65-68°C (4 mm), n_D^{20} 1.4714. Found, %: C 69.26; H 12,80; N 17.78. $C_9H_{20}N_2$. Calculated, %: C 69.18; H 12.90; N 17.93.

α-Truxillic Acid Bis-[N-methyl-N-(3-piperidylpropyl)amide] (XI). Obtained by the general method for the preparation of bis(aminoamides) (see above); yield 55%, mp 149-150°C (from ethyl acetate). Found, %: C 75.22; H 9.19; N 9.93. $C_{36}H_{52}N_4O_2$. Calculated, %: C 75.48; H 9.15; N 9.78.

Diethiodide. A solution of 0.5 g of the base and 0.5 g of iodoethane in 5 ml of methanol was heated in an ampul for 5 h at 60°C. After removal of the solvent, the residue was dissolved in water, and the solution treated with charcoal and evaporated to give 0.45 g of product, mp 120-124°C. Found, %: I 28.72. $C_{36}H_{52}N_4O_2 \cdot 2C_2H_5I$. Calculated, %: I 29.6.

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ANTICONVULSIVE ACTIVITY OF PEPTIDOAMIDOBENZOPHENONES

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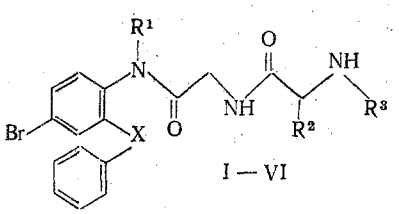
The interest in peptidoamidobenzophenones is due to several factors. In the first place, the molecules of these compounds contain peptide fragments, which can play an important regulatory role in the animal and human organism. In the second place, there is a possibility of selecting a structure of the peptide chain that would be hydrolyzed in the gastrointestinal tract to an unstable intermediate compound, forming a 1,4-benzdiazepine derivative with the second part of the molecule (benzophenone), i.e., a classic tranquilizer [2]. The possibility remains that the presence of a peptide fragment and a benzophenone in a single molecule may promote the appearance of new pharmacological properties of the given substance.

It should be noted that 2-(5-cyano-N-methyl-3,5-dimethyl-3-ase-pentaamido)-5-nitrobenzophenone [11] and N-methyl-N-glycylglycyl-anilide [10] have been introduced into medical practice as tranquilizers in England and Japan, respectively.

In this work we describe the synthesis of peptidoamidobenzophenones of various structures and also present the results of a study of their anticonvulsive activity.



I — VI

R^1 = H(I — V), CH_3 (VI);
R^2 = H(I, II, III, IV, V), CH_3 (III, VI);
R^3 = H(IV), H·HCl (V), $CO_2CH_2C_6H_5$ (I—III, VI)
X = C=O (I, III — VI), CHOH (II)

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EXPERIMENTAL (CHEMICAL)

The course of the reactions and the homogeneity of the chemical compounds was monitored by thin-layer chromatography. R_f was determined in the systems acetone–hexane (2:3, compounds I-III, VI), octanol–water–acetic acid (4:1:1, compounds IV and V) on Silufol-UV-254 plates. The IR spectra were recorded in KBr tablets on a Perkin-Elmer instrument (England).

2-(N-Carbobenzoxyglycyl-glycyl)amido-5-bromobenzophenone (I). To a solution of 2.66 g (0.01 mole) N-carbobenzoxyglycyl-glycine in 10 ml of abs. dimethylformamide and 10 ml of ethyl acetate at −10 to −13°C, 1.3 ml (0.01 mole) of isobutyl chloroformate and then 1.4 ml (0.01 mole) of triethylamine were added. The mixture was mixed for 5-6 min, and 2.76 g (0.01 mole) 2-amino-5-bromobenzophenone in 20 ml abs. chloroform was added, mixed at 5-7°C for 0.5 h, then at 17-20°C for another 0.5 h, and evaporated under vacuum. The residue was dissolved in 50 ml of ethyl acetate and 10 ml of water, the organic layer washed with a 5% solution of $NaHCO_3$ (2 × 10 ml), with water, with 1% hydrochloric acid, and with water to pH 7.0; the ethyl acetate solution was dehydrated with sodium sulfate and evaporated under vacuum. The residue was recrystallized from ethanol. We obtained 4.7 g (89%) I, mp 162-163°C, R_f 0.23. Found, %: C 57.4; H 4.1; N 8.1. $C_{25}H_{22}N_3O_5Br$. Calculated, %: C 57.3; H 4.2; N 8.0. IR spectrum, ν, cm^{-1}: 1635, 1670, 1695 (C=O).

2-(N-Carbobenzoxyglycyl-glycyl)amido-5-bromobenzhydrol (II). Analogously to the preceding, II (86%) was obtained from N-carbobenzoxyglycyl-glycine and 2-amino-5-bromobenzhydrol, mp 149-150°C (from ethanol), R_f 0.15. Found, %: C 57.3; H 4.5; N 8.1. $C_{25}H_{24}N_3O_5Br$. Calculated, %: C 57.1; H 4.6; N 8.0. IR spectrum, ν, cm^{-1}: 1660, 1675 (C=O).

2-(N-Carbobenzoxy-α-alanyl-glycyl)amido-5-benzophenone (III). To a solution of 1.4 g (0.005 mole) N-carbobenzoxy-α-alanyl-glycine in 50 ml abs. chloroform at −17 to −19°C we added 1 g (0.005 mole) of phosphorus pentachloride; the mixture was mixed for 30-40 min, then 1.38 g (0.005 mole) of 2-amino-5-bromobenzophenone in 20 ml of abs. chloroform was added, and mixed for 1 h at 20-22°C. Then 10 ml of water was added at 0-5°C, the reaction mixture was neutralized with a 5% solution of $NaHCO_3$, the organic layer was washed with water, dehydrated with sodium sulfate, evaporated under vacuum, the residue was recrystallized from ethanol, and 2.8 g (82%) III, mp 139-140°C, R_f 0.28 was obtained. Found, %: C 57.8; H 4.5; N 7.9. $C_{26}H_{24}N_3O_5Br$. Calculated, %: C 58.0; H 4.5; N 7.8. IR spectrum, ν, cm^{-1}: 1655, 1685 (C=O).

2-Methyl(N-carbobenzoxy-α-alanyl-glycyl)amino-5-bromobenzophenone (IV). Analogously to the preceding, from N-carbobenzoxy-α-alanyl-glycine and 2-methylamino-5-bromobenzophenone we obtained VI (40%), mp 79-80°C (from ethanol), R 0.58. Found, %: C 58.9; H 4.7. $C_{27}H_{26}N_3O_5Br$. Calculated, %: C 58.7; H 4.7; N 7.6. IR spectrum, ν, cm^{-1}: 1650, 1710 (C=O).

2-(Glycyl-glycyl)amido-t-bromobenzophenone (IV). A suspension of 2 g (0.0038 mole) 2-(N-carbobenzoxyglycyl-glycyl)amido-5-bromobenzophenone (I) in 5 ml of a 20% solution of hydrogen bromide in glacial acetic acid was mixed until dissolution at 20-23°C. Then 20 ml of diethyl ether was added to the solution, the precipitate formed was removed, dissolved in 20 ml of water at 5-8°C, neutralized with 5% $NaHCO_3$, and extraction was performed with chloroform (5 × 10 ml). The organic layer was dehydrated with sodium sulfate, evaporated under vacuum, and 0.7 g (50%) IV, mp 130-131°C, 0.3, was obtained. Found, %: C 52.4; H 4.3; N 10.9. $C_{27}H_{16}N_3O_3Br$. Calculated, %: C 52.3; H 4.1; N 10.8. IR spectrum, ν, cm^{-1}: 1630, 1670 (C=O).

2-(Glycyl-glycyl)amido-5-bromobenzophenone hydrochloride (V). To a solution of 1 g (0.0025 mole) 2-(glycyl-glycyl)amido-5-bromobenzophenone (IV) in 30 ml of ethyl acetate we added 10 ml of diethyl ether, saturated with hydrogen chloride; the precipitate formed was removed, washed with ether, and exposed over sodium hydroxide and phosphoric anhydride for three days. We obtained 1 g (94%) V, mp 134-135°C, R_f 0.3. Found, %: C 48.0; H 4.1; N 9.9. $C_{17}H_{16}N_3O_3Br$. Calculated, %: C 47.9; H 4.0; N 9.8. IR spectrum, ν, cm^{-1}: 1635, 1675 (C=O).

EXPERIMENTAL (BIOLOGICAL)

The experiments were conducted on noninbred white mice weighing 18-20 g. The anticonvulsive activity of compounds I-VI was investigated according to antagonism to corasole, which was injected intramuscularly in doses of 110-130 mg/kg, thiosemicarbazide in doses

of 25-28 mg/kg intraperitoneally, and strychnine in doses of 1.7-2 mg/kg subcutaneously, 30 or 120 min after intraperitoneal or oral administration of the peptidoamidobenzophenone. The prevention of tonic convulsions under maximum electric shock, which was induced by a current with a strength of 50A with exposure 2 sec, was studied. The change in motor activity was judged according to the "rotating rod" test. The mice were placed on a smooth wooden rod 2 cm in diameter, rotating at a speed of 10 rpm. The number of mice that fell off the rod within 30 sec was recorded [4].

Derivatives I-IV, VI were administered to the experimental animals in the form of a Tween emulsion; the derivative V was dissolved in water.

RESULTS AND DISCUSSION

In a single administration, most of the investigated substances eliminated the clonic-tonic convulsions induced by corasole, which, in the opinion of certain authors, correspond to the main components of the petit mal attack in humans [6]. The anticorasole effect is considered as one of the indices accompanying the psychosedative action of the drug. In view of the fact that there is a pronounced correlation between the activity of tranquilizers according to specific models of behavior of the "conflict situation" type and external inhibition, permitting an evaluation of the antineuritic and tranquilizing components of the action with the activity according to the corasole test, the latter is used as a simple and convenient method for evaluating the tranquilizing action of new drugs [3].

According to antagonism to corasole, the compound VI exhibits the greatest activity 30 min after intraperitoneal injection (Table 1) and it does not decrease over the following 120 min of the investigation. The water-soluble derivative V is close to VI in LD_{50} (anticorasole test), but its activity decreases with time. A more prolonged anticonvulsive action is exhibited by compounds I and IV in the case of their intraperitoneal injection.

In the case of oral administration, compounds IV, V, and VI are the most active (Table 2). Although the activity of VI increases threefold after 120 min, for IV and V it decreases. For I and III such an increase comes to 2.2 and 1.4-fold, respectively.

According to the test of prevention of convulsions induced by thiosemicarbazide, the most active is the derivative VI. In the case of intraperitoneal injection, its effect does not decrease with time, while in the case of oral administration it doubles. Compounds IV and V show no change in action with time; I and II decrease their activity after 120 min following intraperitoneal injection, while in the case of oral administration the activity is maintained for the same period of time.

Thiosemicarbazide inhibits GABA synthesis in the brain [5]; it is suggested that substances that suppress the action of thiosemicarbazide affect the activity of GABA-ergic neurons in the cerebellum, which may lie at the basis of the mechanisms of the anticonvulsive effects [7].

Derivatives of peptidoamidobenzophenone exhibit their activity with respect to the elimination of a maximum electroconvulsive attack. Antagonism to the convulsive action of strychnine is only slight for compounds of this series.

Thus, our investigations of the pharmacological properties of the compounds synthesized show that almost all of them, with the exception of II, have a protective effect in mice in the case of convulsions induced by corasole or thiosemicarbazide in doses of 0.3-40 mg/kg.

In an evaluation of tranquilizers, their myorelaxant action, the first characteristic of which is a disruption of motor coordination and equilibrium, can be considered as a side effect. Such disorders in noninbred white mice in the case of oral administration of derivatives I-III were observed at doses greater than 100 mg/kg, and for IV-VI at doses greater than 30 mg/kg [7].

It is known that 1,4-benzdiazepines in acid aqueous solutions are hydrolyzed to the corresponding benzophenones [12]. In weakly acid media the reverse reaction can occur — the formation of a peptide bond of benzdiazepines; this process depends on the nature of the substituents and solvent. Consequently, such substances can serve as precursors of the active tranquilizer molecule and may even possess better pharmacological properties [9].

TABLE 1. Anticonvulsive Activity of Peptidoamidobenzophenone Derivatives after a Single Intraperitoneal Administration to White Mice (ED_{50}, mg/kg)

Compound	Time, min	Antagonism			Prevention of convulsions in max. elec. shock
		with corasole	with thiosemicarbazide	with strychnine	
I	30	34 (27,6—41,8)	33 (24—45,5)	>80	>100
	120	25 (21,9—28,5)	43,2 (29,8—62,4)	>80	>100
II	30	>80	>80	>80	>100
	120	>80	>80	>80	>100
III	30	4,6 (3,4—6,25)	17,5 (11,3—27,2)	45 (25,7—78,7)	57 (28,5—114)
	120	13 (11,3—15)	27 (18—40,5)	32 (18,8—45)	100
IV	30	12,5 (7,8—20)	15 (9,1—25)	>30	—
	120	3,8 (2,5—5,7)	14 (10—20)	>30	—
V	30	2,2 (1,5—3,2)	19 (10,8—33,2)	15 (10,7—21)	22,5 (13,2—38)
	120	4,5 (2,5—8,1)	17 (5,6—51)	34 (20,6—48)	27 (18—40,5)
VI	30	1,1 (0,68—1,8)	1,8 (1,1—3)	12,5 (7,8—20)	32 (18,8—54,4)
	120	1,1 (0,6—1,9)	1,3 (0,8—2,2)	12,5 (8,8—17,5)	55 (27,5—120)

Note. Here and in Table 2 the limits of fluctuations are given in parentheses.

TABLE 2. Anticonvulsive Activity of Peptidoamidobenzophenone Derivatives in a Single Oral Administration to White Mice (ED_{50}, mg/kg)

Compound	Time, min	Antagonism			Prevention of convulsions in max. elec. shock
		with corasole	with thiosemicarbazide	with strychnine	
I	30	38 (22,3—67,6)	58 (41,4—81,2)	>80	>100
	120	17 (13,9—20,7)	44,2 (36,2—54)	>80	>100
II	30	>80	>80	>70	>100
	120	>80	>80	>80	>100
III	30	18,5 (14,2—24)	23 (13,1—40,3)	70 (43,7—113)	>100
	120	13 (6,5—26)	30 (17,6—51)	75 (45,5—123,8)	>100
IV	30	0,7 (0,54—0,9)	10 (5,5—18)	>50	—
	120	6 (3,7—6,24)	13 (7,2—23,4)	>50	—
V	30	1 (0,6—1,5)	11 (6,6—18,2)	81 (25—250)	40 (20—78)
	120	4,7 (2,7—7,8)	11,5 (6,5—20)	48 (25,3—91,2)	49 (71—33,8)
VI	30	1 (0,64—1,55)	2,7 (1,0—6,5)	20 (10—40)	>40
	120	0,3 (0,18—0,8)	1,25 (0,8—19)	22 (12,2—40)	>40

Evidently we can assume that the peptidoamidobenzophenones that we synthesized are precursors of 1,4-benzdiazepines, and their biological activity is due to the activity of the metabolites formed. This is evidenced by the data of [8], in which it was shown that peptidoamidobenzophenones are characterized by cleavage of the peptide bond and that the products of metabolism are cyclized at physiological pH values, with the formation of 1,4-benzdiazepines. Compound II does not possess anticonvulsive activity according to the tests that we conducted, from which it can be concluded that its hydroxyl group prevents cyclization and the formation of an active metabolite.

Analyzing the anticonvulsive activity of derivatives V and VI, we established that it depends largely on the nature of the substituents, the time and method of their introduction.

Derivatives I, III, and VI with a long peptide chain exhibit differing activity, which increases with time, especially in the case of a single oral administration. Compound VI, in which there are two methyl groups, is the most active, while compound I, which does not have any, is two orders of magnitude inferior to it in activity.

Derivatives IV and V differ from one another only in that compound V is a water-soluble salt of IV, which greatly facilitates its use. But its anticonvulsive activity decreases with time, whereas in IV it increases 3.3-fold over a period of 120 min after intraperitoneal injection. Comparing the action of the derivatives I, III, and VI with IV and V, we can assume that the nature of R^3 of the peptide chain influences the anticonvulsive activity even when they are administered orally.

The toxicity of the synthesized peptidoamidobenzophenones is exhibited at doses greater than 600 mg/kg.

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