zene and petroleum ether; easily soluble in alcohol and water. The sulphate is fairly hydroscopic.

Calculated for $(HOCH_2CH_2N(CH_3)_3)_2SO_4$: S, 10.52 per cent.
Found: S, 10.63, 10.77 per cent.

Choline Dihydrogen Phosphate, $HOCH_2CH_2N(CH_3)_3H_2PO_4$.—On agitating a concentrated solution of choline chloride with silver phosphate and filtering from the excess of silver phosphate and silver chloride, a very basic solution was obtained, indicating that the tricholine phosphate was largely hydrolyzed. A dilute solution of phosphoric acid was added to permanent acidity. The ratio of phosphate ions to the choline was then practically 1 : 1. The solution was evaporated under high vacuum at 80–90° to a horny mass. The product was purified by extracting with boiling absolute alcohol in which the phosphate is slightly soluble. By this means beautiful, long white, fairly hydroscopic needle crystals were obtained, which were practicably insoluble in benzene, carbon disulphide, petroleum ether, acetone and ethyl ether; slightly soluble in alcohol; very soluble in water. After washing in ether and drying in a vacuum desiccator the product was analyzed.

Calculated for $HOCH_2CH_2N(CH_3)_3H_2PO_4$: P, 15.42 per cent.
Found: P, 15.48 per cent.

The chemistry of the choline derivatives is being further investigated by the author.

WESLEYAN UNIVERSITY, MIDDLETOWN, CONN.

BACTERICIDAL PROPERTIES OF LECITHINS AND CHOLINE SALTS.

BY R. R. RENSHAW AND K. N. ATKINS.

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There is some disagreement in the literature concerning the bactericidal properties of lecithins. The existence of cobra lecithides, the speculation regarding a possible relation between lecithin and the amboceptors,[1] and, among others, the fact that such a product as fresh milk, containing certain amounts of lecithins, has some germicidal constituent, gives interest to the problem in question. In view of this the authors have attempted to determine what influence, if any, lecithins might have upon the growth of some of the more common organisms. Along with this, since it is possible that the presence of a decomposition product, *i. e.*, choline, might have been responsible for the disagreement, a few of its salts were prepared and their bactericidal properties determined.

The procedure was as follows: Pure cultures of the different organisms were grown at room temperature for about twelve hours in an ordinary broth medium. One loopful of this was transferred to a flask con-

[1] Kyes, *Z. physiol. Chem.*, **41**, 273.

taining about 50 cc. of the same kind of medium. After a thorough shaking this second broth culture was divided among small bottles holding 9 cc. of the medium. In the experiments on milk the raw fresh product was used. To the different bottles of the culture or milk 1 cc. of a 4 per cent. emulsion of lecithin or 4 per cent. solutions of the different choline salts were added. One bottle was used as a control. After the organisms had developed 12 hours, 1 cc. of the medium was withdrawn from each bottle by means of a sterile pipette and transferred to sterile water blanks of either 9 or 99 cc., making a dilution of 10 or 100. The water blanks were then thoroughly shaken and if higher dilutions were to be used, as was true in most cases, 1 cc. of the diluted culture was transferred to a second water blank and again thoroughly shaken. In this way the proper dilution was reached. Anticipating from other experiments the approximate number of bacteria which would develop under given conditions the dilution was so calculated as to allow accurate counting of the plates. Knowing that the number of bacteria in milk kept for 24 hours at 19° would be large (in fact the product was almost sour) the dilution here used was as high as 1,000.000. In all cases 1 cc. of this finally diluted culture was plated out in agar, incubated for 24--48 hours at 37° and counted. The agar medium used contained 1.2 per cent. agar-agar and its acidity was equivalent to 1 per cent. normal acid.

Commercially pure samples of lecithins were repurified by several precipitations from alcoholic solutions by means of acetone and subsequent drying in a vacuum desiccator. The concentration of the emulsions was low, about 0.4 per cent. This, however, is greater than is usually found in biological fluids.

With some of the organisms, especially typhoid, a large number of series were made. The following tables are composed of representative determinations. The figures in each are the average counts of ten to a dozen plates.

In several series the organisms were plated out after incubating 6 or 24 hours. It was found, however, that a twelve-hour period was the best.

B. LACTIS AEROGENES.

	12°.		19°.	
	0 hrs. × 1000.	12 hrs. × 1000.	0 hrs. × 1000.	12 hrs. × 1000.
Control	5.0	8.0	2.5	140.0
Lecithin	5.0	7.0	2.5	135.0
Choline chloride	5.0	8.7	2.5	128.0
Choline phosphate	5.0	5.4	2.5	132.0
STAPH. PYOGENES ALBUS.				
Control	1.4	7.0	1.3	15.0
Lecithin	1.4	6.0	1.3	12.0
Choline chloride	1.4	6.0	1.3	14.0
Choline phosphate	1.4	5.0	1.3	17.0

	12°.		19°.	
Milk.	0 hrs. x 1000.	12 hrs. x 1000.	0 hrs. x 1000.	12 hrs. x 1000,
Control	15.0	160.0	120.0	112,000.0
Lecithin	15.0	160.0	120.0	95,000.0
Lecithin (2)	15.0	100.0	.....	.....
Choline chloride	15.0	140.0	120.0	104,000.0
Choline phosphate	....	.....	120.0	108,000.0
Coli.				
Control	6.0	9.0	20.0	4,000.0
Lecithin	6.0	7.0	20.0	2,400 0
Choline chloride	6.0	10.0	20.0	3,500.0
Choline phosphate	6.0	9.0	20.0	3,200.0
Typhoid.	(15°).			
Control	23.0	290.0	4.8	137.0
Lecithin	23.0	260.0	4.8	113.0
Choline chloride	23.0	270.0	4.8	105.0
Choline phosphate	23.0	270.0	4.8	127.0

As indicated in the tables, there is, in general, a varying retardation in the development of bacteria in the cultures containing lecithins, although this is not marked in some cases. In fact, in looking over the records of individual plates, it was found that in about 10 per cent. of over two hundred plates no change at all was shown, or there was a very slight increase in the development. Nevertheless, we are inclined to attribute slight bactericidal properties to lecithins at these dilutions, although for practical purposes this is negligible.

In general an effect by choline salts was less frequently observed and with them the diminution was less marked.

WESLEYAN UNIVERSITY, MIDDLETOWN, CONN.
DEPARTMENT OF CHEMISTRY.

[FROM THE DIVISION OF CHEMISTRY, HYGIENIC LABORATORY, U. S. PUBLIC HEALTH AND MARINE HOSPITAL SERVICE, WASHINGTON, D. C.].

FURTHER STUDIES ON THE APPLICATION OF THE VOLHARD METHOD TO THE ESTIMATION OF ALKALOIDS.

BY ELIAS ELVOVE.

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It has been pointed out by the writer[1] that the difficulty experienced by many workers in obtaining suitable indicators in the alkalimetric estimation of alkaloids and the necessity for using a comparatively large number of such indicators, each more or less suitable to one or only a few alkaloids, may be completely overcome if we adopt the very simple modification of the usual procedure and substitute hydrochloric acid as

[1] Bull. 54, Hyg. Lab., U. S. Pub. Health and Mar. Hosp. Serv., Wash.