

usually observed. As in the case of other types of local anesthetics, hypnotics protect against nupercaine injected subcutaneously, increasing the lethal dose about 3 times, but on intravenous injection, little protection is obtained.

Absorption of nupercaine when given subcutaneously is very slow, but it is 12 times more toxic than cocaine when given in that manner, and only 6 times more toxic if given intravenously. This indicates that, compared with cocaine, nupercaine attacks the central nervous system more readily than the heart. All the phenomena observed in this study are in accord with that explanation of intoxication from local anesthetics which has already been discussed.⁵

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Preservatives for Bacteriophage Suspensions.

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That bacteriophage possesses therapeutic value in certain types of bacterial infections is becoming more and more evident.¹ The increasing demand for 'phages intended for therapeutic use raises a question, applicable to all biological products, as to the choice of a suitable preservative. While filtration through a sound candle generally fulfills the requirements as to sterility, it is well known that this is not absolute proof against the development of so-called "secondary cultures". Besides this there is also the possibility of accidentally introducing organisms in ampouling the filtrate for shipment.

In choosing preservatives for 'phage products 2 considerations must be borne in mind: (1) The preservative must be bactericidal without in any way impairing the potency of the 'phage and (2) the concentration of the germicide which is employed should be such that it will not be toxic for the patient in the amounts which would be administered with large doses of 'phage. In studies designed to elicit the value of 'phage as a therapeutic agent, a third consideration enters. Here it is important to reduce the concentration to a level which will insure bacteriostasis in the ampoule and will not, on further dilution by body fluids (urine, etc.) after admin-

¹ Schultz, E. W., *California and Western Medicine*, 1932, in press.

istration of the 'phage, exercise an appreciable bactericidal effect *in vivo*.

Of a number of substances tested merthiolate,* metaphen, and potassium mercuric iodide seemed essentially to fulfill the above requirements. Jamieson and Powell² have reported that merthiolate in a concentration of 1-5,000 has little effect on the activity of an antistaphylococcus bacteriophage at room or ice box temperature. They also reported that "bacteriophage containing as much as 1% merthiolate for several weeks produces high titered lysing of fresh cultures."

In the studies reported below a freshly prepared solution of merthiolate prevented growth in a dilution of the powdered reagent of 1-1,000,000 in heavily inoculated broth cultures of *Esch. coli* and *S. aureus*. The bacteriostatic activity of this solution of merthiolate was reduced to 1-100,000 after standing several weeks. Metaphen prevented growth in a dilution of 1-100,000 and potassium mercuric iodide, 1-10,000.

Results obtained with various concentrations of the above agents

TABLE I.

'Phage	Days Incubation	Visible Lysis in Dilution of			
		Merthiolate 1-10,000	Merthiolate 1-100,000	Metaphen 1-100,000	K ₂ HgI ₄ 1-10,000
Antistaph. No. 15 AD-52	0	10 ⁻⁹	10 ⁻⁹	10 ⁻⁹	10 ⁻¹⁰
	5	10 ⁻⁶	10 ⁻⁹	10 ⁻⁸	10 ⁻⁸
	10	10 ⁻³	10 ⁻¹⁰	10 ⁻⁹	10 ⁻⁹
	20	10 ⁻⁴	10 ⁻⁹	10 ⁻⁹	10 ⁻⁹
	30	10 ⁻²	10 ⁻⁸	10 ⁻⁹	10 ⁻⁹
Anticoli No. 2 K-13	0	10 ⁻⁸	10 ⁻⁸	10 ⁻⁸	10 ⁻⁸
	5	10 ⁻⁷	10 ⁻⁸	10 ⁻⁷	10 ⁻⁸
	10	10 ⁻⁷	10 ⁻⁸	10 ⁻⁸	10 ⁻⁷
	20	10 ⁻⁶	10 ⁻⁸	10 ⁻⁸	10 ⁻⁸
	30	10 ⁻⁵	10 ⁻⁸	10 ⁻⁷	10 ⁻⁷
Anticoli No. 77 K-199	0	10 ⁻⁶	10 ⁻⁶	10 ⁻⁶	10 ⁻⁶
	5	10 ⁻⁵	10 ⁻⁷	10 ⁻⁵	10 ⁻⁵
	10	10 ⁻⁶	10 ⁻⁵	10 ⁻⁵	10 ⁻⁴
	20	10 ⁻⁴	10 ⁻⁶	10 ⁻⁶	10 ⁻⁵
	30	10 ⁻³	10 ⁻⁶	10 ⁻⁵	10 ⁻⁵
Anticoli No. 98 K-222	0	10 ⁻⁵	10 ⁻⁵	10 ⁻⁵	10 ⁻⁵
	5	10 ⁻⁴	10 ⁻⁵	10 ⁻⁵	10 ⁻⁴
	10	10 ⁻⁴	10 ⁻⁴	10 ⁻⁴	10 ⁻⁵
	20	10 ⁻³	10 ⁻⁴	10 ⁻⁴	10 ⁻⁴
	30	0	10 ⁻⁴	10 ⁻⁴	10 ⁻⁴

* I am indebted to Dr. W. A. Jamieson of Eli Lilly and Company for supplying me with the merthiolate used in this work.

² Jamieson, W. A., and Powell, H. M., *Am. J. Hyg.*, 1931, **14**, 218.

in antistaphylococcus and anticoli bacteriophages incubated for one month at 37.5°C. are reported in Table I, the activity of the 'phage suspension being determined by periodic titration during this period. No secondary cultures developed in any of the chemically treated suspensions while secondary cultures developed in several of the filtered 'phage controls.

These results indicate that merthiolate can be added in a concentration of 1-100,000, metaphen 1-100,000, or potassium mercuric iodide 1-10,000 to suspensions of 'phages active against *Esch. coli* or *Staph. aureus* without reducing the titer of the 'phage on standing and at the same time preventing the development of secondary cultures.

At 37.5°C. the results reported here with merthiolate do not agree with those reported by Jamieson and Powell for tests carried out at a lower temperature. Results given in Table I show that the activity of the various bacteriophages tested was slowly reduced by merthiolate in a concentration of 1-10,000. Jamieson and Powell reported that merthiolate in a dilution of 1-5,000 did not markedly affect the 'phage in one year at room or ice box temperature. In these tests at 37.5°C. merthiolate in a concentration of 1-1,000 completely inactivated an antistaphylococcus bacteriophage in less than 2 weeks.

By the use of such preservatives it should, therefore, be possible for the physician located at a distance from the bacteriophage supply laboratory to maintain an efficient supply of various polyvalent 'phages available for use in emergency or routine therapeutic use of bacteriophage.

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A Method for the Collection of Blood from Rats.

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Various routes have been used for obtaining blood from rats. Some of these are: the heart, femoral vein, and the tail. All these routes have been found unsatisfactory for our work. The heart puncture is too dangerous since the number of accidents and deaths is very large. The tail was found unsatisfactory since the amount of blood one could get was small. It also exposes the animal to in-