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Synthesis, 5-HT_{1A} and 5-HT_{2A} receptor affinity of new 1-phenylpiperazinylpropyl derivatives of purine-2,6- and pyrrolidine-2,5-diones

Maciej Pawłowski [a,*], Grażyna Chłoń [a], Jolanta Obniska [a], Alfred Zejc [a], Sijka Charakchieva-Minol [b], Maria J. Mokrosz [b]

[a] *Department of Pharmaceutical Chemistry, Collegium Medicum of Jagiellonian University, Medyczna 9, 30-688 Krakow, Poland*
[b] *Institute of Pharmacology, Polish Academy of Sciences, Smętna 12, 31-343 Krakow, Poland*

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Abstract

Two series of 1-phenylpiperazinylpropyl derivatives **10**, **11**, **16**, **17** and **19**–**24**, structurally related to previously described 5-HT_{1A} or 5-HT_{2A} ligands **4** and **1**, respectively, were synthesized and their binding properties were determined. Structural modifications which involved 1,3-diazepine ring opening in **4** (compounds **10**, **11**, **15**, **16**) and replacement of spiroalkyl moiety in **1** by aryl substituent (**19**–**24**) did not improve binding affinity and selectivity of the tested compounds. The results showed, however, that the diazepine ring present in **4** or spiroalkyl ring in **1** are important for high 5-HT_{1A} or 5-HT_{2A} binding affinity and selectivity of these compounds.

Keywords: 1-Phenylpiperazinylpropylpurine-2,6-diones; 1-Phenylpiperazinylpropyl-pyrrolidine-2,5-diones; 5-HT_{1A} and 5-HT_{2A} receptor affinity

1. Introduction

Serotonin (5-hydroxytryptamine, 5-HT) is involved in many physiological and pathophysiological processes in the brain which are mediated by the specific interaction of 5-HT with seven major receptor classes [1]. The 5-HT_{1A} receptors are to date one of the best characterized subtypes and it is generally accepted that they are involved in psychiatric disorders such as depression and anxiety [2]. Several compounds from different chemical classes possess high affinity for 5-HT_{1A} receptors. Among them, some 1-arylpiperazines linked with a terminal cyclic amide fragment via a long-chain, e.g. buspirone or ipsapirone (Fig. 1), are effective antianxiety and antidepressant drugs [3]. Many studies on the structure–affinity relationship of the 1-arylpiperazine class of 5-HT_{1A} receptor ligands have been reported [4–6]. Misztal et al. [7] assumed that the terminal amide fragment in buspirone-like ligands stabilized the 5-HT_{1A} receptor-ligand complex by either π-electron or local dipole–dipole interaction. Also, the results obtained by Mokrosz et al. [8] showed that the basic nitrogen atom and terminal, bulky cycloimide moiety, but not the 2-pyrimidinyl group of buspirone, are directly involved in the formation of the bioactive complex with 5-HT_{1A} receptors.

In previous studies [9] we showed that 3-(ω-aminoalkyl)-5,5-dialkyl (or spirocycloalkyl-1′,5-)hydantoins containing 1-phenylpiperazine or 1-(*o*-methoxyphenyl)-piperazine fragments are recognized by 5-HT_{1A} and 5-HT_{2A} receptors due to the presence of the 1-arylpiperazine fragment; however, the terminal hydantoin moiety plays an important role in the stabilization of the receptor–ligand complex. It has also been found that two selected compounds, 5-cyclopentanespiro-3-[3-(4-phenyl-1-piperazinyl)-propyl]-imidazolidine-2,4-dione (**1**) and its 5-cyclohexanespiro analog (**2**) ($K_i = 34$ and 37 nM, respectively) are new selective 5-HT_{1A} receptor ligands, whereas the 5-cyclohexanespiro-3-[3-(4-*o*-methoxyphenyl-1-piperazinyl)-butyl]-imidazolidine-2,4-dione (**3**) is a new, highly potent 5-HT_{1A} receptor ligand ($K_i = 0.51$ nM) with a moderate affinity for 5-HT_{2A} receptors ($K_i = 213$ nM) [9] (Fig. 1). In addi-

* Corresponding author. Tel.: +48-12-657 0560; fax: +48-12-657 0262.

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tion, 5-HT_{1A} and 5-HT_{2A} receptor affinities of several ω-alkyl-1-arylpiperazines containing a terminal pyrimidopurine or 1,3-diazepinopurine ring system have been reported [10]. Several behavioral models demonstrated that 1,3-dimethyl-10-[3-(4-phenyl-1-piperazinyl)-propyl]-2,4-dioxo-1,3,6,7,8,9-hexahydro-10H-1,3-diazepino-[2,1-*f*]-purine (**4**) and its analog (**5**) may be classified as 5-HT_{1A} postsynaptic antagonists, whereas 1,3-dimethyl-9-[3-(4-phenyl-1-piperazinyl)-butyl]-2,4-dioxo-1,3,6,7,8,9-hexahydropyrimido-[2,1-*f*]-purine (**6**) is a partial agonist of 5-HT_{1A} receptors [10] (Fig. 1).

Although the terminal amide fragment significantly affects binding of 1-arylpiperazine derivatives to serotonin receptors, its role is not clear yet. To evaluate the influence of the 1,3-diazepine ring in **4** and of the spiroalkyl ring in **1** on their binding affinity and selectivity, two series of compounds, i.e. **10**, **11**, **16** and **17** related to **4** and **19**–**24** related to **1**, were synthesized and their $5HT_{1A}$ and 5-HT_{2A} binding constants were determined.

2. Chemistry

The 4-(3-aminopropyl)-l-phenylpiperazine **7** was synthesized based on the described method [11]. The 1,3-dimethyl-7-alkyl-8-bromo-purine-2,6-diones **8** and **9** were prepared according to the published procedures [12]. The 1,3-dimethyl-7-alkyl-8-[3-(4-phenyl-1-piperazinyl)-propylamino]-purine-2,6-diones **10** and **11** were obtained by reaction of the appropriate 1,3-dimethyl-7-alkyl-8-bromo-purine-2,6-diones **8** or **9** with **7** in the presence of anhydrous K_2CO_3 (Scheme 1).

The previously reported ethyl esters of 1,3-dimethyl-8-bromo-purine-2,6-dione-7-acetic acid **12** [13] and 1,3-dimethyl-8-bromo-purine-2,6-dione-7-butyric acid **13** [14] were heated with **7** in the presence of K_2CO_3 in *n*-butanol. In these reactions, the nucleophilic substitution in the 8 position took place and the esters were transformed into the appropriate potassium salts. After treating with HCl solution, the free acids **14** and **15** were isolated. For the synthesis of the esters **16** and **17**

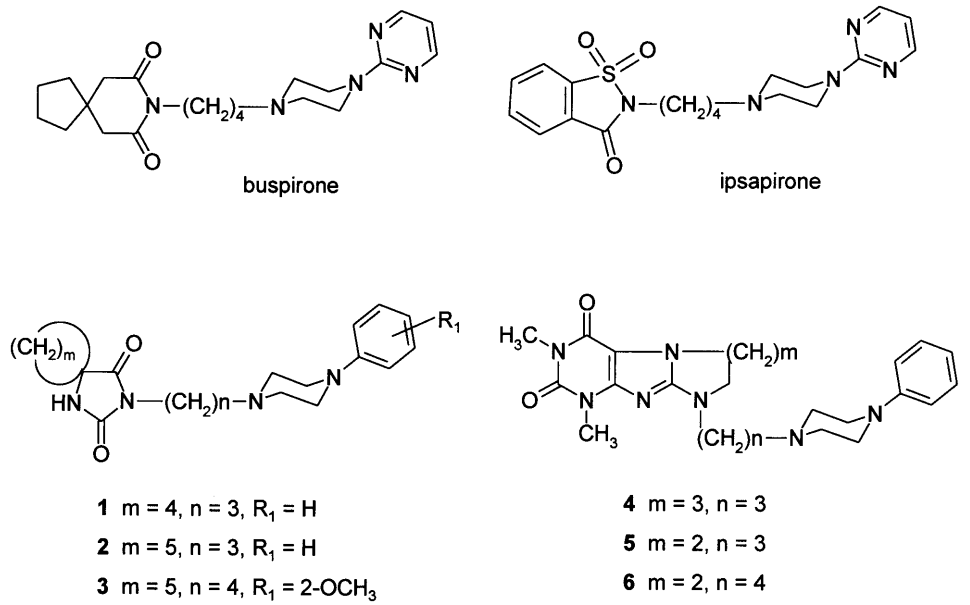
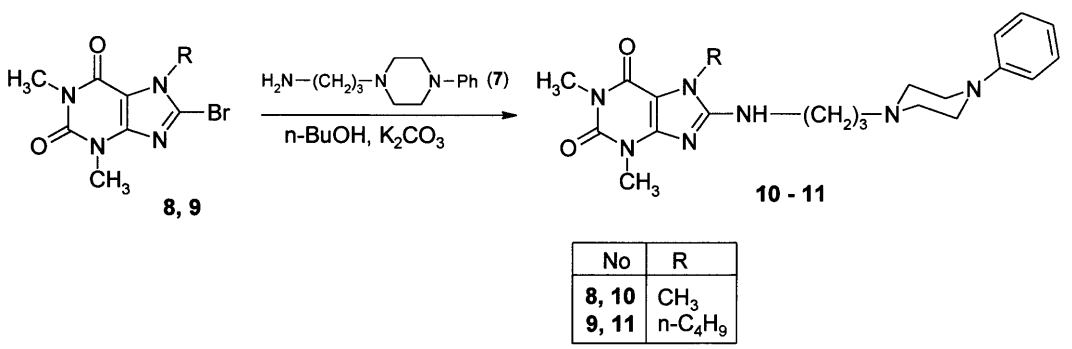
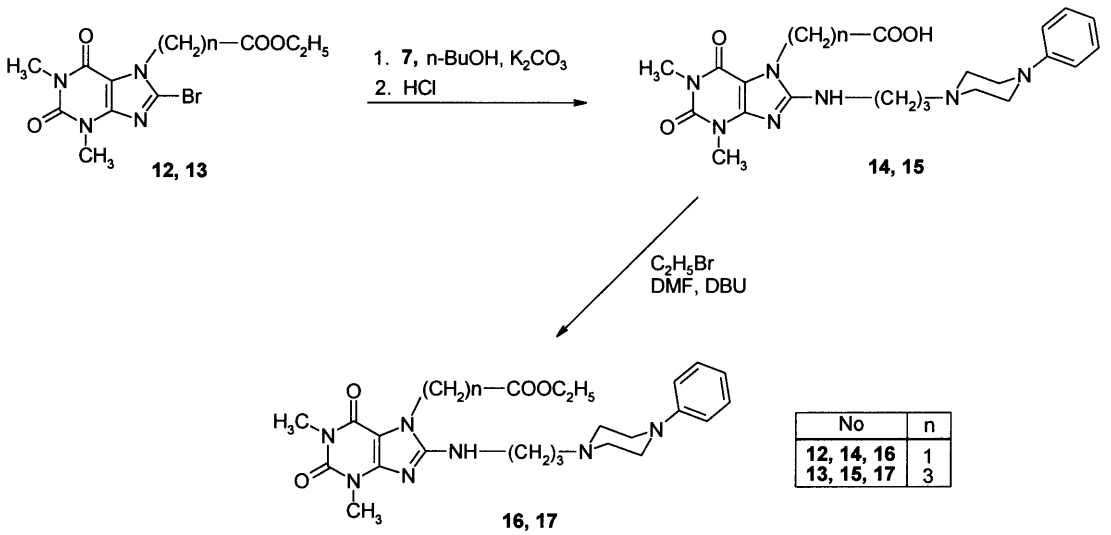


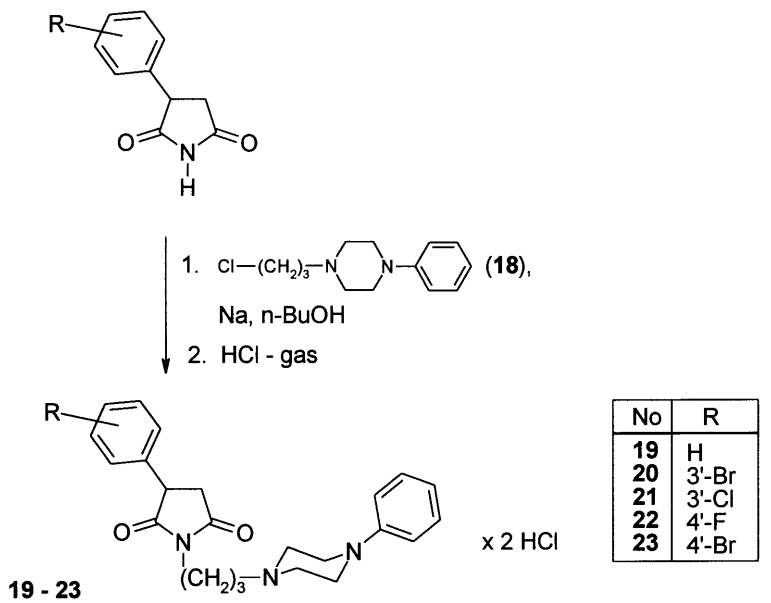
Fig. 1.



Scheme 1.



Scheme 2.



Scheme 3.

compounds **14** and **15** were *O*-alkylated by ethyl bromide in DMF, in the presence of 1,8-diazabicyclo-[5,4,0]-undec-7-ene (DBU) (Scheme 2).

The phenylpropylpiperazine derivatives of 3-aryl-pyrrolidine-2,5-diones **19**–**23** were obtained by several condensation steps. For the synthesis of 1-(3-chloro-propyl)-4-phenylpiperazine **18** the reaction of 1-phenylpiperazine with 1-bromo-3-chloropropane was developed [6].

The N-unsubstituted pyrrolidine-2,5-diones with aryl substituent in 3-position were obtained by intramolecular cyclocondensation of appropriate succinic acid derivatives with ammonia [15,16]. The reaction of 3-aryl-pyrrolidine-2,5-diones with **18** finally led to *N*-[(4-phenyl-1-piperazinyl)-propyl]-3-aryl-pyrrolidine-2,5-diones **19**–**23** which were isolated as hydrochlorides (Scheme 3).

Compound **24** was prepared by cyclocondensation of 2,2-diphenylsuccinic acid [15,16] with **7** (Scheme 4).

The structures of **10**, **11**, **16**, **17** and **19**–**24** were confirmed by examination of their ^{1}H NMR and MS spectra as well as by elemental analyses. For binding studies free bases of **10**, **11**, **16** and **17** were converted into hydrochloride salts.

3. Experimental

3.1. Chemistry

Analytical results for C, H, N of compounds **10**, **11**, **16**, **17** and **19**–**24** were within $\pm 0.4\%$ of their theoretical values. Melting points are uncorrected. UV spectra were recorded on a Perkin–Elmer Lambda 12 UV–Vis spectrometer in 5×10^{-5} mol/l methanolic solutions. ^{1}H NMR spectra (Varian BB 200 MHz NMR spectrometer) were determined in $CDCl_3$ solution, with TMS as internal standard. All chemical shifts are quoted in δ values. MS spectra were obtained with an AMD-604 spectrometer (70 eV). The chromatography was performed with Merck silica gel GF_{254} precoated TLC sheets using the following developing systems: A, 1:1:1 benzene–acetone–methanol; B, 60:10:5 chloroform–methanol–acetic acid; C, 9:11:2 chloroform–isopropanol–NH_3 aq. Spots were detected by their absorption under UV light and visualization with 0.05 mol J_2 in 10% HCl. Some starting materials were commercial products (Aldrich or Fluka).

3.2. Chemical procedures

3.2.1. General procedure for the preparation of the 1,3-dimethyl-7-alkyl-8-[3-(4-phenyl-1-piperazinyl)-propylamino]-purine-2,6-diones (**10** and **11**)

A mixture of the appropriate 1,3-dimethyl-7-alkyl-8-bromo-purine-2,6-diones **8** or **9** (0.01 mol), amine **7** (2.19 g, 0.01 mol), anhydrous K_2CO_3 (2.76 g, 0.02 mol), and *n*-butanol (20 ml) was refluxed for 12 h. Then the reaction mixture was cooled and refrigerated overnight. The precipitated solid product was filtered off, washed with H_2O and then purified by crystallization from 50° ethanol or *i*-propanol.

3.2.2. 1,3,7-Trimethyl-8-[3-(4-phenyl-1-piperazinyl)-propylamino]-purine-2,6-dione hydrochloride (**10**)

Free base of **10** from **8**. Yield 1.64 g (40%); m.p. 190–192°C (*i*-propanol). *Anal.* ($C_{21}H_{29}N_7O_2$) C, H, N. TLC: $R_f = 0.83$ (A), $R_f = 0.42$ (B); UV (methanol): λ_{max} ($\log \varepsilon$) = 294 nm (4.104); ^{1}H NMR: $\delta = 1.85$–1.94 (m, 2H, $CH_2CH_2CH_2$), 2.62–2.72 (m, 6H, $CH_2N(CH_2)_2$), 3.20–3.25 (m, 4H, $(CH_2)_2N$–Ph), 3.36 (s, 3H, N1–CH_3), 3.51 (s, 3H, N3–CH_3), 3.55–3.58 (m, 2H, $NHCH_2$), 3.61 (s, 3H, N7–CH_3); 6.67 (t, $J = 4.3$ Hz, 1H, $NHCH_2$), 6.85–6.95 (m, 3H aromatic, Ph); 7.24–7.32 (m, 2H aromatic, Ph); MS *m*/*z* (%): 411 (54) [M^+], 396 (56) [$M^+ - CH_3$], 305 (34), 279 (100), 250 (38), 236 (78), 222 (14), 208 (5), 203 (8), 195 (5), 194 (4), 188 (3), 175 (27), 161 (73), 132 (26), 77 (6).

Free base of **10** was converted into HCl salt in anhydrous ethanol saturated with HCl gas. The obtained precipitate was crystallized from ethanol.

HCl salt **10**. M.p. 254–257°C (ethanol). *Anal.* ($C_{21}H_{29}N_7O_2$xHCl) C, H, N.

3.2.3. 1,3-Dimethyl-7-n-butyl-8-[3-(4-phenyl-1-piperazinyl)-propylamino]-purine-2,6-dione hydrochloride (**11**)

Free base of **11** from **9**. Yield 1.4 g (31%); m.p. 135–136°C (*i*-propanol). *Anal.* ($C_{24}H_{35}N_7O_2$) C, H, N. TLC: $R_f = 0.90$ (A), $R_f = 0.67$ (B). UV (methanol): λ_{max} ($\log \varepsilon$) = 295 nm (4.032); ^{1}H NMR: $\delta = 0.85$ (t, $J = 7.2$ Hz, 3H, CH_2CH_3), 1.22–1.33 (m, 2H, CH_2CH_3), 1.65–1.73 (m, 2H, $CH_2CH_2CH_3$), 1.85–1.94 (m, 2H, $CH_2CH_2CH_2$), 2.60–2.72 (m, 6H, $CH_2N(CH_2)_2$), 3.21–3.26 (m, 4H, $(CH_2)_2N$–Ph), 3.37 (s, 3H, N1–CH_3); 3.52 (s, 3H, N3–CH_3), 3.55–3.63 (m, 2H, $NHCH_2CH_2$), 3.98 (t, $J = 7.2$ Hz, 2H, N–7CH_2), 6.32 (t, $J = 4.4$ Hz, 1H, $NHCH_2$), 6.89–6.95 (m, 3H aromatic, Ph), 7.24–7.32 (m, 2H, aromatic Ph); MS *m*/*z* (%): 453 (44) [M^+], 438 (42), 321 (100), 292 (17), 278 (48), 264 (4), 250 (2), 208 (7), 203 (6), 195 (5), 194 (2), 189 (3), 175 (23), 161 (59), 132 (19), 77 (7), 57 (5), 43 (3).

HCl salt of **11** was prepared according to the procedure of HCl salt of **10**. M.p. 210–212°C (ethanol). *Anal.* ($C_{24}H_{35}N_7O_2$xHCl) C, H, N.

3.2.4. General procedure for the synthesis of acids **14** and **15**

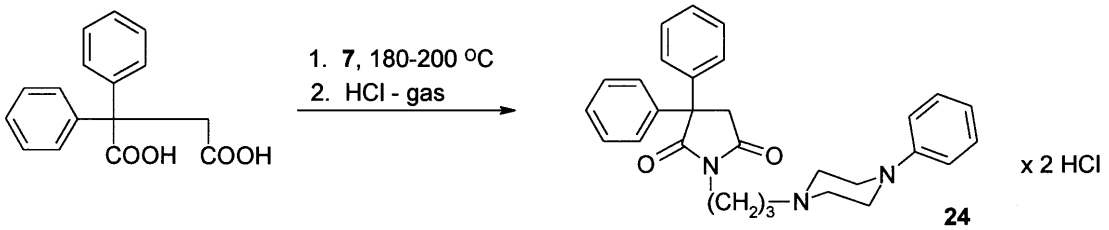
A mixture of the appropriate ester (**12** or **13**) (0.01 mol), amine **7** (2.19 g, 0.01 mol), anhydrous K_2CO_3 (4.14 g, 0.03 mol) and *n*-butanol (30 ml) was refluxed for 5 h. After cooling the precipitated potassium salt was filtered off, dissolved in water (50 ml) and the solution was acidified to pH 2–3 by the addition of 5% HCl. The precipitated acid **14** or **15** was collected, washed with H_2O and cold methanol.

3.2.5. 1,3-Dimethyl-8-[3-(4-phenyl-1-piperazinyl)-propylamino]-purine-2,6-dione-7-acetic acid (**14**)

From **12**. Yield 2.89 g (60%); m.p. 242–244°C (ethanol); *Anal.* ($C_{22}H_{29}N_7O_4$) C, H, N; TLC: $R_f = 0.16$ (A); ^{1}H NMR: $\delta = 1.90$–1.96 (m, 2H, $CH_2CH_2CH_2$), 2.62–2.77 (m, 6H, $CH_2N(CH_2)_2$), 2.81–2.90 (m, 4H, $(CH_2)_2N$–Ph), 3.21 (s, 3H, N1–CH_3), 3.36 (s, 3H, N3–CH_3) 3.42–3.46 (m, 2H, $NHCH_2$), 4.62 (s, 2H, N7–CH_2CO), 6.89–6.95 (m, 3H aromatic, Ph), 7.25–7.33 (m, 2H aromatic, Ph), 8.43 (t, $J = 4.4$ Hz, 1H, $NHCH_2$); MS *m*/*z* (%): 455 (2) [M^+], 437 (59) [$M^+ - H_2O$], 422 (60), 396(4), 305(100), 276(15), 262(6), 248(11), 189(6), 175(64), 161(18), 132(50), 77(4).

3.2.6. 1,3-Dimethyl-8-[3-(4-phenyl-1-piperazinyl)-propylamino]-purine-2,6-dione-7-butyric acid (**15**)

From **13**. Yield 2.17 g (45%); m.p. 209–211°C (ethanol); *Anal.* ($C_{24}H_{33}N_7O_4$) C, H, N; TLC; $R_f = 0.28$ (A); ^{1}H NMR: $\delta = 2.05$–2.1 (m, 4H, $CH_2CH_2CH_2$ and



Scheme 4.

CH_2CH_2CO), 2.20–2.23 (m, 2H, CH_2CH_2CO), 2.90–3.3 7 (m, 10H, $CH_2N(CH_2)_2$ and $(CH_2)_2N$–Ph), 3.37 (s, 3H, N1–CH_3), 3.51 (s, 3H, N3–CH_3), 3.55–3.57 (m, 2H, $NHCH_2$), 4.16 (t, $J = 6$ Hz, 2H, N7–CH_2CH_2), 6.43 (t, 1H, $NHCH_2$), 6.89–6.96 (m, 3H aromatic, Ph), 7.25–7.33 (m, 2H aromatic, Ph); MS m/z (%): 483(56) [M^+], 468(71), 466(5), 450(24), 351(100), 322(13), 308(56), 208(8), 195(8), 194(4), 189(6), 175(75), 161(84), 132(14), 87(2), 77(10).

*3.2.7. General method for the preparation of esters **16** and **17***

To a solution of the suitable acid (**14** or **15**) (0.005 mol) in DMF (20 ml), DBU (0.75 ml, 0.005 mol) was added, followed by ethyl bromide (1.12 ml, 0.015 mol). The mixture was stirred at room temperature (r.t.) for 3 h. Then water (150 ml) was added, and the precipitated product (**16** or **17**) was filtered off, washed with water and purified by crystallization from 96% ethanol.

*3.2.8. Ethyl ester of 1,3-dimethyl-8-[3-(4-phenyl-1-piperazinyl)-propylamino]-purine-2,6-dione-7-acetic acid hydrochloride (**16**)*

Free base of **16** from **14**. Yield 1.52 g (63%); m.p. 156–158°C (ethanol); *Anal.* ($C_{24}H_{34}N_7O_4$) C, H, N; TLC: $R_f = 0.82$ (A); UV: (methanol): λ_{max} (log ε) = 296 nm (4.372); ^{1}H NMR: $\delta = 1.20$ (t, $J = 7.1$ Hz, 3H, CH_2CH_3), 2.03–2.10 (m, 2H, $CH_2CH_2CH_2$), 3.04–3.20 (m, 6H, $CH_2N(CH_2)_2$), 3.14 (s, 3H, N1–CH_3), 3.36 (s, 3H, N3–CH_3), 3.52–3.94 (m, 4H, $(CH_2)_2N$–Ph), 4.12–4.19 (m, 4H, $NHCH_2CH_2$ and CH_2CH_3) 4.96 (s, 2H, N7–CH_2CO), 6.86 (t, $J = 7.1$ Hz, 1H, $NHCH_2$), 6.98–7.02 (m, 5H aromatic, Ph), 7.22–7.66 (m, 2H aromatic, Ph); MS m/z (%): 483(47) [M^+], 468(53), 422(55), 396(4), 351(62), 308(56), 294(4), 208(8), 203(8), 194(3), 189(9), 175(83), 161(53), 132(68), 77(16), 70(100).

HCl salt of **16** was prepared according to the procedure for the HCl salt of **10**. M.p. 218–220°C (ethanol); *Anal.* ($C_{24}H_{34}N_7O_4 \times HCl$) C, H, N.

*3.2.9. Ethyl ester of 1,3-dimethyl-8-[3-(4-phenyl-1-piperazinyl)-propylamino]-purine-2,6-dione-7-butyric acid (**17**)*

From **15**. Yield 1.35 g (53%); m.p. 128–130°C (ethanol); *Anal.* ($C_{26}H_{37}N_7O_4$) C, H, N; TLC: $R_f = 0.92$ (A); UV: λ_{max} (log ε) = 295 nm (4.381); ^{1}H NMR: $\delta = 1.21$–1.28 (t, $J = 7.1$ Hz, 3H, CH_2CH_3), 1.86–1.96 (m, 2H, $CH_2CH_2CH_2$), 2.00–2.03 (m, 2H, N7–$CH_2CH_2CH_2$), 2.31–2.38 (t, $J = 6.6$ Hz, 2H, CH_2CH_2CO), 2.54–2.68 (m, 6H, $CH_2N(CH_2)_2$), 3.20–3.25 (m, 4H, $(CH_2)_2N$–Ph, 3.31 (s, 3H, N1–CH_3), 3.52 (s, 3H, N3–CH_3), 3.56–3.62 (m, 2H, $NHCH_2$), 4.04–4.17 (m, 4H, OCH_2CH_3 and N7–CH_2), 6.25 (t, $J = 5$ Hz, 1H, $NHCH_2$), 6.86–6.95 (m, 3H aromatic, Ph), 7.23–7.31 (m, 2H aromatic, Ph); MS m/z (%): 511 (51) [M^+], 496(65), 466(24), 450(3), 379(100), 350(10), 336(62), 322(2), 208(4), 203(8), 194(3), 189(3), 175(34), 161 (82), 132(21), 115(27).

*3.2.10. 1-(3-Chloropropyl)-4-phenylpiperazine (**18**)*

A mixture of 1-phenylpiperazine (4.99 g, 0.03 mol), 1-bromo-3-chloropropane (6.3 g, 0.04 mol) and anhydrous K_2CO_3 (1.2 g, 0.009 mol) in acetone (20 ml) was stirred for 24 h at r.t. Then the inorganic salt was filtered off and acetone was evaporated to dryness. The residue was dissolved in *n*-hexane, filtered off again and the solvent was evaporated. The crude product of **18** was purified using a silica gel column chromatography eluting system 1:1 chloroform–ethyl acetate.

*3.2.11. General method for the synthesis of imides **19**–**24***

To the sodium butoxide solution, prepared from sodium (0.23 g, 0.01 mol) and *n*-butanol (50 ml), appropriate 3-substituted succinimide (0.01 mol), and **18** (2.83 g, 0.001 mol) were added. The reaction mixture was refluxed for 1 h and left overnight at r.t. Then the solvent was evaporated and the residue was treated with benzene (100 ml) and 20% K_2CO_3 (10 ml). The organic layer was separated, washed with water, dried over anhydrous KCO_3 and filtered off. The solvent was evaporated. The resulting free base was dissolved in ethanol, treated with an excess of ethanol saturated with dry HCl gas and kept in a refrigerator to obtain crystalline products.

*3.2.12. N-[(4-Phenyl-1-piperazinyl)-propyl]-3-phenyl-pyrrolidine-2,5-dione dihydrochloride (**19**)*

Yield 2.61 g (58%); m.p. 191–194°C (ethanol); *Anal.* ($C_{23}H_{29}N_3O_2Cl_2$) C, H, N; TLC: $R_f = 0.79$ (B), $R_f = 0.83$ (C); UV: λ_{max} (log ε) = 205 nm (3.502). ^{1}H NMR: $\delta = 2.06$ (t, $J = 7.6$ Hz, 2H, $CH_2CH_2CH_2$), 2.70–2.82

(dd, $J = 5.3$ Hz, 1H, *H*-imide), 3.15–3.19 (m, 4H, $(CH_2)_2$N–Ph), 3.24–3.37 (q, $J = 9.4$ Hz, 1H, *H*-imide), 3.40–3.43 (d, $J = 6.9$ Hz, 2H, CH_2N), 3.49–3.55 (t, $J = 6.5$ Hz, 4H, N$(CH_2)_2$), 3.76–3.81 (d, $J = 9.9$ Hz, 2H, NCH_2), 4,19 (q, $J = 5.2$ Hz, 1H, C*H*-imide), 6.82–7.36 (m, 10H, aromatic Ph).

*3.2.13. N-[(4-phenyl-1-piperazinyl)-propyl]-3-(3-bromophenyl)-pyrrolidine-2,5-dione dihydrochloride (**20**)*

Yield 3.3 g (63%); m.p. 220–223°C (ethanol); *Anal.* ($C_{23}H_{28}N_3O_2BrCl_2$) C, H, N; TLC: $R_f = 0.74$ (B), $R_f = 0.81$ (C); UV: λ_{max} (log ε) = 207 nm (3.590); λ_{max} (log ε) = 250 nm (3.145); ^{1}H NMR: $\delta = 2.04$ (t, $J = 7.5$ Hz, 2H, $CH_2CH_2CH_2$), 2.76–2.86 (dd, $J = 5.6$ Hz, 1H, *H*-imide), 3.12–3.26 (m, 7H, $(CH_2)_2$N–Ph, *H*-imide, CH_2N), 3.50–3.53 (t, $J = 6.6$ Hz, 4H, N$(CH_2)_2$), 3.76–3.82 (d, $J = 10.2$ Hz, 2H, NCH_2), 4.25 (q, $J = 5.7$ Hz, 1H, *H*-imide), 6.83–7.62 (m, 9H, aromatic Ph).

*3.2.14. N-[(4-phenyl-1-piperazinyl)-propyl]-3-(3-chlorophenyl)-pyrrolidine-2,5-dione dihydrochloride (**21**)*

Yield 2.8 g (58%); m.p. 204–206°C (ethanol); *Anal.* ($C_{23}H_{28}N_3O_2Cl_3$) C, H, N; TLC: $R_f = 0.83$ (B), $R_f = 0.92$ (C); ^{1}H NMR: $\delta = 2.03$ (t, $J = 7.6$ Hz, 2H, $CH_2CH_2CH_2$), 2.78–2.88 (dd, $J = 5.7$ Hz, 1H, *H*-imide), 3.12–3.26 (m, 7H, $(CH_2)_2$N–Ph, *H*-imide, CH_2N), 3.47–3.53 (t, $J = 6.7$ Hz, 4H, N$(CH_2)_2$), 3.76–3.82 (d, $J = 10.6$ Hz, 2H, NCH_2), 4.26 (q, $J = 5.7$ Hz, 1H, *H*-imide); 6.83–7.49 (m, 9H, aromatic, Ph).

*3.2.15. N-[(4-Phenyl-1-piperazinyl)-propyl]-3-(4-fluorophenyl)-pyrrolidine-2,5-dione dihydrochloride (**22**)*

Yield 2.57 g (55%); m.p. 212–214°C (ethanol); *Anal.* ($C_{23}H_{28}N_3O_2FCl_2$) C, H, N; TLC: $R_f = 0.75$ (B), $R_f = 0.80$ (C); ^{1}H NMR: $\delta = 2.05$ (t, $J = 7.5$ Hz, 2H, $CH_2CH_2CH_2$), 2.72–2,81 (dd, $J = 5.5$ Hz, 1H, *H*-imide), 3.13–3.26 (m, 7H, $(CH_2)_2$N–Ph, *H*-imide, CH_2–N), 3.47–3.53 (t, $J = 6.7$ Hz, 4H, N$(CH_2)_2$), 3.76–3.81 (d, $J = 10.9$ Hz, 2H, NCH_2), 4.24 (q, $J = 5.5$ Hz, 1H, *H*-imide); 6.83–7.43 (m, 9H, aromatic, Ph).

*3.2.16. N-[(4-Phenyl-1-piperazinyl)-propyl-3-(4-bromophenyl)-pyrrolidine-2,5-dione dihydrochloride (**23**)*

Yield 2.7 g (51%); m.p. 210–212°C (ethanol); *Anal.* ($C_{23}H_{28}N_3O_2BrCl_2$) C, H, N; TLC: $R_f = 0.69$ (B), $R_f = 0.78$ (C); ^{1}H NMR: $\delta = 2.03$ (t, $J = 7.4$ Hz, 2H, $CH_2CH_2CH_2$), 2.73–2.82 (dd, $J = 5.5$ Hz, 1H, *H*-imide), 3.13–3.27 (m, 7H, $(CH_2)_2$N–Ph, *H*-imide, CH_2–N), 3.46–3.53 (t, $J = 6.3$ Hz, 4H, N$(CH_2)_2$), 3.76–3.81 (d, $J = 10.8$ Hz, 2H, NCH_2), 4.22 (q, $J = 5.5$ Hz, 1H, *H*-imide); 6.83–7.57 (m, 9H, aromatic, Ph).

*3.2.17. N-[(4-Phenyl-1-piperazinyl)-propyl]-3,3-diphenyl-pyrrolidine-2,5-dione dihydrochloride (**24**)*

To a mixture of amine **7** (4.38 g, 0.02 mol) in water (50 ml), 3,3-diphenylsuccinic acid (5.4 g, 0.02 mol) was gradually added. The mixture was then heated on an oil bath with simultaneous distillation of water. After complete removal of water the reaction temperature was raised to 200°C and heating was maintained for 2 h. The crude product, isolated as hydrochloride salt, was purified by recrystallization from ethanol.

Yield 5.15 g (49%); m.p. 202–204°C (ethanol); *Anal.* ($C_{29}H_{33}N_3O_2Cl_2$) C, H, N; TLC: $R_f = 0.89$ (B), $R_f = 0.64$ (C); ^{1}H NMR: $\delta = 2.02$ (t, $J = 7.5$ Hz, 2H, $CH_2CH_2CH_2$), 3.06–3.26 (m, 6H, $(CH_2)_2$N–Ph,CH_2N), 3.37 (s, 2H, CH_2-imide), 3.52–3.59 (t, $J = 6.9$ Hz, 4H, N$CH_2)_2$, 3.75–3.82 (d, $J = 9.8$ Hz, 2H, NCH_2), 6.82–7.43 (m, 15H, aromatic, Ph).

3.3. Pharmacology: binding experiments

Radioligand binding studies were carried out in the rat brain using the hippocampus (5-HT_{1A}) and cortex (5-HT_{2A}) according to published procedures [17]. Radioligands used were [^{3}H]-8-OH-DPAT (190 Ci/mmol, Amersham) and [^{3}H]-ketanserin (60 Ci/mmol, NEN Chemicals) for 5-HT_{1A} and 5-HT_{2A} receptors, respectively.

K_i values were determined on the basis of at least three competition binding experiments in which 10–14 drug concentrations (10^{-10}–10^{-3} M), run in triplicate, were used. K_i values were calculated using an ACCUFIT (Lundon Software) program.

4. Results and discussion

The hydrochlorides **10**, **11**, **16**, **17** and **19**–**24** were evaluated for their affinities for 5-HT_{1A} and 5-HT_{2A} receptors by determining their ability to displace [^{3}H]-8-OH-DPAT in the hippocampus and [^{3}H]-ketanserin in the cortex of rat brain, respectively. The obtained results are summarized in Table 1.

All the evaluated compounds **10**, **11**, **16**, **17** and **19**–**24** showed similar, moderate 5-HT_{1A} and 5-HT_{2A} receptor affinity (K_i within 10^{-7}–10^{-6} M) and rather low 5-HT_{2A}/5-HT_{1A} selectivity. Removal of the 1,3-diazepine ring in **4** resulted in a 12-fold decrease in 5-HT_{1A} affinity of **10**. Its *n*-butyl analog **11**, structurally similar to **4**, showed 5-HT_{1A} binding constant three times lower than the parent compound. Increasing the lipophilicity of the 7-substituent also did not improve binding properties of **16** and **17** (Table 1). In the second series replacement of spiroalkyl fragment of **1** with a 3-phenyl substituent produced **19**, which demonstrated the same weak 5-HT_{1A} affinity as **1**, but seven times lower 5-HT_{2A} binding constant. Introduction of a halogen atom on the 3-phenyl ring in **19** enhanced the 5-HT_{1A} affinity of compounds **20**–**23**, but their 5-HT_{2A} properties remained on the comparable moderate level as for **19** (Table 1). This suggests that the 3-aryl sub

Table 1
5-HT_{1A} and 5-HT_{2A} receptor affinities (K_i) and selectivity ratios ($S_{2,1}$) of compounds **1–3**, **4–6**, **10**, **11**, **16**, **17** and **19–24**

Comp.	$K_i \pm$ SEM (nM) [a]		Selectivity ratio $S_{2.1} = K_i$(5-HT_{2A})/K_i(5-HT_{1A})
	5-HT_{1A}	5-HT_{2A}	
1 [b]	1185 ± 45	34 ± 6	0.029
2 [b]	830 ± 60	37 ± 3	0.045
3 [b]	0.51 ± 0.06	213 ± 4	418
4 [c]	31 ± 2	258 ± 14	8.3
5 [c]	16 ± 2	422 ± 51	26
6 [c]	2.1 ± 0.2	243 ± 46	116
10	385 ± 5	197 ± 6	0.51
11	100 ± 1	614 ± 10	6.14
16	592 ± 72	179 ± 5	0.30
17	457 ± 4	1010 ± 14	2.21
19	1069 ± 50	268 ± 4	0.25
20	177 ± 10	329 ± 21	1.85
21	258 ± 27	388 ± 14	1.50
22	223 ± 9	436 ± 8	1.95
23	510 ± 14	436 ± 7	0.85
24	444 ± 30	842 ± 11	1.89
8-OH-DPAT [b]	1.43 ± 0.21		
Ritanserin [b]		1.14 ± 0.13	

[a] The mean value from at least three independent experiments; Hill slopes $n_H = 0.84$–1.08.
[b] Data taken from Ref. [9].
[c] Data taken from Ref. [10].

stituent in pyrrolidine-2,5-dione is tolerated better than the spiroalkyl fragment at 5-HT_{1A} receptors, but is apparently less favoured at 5-HT_{2A} binding sites.

In conclusion, the present results show that structural modifications in both series of compounds do not improve binding affinity and selectivity in comparison to the parent compounds. They demonstrate, however, that other elements, i.e. 1,3-diazepine ring in **4** or spiroalkyl ring in **1** are important for high 5-HT_{1A} or 5-HT_{2A} binding affinity and selectivity of these compounds.

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References

[1] D. Hoyer, G.R. Martin, Classification and nomenclature of 5-HT receptors: A comment on current issues, Behav. Brain Res. 73 (1995) 263–268.
[2] S.J. Peroutka, A.J. Sleight, B.G. McCarthy, P.A. Pierce, A.W. Schmidt, C.R. Hekmatpanah, The clinical utility of pharmacological agents that act at serotonin receptors, J. Neuropsychiatry Clin. Neurosci. 1 (1989) 253–262.
[3] C.L.E. Broekkamp, D. Leysen, B.W.M.M. Peters, R.M. Pinder, Prospects for improved depressants, J. Med. Chem. 38 (1995) 4615–4633.
[4] R. Perrone, F. Berardi, M. Leopoldo, V. Tortorella, M.G. Fornaretto, C. Caccia, R.A. McArthur, 1-Aryl-4-[(1-tetralinyl)alkyl]piperazines: alkylamido and alkyamino derivatives. synthesis, 5-HT_{1A} receptor affinity and selectivity, J. Med. Chem. 39 (1996) 3195–3202.
[5] B.J. van Steen, I. van Wijngaarden, M.T.M. Tulp, W. Soudijn, Structure–affinity relationship studies on 5-HT_{1A} receptor ligands 1. Heterocyclic phenylpiperazines with N4-alkyl substituents, J. Med. Chem. 36 (1993) 2751–2760.
[6] J.L. Mokrosz, M.H. Paluchowska, E. Chojnacka-Wojcik, M. Filip, S. Charakchieva-Minol, A. Dereń-Wesotek, M.J. Mokrosz, Structure–activity relationship studies of central nervous system agents 13. 4-[3-(Benzotriazol-1-yl)propyl]-1-(2-methoxyphenyl) piperazine, a new putative 5-HT_{1A} receptor antagonist, and its analogs, J. Med. Chem. 37 (1994) 2754–2760.
[7] S. Misztal, A. Bojarski, M. Maćkowiak, J. Boksa, Z. Bielecka, J.L. Mokrosz, Structure–activity relationship studies of CNS agents 6. Effect of terminal amide fragment on 5-HT_{1A} and 5-HT_{2A} receptor affinity for *N*-[3-(4-aryl-1-piperazinyl)-propyl]derivatives of 3,4-dihydroquinolin-2-(1*H*)-one and its isomeric isoquinolinones, Med. Chem. Res. 2 (1992) 82–87.
[8] J.L. Mokrosz, A. Dereń-Wesotek, E. Tatarczyfiska, B. Duszyfiska, A.J. Bojarski, M.J. Mokrosz, E. Chojnacka-Wójcik, 8-[4-[2-(1,2,3,4-Tetrahydroisoquinolinyl)]butyl]-8-azaspiro[4,5]-decane-7,9-dione: a new 5-HT_{1A} receptor ligand with the same activity profile as buspirone, J. Med. Chem. 39 (1996) 1125–1129.
[9] H. Byrtus, M. Pawłowski, S. Charakchieva-Minol, B. Duszyńska, M.J. Mokrosz, J.L. Mokrosz, A. Zejc, 3-(ω-Aminoalkyl)-5,5-dialkyl (or spirocycloalkyl-l′,5) hydantoins as new 5-HT_{1A} and 5-HT_{2A} receptor ligands, Arch. Pharm. Pharm. Med. Chem. 329 (1996) 283–290.
[10] E. Chojnacka-Wojcik, A. Klodzińska, A. Drabczyńska, M. Pawłowski, S. Charakchieva-Minol, G. Chtoń, M. Gorczyca, A new putative 5-HT_{1A} receptor antagonist of the 1-arylpiperazine class of ligands, Eur. J. Med. Chem. 30 (1995) 587–592.

[11] R.A. Glennon, N.A. Naiman, R.A. Lyon, M. Titeler, Arylpiperazine derivatives as high affinity 5-HT_{1A} serotonin ligands, J. Med. Chem. 31 (1988) 1968–1971.
[12] V.B. Kalcheva, T.M. Apostolova, V.Z. Anakieva, A Procedure for the preparation of *N*-(ω-haloalkyl)xanthines based on phase-transfer catalysis, J. Prakt. Chem. 327 (1985) 165–168.
[13] F. Cacace, R. Criserà, M. Zifferero, Preparazione di alcuni derivati degli acidi 8-Br-teofillin 7-acetico e 8-alchilammino-teofillin-7-acetici, Ann. Chim. (Roma) 46 (1956) 99–104.
[14] J. Karolak-Wojciechowska, M. Pawłowski, Synthesis, crystal and molecular structure of 1,3-dimethyl-10-benzyl-2,4,9-trioxo-1,3,6,7,8,10-hexahydro-1,3-diazapino-[2,1-*f*]-purine, J. Crystallogr. Spectrosc. Res. 20 (1990) 477–482.
[15] C.A. Miller, L.M. Long, Anticonvulsants 1. An investigation of N-R-α-R_1-α-phenylsuccin imides, J. Am. Chem. Soc. 73 (1951) 4895–4898.
[16] J. Lange, J. Tapszewicz, Synthesis and properties of phenylsuccinic acid VII. New phenylsuccinimide derivatives with anticonvulsant properties, Acta Polon. Pharm. 3 (1978) 289–294.
[17] A.J. Bojarski, M.T. Cegta, S. Charakchieva-Minol, M.J. Mokrosz, M. Maćkowiak, S. Misztal, J.L. Mokrosz, Structure–activity relationship studies of CNS agents. Part 9: 5-HT_{1A} and 5-HT_{2A} receptor affinity of some 2- and 3-substituted 1,2,3,4-tetrahydrocarbolines, Pharmazie 48 (1993) 289–294.