Eur J Med Chem (1996) 31, 151–157

Short communication

Synthesis and antiamnesic activity of a series of *N*-acylprolyl-containing dipeptides

TA Gudasheva[1], TA Voronina[2], RU Ostrovskaya[2], GG Rozantsev[1], NI Vasilevich[1], SS Trofimov[2], EV Kravchenko[2], AP Skoldinov[1], SB Seredenin[2]

[1]*Chemistry Department, Institute of Pharmacology, Russian Academy of Medical Science;*
[2]*Pharmacology Department, Institute of Pharmacology, Russian Academy of Medical Science, Baltijskaya, 8, Moscow 125315, Russia*

(Received 16 May 1995; accepted 30 October 1995)

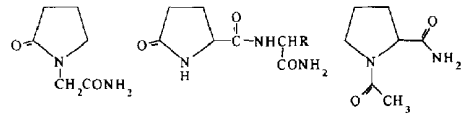
Summary — Esters and amides of a series of *N*-acylprolyl-containing dipeptides were synthesized. It was established that these substances possess the ability to prevent memory decline evoked by maximal electroshock (MES) in a passive avoidance step-through paradigm. These *N*-acylprolyl-containing dipeptides were designed as analogues of pyroglutamyl-containing dipeptides, which we previously demonstrated to be highly active nootropics. Among the structure–activity relationships explored were the effect of *N*-acyl-substitution size, C-terminal substitution and the nature of the second amino acid. The optimal *N*-acyl moiety was the *N*-phenyl-acetyl group, while the optimal C-terminal substitution-esters were those derived from low alkyl alcohols. The optimal second amino acids were Asp, Glu or their fragments, Gly, β-Ala, GABA. Compound **1** (*N*-phenylacetylprolylglycine ethyl ester) was selected for further evalution in impaired cognitive functions. It was supposed that esters and unsubstituted amides of *N*-acylprolylglycines are prodrugs, which convert to the bioactive cyclo-(Pro-Gly) by virtue of enzymatic or chemical lability within the body.

***N*-acylprolyl-containing dipeptide / cyclo-(Pro-Gly) / antiamnesic activity / structure–activity relationship**

Introduction

Many substances are known to affect intellectual performance in humans. Nootropic agents constitute a promising group of these substances. Piracetam, *N*-carbamidomethylpyrrolidone-2, is the prototype of nootropic drugs and exerts a direct effect on integrative brain functions [1, 2].

Several years ago [3–5] we reported the design and synthesis of a novel class of potent, specific, orally effective peptide analogues of the nonpeptide nootropic piracetam. The peptide design was based on the conception that piracetam is an exogenic analogue of an endogenous neuropeptide with an *N*-terminal pyroglutamic acid. Designed compounds are pyroglutamyl-containing dipeptide amides (PGDA). PGDA were shown to be able to facilitate the performance in the passive avoidance test and to possess cognition-enhancing activity in other paradigms [6–9]. These substances (and piracetam) demonstrated neither stimulating nor depressing influences on locomotor activity. The threshold doses of PGDA are 0.01–1 mg/kg (ip).



Piracetam PGDA *N*-Acetylprolinamide

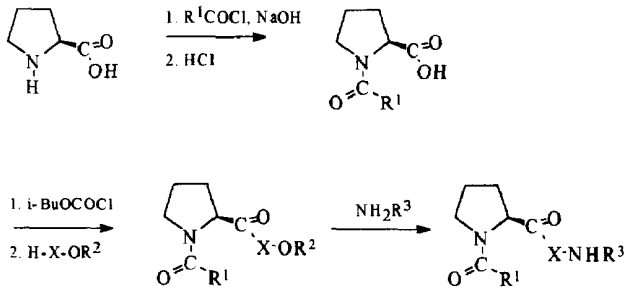
Surprisingly, the most active peptide analogue of piracetam, pyroglutamylasparaginamide [4], stereoselective on both amino acids, was identical to the *N*-end terminal sequence of the major arginine-vasopressin metabolite [pGlu[4], Cyt[6]]AVP (4-9); the latter was more potent and more efficacious on memory than the parent hormone [10]. Previously, we also reported on the cognition-activating effects of *N*-acetylprolinamide [11]. Only the (*S*)-(–)-enantiomer of this compound improved the performance in a passive avoidance test in rats. We considered *N*-acylprolinamide as the topological analogue of the pyroglutamylamide moiety of PGDA, because both compounds have the pyrrolidine ring system and the carbonyl oxygen closely located.

We have continued the search for new agents with cognitive enhancing properties. In this paper, we

describe the chemistry and structure–activity relationships of amides and esters of *N*-acylprolyl-containing dipeptide series. These compounds were designed as topological analogues of pyroglutamyl-containing dipeptides, where the pyroglutamic acid moiety was replaced by *N*-acylprolyl moiety.

Chemistry

The general synthetic route for *N*-acylprolyl-containing dipeptides is presented in scheme 1. *N*-Acylprolines were prepared by acylation of proline with the appropriate acyl chloride according to Schotten-Baumann [12]. A coupling of *N*-acylproline with the corresponding amino acid ester was carried out by the mixed anhydride method using isobutyl chlorformate in the presence of *N*-methylmorpholine. Amides of dipeptides were synthesized by aminolyses of the corresponding esters. *N*-Phenylacetylprolylglycine was obtained by alkaline hydrolysis of the ethyl ester of *N*-phenylacetylprolylglycine. Diastereomeric purity of the prepared compounds was not below 98% according to ^{1}H-NMR spectroscopy. An observed doubling of signals is caused by a presence of rotamers about the *N*-acylproline amide bond (Gudasheva TA et al, manuscript in preparation).



(1-6, 11, 13, 14, 16, 17, 19) (2, 7, 9, 12, 15, 18)

1: $R^1 = CH_2C_6H_5$; X = Gly; R^2 = Et
2: $R^1 = C_6H_5$; X = Gly; R^2 = Et
3: $R^1 = (CH_3)_2CHCH_2$; X = Gly, R^2 = Et
4: $R^1 = CH_3(CH_2)_4$; X = Gly; R^2 = Et
5: R^1 = 1-Ad; X = Gly; R^2 = Et
6: $R^1 = C_6H_5(CH_2)_3$; X = Gly; R^2 = Et
7: $R^1 = CH_2C_6H_5$; X = Gly; R^3 = Et
9: $R^1 = CH_2C_6H_5$; X = Gly; R^3 = Et
11: $R^1 = CH_2C_6H_5$; X = β-Ala; R^2 = Et
12: $R^1 = NH_2C_6H_5$; X = β-Ala; R^2 = H
13: $R^1 = CH_2C_6H_5$; X = GABA; R^2 = Me
14: $R^1 = CH_2C_6H_5$; X = AspOEt; R^2 = Et
15: $R^1 = CH_2C_6H_5$; X = Asn; R^3 = H
16: $R^1 = CH_2C_6H_5$; X = GluOEt; R^2 = Et
17: $R^1 = CH_2C_6H_5$; X = Ala; R^2 = Et
18: $R^1 = CH_2C_6H_5$; X = Leu; R^3 = H
19: $R^1 = CH_2C_6H_5$; X = Val; R^2 = Et

Scheme 1. Synthetic scheme to compounds **1–9** and **11–19**.

Pharmacology

The testing of new chemical entities for potential cognition-activation properties was conducted in two phases. At first, all the compounds were evaluated for their ability to prevent the memory decline evoked by maximal electroshock (MES) in the passive avoidance step-through paradigm in rats [13–15].

Each substance was administered intraperitoneally 15 min before passive avoidance training. Amnesia was produced by transcorneal MES immediately after the learning session. The retention was tested 24 h later by measuring the latency time for the entering the dark compartment.

In the second phase, all compounds were studied from the point of view of their gross effects on the central nervous system (CNS) [16]. The aim of this stage was to exclude any substance having psychostimulant or depressant activity, as well as anticonvulsive activity. These data could be of great importance for revealing false non-cognitive effects of the substances tested.

All the compounds tested were inactive in gross behavioral tests. Most of the substances under study were able to reverse MES-induced amnesia.

Results and discussion

Amnesia reversal. Structure–activity relationships (SAR)

Since compound **1** was regarded as the basis for further studies, its active dose range was evaluated (see table I). This compound was found to be active over a broad, ten-fold dose range (0.1–1.0 mg/kg ip or per os). The activity of other compounds was examined at a dosage equal to the minimal effective dose of compound **1**. The compounds tested have demonstrated significant antiamnesic effects at a dose of 0.1 mg/kg (ip). *N*-Acylprolyl-containing dipeptides studied were found to be 2000 times more active than piracetam (its minimal effective dose was 200 mg/kg ip) and showed similar activity when compared to nootropic pyroglutamyl-containing dipeptides [3]. *N*-Phenylacetyl-L-prolylglycine was considered as the main model compound because its glycine residue can be regarded as a fragment of the 20 naturally occurring mammalian amino acids.

Table II illustrates in detail *N*-substitution effects and table III that of C-substitution. Table IV describes

Table I. Dose–response relationship for *N*-phenylacetyl-L-prolylglycine ethyl ester (compound **1**) and piracetam revealed in passive avoidance test with MES amnesia.

Compound	*Dose (mg/kg) per os*	*Antiamnesic activity (%)*
1	0.1	19.0*
	0.3	19.0**
	0.4	23.0*
	0.5	43.0*
	0.7	52.0*
	0.9	36.0*
	1.0	10.0*
	1.2	16.0
Piracetam	200	−1.8
	300	22.9*
	400	59.1*
	800	58.0*
	1000	29.7

*$P < 0.05$; **$P < 0.01$ in comparison with controls (*U* test).

the effect of glycine substitution by other amino acids. Several SAR generalizations can be made from the data contained in tables II–IV:

1. The size and the nature of the *N*-acyl moiety have a minor influence on the activity of compounds in series of *N*-acylprolylglycine esters.
2. The nature of *C*-substitution has a strong influence on *N*-phenylacetylprolylglycine's antiamnesic activity. Esters (compounds **1**, **11** and **14**) and unsubstituted amides (compounds **7**, **12** and **15**) were found to be active, whereas substituted amides and peptides with a free C-terminal carboxyl group failed to be active (compounds **8–10**).
3. Gly can be replaced by hydrophylic amino acids such as Asp, Glu or their fragments, β-Ala and GABA accordingly (compounds **11–16**) without loss of activity. Gly substitution by hydrophobic amino acids Val, Leu and even Ala results in activity loss.

The SAR generalizations 1 and 2 can probably be explained as follows. The nature and size of the *N*-acyl substituent in the series of *N*-acylprolylglycines studied failed to affect the antiamnesic activity, and

Table II. Antiamnesic activity of *N*-acylprolylglycine ethyl esters R^1CO-Pro-Gly-OC_2H_5.

Compound	*R^1*	*Latency (s) (mean ± SEM)*			*Antiamnesic activity (%)(dose 0.1 mg/kg, ip)*
		Control	*Amnesia*	*Amnesia + substance*	
1	$C_6H_5CH_2$	110.5 ± 18.4	25.7 ± 7.2	69.1 ± 14.9	51*
2	C_6H_5	95.2 ± 19.2	3.2 ± 0.4	37.0 ± 19.7	37**
3	$(CH_3)_2CHCH_2$	138.0 ± 13.3	78.0 ± 7.8	95.0 ± 5.1	28*
4	$CH_3(CH_2)_4$	134.1 ± 17.2	60.3 ± 11.2	76.1 ± 4.9	20*
5	1-Ad	111.0 ± 18.3	19.0 ± 8.4	32.0 ± 2.4	13*
6	$C_6H_5(CH_2)_3$	111.0 ± 18.3	19.0 ± 1.8	31.0 ± 1.8	13*

*$P < 0.05$; **$P < 0.01$ in comparison with controls (*U* test).

Table III. Antiamnesic activity of *N*-phenylacetylprolylglycine derivatives $C_6H_5CH_2CO$-Pro-Gly-R^2.

Compound	*R^1*	*Latency (s) (mean ± SEM)*			*Antiamnesic activity (%)(dose 0.1 mg/kg, ip)*
		Control	*Amnesia*	*Amnesia + substance*	
1	OC_2H_5	110.5 ± 18.4	25.7 ± 7.2	69.1 ± 14.9	51*
7	NH_2	153.0 ± 13.5	19.0 ± 8.7	103.5 ± 17.4	62**
8	OH	103.0 ± 17.3	52.0 ± 11.2	57.0 ± 12.4	9.8
9	$NHCH_3$	103.0 ± 17.3	52.0 ± 11.2	48.0 ± 12.8	−7.6
10	$N(CH_3)_2$	111.2 ± 19.3	19.0 ± 7.3	1.2 ± 15.8	23.0

*$P < 0.05$; **$P < 0.01$ in comparison with controls (*U* test).

Table IV. Antiamnesic activity of esters and amides of *N*-phenylacetyl-L-proline-containing dipeptides $C_6H_5CH_2$-Pro-X.

Compound	*R'*	*Latency (s) (mean ± SEM)*			*Antiamnesic activity (%)(dose 0.1 mg/kg, ip)*
		Control	*Amnesia*	*Amnesia + substance*	
11	β-Ala-OEt	114.0 ± 8.8	15.8 ± 8.8	76.9 ± 18.3	66**
12	β-Ala-NH_2	98.0 ± 18.2	26.0 ± 11.2	64.2 ± 15.5	52*
13	GABA-OMe	139.1 ± 19.3	60.3 ± 13.4	96.0 ± 16.3	45*
14	Asp$(OEt)_2$	153.9 ± 17.6	41.0 ± 13.2	80.0 ± 14.4	34*
15	Asn-NH_2	133.0 ± 24.0	28.4 ± 7.4	55.2 ± 11.1	26*
16	Glu$(OEt)_2$	111.0 ± 18.3	19.0 ± 8.4	73.1 ± 13.8	58*
17	Ala-OEt	139.0 ± 19.3	60.3 ± 13.4	69.0 ± 14.3	10
18	Leu-NH_2	139.0 ± 19.3	60.3 ± 13.4	66.0 ± 13.7	8
19	Val-OEt	133.0 ± 24.0	40.8 ± 15.1	40.2 ± 14.6	0

*$P < 0.05$; **$P < 0.01$ in comparison with controls (*U* test).

this could be due to its probable enzymatic elimination in vivo followed by the formation of an active metabolite. Since the C-end terminal substituents (esters and unsubstituted amides) that enhance the antiamnesic activity are exactly those known to promote spontaneous dipeptide cyclization, we can propose diketopiperazine, cyclo-(Pro-Gly) as an active product of *C*-substituted *N*-acylprolylglycine metabolism.

These speculations are supported by the data of our additional experiments, which revealed the ability of cyclo-(Pro-Gly) to demonstrate antiamnesic activity: AA 76% ($P < 0.05$) with dosage of 0.1 mg/kg ip and AA 65% ($P < 0.05$) with a dosage of 1 mg/kg ip. Moreover, this diketopiperazine was detected in rat brain following the systemic administration of the compound **1** by high-performance liquid chromatographic and gas chromatographic methods (SS Boyko et al, unpublished results).

The antiamnesic activity observed by cyclo-(Pro-Gly) could be attributed to the fact that this compound is the covalent analogue of the proposed endogenous nootropic dipeptide pGlu-Asn-NH_2 in its pseudocyclic conformation [17]. The relationship between the active *N*-acylprolyl-containing dipeptides and this dipeptide is well demonstrated by SAR generalization 3. Furthermore, only those dipeptides were found active (see table IV), which had the second amino acid analogous (Asn, Asp) or homologous (Glu) to that of pGlu-Asn-NH_2 or its fragment (Gly, β-Ala, GABA).

Conclusion

We have developed a new series of nootropic dipeptides, namely *N*-acylprolyl-containing dipeptides. The SAR generalizations in this series make it possible to assume that esters and unsubstituted amides of *N*-acyprolylglycines can be considered as prodrugs, which convert to the bioactive bicyclic compound cyclo-(Pro-Gly) in the body. One of the most active, and moreover technologically available compounds, *N*-phenylacetylprolylglycine ethyl ester was selected for further development as a potential nootropic drug.

Experimental protocols

Chemistry

Melting points were determined on a capillary melting point apparatus in open capillary tubes and are uncorrected. The structures of the compounds were confirmed by elemental analysis an ^1H NMR spectroscopy. Microanalyses agree with calculated values within ±0.4%. The NMR spectra were obtained on a Bruker AC-250 spectrometer using tetramethylsilane as an internal standard. The NMR peaks were designated as follows: s, singlet; d, doublet; t, triplet; q, quartet; m, multiplet; br, broad. Specific optical rotations were recorded by automatic polarimeter Perkin-Elmer 241. The TLC was carried out on Merck silica-gel 60 F 254 plates and spots were developed in an iodine chamber or under UV light. For column chromatography, E Merck silica-gel 60, 230-400 mesh was used. If required, all solvents used in reaction mixtures were dried and purified by standard procedures.

General synthetic method for the preparation of N-acylprolines
To a well-stirred solution of L-proline (50.0 mmol) in 2 N NaOH (25 mL), 4 N NaOH (12.5 mL) and acyl chloride (50.0 mmol) were added dropwise from different drop funnels at a temperature of about 0–4 °C. The reaction mixture was stirred for 15 min, extracted by ethylacetate to remove chloride and then it was acidified with 1 N HCl to pH ≈ 3. The resulting oil was extracted with chloroform, the combined organic extract was dried ($MgSO_4$) and the solvent was removed. Ether was added to the residue and the mixture was allowed to stand overnight at 0 °C. The crystals of *N*-acylproline were separated.

N-Phenylacetyl-L-proline. Yield 86%, mp 150–152 °C, $[\alpha]_D^{20}$ –60.5° (c 0.4, DMF), R_f 0.66 (dioxane/water 9:1). NMR ($CDCl_3$): δ 1.77–2.29 (m, $C^{\beta}H_2$-$C^{\gamma}H_2$ Pro, 4H); 3.40–3.63 (m, $C^{\delta}H_2$ Pro, 2H); 3.73 and 3.63 (each s, CH_2-C_6H_5, 2H); 4.56 and 4.38 (each dd, $C^{\alpha}H$ Pro, 1H); 7.18–7.39 (m, CH_2-C_6H_5, 5H); 11.38 (br s, COOH, 1H). Lit data [18]: mp 150–152 °C, $[\alpha]_D^{19}$ –68.9° (c 1.0, MeOH).

N-Benzoyl-L-proline. Yield 60%, mp 152–154 °C, $[\alpha]_D^{20}$ –68.5° (c 0.4, $CHCl_3$), R_f 0.13 (chloroform/methanol 9:1). Anal ($C_{12}H_{13}NO_3$) C, H, N. Lit data [19]: mp 154–156 °C, $[\alpha]_D^{25}$ –97° (c 1.0, MeOH).

N-Isovaleryl-L-proline. Yield 43%, oil, R_f 0.66 (*n*-C_4H_9OH/AcOH/H_2O 5:1:2), $[\alpha]_D^{20}$ –129.3° (c 0.6, $CDCl_3$). NMR ($CDCl_3$): δ 0.99 (d, $CH(CH_3)_2$, 6H); 2.25 (m, $CH(CH_3)_2$, 1H); 2.3 (m, $CH_2CH(CH_3)_2$, 2H); 1.9–2.65 (m, $C^{\beta}H_2$-$C^{\gamma}H_2$, 4H); 3.45–3.70 (m, $C^{\delta}H_2$ Pro, 2H); 8.85 (br s, COOH, 1H). Anal ($C_{10}H_{17}NO_3$) C, H, N.

N-Caproyl-L-proline. Yield 54%, oil, R_f 0.66 (*n*-C_4H_9OH/AcOH/H_2O 5:1:2), $[\alpha]_D^{20}$ –298.0° (c 0.47, DMF). Anal ($C_{11}H_{19}NO_3$) C, H, N.

N-(1-Adamantoyl)-L-proline. Yield 76%, mp 172–174 °C (from EtOH), R_f 0.78 (dioxane/water 9:1), R_f 0.87 (*n*-C_4H_9OH/AcOH/H_2O 4:1:1), $[\alpha]_D^{20}$ –111.46° (c 0.8, $CHCl_3$). NMR (DMSO-d_6): δ 1.62–2.0 (m, Ad, 15H), 1.8–2.1 (m, under Ad, $C^{\beta}H_2$-$C^{\gamma}H_2$ Pro, 4H); 3.73 (br s, $C^{\beta}H_2$ Pro, 2H); 4.21 (br s, $C^{\alpha}H$ Pro, 1H); 12.16 (br s, COOH, 1H). Anal ($C_{16}H_{23}NO_3$) C, H, N.

N-Phenyl-n-butyryl-L-proline. Yield 81%, oil, R_f 0.50 (*n*-C_4H_9OH/AcOH/H_2O 5:1:2), $[\alpha]_D^{20}$ –27.0° (c 0.4, DMF). NMR (DMSO-d_6): δ 1.6–2.2 (m, $C^{\beta}H_2$-$C^{\gamma}H_2$ Pro, 4H); 1.81, 2.26, 2.59 (m and two t $(CH_2)_3$, 6H); 3.3–3.54 (m, $C^{\delta}H_2$ Pro, 2H); 4.22 and 4.40 (each dd, $C^{\alpha}H$ Pro, 1H); 7.05–7.33 (m, Ar, 5H); COOH in the solvent blind peak. Anal ($C_{15}H_{19}NO_3$) C, H, N.

General synthetic method for the preparation of N-acylprolyl-dipeptide esters

To a well-stirred solution of *N*-acylproline (10.0 mmol) in 100 mL chloroform (or ethylacetate), *N*-methylmorpholine (10.0 mmol) and isobutyl chloroformate (10.0 mmol) were added dropwise at –10 °C. After 2 min a mixture of hydrochloride alkyl ester of amino acid (10.0 mmol) and *N*-methylmorpholine (10.0 mmol) in dimethylformamide (20 mL) were added. Stirring continued for 30 min at –5 °C and then the mixture was allowed to stand for 1 h. The precipitate was separated by filtration, the solvent was evaporated in vacuo, the residue was dissolved in chloroform and washed with 5% solution of $NaHCO_3$, water, 1 N HCl, water, dried with Na_2SO_4. After filtration the solvent was evaporated to dryness. To the residue was added ether and the resulting crystals of ester were separated, or the residue was purified by column chromatography using chloroform and the mixture chloroform/ethanol as eluent.

N-Phenylacetyl-L-prolylglycine ethyl ester ***1***. Yield 54%, mp 96–97 °C, $[\alpha]_D^{20}$ –120° (c 0.4, $CHCl_3$), R_f 0.80 (chloroform/water, 9:1). NMR (DMSO-d_6): δ 1.18 (t, CH_3CH_2O, 55% 5H); 1.17 (t, CH_3CH_2O, 45% 3H); 1.65–2.35 (m, $C^{\beta}H_2$-$C^{\gamma}H_2$ Pro, 4H); 3.2–3.4 (m, $C^{\delta}H_2$ Pro, 2H); 3.40 (s, CH_2-C_6H_5, 45% 2H); 3.67 (s, CH_2-C_6H_5, 55% 2H); 3.80 (d, $C^{\alpha}H_2$ Gly, J = 5.9 Hz, 55% 2H); 3.86 (d, J = 5.9 Hz, $C^{\alpha}H_2$ Gly, 45% 2H); 4.08 (q, CH_3-CH_2-O, 55% 2H); 4.09 (q, CH_3-CH_2-O, 45% 2H); 4.32 (dd, $C^{\alpha}H$ Pro, 55% 1H); 4.48 (dd, $C^{\alpha}H$ Pro, 45% 1H); 7.1–7.6 (m, CH_2-C_6H_5); 8.29 (t, J = 5.9 Hz, NH Gly, 55% 1H); 8.63 (t, J = 5.9 Hz, NH Gly, 45% 1H). Anal ($C_{17}H_{22}N_2O_4$) C, H, N.

N-Benzoyl-L-prolylglycine ethyl ester ***2***. Yield 76%, mp 63–65 °C (after triturating with ether), $[\alpha]_D^{20}$ –148.0° (c 0.4, $CHCl_3$), R_f 0.71 (chloroform/methanol 9:1). NMR (DMSO-d_6); δ 1.18 and 1.09 (each t, CH_3CH_2O, 3H); 1.73–2.28 (m, $C^{\beta}H_2$-$C^{\gamma}H_2$ Pro, 4H); 3.3–3.4 (m, $C^{\delta}H_2$ Pro, 2H); 3.60, 3.74 and 3.85 (two dd and d, $C^{\alpha}H_2$ Gly, 2H); 4.10 and 4.13 (each q, CH_3CH_2O, 2H); 4.47 and 4.48 (each dd, $C^{\alpha}H$ Pro, 1H); 7.33–7.62 (m, C_6H_5, 5H); 8.36 and 8.40 (each t, NH, 1H). Anal ($C_{16}H_{20}N_2O_4$) C, H, N.

N-Isovaleryl-L-prolylglycine ethyl ester ***3***. Yield 49%, oil (column chromatography, $CHCl_3$), R_f 0.55 (chloroform/methanol 9:1), $[\alpha]_D^{20}$ –113.4° (c 0.3, $CHCl_3$). NMR ($CDCl_3$); δ 0.99 (d, J = 5.97 Hz, $CH(CH_3)_2$, 6H); 2.22 (m, $CHMe_2$, 1H); 1,27 (t, J = 7.16 Hz, OCH_2CH_3, 3H); 4.18 (q, OCH_2CH_3, 2H); 1.75–2.60 (m, $C^{\beta}H_2$-$C^{\gamma}H_2$ Pro, 4H); 3.35–3.70 (m, CH_2 Pro, 2H); 3.96 and 4.02 (each dd, $C^{\alpha}H_2$ Gly, 2H), 3.85–4.10 (m, CH_2CHMe_2, 2H); 6.50 and 7.59 (each t, NH Gly, 1H). Anal ($C_{14}H_{24}N_2O_4$) C, H, N.

N-Caproyl-L-prolylglycine ethyl ester ***4***. Yield 54%, oil (column chromatography, $CHCl_3$), R_f 0.8 (dioxane/water 9:1), $[\alpha]_D^{20}$ –216° (c 0.2, $CHCl_3$). NMR (DMSO-d_6): δ 0.90 and 0.91 (each t, $CH_3(CH_2)_4$, 3H); 1.19 (t, CH_3CH_3O, 3H); 1.27, 1.50 and 2.25 (two m and t, $CH_3(CH_2)_4$, 8H); 1.70–2.20 (m, $C^{\beta}H_2$-$C^{\gamma}H_2$ Pro, 4H); 3.35–3.50 (m, $C^{\delta}H_2$ Pro, 2H), 3.78 and 3.82 (each d, $C^{\alpha}H_2$ Gly, 2H); 4.08 (q, CH_3CH_2O, 2H); 4.30 and 4.36 (each dd, $C^{\alpha}H$ Pro, 1H); 8.15 and 8.36 (each t, NH Gly, 1H). Anal ($C_{15}H_{26}N_2O_4$) C, H, N.

N-(1-Adamantoyl)-L-prolylglycine ethyl ester 5. Yield 81% mp 177–179 °C (trituration with ether), R_f 0.93 ($CHCl_3$/EtOH 2:3), $[\alpha]_D^{20}$ –66.2° (c 0.6, $CHCl_3$). NMR (DMSO-d_6): δ 1.18 (t, CH_3CH_2O, 3H); 1.66, 1.88 and 1.96 (m, Ad); 1.6–2.0 (m, $C^{\beta}H_2$-$C^{\gamma}H_2$ Pro under Ad); 3.23–3.37 (m, $C^{\delta}H_2$ Pro, 2H); 3.72 and 3.84 (each dd, $C^{\alpha}H_2$ Gly, J = 16.5 Hz, 2H); 4 08 (q, CH_3CH_2O, 2H); 4.9 (br m, $C^{\alpha}H$ Pro, 1H); 8.07 (br t, NH Gly, 1H). Anal ($C_{20}H_{30}N_2O_4$) C, H, N.

N-Phenyl-n-butyryl-L-prolylglycine ethyl ester ***6***. Yield 84%, oil (column chromatography, $CHCl_3$), R_f 0.87 (dioxane/water 9:1); R_f 0.75 ($CHCl_3$/EtOH 9:1), $[\alpha]_D^{20}$ –90.1° (c 0.8, $CHCl_3$). NMR (DMSO-d_6): δ 1.18 (t, CH_3CH_2O, 3H); 1.64–2.23 (m, $C^{\beta}H_2$-$C^{\gamma}H_2$ Pro, 4H); 1.79, 2.28 and 2.59 (m, two t, $(CH_2)_3$, 6H); 3.2–3.6 (m, $C^{\delta}H_2$ Pro, 2H); 3.78 and 3.81 (each d, $C^{\alpha}H_2$ Gly, 2H); 4.07 and 4.09 (each q, CH_3CH_2O, 2H); 4.33 and 4.36 (each dd, $C^{\alpha}H$ Pro, 1H); 7.04–7.35 (m, C_6H_5, 5H); 8.18 and 8.47 (each t, NH Gly, 1H). Anal ($C_{19}H_{26}N_2O_3$) C, H, N.

N-Phenylacetyl-L-prolyl-β-alanine ethyl ester ***11***. Yield 98%, oil, R_f 0.52 (chloroform/methanol 9:1) $[\alpha]_D^{20}$ –92.25° (c 0.3, $CHCl_3$). NMR (DMSO-d_6): δ 1.17 (t, CH_3CH_2O, 68% 3H); 1.13 (t, CH_3CH_2O, 32% 3H); 1.72–2.2 (m, $C^{\beta}H_2$-$C^{\gamma}H_2$ Pro, 4H); 2.42 (m, $C^{\alpha}H_2$ β-Ala, 2H); 3.2–3.3 (m, $C^{\delta}H_2$ Pro, 2H); 3.40 (s, CH_2-C_6H_5 under HDO, 32% 2H); 3.66 (s, CH_2-C_6H_5, 68% 2H); 4.01 (q, CH_3CH_2O, 32%, 2H); 4.04 (q, CH_3CH_2O, 68% 2H); 4.41 (m, $C^{\beta}H_2$, β-Ala, 2H);4.21 (dd, $C^{\alpha}H$ Pro, 1H); 7.1–7.36 (m, CH_2-C_6H_5, 5H); 7.93 (t, NH, 1H). Anal ($C_{18}H_{24}N_2O_4$) C, H, N.

N-Phenylacetyl-L-prolyl-γ-aminobutyric acid methyl ester ***13***. Yield 86%, oil (column chromatography, $CHCl_3$), R_f 0.65 ($CHCl_3$/EtOH 9:1); $[\alpha]_D^{20}$ –93.6° (c 0.4, $CHCl_3$). NMR

(DMSO-d_6): δ 1.63 (m, $C^{\beta}H_2$ GABA, 2H); 1.65–2.16 (m, $C^{\beta}H_2$-$C^{\gamma}H_2$ Pro, 4H); 2.29 (m, $C^{\alpha}H_2$ GABA, 2H); 3.05 (m, $C^{\gamma}H_2$ GABA, 2H); 3.2–3.4 (m, $C^{\delta}H_2$ Pro under HDO, 2H); 3.58 (s, OCH_3, 3H), 3.66 (s, CH_2-C_6H_5, 2H); 4.20 and 4.40 (each dd, $C^{\alpha}H$ Pro 1H); 7.02–7.37 (m, C_6H_5, 5H); 7.85 and 8.20 (each t, NH, 1H). Anal ($C_{18}H_{24}N_2O_4$) C, H, N.

N-Phenylacetyl-L-prolyl-L-glutamic acid diethyl ester ***16***. Yield 69%, oil, R_f 0.9 (dioxane/water 9:1), R_f 0.7 (chloroform/methanol 3:1), $[\alpha]_D^{20}$ –45.9° (c 0.3, $CHCl_3$). NMR ($CDCl_3$): δ 1.25 and 1.27 (each t, 2 CH_3CH_2O, 6H); 1.76–2.49 (m, $C^{\beta}H_2$-$C^{\gamma}H_2$ Pro, $C^{\beta}H_2$-$C^{\gamma}H_2$ Glu, 8H); 3.39–3.92 (m, $C^{\delta}H_2$ Pro, 2H); 3.71 (s, CH_2-C_6H_5, 2H); 4.13 and 4.19 (each q, $2CH_3CH_2O$, 4H); 4.35 and 4.49 (each m, $C^{\alpha}H$ Glu, 1H); 4.49 and 4.61 (each dd, $C^{\alpha}H$ Pro, 1H); 7.15–7.38 (m, C_6H_5, 5H); 7.30 and 7.43 (each d, NH Glu, 1H). Anal ($C_{22}H_{30}N_2O_6$) C, H, N.

N-Phenylacetyl-L-prolyl-L-asparaginic acid diethyl ester ***14***. Yield 95%, oil (column chromatography, $CHCl_3$/C_2H_5OH), R_f ($CHCl_3$/C_2H_5OH 9:3), $[\alpha]_D^{20}$ –38.0° (c 2.2, $CHCl_3$), NMR ($CDCl_3$): δ 1.23 (t, J = 7.16 Hz, CH_3CH_2O, 90% 3H); 1.24 (t, J = 7.16 Hz, CH_3CH_2O, 10% 3H); 4.10 (q, CH_3CH_2O, 90% 2H); 4.12 (q, CH_3CH_2O, 10% 2H); 1.25 (t, J = 7.14 Hz, CH_3CH_2O, 90% 3H); 1.26 (t, J = 7.14 Hz, CH_3CH_2O, 10% 3H); 4.19 (q, CH_3CH_2O, 90% 2H); 4.21 (q, CH_3CH_2O, 10% 2H); 1.75–2.40 (m, $C^{\beta}H_2$-$C^{\gamma}H_2$ Pro, 4H); 3.45–3.65 (m, $C^{\delta}H_2$ Pro, 2H); 4.58 (dd, J = 8.00 Hz, J = 2.59 Hz, $C^{\alpha}H$ Pro, 1H); 2.78, 2.95 (dd, AB-part of ABX, J_{AB} = 17.04 Hz, J_{AX} = 4.88 Hz, J_{BX} = 4.88 Hz, $C^{\beta}H_2$ Asp, 90% 2H); 2.80, 3.00 (dd, $C^{\beta}H_2$ Asp, 10% 2H); 4.81 (dt, $J_{CH,NH}$ = 8.50 Hz, $C^{\alpha}H$ Asp, 1H); 7.5 (d, J = 8.50 Hz, NH Asp, 90% 1H); 7.03 (d, J = 8.40 Hz, NH Asp, 10% 1H); 3.70 (s, $CH_2C_6H_5$, 2H); 7.20–7.36 (m, C_6H_5, 5H). Anal ($C_{21}H_{28}N_2O_6$) C, H, N.

N-Phenylacetyl-L-prolyl-L-alanine ethyl ester ***17***. Yield 78%, mp 48–51 °C (hygroscopic), R_f 0.75 (dioxane/water 10:1), $[\alpha]_D^{20}$ –99.2° (c 0.6, $CHCl_3$). NMR (DMSO-d_6): δ 1.16 (t, CH_3-CH_2O, 3H); 1.27 and 1.31 (each d, CH_3 Ala, 3H); 1.68–2.27 (m, $C^{\beta}H_2$-$C^{\gamma}H_2$ Pro, 4H); 3.46–3.61 (m, $C^{\delta}H_2$ Pro, 2H); 3.65 (s, CH_2 Ar, 2H); 3.98–4.14 (q, CH_3-CH_2O, 2H); 4.34 and 4.48 (each dd, $C^{\alpha}H$ Pro, 1H); 7.12–7.36 (m, C_6H_5, 5H); 8.28 and 8.60 (each d, NH Ala 1H). Anal ($C_{18}H_{24}N_2O_4$) C, H, N.

N-Phenylacetyl-L-prolyl-L-valine ethyl ester ***19***. Yield 72%, oil, R_f 0.64 (dioxane/water 9:1), $[\alpha]_D^{20}$ –99.3° (c 0.35, $CHCl_3$). NMR ($CDCl_3$): δ 0.83 and 0.86 (each d, J = 6.9 Hz, $C^{\beta}H(CH_3)_2$ Val, 90% 6H); 0.89 and 0.95 (each d, J = 6.9 Hz, $C^{\beta}H(CH_3)_2$ Val, 10% 6H); 1.27 (t, CH_3CH_2O, 90% 3H); 1.28 (t, CH_3CH_2O, 10% 3H); 1.7–2.5 (m, $C^{\beta}H_2$-$C^{\gamma}H_2$ Pro, 4H); 2.25 (m, $C^{\beta}H$ Val, 1H); 3.4–3.7 (m, $C^{\delta}H_2$ Pro, 2H); 3.7 (s, CH_2Ar, 2H); 4.18 (q. CH_3-CH_2O, 2H); 4.38 (dd, $C^{\alpha}H$ Val, J = 8.4 Hz, J = 4.9 Hz, 90% 1H); 4.54 (dd, $C^{\alpha}H$ Val, J = 8.4 Hz, J = 4.9 Hz, 10% 1H); 4.68 (dd, $C^{\alpha}H$ Pro, 1H); 7.28 (m, C_6H_5, 5H); 7.44 (d, J = 8.4 Hz, NH Val, 90% 1H); 6.48 (d, J = 8.4 Hz, NH Val, 10% 1H). Anal ($C_{20}H_{28}N_2O_4$) C, H, N.

General synthetic method for the preparation of N-alcylprolyl-dipeptide amides

A solution of *N*-acylprolyldipeptide ester (5 mmol) in 50 mL of methanol was cooled to 0 °C. Amine (dried through NaOH trap) was then bubbled through the solution for 30 min. The solution was maintained at room temperature overnight. Methanol was evaporated in vacuo, and the residue was purified by recrystallization or column chromatography.

N-Phenylacetyl-L-prolylglycine amide ***2***. Yield 95%, oil (column chromatography, $CHCl_3$/MeOH), R_f 0.36 ($CHCl_3$/MeOH 9:1), $[\alpha]_D^{20}$ –58.5° (c 0.2, $CHCl_3$3). NMR ($CDCl_3$): δ 1.8–2.3 (m, $C^{\beta}H_2$-$C^{\gamma}H_2$ Pro, 4H); 3.3–3.5 (m, $C^{\delta}H_2$ Pro, 2H); 3.55–3.75 (m, $C^{\alpha}H_2$ Gly, 2H); 3.66 (s, $CH_2C_6H_5$, 2H); 4.07 (dd, $C^{\alpha}H$ Pro, 1H); 4.37 (t, NH Gly, 1H); 5.63 and 7.86 (each s, NH_2, 2H); 7.2–7.4 (m, C_6H_5, 5H). Anal ($C_{15}H_{19}N_3O_3$), C, H, N.

N-Phenylacetyl-L-prolylglycine methylamide ***9***. Yield 99%, mp 185–186 °C (ether), R_f 0.66 (dioxane/water 9:1), $[\alpha]_D^{20}$ –36.0° (c 0.5, $CHCl_3$). NMR (DMSO-d_6): δ 1.66–2.24 (m, $C^{\beta}H_2$-$C^{\gamma}H_2$ Pro, 4H); 2.49 (d, $NHCH_3$, 85% 3H); 2.60 (d, $NHCH_3$, 15% 3H); 3.61 and 3.63 (each m, $C^{\alpha}H_2$ Gly, 85% 2H); 3.52 and 3.62 (each m, $C^{\alpha}H_2$ Gly, 15% 2H); 3.40–3.60 (m, $C^{\beta}H_2$ Pro, 2H); 3.70 (s, CH_2Ar, 85% 2H); 3.68 (s, CH_2Ar, 15% 2H); 4.23 (dd, $C^{\alpha}H$ Pro, 85% 1H); 4.44 (dd, $C^{\alpha}H$ Pro, 15% 1H); 7.16–7.36 (m, C_6H_5, 5H); 7.58 (q, NHMe, 85% 1H); 7.84 (q, NHMe, 15% 1H); 8.38 (t, NH Gly, 85% 1H); 8.36 (t, NH Gly 15% 1H). Anal ($C_{16}H_{21}N_3O_3$) C, H, N.

N-Benzoyl-L-prolylglycine amide. Yield 76%, mp 64–67 °C (amorphous), R_f 0.34 ($CHCl_3$/EtOH 9:1), $[\alpha]_D^{20}$ –47.9° (c 0.45, $CHCl_3$). NMR (DMSO-d_6): δ 1.68–2.20 and 2.05, 2.30 (m, $C^{\beta}H_2$-$C^{\gamma}H_2$ Pro, 4H); 3.3–3.45 (m, $C^{\delta}H_2$ Pro under HDO, 2H); 3.53–3.75 (m, $C^{\alpha}H_2$ Gly, 2H); 4.20 and 4.40 (each m, $C^{\alpha}H$ Pro, 1H); 6.95–7.65 (m, C_6H_5 and NH_2, 7H); 8.05 and 8.41 (each t, NH Gly. 1H). Anal ($C_{14}H_{17}N_3O_3$) C, H, N.

N-Phenylacetyl-L-prolyl-β-alanine amide ***12***. Yield 61%, oil (column chromatography, $CHCl_3$), R_f 0.28 ($CHCl_3$/MeOH 9:1), $[\alpha]_D^{20}$ –22.8° (c 0.33, $CHCl_3$). NMR (DMSO-d_6): δ 1.69–2.2 (m, $C^{\beta}H_2$-$C^{\gamma}H_2$ Pro, 4H); 2.16–2.31 (m, $C^{\alpha}H_2$ β-Ala, 2H); 3.1–3.3 (m, $C^{\beta}H_2$ β-Ala, 2H); 3.3–3.45 (m, $C^{\delta}H_2$ Pro, 2H); 3.66 (s, CH_2-C_6H_5, 2H); 4.22 and 4.41 (each dd, $C^{\alpha}H$ Pro, 1H); 6.84, 7.36 and 6.86, 7.38 (each br s, NH_2, 2H); 7.12–7.35 (m, CH_2-C_6H_5, 5H); 7.89 and 8.22 (each t, NH β-Ala, 1H). Anal ($C_{16}H_{21}N_3O_3$) C, H, N.

N-Phenylacetyl-L-prolyl-L-asparagine amide ***15***. Yield 89%, mp 170–172 °C (EtOH/$CHCl_3$/pentane 1:1:1), R_f 0.24 ($CHCl_3$/EtOH 9:3), $[\alpha]_D^{20}$ –55.7° (c 1.4, DMSO). NMR (DMSO-d_6): δ 1.60–2.30 (m, $C^{\beta}H_2$-$C^{\gamma}H_2$ Pro, 4H); 2.35–2.50 (m, $C^{\beta}H_2$ Asn, 2H); 3.63 (s, CH_2-C_6H_5, 2H); 4.36 (m, $C^{\alpha}H$ Pro, 1H); 4.4–4.6 (m, $C^{\alpha}H$ Asn, 1H); 6.80–7.60 (m, C_6H_5, 5H); 6.8–7.1 (s, NH_2, 4H); 8.15, 8.25, 8.35 (each d, NH Asn, 1H). Anal ($C_{17}H_{22}N_4O_4$) C, H, N.

N-Phenylacetyl-L-prolyl-L-leucine amide ***18***. Yield 83%, mp 174–175 °C, R_f 0.5 ($CHCl_3$/EtOH 9:1), $[\alpha]_D^{20}$ –101.4° (c 0.4, $CHCl_3$). NMR (DMSO-d_6): δ 0.82 and 0.88 (each d, $C^{\beta}H$-$(CH_3)_2$ Leu, 6H); 1.50 (m, $C^{\beta}H$ Leu, 1H); 1.31–1.93 and 1.7–2.40 (each m, $C^{\beta}H_2$-$C^{\gamma}H_2$ Pro, 4H); 3.43–3.67 (m, $C^{\delta}H_2$ Pro, 2H); 3.69 (s, CH_2Ar, 2H); 4.17 and 4.34 (each m, $C^{\alpha}H$ Leu, 1H); 4.28 and 4.58 (each dd, $C^{\alpha}H$ Pro, 1H); 7.0 and 7.13 (each s, NH_2, 2H); 7.15–7.35 (m, C_6H_5); 7.10–7.45 (m, two s, C_6H_5 and NH_2); 7.86 and 8.27 (each d, NH Leu, 1H). Anal ($C_{19}H_{27}N_3O_3$·0.5 H_2O) C, H, N.

N-Phenylacetyl-L-prolylglycine dimethylamide ***10***

To a solution of 1.6 g (5.5 mmol) *N*-phenylacetyl-L-prolylglycine in 30 mL DMF, 0.68 mL (5.5 mmol) ethylmorpholine and 0.72 mL (5.5 mmol) isobutyl chloroformate were added under stirring at –10 °C. In 2 min gaseous $NHMe_2$ was bubbled to saturation through the reaction mixture. This was further stirred

for 30 min at −5 °C and then evaporated in vacuo. The residue was dissolved in $CHCl_3$, and the solution was washed with 5% aqueous $NaHCO_3$, water, 1 N HCl and water again. The dry ($MgSO_4$) organic phase was concentrated by rotary evaporation and dimethylamide **10** was obtained (1.3 g, 78%) as a syrup. R_f 0.68 (dioxane/water 9:1), $[\alpha]_D^{20}$ −147.1° (c 0.1, $CHCl_3$). NMR (DMSO-d_6): δ 1.71–2.06 (m, $C^{\beta}H_2$-$C^{\gamma}H_2$ Pro, 4H); 2.83, 2.93 and 2 84, 2.96 (each s, $N(CH_3)_2$, 6H); 3.3–3.6 (m, $C^{\delta}H_2$ Pro, 2H); 3.67 (s, CH_2Ar, 2H); 3.89 and 3.95 (each d, $C^{\alpha}H_2$ Gly, 2H); 4.37 and 4.52 (each dd, $C^{\alpha}H$ Pro, 1H); 7.15–7.34 (m, CC_6H_5, 5H); 7.88 and 8.24 (each t, NH Gly, 1H). Anal ($C_{17}H_{23}N_3O_3$) C, H, N.

N-Phenylacetyl-L-prolylglycine **8**

A suspension of 1.06 g (3.5 mmol) of *N*-phenylacetyl-L-prolylglycine, ethyl ester **1** in 5 mL of 1 N NaOH was stirred at room temperature for 3 h to obtain a solution. It was then acidified with 2 N HCl to ~ pH 3. The solution was evaporated in vacuo to obtain an oil. The oil was dissolved in 15 mL chloroform, the unsolubilized part was removed by filtration and the filtrate was evaporated. Ether was added to the residue and the solid was filtered and dried in vacuo at room temperature to obtain 0.9 g (89.9%) of the product, mp 159–160 °C, R_f 0.54 (dioxane/water 9:1), $[\alpha]_D^{20}$ −85.8° (c 0.5; $CHCl_3$). NMR spectrum in DMSO-d_6: δ 1.8–2.25 (m, $C^{\beta}H_2$-$C^{\gamma}H_2$ Pro, 4H); 3.36–3.63 (m, $C^{\delta}H_2$ Pro, 2H); 3 64 and 3.68 (each s, CH_2-C_6H_5 2H); 3.86, 4.00 and 3.83, 4.02 (each dd, $C^{\alpha}H_2$ Gly, 2H); 4.57 and 4.44 (each dd, $C^{\alpha}H$ Pro, 1H); 7.11–7.38 (m, C_6H_5, 5H); 7.52 and 7.32 (each t, NH 1H); 12.6 (br s. COOH 1H). Anal ($C_{15}H_{18}NO_4$) C, H, N.

Pharmacology

Amnesia-reversal testing

These experiments were carried out on adult male outbred rats (Krjukovo, Moscow region) weighing 180–240 g. The step-through passive-avoidance test [16] was used for estimating memory retention in rats in the apparatus Lafayett Instrument Co (USA) with lighted platform (25 × 7 cm) and dark compartment (40 × 40 × 40 cm) according to Ader et al [20]. A rat was placed in a well-lit start platform and was oriented away from the dark compartment, which had an electrified grid floor. When the animal entered the dark compartment through a guillotine square door (6.5 × 6.5 cm) it received eight avoidable painful footshocks (0.45 mA). The rat was then removed from the chamber. On the retention test performed 24 h after training the animal was again placed on the lit platform. The latency to enter into the dark compartment was registered. Maximal electroconvulsive shock (MES, 70 V, 300 ms) was used immediately after training. MES caused amnesia of passive avoidance and rats entered into the dark compartment with a short latency period. Compounds dissolved in saline or saline alone were injected intraperitoneally 15 min before the trial. Antiamnesic activity (AA) was calculated according to the formula [21]:

$$\%AA = \frac{L(MES + compound) - L(MES)}{L(control) - L(MES)} \cdot 100\%$$

Where L(MES) is the average latent time to enter the dark compartment for animals exposed to MES 24 h ago, L(MES + compound) is the average latent time to enter the dark compartment for animals received the compounds and exposed to MES, and L(control) is the average time to enter the dark compartment for saline-treated animals with sham MES.

Statistical analysis was carried out using the Mann–Whitney *U*-test.

The influence on locomotor activity

An Opto-varimex multichannel motor activity recorder (Columbus Instrument Co, USA) was used for evaluation of a possible sedative or stimulant effect of compounds. Substances were administered intraperitoneally; 15 min later the rat was placed into the cage of the recorder and its horizontal activity during 5 min was registered.

Anticonvulsant activity

The maximal ECS (70 V, 300 ms) inducing tonic-clonic seizures in rats was used to study a possible anticonvulsant compounds activity. The substances or vehicle were injected intraperitoneally 15 min before testing.

References

1 Giurgea C (1982) *Drug Develop Res* 2, 441–446
2 Gouliaev AH, Senning A (1994) *Brain Res Rev* 19, 180–222
3 Gudasheva TA, Ostrovskaya RU, Trofimov SS et al (1985) *Khim-Farm Zh (Russia)* 19, 1322–1329
4 Gudasheva TA, Ostrovskaya RU, Trofimov SS et al (1988) *Khim-Farm Zh (Russia)* 22, 271–275
5 Gudasheva TA, Ostrovskaya RU (1994) *Can J Physiol Pharmacol* 72 Suppl 1, 418
6 Ostrovskaya RU, Gudasheva TA, Trofimov SS et al (1987) *Bull Exp Biol Med* 104, 1561–1564 (English version)
7 Chepkova AN, Doreulee NV, Trofimov SS, Gudasheva TA, Ostrovskaya RU, Skrebitskiy VG (1995) *Neurosci Lett* 188, 161–166
8 Romanova GA, Gudasheva TA, Dobrinin VP, Ostrovskaya RU (1992) Eksper *i Klinic Farmacol (Russia)* 55, 6–8
9 Ostrovskaya RU, Gudasheva TA, Chepkova AN, Trolmov SS (1995) *Behav Pharmacol* 6, 628
10 Burbach JPH, Kovacs GL, De Wied D, Van Nispen JW, Greven HM (1983) *Science* 221, 1310–1312
11 Gudasheva TA, Vasilevich NI, Zolotov NN et al (1991) *Khim-Farm Zh (Russia)* 25, 12–15
12 Winitz M, Bloch-Frankenyhal L, Lzumiya N, Birnbaum SM, Baker CC, Greenstein JD (1956) *J Am Chem Soc* 78, 2423–2427
13 Bures J, Buresova O (1963) *J Comp Physiol Psychol* 56, 268–272
14 Voronina TA (1992) *Sov Med Rev G Neuropharmacology* 2, 51–106
15 Voronina TA, Kutepova OA (1988) *Drug Develop Res* 14, 353–358
16 Irwin S (1964) *In: Animal and Clinical Pharmacologic Techniques in Drug Evalution* (Nodine JU, Siegler PE, eds), Chicago Year Book Medical Publishers, 36–54
17 Gudasheva TA, Rozantsev GG, Ostrovskaya RU et al (1995) *Khim-Farm Zh (Russia)* 29, 15–18
18 Abraham DJ, Gazze DM, Kennedy PE, Mokotoff M (1984) *J Med Chem* 27, 1549–1559
19 Davies JS, Thomas WA (1978) *J Chem Soc Perkin Trans II* 1157–1163
20 Ader R, Weijnen JAWM, Moleman P (1972) *Psychonomic Sci* 26, 125–128
21 Butler DE, Nordin IC, L'Italien YJ, Zweisler L, Poschel PH, Marriot JC (1984) *J Med Chem* 27, 684–691