrome	90	12½*	7.2+	—
Merthiolate	4,220	2,660	1.6	14.0	Merthiolate	4,220	25*	169.0+	—

*Failed to kill. A more concentrated solution could not be prepared.

For purposes of comparison results of tests on other germicides are included (Table II). It may be seen that the dyes rate very poor in comparison to some well-known germicides of a non-dye nature.

Summary. The dyes as a group are extremely poor germicidal agents. Only the triphenyl methane dyes were capable of killing *E. typhosa* and *Staph. aureus.* Brilliant green was the only dye that killed *E. typhosa* while several killed *Staph. aureus* but their extremely high toxicity to tissue showed that they are inferior to the better known non-dye germicides. Further work on the dyes is now in progress.

11770

The Relation of the Nervous System to Skin Potentials in the Intact Frog.

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Considerable has been written about the relation of nerve stimulation to the action potential across the frog's skin; but, to our knowledge, little is known concerning the influence of the brain and spinal cord on the resting potential of the intact animal. Roeber[1] was the first to note a definite negative variation across the frog's skin following nerve stimulation. Later Engelmann,[2] Hermann[3] and Bayliss and Bradford[4] showed an association between the degree of response to nerve stimulation and the secretory activity of the gland cells of the skin, as well as seasonal variations. Furthermore, Orbili[5] investigated in a similar way the influence of various concentrations of salts. Although these observations are of significance, it should be pointed out that they were performed either on the isolated nerve skin or after the frogs were pithed or curarized.

Since our observations show that injury to the brain and spinal cord causes definite changes in the resting potential across the skin in the intact animal we feel justified in reporting our results. Although Francis and Pumphrey[6] report no pronounced difference be-

1 Roeber, H., *Arch. f. Anat. u. Physiol.*, 1869, 633.

2 Engelmann, W., *Pfluger's Arch.*, 1872, **6**, 97.

3 Hermann, L., *Pfluger's Arch.*, 1878, **17**, 291.

4 Bayliss, W. H., and Bradford, J. R., *J. Physiol.*, 1886, **7**, 217.

5 Orbili, A. L., *Z. f. Biol.*, 1910, **54**, 329.

6 Francis, W. L., and Pumphrey, R. J., *J. Exp. Biol.*, 1933, **10**, 379.