It seems that the ketogenic amino acids increased the acetylcholine synthesis less than the non-ketogenic amino acids.

Summary. 1. The effect of amino acids, some polypeptides, and a few decomposition products on the synthesis of acetylcholine was investigated. 2. Most of the amino acids, carnosine, glutathione, and creatine increased the synthesis. 3. Glycine, leucine, and creatinine did not modify the synthesis. 4. Ammonia decreased the amount of acetylcholine synthesized.

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Effect of Some Isocyclic, Aromatic, and Heterocyclic Compounds on Acetylcholine Synthesis.*

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In the presence of body fluids of patients with myasthenia gravis less acetylcholine is synthesized than in the presence of body fluids of healthy subjects.[1,2,3] In the following it was ascertained whether some isocyclic, aromatic hydrocarbons, and heterocyclic compounds modify the synthesis of acetylcholine. These substances were chosen because some of them occur in the animal organism as intermediate or end product of the metabolism, some are widely administered for their therapeutic effect, and some are known to modify the activity of various enzymes.

Experimental. The effect of the substances on the synthesis of acetylcholine was studied by the method described previously.[2] The amounts of acetylcholine synthesized are given in Table I. Isocyclic (cyclohexane, inositol) and aromatic hydrocarbons (benzol, toluol, benzoic acid, naphthalene) either did not modify the synthesis of acetylcholine or decreased it only in higher concentrations (10^{-2} mols). Hydroxyl derivatives (phenol, *p*-aminophenol, hydroquinone, salicylic acid, α-naphthol, β-naphthol), aldehydes (benzaldehyde), and ketones (camphor, penicillin) decreased the synthesis in concentrations from 10^{-5} mol. Conjugated products either did not modify the synthesis or decreased it slightly (potassium phenol sulfonate, phenacetine, diphenylamine, benzidine, acetylsalicylic acid, sulfonamides). From the heterocyclic compounds indol decreased the synthesis in concentrations from 10^{-5} mol. Substitution in β position decreased this property of indol: skatol was somewhat less active depressor, indol-3-acetic acid was nearly inactive, *dl*-tryptophane was inactive, and *l* (—) tryptophane, the naturally occurring amino acid, increased the ability of frog brain to synthesize acetylcholine. Quinoline decreased the synthesis in concentrations from 10^{-5} mol.; carbazole decreased it to a lesser degree.

Discussion. The substances used differ in their chemical structure and their ability to enter into various chemical reactions but are known to inactivate proteins. Therefore, it is possible that the decrease of acetylcholine synthesis was due to a partial inactivation of the protein component of the enzyme involved in the synthesis. Many of the substances used are readily combined in the animal organism forming less toxic compounds. Conjugated substances depressed the synthesis of acetylcholine to a lesser degree than did the corresponding acids.

A correlation between the decrease of synthesis of acetylcholine *in vitro* and the physio-

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1 Torda, C., and Wolff, H. G., *Science*, 1943, **98**, 242.

2 Torda, C., and Wolff, H. G., *J. Clin. Invest.*, 1944, **23**, 649.

3 Torda, C., and Wolff, H. G., *Science*, 1944, **100**, 200.

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

Substance	No. of exp.	Amt of total acetylcholine synthesized in % of control*: Amt (mg) of substances added to 100 mg frog brain: 3	0.3	0.03	0.003
Inositol	11	98	99	98	105
Cyclohexane	8	88	98	100	101
Benzol	10	84	94	97	98
Toluol	7	109	108	100	104
Benzoic acid	8	80	90	96	103
Benzaldehyde	8	44	65	77	95
m-dinitrobenzene	6	58	75	80	90
Phenol	9	30	55	77	111
Potassium phenol sulfonate	8	104	97	99	102
Phenacetin	8	66	103	105	99
Sulfanilamide	8	113	117	114	107
Sulfapyridine	8	109	106	104	105
Sulfathiazole	8	100	106	107	104
Sulfadiazine	8	99	100	96	97
Diphenylamine	8	80	105	112	114
Benzidine HCl	8	67	97	105	106
p-aminophenol	9	52	69	111	109
Hydroquinone	10	40	71	98	98
Salicylic acid	9	35	67	79	87
Acetylsalicylic acid	9	113	105	105	102
Naphthalene	8	82	97	104	106
α-naphthol	8	21	50	81	93
β-naphthol	8	22	50	84	100
Quinoline	8	54	66	76	83
Indol	9	41	48	88	95
Skatol	8	51	57	88	99
Indol-3-acetic acid	7	75	100	103	103
dl-tryptophane	10	108	105	102	98
l(—)tryptophane	11	132	114	117	103
Carbazole	9	82	87	92	99
Camphor	11	31	78	96	105
		Amt of penicillin added in Oxford units: 15,000	1,500	150	15
Penicillin (Merck)	5	15	58	90	94

* The S.E. of the mean for each value was less than ±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.

logical effects of the substances in the living animal is not yet possible. However, symptoms occurring during intoxication by phenol, salicylic acid, camphor, and benzol have been described as resembling the symptoms occurring during atropine poisoning.[4] It is conceivable that some of the symptoms are due to a lack of acetylcholine.

Sulfonamides and acetylsalicylic acid, in the concentrations used, did not modify the synthesis of acetylcholine. Penicillin, an unsaturated ketone, depressed the synthesis probably because of its ability to react with the sulfhydryl group,[5] an active group of the enzyme involved in the synthesis of acetylcholine.

[4] Goodman, L., and Gilman, A., *The Pharmacological Bases of Therapeutics*, Macmillan Co., New York, 1941.

[5] Geiger, W. B., and Conn, J. F., *J. Am. Chem. Soc.*, 1945, **67**, 112.