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Novel Buspirone-Like 5-HT$_{1A}$ Receptor Ligands

by K.J. Krajewski[1], A. Leś[1,2], J. Cybulski[1], Z. Chilmonczyk[3], A. Bronowska[2] and A. Szelejewska-Woźniakowska[1]

[1]*Pharmaceutical Reaserch Institute, 8 Rydygiera Str., 01-793 Warszawa, Poland*
[2]*Departament of Chemistry, University of Warsaw, 1 Pasteura Str., 02-093 Warszawa, Poland*
[3]*Drug Institute, 30/34 Chełmska Str., 00-725 Warszawa, Poland*

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Four novel buspirone-like compounds were synthesized and their affinity to the serotonin 5-HT$_{1A}$ receptor was determined. Two compounds bind to 5-HT$_{1A}$ receptor with a high affinity and two other compounds exhibit a low affinity to this receptor. A simple two-parameter polynomial correlation was suggested to express the structure-activity relationships.

Key words: anxiolytics, pharmacophores, conformational analysis, structure-activity relationships

Buspirone [1] (Scheme 1, **1**) is an 1-arylpiperazine derivative with a high affinity to the 5-HT$_{1A}$ receptor [2] and with a high selectivity to the other serotonin receptor subtypes. Buspirone, classified as 5-HT$_{1A}$ partial agonist, was introduced for clinical use in the middle of the eighties. Recent investigations have led to the discovery of many new selective 5-HT$_{1A}$ receptor ligands, exhibiting anxiolytic activity with similar to buspirone pharmacological profile, *e.g.* tandospirone [3] (Scheme 1, **2**) or different from it, *e.g.* SUN 8399 [4], lesopitrone [5], FG-5893 [6], (S)-WAY-100135 [7]. Simultaneously, theoretical studies [8] were undertaken to explain the structure-affinity or structure-intrinsic activity relationships. The 3D-molecular structure of 5-HT$_{1A}$ receptor is incomplete yet. Till now only a few topographic 5-HT$_{1A}$ receptor models exist [9–16] and it is hard to decide which of them is the best one.

RESULTS AND DISCUSSION

In a search of 5-HT$_{1A}$ serotonin receptor ligands, new buspirone-like analogues **4**–**7** were obtained in our Institute (Scheme 1). The compounds **3**–**7** (the compound **3** was designed earlier by Glennon [17]) were screened for *in vitro* binding affinities for rat hippocampus 5-HT$_{1A}$ receptor labelled with [^{3}H]-8-hydroxy-2-(di-n-propylamino)tetralin. Binding affinity data (IC$_{50}$ and K$_i$) are summarized in Table 1.

The compounds **4**–**7** were obtained by the general method according to the Scheme 1. 2-Chloroheteroaromatic derivatives were treated with piperazine or 2-methylpiperazine to give **8**–**11** arylpiperazines, which were subsequently condensed with N-(4-bromobutyl)-phthalimide under phase transfer conditions, in the presence of 18-crown-6, potassium carbonate and toluene as a solvent, to give finally compounds (**4**–**7**).

Table 1. The $5HT_{1A}$ binding data for compounds **3**–**7**.

Compound	$5HT_{1A}$ binding	
	IC_{50} (nM)	K_i (nM)
3	n.a.	36
4	217	126
5	n.a.	39
6	24	13
7	1192	590

n.a. – not available; IC_{50} – concentration of unlabelled ligands causing 50% inhibition of specific [^{3}H]-8-hydroxy-2-(di-n-propylamino)tetralin ([^{3}H]8-OH-DPAT) binding (rat hippocampus). The K_i value was calculated as $K_i = IC_{50}/(1 + [L]/K_D)$, where [L] is the concentration of the hot ligands (~1 nM), and K_D is dissociation constant (0.98 nM).

Due to the lack of the X-ray crystal structure analysis for the compounds examined, a conformational analysis has been carried out by theoretical methods with the aid of molecular mechanics implemented in the Sybyl 6.2 package. The internal Tripos force field was used to obtain the molecular energy. For molecular geometry optimization and the conformational analysis, the conjugate gradient method was used. For compounds **3**, **5**, **7**, four isomeric structures were constructed. Each of them corresponds to a different conformation of the piperazine ring with substituents located at the axial or equatorial positions. For the compounds **4** and **6** eight isomeric structures were constructed, because the 3-methyl substituent can adopt two different locations ("axial" or "equatorial") at the piperazine ring. Each of the structures was optimized in the course of the minimal internal energy search. In the next step the optimal position of the aromatic and piperazine ring was kept and the five aliphatic bonds were rotated (scanned with the increment of 30 degrees under the Systematic Search option). The rotamers were rejected, if their internal energy exceeded by 4 kcal mol^{-1} the energy of the optimal structure. This algorithm generated a large number of conformations (about 1×10^6 for the compounds **3**, **5** and **7**, and about 2×10^6 for the compounds **4** and **6**) which, after a suitable sorting, were aligned according to the internal energy values. The ten lowest-energy conformations were selected for a subsequent optimization process. Finally, the resulted conformation with the minimal internal energy was chosen. These procedures were repeated (usually 3–4 times), until the minimal internal energy converged within some preselected threshold. In total 3–8 millions conformations were analysed per compound. The above algorithm is expected to generate conformations that are close to the optimal structure. Twenty low-energy conformations for each of compounds **3**, **5**, and **7** (5 low-energy structures for each of 4 conformations of the piperazine ring, *i.e.* where the nitrogen atoms can bear the substituents placed at the "axial" or "equatorial" positions with respect to the C_2-axis perpendicular to the ring) and forty low-energy conformations for compounds **5** and **6** (the number of structures was doublet due to the "axial" and "equatorial" positions of the methyl group) were selected for the subsequent analysis. On the basis of the 3-point pharmacophore model [16] we attempted to find relationships between the af-

Scheme 1

HetAr-Cl + H–N(piperazine, R)N–H →(i) HetAr–N(piperazine, R)N–H

8, **9**, **10**, **11**

→(ii)

4, **5**, **6**, **7**

4, **8** - R = CH_3, HetAr = 2-pirymidynyl-; **5**, **9** - R = H, HetAr = 2-quinolinyl-
6, **10** - R = CH_3, HetAr = 2-quinolinyl-; **7**, **11** - R = H, HetAr =2-(4,6-dimethoxy-1,3,5-triazinyl)-

i (for compounds **8**, **9**, **10**) - EtOH, K_2CO_3, reflux

i (for compounds **11**) - CH_2Cl_2, reflux

ii - N-(4-bromobutyl)phthalimide, 18-crown-6, K_2CO_3, toluene, reflux

4 **8** **9**

5 **10** **11**

6 **1**

7 **2**

3

finity (for 5-HT_{1A} receptor) and the theoretical 3D-structures for the compounds **3**–**7**. We chose this model because it includes structural features characteristic for compounds **3**–**7**, such as the heteroaromatic ring, the basic nitrogen atom in the piperazine ring and the carbonyl group of imide moiety. Chilmonczyk's pharmacophore model was constructed based on the crystal structures of several the high-affinity 5-HT_{1A} ligands, which were structurally close-related to compounds **3**–**7**, using the potent 5-HT_{1A} receptor ligand (+)-LSD (K_i = 2.5 nM, [18]) as a template. This pharmacophore possesses three binding centres: the aromatic ring centroid, the basic nitrogen atom and the imide oxygen atom (Fig. 1).

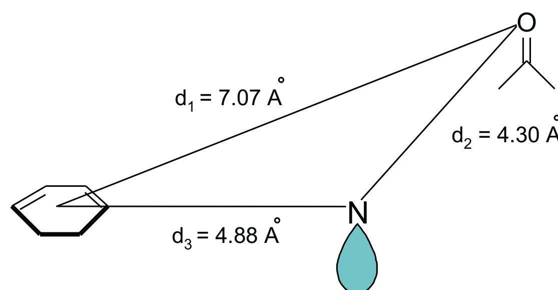

Figure 1. A 3-point pharmacophore model of 5-HT_{1A} ligands.

For each of **3**–**7** compounds three low-energy conformers were selected. Subsequently, these conformers were compared to the Chilmonczyk's pharmacophore with the aid of the RMS quantity, *i.e.* the criterion of geometrical similarity:

$$RMS = \sqrt{\sum_{i=1}^{n=3}(d_{1i} - 7.07)^2 + \sum_{i=1}^{n=3}(d_{2i} - 4.30)^2 + \sum_{i=1}^{n=3}(d_{3i} - 4.88)^2} \quad (1)$$

The d_{1i}, d_{2i} and d_{3i} (for i = 1, 2, 3 low-energy conformers) correspond to the d_1, d_2 and d_3 distances shown in Figure 1. The RMS values were correlated with the K_i values that characterize the affinity to the 5-HT_{1A} receptor. In Figure 2 one can see a plausible correlation obtained with a 2-parameter polynomial fit.

The monotonic non-linear K_i *vs* RMS correlation presented in Figure 2 suggests that one can expect low K_i values for low RMS values. For large RMS values fairly low affinities towards the 5-HT_{1A} receptor can be expected. It is interesting to notice that the combined approach, where the pharmacophore model [16] was used in a connection to a simple, non-linear structure-activity correlation, can be a useful tool for the activity predictions of new compounds (structurally related to **3**–**7**). However, there are some drawbacks of this approach. The affinity data (Table 1) show that the introduction of the methyl substituent into the piperazine ring, close to the central nitrogen atom, drops almost 3 times the affinity of the resulted compound **4** (as compared to **3**). A similar reduction of activity upon methylation of tandospirone **(2)** (Scheme 1) was observed earlier [3]. The opposite result was obtained, while the pyrimidine moiety was replaced by the quinoline moiety, as in the **5**/**6** pair of com-

pounds. The presence of the methyl group in the piperazine ring (compound **6**) improved the affinity towards 5-HT$_{1A}$ receptor almost 3 times, compared to **5**. The reason of affinity reduction (Table 1) caused by the replacement of the quinoline moiety **5** by the 2-(4,6-dimethoxy-1,3,5-triazinyl) moiety **7** is unclear at this moment.

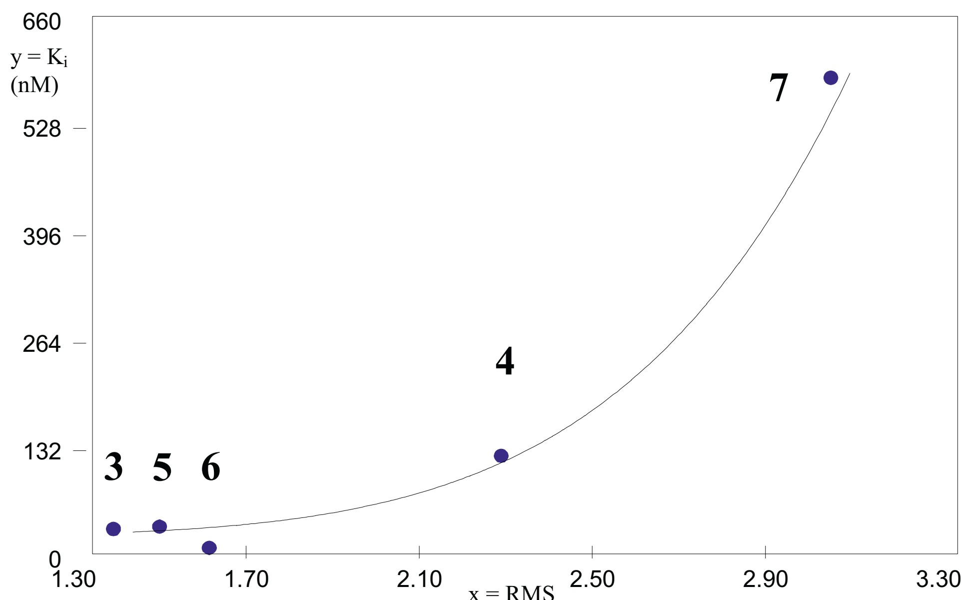

Figure 2. The 2-parameter polynomial fit: $y = a_0 + a_1 \cdot x^6$, where $a_0 = 21.66$, $a_1 = 0.70$, $r^2 = 0.997$.

EXPERIMENTAL

Melting points were determined on a Boetius apparatus (Carl Zeiss Jena) and are uncorrected. IR spectra were recorded on a Perkin Elmer 1725X spectrophotometer. ^{1}H NMR and ^{13}C NMR spectra were obtained using a Varian Gemini 2000 (200 MHz) with TMS as an internal standard; chemical shifts are given in δ (ppm) and coupling constants (*J*) in Hz. GC/MS spectra were recorded on a Hewlett-Packard GC model 5890 with 5970 mass detector by the electron impact (EI) method. 2-Methylpiperazine, piperazine, 2-chloro-4,6-dimethoxy-1,3,5-triazine, N-(4-bromobutyl)phthalimide, 2-chloropyrimidine were commercial products (Aldrich, Fluka). Reagents and solvents were purchased from common commercial suppliers. For column chromatography, Merck Kieselgel 100 (70–230 mesh) was used. 1-(2-Quinolinyl)-piperazine (**9**) [19] and 1-(2-pyrimidinyl)-3-methylpiperazine (**8**) [3] were synthesized by published procedures with some modifications.

Synthesis: General procedure for the preparation of compounds 8–10 from the corresponding 2-chloroheteroaromatic and piperazine or 2-methylpiperazine. The mixture of 2-chloroheteroaromatic (0.1 mol), piperazine or 2-methylpiperazine (0.3 mol), anhydrous K_2CO_3 (0.012 mol) in 99.8 % ethanol (~300 ml) was stirred and refluxed for 3 h (for compound **10** the reaction mixture was stirred and refluxed for 20 h). The hot reaction mixture was filtered and the filter cake washed with ethanol. The combined filtrates were concentrated under reduced pressure. Crude product was dissolved in chloroform and washed with water. Then, organic layer was extracted of 2 M HCl aq. Acid aqueous layer was separated and neutralized by 2 M NaOH and washed with chloroform. The combined organic layers were dried and concentrated to give product.

1-(2-Pyrimidinyl)-3-methylpiperazine (8) [3]. From 2-chloropyrimidine and 2-methylpiperazine. Yield 93% (16.5 g), yellow oil.

1-(2-Quinolinyl)-piperazine (9) [19,20]. From 2-chloroquinoline and piperazine. Crude product was crystallized from 99.8% ethanol to give 8.3 g of 1-(2-quinolinyl)-piperazine. Yield 32%. M.p. = 117–120°C (118–120°C [20]).

1-(2-Quinolinyl)-3-methylpiperazine (10). From 2-chloroquinoline and 2-methylpiperazine, yield 44% (10 g). M.p. = 70–72°C. ^{1}H NMR ($CDCl_3$): δ = 1.18 (d, J = 7 Hz, 3H, CH_3), 2.53–2.65 (m, 1H, piperazine H), 2.83–3.22 (m, 4H, piperazine H), 4.27–4.48 (m, 2H, piperazine H), 6.97 (d, J = 9 Hz, 1H, quinoline H), 7.16–7.28 (m, 1H, quinoline H), 7.46–7.75 (m, 3H, quinoline H), 7.88 (d, J = 9 Hz, 1H, quinoline H); ^{13}C NMR ($CDCl_3$): δ = 157.28, 147.81, 137.38, 129.45, 127.12, 126.58, 123.03, 122.33, 109.49, 52.14, 50.59, 45.56, 45.13, 19.38; MS (70 eV): m/z (%) = 227 (19)[M^+], 183 (26), 171 (100), 157 (36), 128 (30); IR (KBr): ν = 3282, 3054, 2934, 1617, 1602, 1557, 1233 cm^{-1}. Anal. Calcd. for $C_{14}H_{17}N_3$: C, 73.97; H, 7.54; N, 18.49%. Found: C, 73.68; H, 7.40; N, 18.31%.

1-[2-(4,6-Dimethoxy-1,3,5-triazynyl)]piperazine (11). A solution of (0.02 mol) 2-chloro-4,6-dimethoxy-1,3,5-triazine in dichloromethane (30 ml) was added dropwise into a solution of piperazine (0.08 mol) in dichloromethane (30 ml) for 30 minutes at room temperature. Then, the reaction mixture was stirred for 30 minutes. After cooling below 10°C, an aqueous sodium hydroxide solution (5%) was added. The organic layer was separated and washed with water and extracted with 2 M HCl. Acid-aqueous layer was separated, neutralized by 5% NaOH and washed with chloroform (2 × 25 ml). The combined organic layers were dried and concentrated to give 1-[2-(4,6-dimethoxy-1,3,5-triazynyl)]-piperazine (4.2 g, 95%). M.p. = 134– 136°C. ^{1}H NMR ($CDCl_3$): δ = 1.79 (s, 1H, NH), 2.84–2.94 (m, 4H, piperazine H), 3.79–3.88 (m, 4H, piperazine H), 3.95 (s, 6H, OCH_3); ^{13}C NMR ($CDCl_3$): δ = 173.05, 167.16, 55.06, 53.45, 44.08; MS (70 eV): m/z (%) = 225 (19)[M^+], 195 (19), 183 (33), 169 (35), 157 (100), 140 (17), 126 (20), 83 (25), 69 (42), 56 (50), 42 (44); IR (KBr): ν = 3253, 2931, 1592, 1545, 1479, 1454, 1284 cm^{-1}. Anal. Calcd. for $C_9H_{15}N_5O_2$: C, 48.00; H, 6.71; N, 31.09%. Found: C, 47.72; H, 6.85; N, 30.89%.

General procedure for the preparation of compounds 4–7 from the corresponding arylpiperazine and N-(4-bromobutyl)phthalimide. The mixture of arylpiperazine (0.005 mol), N-(4-bromobutyl)phthalimide (0.006 mol), anhydrous K_2CO_3 (0.012 mol) and 18-crown-6 (1 mg) in toluene (40 ml) was stirred and refluxed for 20 h. Then, the reaction mixture was filtered and the filter cake washed with toluene. The combine filtrates were washed with water (2 × 20 ml) and with aqueous 1 M HCl solution (3 × 10 ml). The acid aqueous layer was separated, basified by 5% aqueous NaOH solution to pH ~ 9 and extracted with chloroform (2 × 20 ml). The organic layer was dried and concentrated, afterwards the crude product was purified by column chromatography with chloroform.

General procedure for the preparation of hydrochlorides and hydrobromides. Free bases were converted into hydrochlorides with 2.02 M ethanolic HCl and into hydrobromides with mixture of 48% hydrobromic acid in THF (1:10).

N-[4-(2-Methyl-4-pyrimidin-2-yl-piperazin-1-yl)-butyl]phthalimide (4). From 1-(2-pyrimidinyl)-3-methylpiperazine [3] and N-(4-bromobutyl)phthalimide, yield 1 g (29%), yellow oil. ^{1}H NMR ($CDCl_3$): δ = 1.10 (d, J = 6.2 Hz, 3H, H13), 1.44–1.82 (m, 4H, H15, H16), 2.20–2.52 (m, 3H, H14, H9), 2.71–3.04 (m, 3H, H11, H8), 3.19–3.35 (m, 1H, H12), 3.72 (t, J = 7.1 Hz, 2H, H17), 4.23–4.39 (m, 2H, H8, H12), 6.46 (t, J = 4.6 Hz, 1H, H5), 7.68–7.73 (m, 2H, H22, H22′), 7.82–7.86 (m, 2H, H21, H21′), 8.28 (d, J = 4.6 Hz, 2H, H4, H6); ^{13}C NMR ($CDCl_3$): δ = 168.53 (C19, C19′), 161.65 (C2), 157.78 (C6, C4), 133.97 (C22, C22′), 132.21 (C20, C20′), 123.24 (C21, C21′), 109.66 (C5), 54.75 (C9), 52.91 (C14), 50.54 (C8), 50.14 (C11), 43.73 (C12), 37.80 (C17), 26.63 (C16), 23.30 (C15), 15.74 (C13); IR (neat): ν = 3456, 2934, 1765, 1704, 1586,1549, 1436, 1367, 1265, 1043, 803, 723, 638, 528 cm^1; MS (70 eV): m/z (%) = 379 (15)[M^+], 364 (20), 271 (100), 259 (80), 245 (7), 202 (12), 191 (83), 176 (5), 160 (38), 134 (16), 122 (23), 108 (7), 96 (10), 80 (12), 70 (11), 56 (14).

4•Hydrochloride. White powder, m.p. = 207–209°C (EtOH). ^{1}H NMR (DMSO): δ = 1.40 (d, J = 5.3, H13), 1.54–1.90 (m, 4H, H15, H16), 2.90–3.88 (m, 9H, H9, H8, H11, H12, H14, H17), 4.48–4.68 (m, 2H, H8, H12), 6.73–6.83 (m, 1H, H5), 7.76–7.91 (m, 4H, H22, H22′, H21, H21′), 8.42–8.52 (m, 2H, H4, H6), 11.60 (s, 1H, H^+); IR (KBr): ν = 3440, 2946, 2627, 1770, 1712, 1625, 1550, 1437, 1404, 1347, 1213, 1058, 1004, 930, 800, 726, 532 cm^{-1}; Anal. Calcd. for $C_{21}H_{25}N_5O_2{\cdot}1.9HCl{\cdot}1.5H_2O$: C, 53.00; H, 6.33; N, 14.72; Cl, 14.15%. Found: C, 52.80; H, 6.25; N, 15.18; Cl, 13.96%.

N-[4-(4-Quinolin-2-yl-piperazin-1-yl)butyl]phthalimide (5). From 1-(2-quinolinyl)piperazine [19] and N-(4-bromobutyl)phthalimide, yield 0.6 g (40%), m.p. = 109–112°C. ^{1}H NMR ($CDCl_3$): δ = 1.46–1.88 (m, 4H, H18, H19), 2.37–2.61 (m, 6H, H13, H15, H17), 3.70–3.81 (m, 6H, H12, H16, H20), 6.90 (d, J = 9.0 Hz, 1H, H3), 7.16–7.27 (m, 1H, H6), 7.47–7.90 (m, 8H, H4, H5, H7, H8, H24, H24′, H25, H25′); ^{13}C NMR ($CDCl_3$): δ = 169.20 (C22, C22′) 158.14 (C2), 148.58 (C9), 138.09 (C4), 134.62 (C25, C25′), 132.80 (C23, C23′), 130.80 (C7), 127.87 (C5), 127.31 (C8), 123.88 (C24, C24′), 123.74 (C6), 123.00 (C10), 110.16 (C3), 58.70 (C17), 53.77 (C13, C15), 45.65 (C12, C16), 38.42 (C20), 27.17 (C19), 24.07 (C18); IR (KBr): ν = 2920, 1770, 1715, 1620, 1510, 1400, 1230 cm^{-1}; MS (70 eV): m/z (%) = 414 (3)[M+], 270 (15), 171 (40), 158 (13), 157 (100), 145 (12), 129 (22), 128 (26), 123 (11), 95 (9).
5•Hydrochloride. White powder, yield 80%, m.p. = 185–188°C. ^{1}H NMR ($CDCl_3$): δ = 1.69–2.02 (m, 4H, H18, H19), 2.90–3.24 (m, 6H, H13, H15, H17), 3.69–3.75 (m, 2H, H20), 4.00–4.28 (m, 4H, H12, H16), 6.98 (d, J = 9 Hz, 1H, H3), 7.24–7.37 (m, 1H, H6), 7.50–7.86 (m, 7H, H5, H7, H8, H24, H24′, H25, H25′), 7.96 (d, J = 9 Hz, 1H, H4); IR (KBr): ν = 3090, 2925, 2855, 1770, 1618, 1603, 1509, 1435, 1400 cm^{-1}; Anal. Calcd. for $C_{25}H_{26}N_4O_2 \cdot HCl \cdot 2H_2O$: C, 61.66; H, 6.41; N, 11.50; Cl, 7.28%. Found: C, 61.92; H, 6.20; N, 11.85; Cl, 7.50%.
N-[4-(2-Methyl-4-quinolin-2-yl-piperazin-1-yl)-butyl]phthalimide (6). From 1-(2-quinolinyl)-3-methylpiperazine and N-(4-bromobutyl)phthalimide, yield 1.1 g (38%), yellow oil. ^{1}H NMR ($CDCl_3$): δ = 1.11 (d, J = 6 Hz, 3H, H17), 1.43–1.89 (m, 4H, H19, H20), 2.25–2.60 (m, 3H, H13, H18), 2.73–3.03 (m, 3H, H12, H15), 3.22–3.38 (m, 1H, H16), 3.72 (t, J = 7 Hz, 2H, H21), 4.10–4.24 (m, 2H, H12, H16), 6.95 (d, J = 9 Hz, 1H, H3), 7.15–7.26 (m, 1H, H6), 7.46–7.58 (m, 2H, H5, H7), 7.66–7.74 (m, 3H, H8, H26, H26′), 7.80–7.87 (m, 3H, H4, H25, H25′); ^{13}C NMR ($CDCl_3$): δ = 168.47 (C23, C23′), 157.31 (C2), 147.97 (C9), 137.36 (C4), 133.91 (C26, C26′), 132.15 (C24, C24′), 129.46 (C7), 127.19 (C5), 126.61 (C8), 123.18 (C25, C25’), 123.00 (C10), 122.22 (C6), 109.42 (C3), 54.81 (C13), 52.88 (C18), 51.72 (C12), 50.62 (C15), 45.15 (C16), 37.75 (C21), 26.59 (C20), 23.30 (C19), 15.96 (C17); IR (neat): ν = 3461, 2942, 2806, 1770, 1714, 1618, 1555, 1508, 1398, 1332, 1273, 1238, 1048, 808, 720, 531 cm^{-1}; MS (70 eV): m/z (%) = 426 (6) [M^+-2], 413 (3), 284 (20), 271 (100), 258 (30), 211 (4), 202 (10), 183 (4), 169 (7), 160 (15), 129 (5), 56 (3); MS (LSIMS(+)): m/z (%) = 451 (7)[M + Na], 429 (100)[M. + H^+].
6 • Hydrochloride. White powder, m.p. 227–228°C (EtOH/ THF). ^{1}H NMR (DMSO): δ = 1.48 (d, J = 5.3, H17), 1.56 – 1.93 (m, 4H, H19, H20), 3.10–4.15 (m, 9H, H13, H15, H18, H12, H16, H21), 4.46–5.02 (m, 2H, H12, H16), 7.36–7.59 (m, 2H, H3, H6), 7.64–7.79 (m, 1H, H7), 7.80–8.45 (m, 7H, H4, H5, H8, H25, H25′, H26, H26′), 11.65 (s, 1H, H^+); IR (KBr): ν = 3427, 2936, 2456, 1770, 1712, 1646, 1605, 1508, 1434, 1399, 1063, 929, 811, 724 cm^{-1}); Anal. Calcd. for $C_{26}H_{28}N_4O_2 \cdot 1.5HCl \cdot 1.1H_2O$: C, 62.00; H, 6.35; N, 11.14; Cl, 10.57%. Found: C, 61.85; H, 6.13; N, 11.31; Cl, 10.27%.
N-{4-[4-(2-(4,6-Dimethoxy)-1,3,5-triazinyl)-)piperazin-1-yl]butyl}phthalimide (7). From 1-[2-(4,6-dimetoxy-1,3,5-triazynyl)]piperazine and N-(4-bromobutyl)phthalimide, yield 0.6 g (25%), white crystals, m.p. 151–153°C (THF). ^{1}H NMR ($CDCl_3$): δ = 1.51–1.62 (m, 2H, H9), 1.67–1.78 (m, 2H, H10), 2.33–2.50 (m, 6H, H5, H7, H8), 3.76 (t, J = 7 Hz, 2H, H11), 3.79–3.90 (m, 4H, H6, H4), 3.94 (s, 6H, H1, H1′), 7.69–7.77 (m, 2H, H15, H15′), 7.80–7.88 (m, 2H, H14, H14′); ^{13}C NMR($CDCl_3$): δ = 173.05 (C2, C2′), 169.17 (C12, C12′), 167.16 (C3), 134.62 (C15, C15′), 132,78 (C13,C13′), 123.86 (C14, C14′), 58.49 (C8), 55.06 (C1, C1′), 53.45 (C5, C7), 44.08 (C4, C6), 38.36 (C11), 27.05 (C10), 24.66 (C9); IR (KBr): ν = 2947, 2816, 1767, 1703, 1595, 1526, 1485, 1370, 1273, 1093, 1040, 811, 723, 531 cm^{-1}; MS (70 eV): m/z (%) = 426 (99)[M^+], 410 (7), 269 (50), 256 (57), 244 (90), 208 (32), 182 (40), 169 (100), 159 (41), 123 (31), 111 (22), 82 (25), 56 (50).
7 • Hydrobromide. White powder, m.p. 197–198°C (MeOH / THF). ^{1}H NMR ($CDCl_3$): δ = 1.73–1.90 (m, 2H, H10), 1.92–2.12 (m, 2H, H9), 2.72–2.98 (m, 2H, H5, H7), 3.09–3.26 (m, 2H, H8), 3.53–4.06 (m, 12H, H1, H1′, H4, H5, H6, H7, H11), 4.82–5.00 (m, 2H, H4, H6), 7.72–7.87 (m, 4H, H14, H14′, H15, H15′), 12.10 (s, 1H, H^+); IR (KBr): ν = 3032, 2952, 2542, 1769, 1712, 1589, 1532, 1470, 1445, 1365, 1271, 1172, 1110, 1049, 943, 813, 723, 529 cm^{-1}; Anal. Calcd. for $C_{21}H_{26}N_6O_4 \cdot HBr$: C, 49.71; H, 5.36; N, 16.56; Br, 15.74%. Found: C, 49.50; H, 5.41; N, 16.51; Br, 15.59%.
The numbering of atoms **4–7** is presented in Scheme 1.
5-HT$_{1A}$ – serotonergic binding assay: Binding was determined using membranes prepared from rat hippocampus (Wistar breed). The procedure used in radioligand binding assay has been already published [21]. Hippocampus was homogenized in 15 ml of 50 mM Tris-TRISMA (pH 7.7 at 20°C) using Ultra-Turrax T25 homogenizer. Homogenates were centrifuged twice for 20 min at 28000 g with resus-

pension of the pellet in the fresh buffer. After the second centrifugation, the pellet was resuspended in homogenization buffer and the suspension incubated 15 min at 37°C. After it had been centrifugated, the final pellet was resuspended in Tris-TRISMA containing 0.125% $CaCl_2$, 0.1% ascorbic acid, and 0.16% pargyline (1 ml buffor/5 mg tissue). Each assay tube contained 100 μL of drug solution, 100 μL of [^{3}H]8-OH-DPAT to achieve a final concentration ~1 nM. The assay samples (in three times repetition) were incubated for 15 min at 37°C, and the incubation was terminated by rapid filtration under vacuum through Whatman GF/B filters, washed three times ice-cold Tris-TRISMA buffer, using Brandel Harvester. Retained radioactivity was measured by introducing dry filters in scintillation liquid and counting in a LS 6000 TA (Beckman) scintillation counter.

Molecular modelling: All computations were carried out on the INDIGO 2 (Silicon Graphics) workstation. The molecular modelling and conformational analysis were carried out using a Sybyl 6.2 package ((1995) Tripos Inc., St.Louis, Missouri, U.S.A) under the Polish Country Wide Licence managed by the ICM computer centre of Warsaw University.

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