SYNTHESIS AND BIOLOGICAL STUDY OF PYRACETAM DERIVATIVES AND RELATED COMPOUNDS

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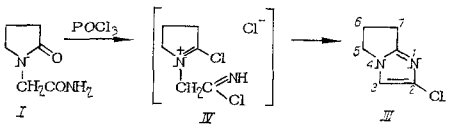
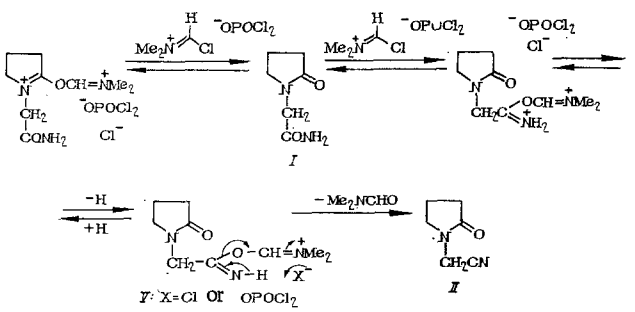
Pyracetam (I) belongs to the new group of psychotropic preparations, viz., the so-called nootropes. Compounds of this group activate the function of the central nervous system (CNS) and increase the resistance of the brain to hypoxia and toxic effects. Pyracetam has found wide application as a medicinal agent in neurological, psychiatric, and narcological practice.

In searching for new nootropic preparations we have carried out the synthesis and pharmacological study of a number of compounds obtained by modification of the pyracetam molecule. To achieve this modification in the present research we used methods that are widely employed in the chemistry of amides and lactams and are based on activation of their amide function by conversion to an amidoacetal function.

Activation of the lactam function of I itself with retention of the carbamido group is hardly possible. It therefore seemed expedient to use 1-cyanomethyl-2-pyrrolidone (II) as the starting compound; the 1 position of this lactam contains a small cyanomethyl substituent, which in subsequent stages of the synthesis can be converted to an N-carbamidomethyl group, which is a fragment of I. The currently extremely accessible I was used as the starting compound for the synthesis of II in the present research. We found that the conversion of I to II cannot be realized by means of ordinary dehydrating agents such as phosphorus pentoxide, polyphosphoric acid, and phosphorus oxychloride. Dehydration (along with replacement of the oxo group by chlorine) occurs formally in the latter case but gives a pyrrolino[1,2-a]imidazole derivative (III) rather than the expected nitrile II. Cyclization probably proceeds through amide chloride-imide chloride derivative IV:



The structure of III follows unambiguously from its PMR spectrum ($CDCl_3$), in which signals of 6-CH_2, 7-CH_2, and 5-CH_2 groups at 2.61 (m), 2.77 (m), and 3.95 ppm, respectively, and a 3-CH singlet at 6.76 ppm are observed. The synthesis of nitrile II was accomplished by the method in [1] by reaction of amide I with the Vilsmeier reagent; II was obtained smoothly in good yield in this case. The reaction evidently proceeds via initial attack at the carbonyl group of the primary amide function. The product of attack at the lactam carbonyl may also participate in the reverse process; however, irreversible stabilization of cation V, which proceeds with splitting out of dimethylformamide, shifts the equilibrium to favor the formation of cyanomethyl derivative II:



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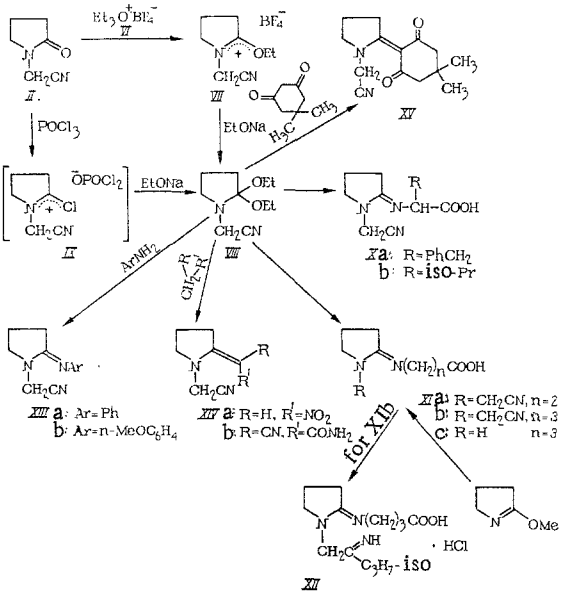
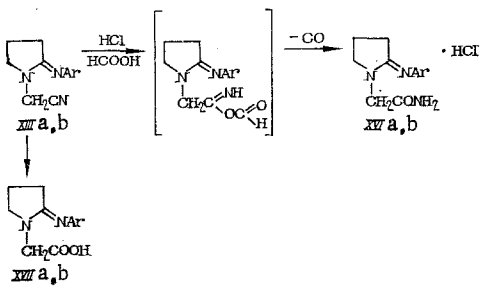
Alkylation of nitrile II with triethyloxonium tetrafluoroborate (VI) with subsequent exchange decomposition of intermediate tetrafluoroborate VII with sodium ethoxide leads to N-cyanomethyl-2-pyrrolidone diethylacetal (VIII). The same acetal can be obtained (although in considerably lower yield) by O-acylation of lactam II with phosphorus oxychloride with subsequent reaction of intermediate amide chloride IX with sodium ethoxide. Acetal VIII, like other lactam acetals, is an extremely reactive compound and reacts readily with various amines and compounds that have an active methylene link to give amidines and enamines, respectively.



In the first stage acetal VIII was subjected to reaction with a number of amino acids (we assumed that the presence of an amino acid fragment might promote better penetration of the subtances through the biological barriers through the utilization of "amino acid transport," at least for those compounds that contain α-amino acid fragments). We selected reaction conditions that made it possible to carry out amide synthesis without appreciable esterification of the acidic group, and the corresponding amidino acids (Xa, b, and XIa, b) were isolated in satisfactory yields. It should be noted that the NH analog (XIc) of amidino acid XIb, which was obtained by reaction of O-methylbutyrolactim with γ-aminobutyric acid, was also synthesized for biological testing. It should also be noted that the CN group of XIb is readily converted to an imido ester when XIb is heated in isopropyl alcohol in the presence of alcoholic HCl. As a consequence of this, the preparation of the hydrochloride of XIb must be carried out with adequate cooling, for otherwise the yield of XIb•HCl decreases due to a side reaction involving the formation of imido ester XII. Lactam acetal VIII also reacts readily with aromatic amines to give N-arylamidines (XIIIa, b) and with C–H acids such as nitromethane, cyanoacetamide, and dimedone to give enamines (XIVa, b and XV).

We were able to obtain N-aryliminopyrrolidines XVIa, b, which contain a carbamidomethyl group in the 1 position of the ring, by a relatively new method of conversion of a cyano group to a carbamido group [2] by passing gaseous hydrogen chloride into solutions of the nitriles in formic acid. The hypothetical scheme of this process is presented below.



Treatment of amidines XIIIa, b with concentrated HCl at room temerpature leads to N-carboxymethyl derivatives XVIIa, b.

TABLE 1. Physicochemical Properties of the Synthesized Compounds

Compound	mp or bp, °C/mm (mercury column)	Found, %				Empirical formula	Calculated, %				Yield, %
		C	H	N	Cl		C	H	N	Cl	
II	117/3	57,90	6,47	22,58	—	$C_6H_8N_2O$	58,06	6,45	22,58	—	77
III	73—6	50,50	4,98	19,40	25,15	$C_6H_7N_2Cl$	50,34	4,95	19,65	24,86	84
VII	64—5	39,69	5,51	11,66	—	$C_8H_{13}N_2OBF_4$	40,03	5,46	11,67	—	86
VIII	92—5/2	60,65	9,32	14,60	—	$C_{10}H_{18}N_2O_2$	60,58	9,15	14,13	—	82*
Xa†	203—4	64,55	6,55	15,52	—	$C_{15}H_{17}N_3O_2 \cdot 0{,}5H_2O$	64,27	6,47	14,99	—	82
Xb	199—200	59,20	7,90	19,01	—	$C_{11}H_{17}N_3O_2$	59,17	7,67	18,82	—	72
XIa‡	129—31	47,50	7,40	19,02	—	$C_9H_{13}N_3O_2 \cdot 1{,}75H_2O$	47,67	7,34	18,53	—	58
XIb·HCl	185—6	48,87	6,72	17,19	14,60	$C_{10}H_{16}N_3O_2Cl$	48,88	6,56	17,10	14,43	76
XIc	155—8	...	...	16,28	—	$C_8H_{14}N_2O_2$	...	...	16,46	—	58
XII	179—81	51,10	7,95	14,10	11,45	$C_{13}H_{24}N_3O_3Cl$	51,06	7,91	13,74	11,59	16
XIIIa	41—3	72,26	6,54	21,04	—	$C_{12}H_{13}N_3$	72,33	6,58	21,09	—	89
XIIIa·HCl	139—41	...	...	18,13	15,18	$C_{12}H_{14}N_3Cl$	...	...	17,83	15,04	68 **
XIIIb	75—6	68,25	6,48	18,54	—	$C_{13}H_{15}N_3O$	68,10	6,59	18,33	—	96
XIVa	101—3	50,64	5,30	25,44	—	$C_7H_9N_3O_2$	50,29	5,43	25,14	—	35
XIVb	>220	56,45	5,46	29,81	—	$C_9H_{10}N_4O$	56,83	5,30	29,46	—	79
XV	124—5	68,06	7,46	11,10	—	$C_{14}H_{18}N_2O_2$	68,27	7,37	11,37	—	45
XVIa	169—70	57,16	6,52	16,52	13,84	$C_{12}H_{16}N_3OCl$	56,80	6,36	16,56	13,97	78
XVIb	238—40	55,10	6,40	14,82	12,24	$C_{13}H_{18}N_3O_2Cl$	55,02	6,39	14,88	12,49	70
XVIIa·HCl	192—5	56,24	5,58	11,23	13,61	$C_{12}H_{15}N_2O_2Cl$	56,58	5,94	11,00	13,92	98
XVIIb	112—6	58,49	6,45	10,66	—	$C_{13}H_{16}N_2O_3 \cdot H_2O$	58,63	6,81	10,52	—	20

*Method A.

†Found, %: H_2O 3.24. Calculated, %: H_2O 3.32.

†Found, %: H_2O 14.29. Calculated, %: H_2O 13.89.

**Based on XIIIa.

Note: The compounds were crystallized: VII, Xa, and XIb•HCl from alcohol, III from ether, Xb, XIa, XII, XIIIa•HCl, XIVa, and XVIa, b from isopropyl alcohol, XIc and XIVb from N,N-dimethylformamide, XIIIa, b from hexane, XV and XVIIb from ethyl acetate, and XVIIa•HCl from acetonitrile.

TABLE 2. Spectral Characteristics of the Compounds Obtained

Compound	IR spectrum, ν, cm^{-1}			UV spectrum, λ_{max}, nm (log ε)
	C=O	C=N	NH, NH_2, OH	
II*	1690			
III		1510		213 (3,72)
XIb†	1755	1645	3160, 3300	213 (3,62); 250 (3.47)
XIc	1550 (COO^-)	1680	2540–2720 (br)	
XII	1740	1680		
XIIIa		1637		203 (4,29); 227 (4,16)
XIIIb		1652		203 (4,22); 231 (4,12) sh 255 (3.89)
XIVb‡	1640		3160, 3270, 3310, 3390, 3480	
XV	1640, 1580			
XVIa	1685	1645	3130, 3230, 3270	207 (4,16); 233 (4,05)
XVIb	1690	1665	3240, 3080	

*2220 (C≡N, w).
†For many of the compounds the CN group is not observed in the IR spectra; this is generally characteristic for α-amino nitriles [3].
‡2180 (C≡N).

The structures of the compounds obtained were confirmed by IR and UV data (see Table 2) and, in a number of cases, by data from the mass spectra.

EXPERIMENTAL CHEMISTRY

The IR spectra of mineral oil pastes of the compounds were recorded with a Perkin-Elmer 457 spectrometer (Sweden). The UV spectra of solutions in ethanol or methanol were recorded with a Hitachi EPS-3T spectrophotometer (Japan). The PMR spectrum of a solution of III in deuterochloroform was obtained with a JMN-4H-100 spectrometer with tetramethylsilane as the internal standard. The mass spectra were recorded with an MAT-112 spectrometer at an ionizing voltage of 50 eV and an ionization-chamber temperature of 140°C.

The purity of the substances was monitored by chromatography on Silufol UV-254 plates (Czechoslovakian SSR). The yields and properties of the synthesized substances are presented in Table 1, and the IR and UV spectra are presented in Table 2.

N-Cyanomethyl-2-pyrrolidone (II). A 19.2-ml (220 mmole) sample of phosphorus oxychloride was added dropwise at 0°C to a solution of 18.6 ml (240 mmole) of N,N-dimethylformamide in 400 ml of acetonitrile, after which the reaction mixture was stirred at 0°C for 1 h, and 28.4 g (200 mmole) of I was added in portions. The mixture was heated at 50°C for 2.5 h, after which it was cooled to 0°C, and 36 ml (440 mmole) of pyridine was added dropwise at 0-5°C. The mixture was then stirred at 0°C for 20 min, 19.7 ml (220 mmole) of concentrated hydrochloric acid was added dropwise at 0-5°C, 20 ml of water was added, and the mixture was extracted with 500 ml of chloroform. The aqueous layer was washed twice with 100-ml portions of chloroform, and the combined chloroform extracts were dried with potassium carbonate and evaporated *in vacuo*. The residue was distilled to give II.

2-Chloro-5H,6,7-dihydropyrrolo[1,2-a]imidazole (III). A 3-g (20.8 mmole) sample of I was heated in 35 ml of freshly distilled phosphorus oxychloride at 60°C for 3 h, after which the mixture was evaporated *in vacuo*, and the residue was dissolved in 10 ml of ice water. The aqueous solution was neutralized to pH ~ 6.0 with 10% sodium carbonate and extracted with chloroform. The solvent was removed, and the residue was triturated with ether to give III.

1-Cyanomethyl-2-ethoxy-1,2-dehydropyrrolidinium Tetrafluoroborate (VII). A solution of 49.5 g (0.400 mole) of II was added dropwise at 8°C to a solution of 90.4 g (0.477 mole) of VI in 200 ml of methylene chloride, and the mixture was stirred at room temperature for 5 h. It was then evaporated *in vacuo*, and the residue was triturated with isopropyl alcohol to give VII.

1-Cyanomethyl-2,2-diethoxypyrrolidine (VIII). A) A 6-g (25 mmole) sample of VII was added in portions at 5-10°C to a solution of sodium ethoxide obtained from 0.6 g (26.1 mmole) of sodium and 35 ml of absolute alcohol, and the mixture was stirred at 5-10°C for 2 h. The resulting precipitate was removed by filtration, the mother liquor was evaporated *in vacuo*, and the residue was distilled to give VIII.

B) A 4.38-ml (60 mmole) sample of phosphorus oxychloride was added dropwise to a solution of 7.5 g (60 mmole) of II in 60 ml of benzene, and the mixture was stirred at room temperature for 4.5 h and evaporated *in vacuo*. The residue was dissolved in 10 ml of absolute alcohol, and the solution was added dropwise to a solution of sodium ethoxide obtained from 1.4 g (60 mmole) of sodium and 25 ml of alcohol at 0-3°C. The reaction mixture was stirred for 0.5 h and evaporated *in vacuo*, and the residue was washed with dry ether and removed by filtration. The mother liquor was evaporated *in vacuo*, and the residue was distilled to give VIII (in 5% yield).

1-Cyanomethyl-2-[N-(1-carboxy-2-phenylethyl)]iminopyrrolidine (Xa). A mixture of 1.7 g (10 mmole) of phenylalanine and 2.3 g (11.6 mmole) of VIII in 30 ml of absolute alcohol was stirred at room temperature for 2 h, after which 1 ml of VIII was added, and the mixture was stirred for another hour. The reaction mixture was filtered, the mother liquor was evaporated *in vacuo*, and the residue was triturated successively with dry ether and acetone to give Xa.

1-Cyanomethyl-2-[N-(1-carboxy-2,2-dimethylethyl)]iminopyrrolidine (Xb). This compound was similarly obtained from VIII and valine.

1-Cyanomethyl-2-[N-(γ-carboxyethyl)]iminopyrrolidine (XIa). A mixture of 0.9 g (10 mmole) of β-alanine and 2.3 g (11.6 mmole) of VIII in 20 ml of absolute alcohol was stirred at room temperature for 2 h, after which 1.0 g of VIII was added, and the mixture was stirred at the same temperature for another 1.5 h. The reaction mixture was filtered, and the filtrate was evaporated *in vacuo*. The residue was triturated with N,N-dimethylformamide to give XIa.

1-Cyanomethyl-2-[N-(γ-carboxypropyl)]iminopyrrolidine Hydrochloride (XIb•HCl). A mixture of 1.5 g (14.6 mmole) of γ-aminobutyric acid and 3.5 g (17.7 mmole) of VIII in 30 ml of absolute alcohol was stirred at room temperature for 2 h, after which 1 g of VIII was added, and the mixture was stirred for another hour. The reaction mixture was evaporated *in vacuo* to give XIb. To obtain the hydrochloride (XIb•HCl) XIb was dissolved in 75 ml of isopropyl alcohol, the solution was filtered and the filtrate was cooled to 0-5°C and treated dropwise with an alcohol solution of hydrogen chloride to pH 3.0. The precipitated XIb•HCl was removed by distillation.

1-(β-Isopropoxy-β-imino)ethyl-2+[N-(γ-carboxypropyl)]iminopyrrolidine Hydrochloride (XII). A 10.0-g (47.8 mmole) sample of XIb was dissolved in 300 ml of isopropyl alcohol, the solution was filtered, and the filtrate was treated with an alcohol solution of hydrogen chloride to pH 3.0 and refluxed for 1 h. The reaction mixture was evaporated, and the residue was washed with ethyl acetate and dissolved in 10 ml of isopropyl alcohol. The solution was cooled to −5 to −7°C, and the precipitated XII was removed by filtration. Mass spectrum: M^+ 269.

2-Carboxypropyliminopyrrolidine (XIc). A mixture of 6.2 g (60 mmole) of γ-aminobutyric acid and 7.2 ml (66 mmole) of O-methylbutyrolactim in 45 ml of methanol was stirred at room temperature for 3 h, 1 ml of O-methylbutyrolactim was added, and the mixture was stirred for another 30 min. The reaction mixture was filtered, and the filtrate was evaporated *in vacuo*. The residue was triturated with ether and dissolved in methanol, and the solution was cooled. The precipitated XIc was removed by filtration. Mass spectrum: $M^{+\bullet}$ 170, $(M - H_2O)^{+\bullet}$ 152, $M - H_2O - H^{\bullet})^+$ 151, $(M - COOH)^+$ 125, $(M - COOH - CH_2)^+$ 111, $(M - COOH - C_2H_4)$ 97, and $(M - COOH - C_3H_6)$ 83.

1-Cyanomethyl-2-(N-phenylimino)pyrrolidine (XIIIa). A mixture of 4.1 g (44.0 mmole) of aniline and 10.0 g (50.5 mmole) of VIII in 150 ml of dry benzene was stirred at room temperature for 4.5 h, after which 5.0 g (25.3 mmole) of VIII was added, and the mixture was refluxed for 45 min. The reaction mixture was evaporated *in vacuo*, and the residue was triturated with petroleum ether to give XIIIa. The hydrochloride (XIIIa•HCl) was obtained by dissolving XIIIa in isopropyl alcohol and treating the solution with an alcohol solution of hydrogen chloride to pH 4.0.

1-Cyanomethyl-2-[N-(p-anisidino)imino]pyrrolidine (XIIIb). A mixture of 1.2 g (10 mmole) of p-anisidine and 2 g (10 mmole) of VIII in 20 ml of dry xylene was heated at 85-90°C for 3 h, after which 0.5 g (2.5 mmole) of VIII was added, and the mixture was heated for another 2 h. A 0.8-g (4.1 mmole) sample of VIII was added, and the mixture was heated for another hour. The reaction mixture was evaporated *in vacuo*, and the residue was triturated with water to give XIIIb.

1-Cyanomethyl-2-nitromethylenepyrrolidine (XIVa). A mixture of 11 g (55.5 mmole) of VIII and 3.7 g (60 mmole) of nitromethane in 100 ml of absolute alcohol was refluxed for 1 h, after which it was cooled, and the precipitated XIVa was removed by filtration.

1-Cyanomethyl-2-(2-cyano-2'-carbamido)methylenepyrrolidine (XIVb). A mixture of 0.7 g (8.3 mmole) of cyanoacetamide and 1.7 g (8.6 mmole) of VIII in 20 ml of absolute alcohol was stirred at room temperature for 15 min, after which it was refluxed for 40 min. The reaction mixture was cooled and XIVb was removed by filtration.

1,3-Dioxo-2-(N-cyanomethyl-2-pyrrolidinene)-5,5-dimethylcyclohexane (XV). A mixture of 2.6 g (13 mmole) of VIII and 1.4 g (10 mmole) of dimedone in 20 ml of absolute alcohol was stirred at room temperature for 6 h with the addition of 1 ml of VIII every 1.25 h. The reaction mixture was evaporated *in vacuo*, and the residue was triturated with ether to give XV.

1-Carbamidomethyl-2-(N-phenylimino)pyrrolidine Hydrochloride (XVIa). Dry hydrogen chloride was passed through a solution of 1.2 g (6.0 mmole) of XIIIa in 4.5 ml of formic acid at room temperature for 4 h, after which the reaction mixture was evaporated *in vacuo*, and the residue was triturated with isopropyl alcohol to give XVIa.

1-Carbamidomethyl-2[N-(p-anisidiono)imino]pyrrolidine Hydrochloride (XVIb). This compound was obtained from XIIIb by the method used to prepare XVIa.

1-Carboxymethyl-2-(N-phenylimino)pyrrolidine Hydrochloride (XVIIa•HCl). A 2.5-g (12.6 mmole) sample of XIIIa was stirred at room temperature in 20 ml of concentrated HCl for 24 h, after which the reaction mixture was evaporated *in vacuo*, and the residue was triturated with ethyl acetate to give XVIIa•HCl.

1-Carboxymethyl-2-[N-(p-anisidino)imino]pyrrolidine (XVIIb). A 0.6-g (2.6 mmole) sample of XIIIb was stirred at room temperature in 5 ml of concentrated HCl for 14 h, after which the reaction mixture was made alkaline to pH 7.0 with 2 N NaOH and extracted with chloroform. The chloroform extract was dried with calcine Na_2SO_4 and evaporated *in vacuo*, and the residue was triturated with ether to give XVIIb.

EXPERIMENTAL PHARMACOLOGY

Models of hypoxic states are widely used to predict the nootropic activity in experiments on animals. The existence of antihypoxic activity can be regarded as an index of potential nootropic activity. Antihypoxic activity has been established both experimentally and clinically for pyracetam [4-6].

In the present research the antihypoxic activity of the compounds was studied in experiments on mice with masses of 20-22 g with the utilization of two experimental models. Acute hypoxic hypoxia was induced by placing each mouse in a hermetically sealed 250-ml vessel [7]. To produce hemic hypoxia sodium nitrite (200 mg/kg subcutaneously), which induces methemoglobinemia [7], was administered to the mice (with masses of 20-25 g). The investigated compounds were administered intraperitoneally in doses of 100, 250, 500, and 1000 mg/kg 60 min prior to placing the mice in the hermetically sealed chamber or administration of sodium nitrite. The antihypoxic activity was judged from the lifetimes (in minutes) of the test animals as compared with control animals (injected with sodium chloride).

To judge the effect of the compounds on GABA-ergic systems we studied their ability to prevent spasms induced by thiosemicarbazide (TSC), which is an inhibitor of glutamatedecarboxylase — an enzyme that participates in the synthesis of γ-aminobutyric acid (GABA). In these experiments the compounds were administered intraperitoneally in doses of 250, 500, and 1000 mg/kg 20 min after subcutaneous administration of TSC (20 mg/kg). The latent period of the onset of spasms (in minutes) and the time of death of the animals were recorded. Data that indicate that inhibiting mediators of the central nervous system (CNS), including GABA, increase the latent period of the development of a painful syndrome in experiments on animals are known [8]. In this connection the existence of analgesic activity for the investigated pyracetam derivatives was determined in experiments on mice by the "hot-plate" method [9]. The compounds were administered intraperitoneally in doses of 100-500 mg/kg. The acute toxicities, including LD_{50}, were also studied in experiments on mice.

To characterize the central activity we studied the effect of the compounds that display a protective antihypoxic effect on the bioelectrical activity of the brain [electroencephalogram (EEG)] and the conditioned avoidance reflexes. The electroencephalographic studies were carried out in acute experiments on cats and rabbits with recording of the biopotentials from different regions of the cerebral cortex. The compounds were administered intravenously in doses of 100-200 mg/kg. The conditioned reflexes were studied with male rats with masses of 200-250 g by the method of electrodefensive conditioned avoidance reflexes. The compounds were administered intraperitoneally in a dose of 500 mg/kg 60 min prior to the experiments.

RESULTS AND DISCUSSION

The pharmacological activity was studied in the case of Xa, b, XIa-c, XIVa, b, XVIa, b, and XVIIa.

It was established that XIVa, b, XIa-c, and XVIa, b increased the resistance of the organism of the mice to oxygen deficiency in the acute hypoxic hypoxia model. The most active compound with respect to this index was XIb, which increased the lifetimes of the mice in the hermetically sealed chamber in doses of 250, 500, and 1000 mg/kg to 45 (40.2-49.8), 67.7 (59.8-75.6), and 130.3 (121.8-138.8) min, respectively [as compared with 28.2 (25.6-30.8) min in control animals]. With respect to antihypoxic activity XIb is considerably superior to pyracetam, which in a dose of 1000 mg/kg under similar conditions increased the lifetime to 41 (37.6-44.4) min.

Like pyracetam, none of the investigated compounds had protective activity against hemic hypoxia.

Activity was observed for XIb and XVIb in a study of the effect on TSC-induced spasms. Compound XIb in doses of 500 and 1000 mg/kg administered intraperitoneally increased the latent period of the onset of spasms in mice from 45.6 (38.6-52.6) min in control animals to 62.3 (57.3-67.3) min and 72.2 (56.2-88.2) min, respectively.

Death of the mice occurred 50.2 (39.2-61.2) min after injection of only TSC, as compared with 77.4 (64.4-90.4) min and 83.2 (70.2-86.7) min after prior injection of XIb. Compound XVIb was less active. In experiments with TSC pyracetam in doses of 1000 and 2000 mg/kg administered intraperitoneally gave a weakly expressed antispasmodic effect.

In a study of the analgesic activity we established that neither the investigated compounds nor pyracetam had this type of activity.

In the electroencephalographic investigations XIb, c, and XIVb, which have the most pronounced antihypoxic activity, in doses of 100-200 mg/kg administered intravenously did not induce changes in the spontaneous bioelectrical activity of different regions of the cerebral cortex and did not affect the reactions of the biopotentials induced by the applications of functional stresses that characterize the state of the functional lability of the cortical neurons and the excitability of the ascending activating system of the brain. According to the results of the electrophysiological experiments, the indicated compounds did not differ from pyracetam, which in doses from 100 to 2000 mg/kg did not induce changes in the EEG.

Compound XIb had low stimulating activity in experiments with conditioned avoidance reflexes: In a dose of 500 mg/kg administered intraperitoneally it caused a decrease in the latent period of the reflex by a factor of 1.5-2 as compared with a control animal. Compounds XIc and XIVb did not affect execution of a conditioned reaction of active avoidance. Pyracetam in a single dose of 1000 mg/kg caused a facilitation effect — it decreased the latent period of the reflex by a factor of 2-2.5.

The LD_{50} in the case of intraperitoneal administration to mice were as follows (in milligrams per kilogram): 475 for XVIa, 525 for XVIb, 625 for XIVb, more than 1500 for Xa, b, XIVa, and XIa, c, 2000 for XVIIa, and 2500 for XIb.

Thus, six derivatives of pyracetam have an antihypoxic effect, which is one of the indices of nootropic activity. The most pronounced antihypoxic effect, which exceeds the analogous effect of pyracetam by a factor of approximately three, is observed for XIb. This compound also displays antagonism with respect to the spasmodic effect of thiosemicarbazide, and this makes it possible to imagine the possibility of the participation of GABA-ergic structures in the realization of its effects. It should be noted that XIb has the lowest toxicity of all of the investigated pyracetam derivatives.

LITERATURE CITED

1. T. M. Bargar and C. M. Riley, Synthet. Commun., 10, 479-87 (1980).
2. F. Becke, H. Fleig, and P. Pässler, Justus Liebigs Ann. Chem., 749, 198-201 (1971).
3. L. A. Kazitsyna, B. V. Lokshin, and O. A. Glushkova, Zh. Obshch. Khim., 32, No. 5, 1391-1395 (1962).
4. C. Guirgea, F. Mouraviff-Lesuisse, and R. Lemans, Rev. Neurol., 122, 484-486 (1970).
5. A. J. Stegink, Arzneim.-Forsch., 22, 975-977 (1972).
6. L. F. Roshchina and R. U. Ostrovskaya, Farmakol. Toksikol., No. 2, 210-213 (1981).
7. M. V. Korablev and P. I. Lukienko, Antihypoxic Agents [in Russian], Minsk (1976).

8. E. I. Danilova, V. N. Grafova, and G. N. Kryzhanovskii, Byull. Eksp. Biol., No. 6, 525-528 (1979).
9. G. Woolfe and A. D. McDonald, J. Pharmacol. Exp. Ther., 80, 300-306 (1944).

QUATERNARY AMMONIUM SALTS WITH A LABILE $\overset{+}{N}$–C BOND AS PRECURSORS FOR DRUGS.

IV. ALKALINE HYDROLYSIS OF BUDIFIN

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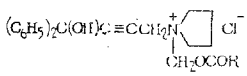
In continuation of studies on the alkaline hydrolysis of quaternary ammonium salts of drugs (QA) containing an acyloxymethyl group [1], we have studied the hydrolysis of some budifin derivatives, of general formula

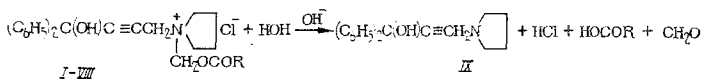


synthesized previously [2], in order to prolong the effects of the central m-cholinolytic drug.

Since the structure of R (Table 1) in this series has been varied quite extensively, we hoped, as a result of studying the reaction kinetics, to arrive at definite conclusions concerning the effects of the structure of the acyloxymethyl group on the rate of hydrolysis. The aim of the investigation was also to further refine our ideas as to the mechanism of this reaction.

Hydrolysis was effected at a given pH and temperature of 37°C under constant pH conditions, as described in the preceding communication [1].

A study of the kinetics of hydrolysis of the QA up to 60-80% reaction showed that the reaction



is described, as in the case of previously-studied cyclosyl derivatives, by a kinetic equation which is first order in alkali

$$k_{hydr.H} = k_0 + k_2 \cdot [OH^-], \quad (1)$$

TABLE 1. Dependence of Rate of Alkaline Hydrolysis on the Structure of the Acyl Group in the Series of Compounds I-VIII

Compound	R	$K_{hydr.H}$, $min^{-1} \cdot 10^3$ (pH 8.5, 37°C)	$\tau_{1/2}$, min	k_2, liter · $mole^{-1} \cdot sec$	r
I	OCH_3	64,2	10,8	114,39	0,9983
II	CH_3	75,2	9,2	133,90	0,9984
III	C_2H_7	41,4	16,7	73,77	0,9976
IV	$C(CH_3)=CH_2$	39,2	17,7	69,90	0,9960
V	$C(CH_3)_3$	6,9	101,9	12,12	0,9978
VI	$(CH_2)_5CH_3$	30,3	22,8	54,09	0,9960
VII	$(CH_2)_7CH_3$	66,2	10,5	118,04	0,9977
VIII	C_6H_5	127,1 at pH 9,0	...	62,95	0,9988

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