peutic possibilities of the lactone, these and the previously reported observations[2] are of considerable interest. Hypophyseal extracts of animal origin have not proved very satisfactory as a means of establishing normal ovarian activity in women with functional disturbances. In certain situations estrogens have been apparently beneficial, presumably because of their pituitary-stimulating effect. As a pituitary stimulant, the lactone should be very much more useful than the estrogens, first, because it is practically non-estrogenic and would not be expected to cause such undesirable side effects as excess cervical secretion, breast changes and endometrial hyperplasia. Secondly, since, unlike estrone, it acts directly on the pituitary and is not influenced by the steroids (*viz.*, progesterone and testosterone[2]), which depress estrogen inactivation, the response to it would presumably be uninhibited by the patient's own secretion of steroid hormones. Finally, according to the above experiments, its prolonged administration should have no depressant effects upon pituitary or ovarian activity. As an inhibitor of F.S.H., on the other hand, it would presumably be ineffective.

Summary. Westerfeld's lactone, an oxidative inactivation product of crystalline estrone, stimulates the release of luteinizing hormone from the pituitaries of immature female rats, eliciting this response in smaller dosages than does estrone. Unlike estrone, it does not, upon prolonged administration, prevent the secretory or cytological changes in the pituitary which follow removal of the ovaries. Therapeutic implications are discussed.

15045 P

Effect of Fatty Acids on Acetylcholine Synthesis.*

CLARA TORDA AND HAROLD G. WOLFF.

From the New York Hospital and the Department of Medicine (Neurology) and Psychiatry, Cornell University Medical College, New York City.

More acetylcholine is synthesized by the frog brain *in vitro* in the presence of serum than in its absence.[1,2] Performance of muscle work modifies the ability of serum to increase the acetylcholine synthesis.[3] Since muscle may utilize fats during performance of work[4-19] the

* This investigation was aided by a grant from the John and Mary R. Markle Foundation.

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TABLE I.
Effect of the Substances on the Synthesis of Acetylcholine.

Substance	Amt of acetylcholine synthesized in % of control*				
	Amts of the substances (mg) added to 100 mg frog brain				
	3	0.3	0.03	0.003	0.0003
Glycerol	101	98	100	103	100
Saturated Fatty Acids:					
Formic acid	88	93	91	96	101
Acetic acid	102	101	104	102	102
Propionic acid	122	118	106	101	97
Butyric acid	131	119	110	102	102
Valeric acid	155	111	106	96	99
n-caproic acid	106	117	110	100	101
n-caprylic acid	64	96	97	103	101
Capric acid	53	80	104	98	102
Lauric acid	48	71	98	97	99
Palmitic acid	102	105	99	100	103
Stearic acid	91	102	97	103	100
Unsaturated Fatty Acids:					
Oleic acid	81	94	105	101	98
Linoleic acid	58	77	93	97	99
Aldehydes:					
Acetaldehyde	20	49	75	88	92
Glyceraldehyde	70	90	102	100	99
Ketones:					
Acetone	96	110	104	97	102
Acetoacetic acid	105	101	103	106	98
β-OH butyric acid	73	96	98	101	103

* Each value represents the average of 8 separate experiments. The S.E. of the mean for each value was less than $\pm 5\%$. The amount of acetylcholine synthesized in μg per 100 mg frog brain, followed by the S.E. of the mean, was: 1.50 ± 0.044.

effect of fatty acids on the acetylcholine synthesis was investigated in the following.

Method. The effect of the substances on the acetylcholine synthesis was studied by the method described previously.[2] Mixtures containing varying amounts of the substances (pH corrected to 7.4), 100 mg minced fresh frog brain, 3 mg physostigmine salicylate, 4.8 mg glucose, and 3 cc Ringer's solution were shaken and incubated aerobically for 4 hours at 37°C. After incubation the amounts of acetylcholine synthesized were assayed biologically on the sensitized rectus abdominis muscle of the frog.

Results. The amounts of acetylcholine synthesized are given in Table I. Butyric-, propionic-, valeric-, and *n*-caproic acids increased the synthesis. These results suggest that during utilization of butyric- and propionic acids by the muscle these acids may exert a beneficial effect on the locally occurring acetylcholine synthesis.

Higher fatty acids decreased the synthesis of acetylcholine; palmitic-, and stearic acids seemed to have no effect on the synthesis, however, the lack of effect may have been due to the low solubility of these acids.

Endproducts of fat and carbohydrate metabolism (acetic acid, acetoacetate, acetone) did not modify the acetylcholine synthesis.

The synthesis was decreased in the presence of the unsaturated acids, aldehydes, and the ketone β-hydroxy butyric acid. This decrease of acetylcholine synthesis, if it occurs in the body, may contribute to the genesis of easy fatiguability observed during administration of large amounts of β-hydroxy butyric acid or during severe ketosis.

These results suggest that unsaturated and higher fatty acids contained in the blood may depress the synthesis of acetylcholine but the lower fatty acids occurring in the muscle either increase it or do not modify the synthesis.

Summary. 1. The effect of fatty acids and related substances on the synthesis of acetylcholine was investigated. 2. The synthesis was not modified in the presence of glycerol, acetic-, palmitic-, and stearic acid, acetone,

and acetoacetic acid. 3. The synthesis increased in the presence of butyric-, propionic-, valeric-, and *n*-caproic acid. 4. The synthesis decreased in the presence of formic-, *n*-caprylic-, capric-, lauric-, oleic-, linoleic acid, glyceraldehyde, acetylaldehyde, and β-hydroxy butyric acid.

15046

Modification of Serological Response to Infection with Murine Typhus by Previous Immunization with Epidemic Typhus Vaccine.

HARRY PLOTZ* AND KENNETH WERTMAN.

From the Division of Virus and Rickettsial Diseases, Army Medical School, Washington, D.C.

It is well known that epidemic typhus cannot be differentiated from murine typhus by means of the non-specific Weil-Felix test since a high OX-19 agglutination titer may occur in both diseases. However, within the past few years, through improved technics and experimentation, both the specific complement fixation and agglutination tests have become of great value in differentiating these diseases. As was first shown in this laboratory, there is an antigenic fraction (soluble antigen) common to both types of rickettsiæ which reacts equally well in the complement fixation test with epidemic or murine antibodies.[1,2,3] However, by washing and centrifugation this antigen can be eliminated, leaving the purified rickettsial bodies. These latter constitute a highly specific antigen in the complement fixation test. Furthermore, these rickettsial suspensions have been found to be of great value as agglutinogen in the agglutination test. With technical improvements, recently described, this test has also been found to be of considerable value as a means of differentiating epidemic from murine infections.[4] As illustrations of the type of serological reactions found see Tables I and II which are representative cases selected from our series where the agent was isolated and identified and where serial serum specimens were available for examination.

It is noted that in epidemic typhus high complement-fixing titers are obtained with the purified epidemic rickettsial antigen and low or negative results with a purified murine rickettsial antigen. Conversely, in murine typhus high titers are obtained with a purified murine rickettsial antigen and low or negative results with a purified epidemic antigen. With regard to the rickettsial agglutination results higher titers are obtained with the homologous antigen than with the heterologous. However, it is to be noted that while there is cross agglutination the difference in titers is sufficient to permit clear differentiation.

During the past year we have received specimens from 147 cases of murine typhus occurring in soldiers stationed in the United States. Of these 135 gave a typical murine serological response comparable to that cited in Table II. However, 12 cases have given an unusual response. Table III records the serological results found in these cases.

In an attempt to determine the factors producing these atypical responses the 12 cases

* Member of the U. S. A. Typhus Commission.

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