Synthesis and 5-HT$_{2A}$ Antagonist Activity of Derivatives of the Novel Heterocycles Indolo[3,2-*d*]pyrrolo[3,2-*g*]azecine and Benzo[*d*]pyrrolo[3,2-*g*]azecine compared to the Benz[*d*]indolo[2,3-*g*]azecine Derivative LE 300

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Summary

An indolo[3,2-*d*]pyrrolo[3,2-*g*]azecine and a benzo[*d*]pyrrolo[3,2-g]azecine analogue of the potent dopamine receptor antagonist LE 300 (7-methyl-6,7,8,9,14,15-hexahydro-5*H*-benz[*d*]indolo[2,3-*g*]azecine) have been prepared in multi-step reactions via C-N bond cleavage of corresponding quaternary *N*-methylquinolizinium iodides. LE 300, the target compounds and two precursor quinolizines have been tested in vitro for antagonist activity at 5-HT$_{2A}$ receptors (rat tail artery) and H$_1$ receptors (guinea-pig ileum), respectively. LE 300 and compound **19** (3,6-dimethyl-4,5,6,7,8,13-hexahydro-3*H*-benzo[*d*]pyrrolo[3,2-*g*]azecine) competitively inhibited 5-HT-induced contractions with similar nanomolar potency (pA$_2$ = 8.32 and 8.01, respectively) but were less active than the reference antagonist ketanserin (pA$_2$ = 9.55). Compound **19** displayed moderate H$_1$-antihistaminic activity in the guinea-pig ileum assay (pA$_2$ = 7.37).

Introduction

There is an ever growing awareness of the role of serotonin (5-hydroxytryptamine, 5-HT) in the regulation of several physiological functions including affective behaviour, memory, and thermoregulation[1]. These diverse biological activities are mediated by a number of serotonin receptor subtypes. Disregulation of the serotonergic system would provoke the pathogenesis of many disease states[2]. Among the family of 5-HT-receptor subtypes, the 5-HT$_{2A}$ receptor has received particular attention because of its thorough characterization in the human body with respect to distribution and function. Extensive stimulation of 5-HT$_{2A}$ receptors would result in psychiatric symptoms, vasoconstriction, and platelet aggregation, respectively. Therefore, selective 5-HT$_{2A}$ receptor antagonists have received particular therapeutic interest for the treatment of mental illness and cardiovascular complications[3]. In some countries, the mixed 5-HT$_{2A}$/α_1-receptor blocker ketanserin and the 5-HT$_{2A}$-selective antagonist sarpogrelate[4] (Chart 1) have been introduced into the therapy of hypertension and ischemia associated with thrombosis, respectively. Furthermore, several mixed D$_2$/5-HT$_{2A}$-receptor antagonists, *e.g.*, risperidone and clozapine (Chart 1) belong to the modern arsenal of atypical neuroleptics which possess a beneficial side-effect profile.

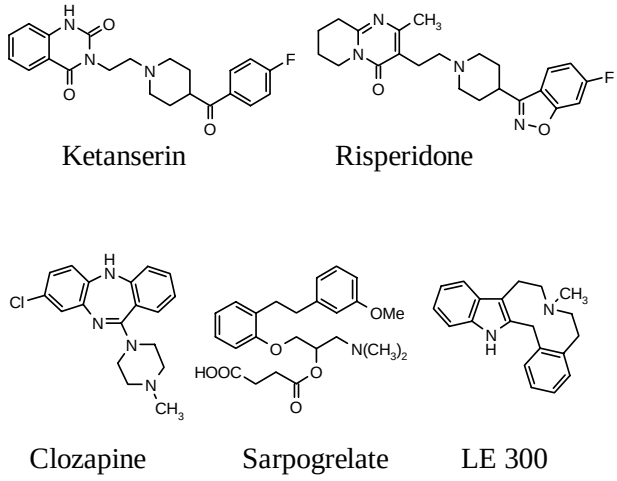

Chart 1. Structures of LE 300[5] and therapeutically relevant 5-HT$_{2A}$-receptor antagonists.

We have recently reported on some novel heterocycles which potently interact with dopamine and 5-HT receptors. These compounds represent hybrid molecules of tryptamine and phenethylamine, linked through the moderately rigid azecine ring system[5]. In radioligand binding studies the most prominent member of this series, compound LE 300 (Chart 1) displays subnanomolar affinity for the rat striatal D$_1$ receptor and a well-balanced binding profile at D$_2$ and 5-HT$_{2A}$ receptors, respectively. Looking for derivatives of LE 300 and related compounds with enhanced selectivity for the 5-HT$_{2A}$ receptor, we decided to replace the benzene or indole ring in LE 300 by introduction of a pyrrole or pyrazole nucleus instead. In a first approach the biological activity of

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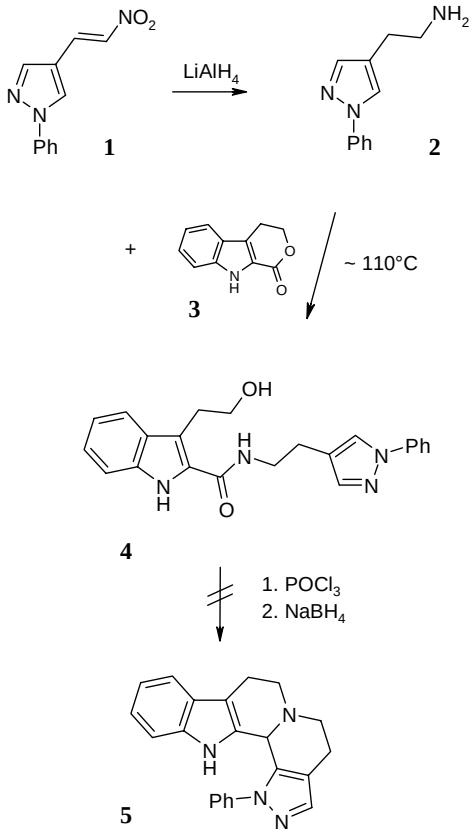
the new compounds **12**, **14**, **17**, **19** and the lead LE 300 was characterized in functional in vitro assays on rat tail artery (5-HT_{2A} receptors) and guinea-pig ileum (H_1 receptors), respectively. These receptors were selected for a preliminary functional characterization because ketanserin, the reference 5-HT_{2A}-receptor antagonist, displays nanomolar affinity for these two sites[6]. The interaction of **12**, **14**, **17**, **19** and related compounds, which are beyond the scope of this report, with dopamine receptor subtypes will be published elsewhere.

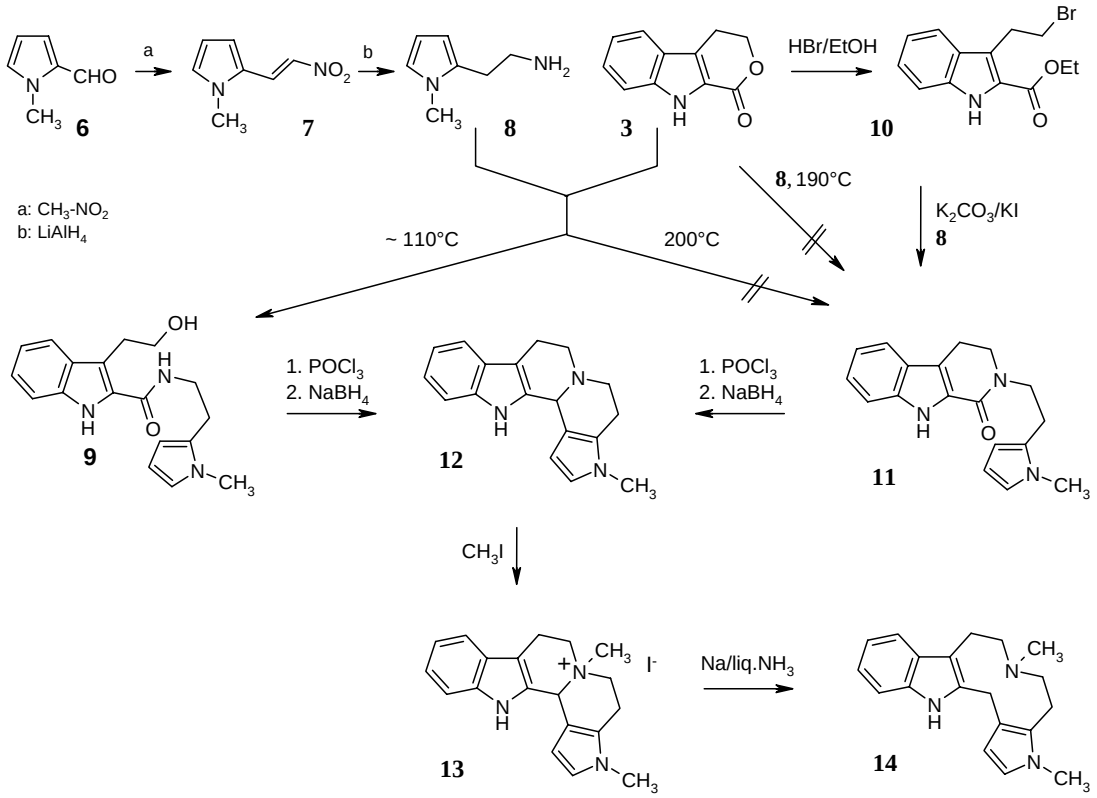
Results and Discussion

Chemistry

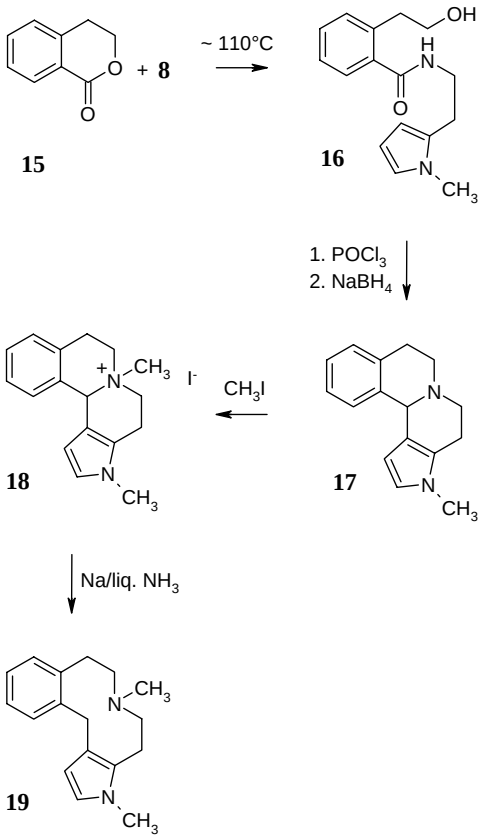
The syntheses of intermediates and target compounds were performed by the reactions illustrated in Schemes 1–3. Compound **4** was prepared in a moderate yield by condensing the pyrazolylethylamine **2** with lactone **3**[7] in toluene (Scheme 1). The required amine **2**, in turn, was prepared by condensing 1-phenyl-1*H*-pyrazole-4-carbaldehyde[8] with nitromethane in the presence of piperidinium acetate as catalyst, followed by the reduction of the resulting nitroethenylpyrazole **1** with lithium aluminium hydride. Trials to convert compound **4** to the polycyclic derivative **5** using phosphorus oxychloride, followed by reduction with sodium borohydride, were unsuccessful. However, synthesis of the isosteric indolopyrroloquinolizine **12** was achieved by two different pathways (Scheme 2). The first pathway involved the condensation of the 1-methylpyrrol-2-ylethylamine **8** with the lactone **3** followed by cyclization of the formed **9** with phosphorus oxychloride according to *Bischler-Napier-*



Scheme 1. Unsuccessful attempt to synthesize compound **5**.



Scheme 2. Synthesis of quinolizine **12** and azecine **14**.



Scheme 3. Synthesis of quinolizine **17** and azecine **19**.

alski reaction conditions[9–11], and *in situ* reduction of the resulting quinolizinium intermediate using sodium borohydride. The second pathway involved the conversion of **3** to the corresponding bromo esters **10** using dry hydrogen bromide and the appropriate dry alcohol at 0 °C[12]. Condensing **10** with the amine **8** in the presence of potassium carbonate and traces of potassium iodide under nitrogen[13] led to the lactam **11**. Conversion of the latter into **12** was performed by a modified procedure of Lehmann et al.[14]. It comprised cyclization of the lactam **11** with phosphorus oxychloride in toluene followed by reduction of the resulting quinolizinium salt with sodium borohydride *in situ*. The purity of the product was much better than that obtained from the first pathway. However, both pathways resulted in poor yields. It is worth mentioning that compound **11** could not be obtained by heating **3** and **8**, as previously described for analogous compounds[14]. Conversion of **12** into the target indolopyrroloazecine **14** was effected by initial quaternization with methyl iodide followed by reductive cleavage of the resulting **13** with sodium in liquid ammonia according to *Birch* reduction conditions[15]. Furthermore, synthesis of the benzene analogues, namely benzopyrroloquinolizine **17** and benzopyrroloazecine **19** was carried out as outlined in Scheme 3. Thus, the amine **8** was allowed to react with isochromanone[16] **15** to yield the intermediate amide **16**. Sequential cyclization of **16** with phosphorus oxychloride, followed by reduction with sodium borohydride, quaternization with methyl iodide and reductive cleavage with sodium in liquid ammonia yielded the desired compound **19**.

In Vitro Pharmacology

Previous radioligand binding experiments have shown that LE 300 (pK_i = 7.79[5]) is about 6- up to 51-fold less potent than the reference 5-HT_{2A}-receptor antagonist ketanserin (pK_i = 9.41[17], 8.46[18], 8.93[19], 8.80[20]). The antagonist potency ratio of 1:17 determined in our functional rat tail artery assay perfectly matches with this range although the absolute affinity tends to be moderately higher (pA_2 = 8.32 *versus* 9.55, see Table 1). Such a discrepancy between binding experiment and functional assay may result from different experimental conditions and is frequently observed. Replacing one of the two aromatic portions of LE 300 by a *N*-methylpyrrole nucleus leads to a slightly less potent congener (**19**) and the micromolar antagonist **14**. Obviously the *N*-methylpyrrole moiety is a bioisosteric replacement for the indole nucleus but not for the benzene ring of LE 300. The interaction between the potent 5-HT_{2A}-receptor antagonists ketanserin, LE 300, and **19** and the reference agonist 5-HT is of competitive nature as indicated by the linear Schild plot regression lines with slope unity (Figure 1, data for ketanserin not shown). Both rigid quinolizines, *viz.* **12** and **17**, are comparably weak antagonists. However, it is striking that **12** is a somewhat more potent than the related **14** while **17** is 3000-fold less active than **19** (Table 1). LE 300 displays approximately the same potency as sarpogrelate[6] does.

Table 1. Affinity of **12**, **14**, **17**, **19**, and reference antagonists for the 5-HT_{2A} receptor in the rat tail artery assay.

Compd.	N[a]	$pA_2 \pm SEM$ $\pm SEM$ (%)	E_{max}(5-HT)[b]	c (μM)[c]
12	6	6.74 ± 0.06	93 ± 2	1
14	6	6.13 ± 0.05	72 ± 3	10
17	4	4.49 ± 0.10	70 ± 4	100
19	20	8.01 ± 0.03	8898[d]	0.01–1
LE 300	22	8.32 ± 0.02	8998[d]	0.01–1
Ketanserin[e]	36	9.55 ± 0.02	8891	0.00047–0.047
Sarpogrelate[e]	16	8.46 ± 0.04	8595	0.01–1

[a] Number of single values. [b] Relative maximum effect of 5-HT in the presence of antagonist. A range is given for compounds tested at different concentrations. [c] Concentration(s) of antagonist. [d] See Figure 1. [e]Data from Pertz and Elz[6]. Schild plot slope was not significantly different from unity.

In contrast to sarpogrelate, ketanserin is endowed with potent H_1-antihistaminic activity which is comparable with the potency of mepyramine (pyrilamine), a competitive reference H_1-receptor antagonist (Table 2). Compounds endowed with prominent 5-HT_{2A}-receptor antagonism (LE 300 and **19**, respectively) also display H_1-receptor-blocking potency in the range of 50 nM whereas **12**, **14**, and **17** possess low affinity for this site.

It is concluded that a certain degree of flexibility between the two aromatic moieties of the molecules presented in this preliminary study is beneficial for interaction with both the 5-HT_{2A} and the histamine H_1 receptor. This trend has also

Table 2. Affinity of **12**, **14**, **17**, **19**, and reference antagonists for the H_1 receptor in the guinea-pig ileum assay.

Compd.	N[a]	$pA_2 \pm SEM$	E_{max}(Hist.)[b] $\pm$ *SEM* (%)	c (μM)[c]
12	4	5.31 ± 0.09	94 ± 2	10
14	5	5.46 ± 0.16	94 ± 2	10
17	8	5.49 ± 0.04	[d]	10 and 100
19[e]	9	7.32 ± 0.04	91102	0.1–10
LE 300[e]	25	7.24 ± 0.02	97103	0.1–10
Ketanserin[f,g]	23	8.85 ± 0.08	97101	0.01–1
Sarpogrelate[e,g]	14	5.47 ± 0.06	90102	10–100
Mepyramine[e,g]	29	9.07 ± 0.04	95101	0.0003–0.1

[a] Number of single values. [b] Relative maximum effect of histamine in the presence of antagonist. A range is given for compounds tested at different concentrations. [c] Concentration(s) of antagonist. [d] 106 ± 5% (10 μM) and 69 ± 2% (100 μM) (N = 4 each). [e] Schild plot slope was not significantly different from unity. [f] Schild plot slope was significantly smaller than unity (0.82 ± 0.03, $P < 0.001$). [g] Data from Pertz and Elz[6].

been observed in a previous study of LE 300 and some analogues vis-à-vis the dopamine receptor subtypes[5]. Supported by the finding that the *N*-methylpyrrolo analogue **19** displays 5-HT_{2A}-receptor affinity comparable with that of LE 300, it is expected that structurally optimized analogues of LE 300 may possess either enhanced 5-HT_{2A}-receptor activity or, with regard to 5-HT_{2A} receptors or dopamine receptor subtypes, a more clear selectivity profile.

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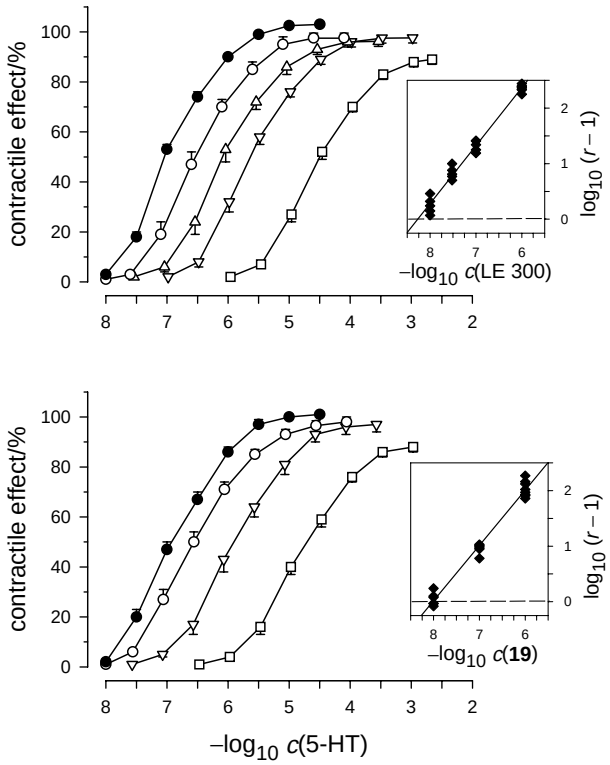

Figure 1. Competitive antagonism of 5-HT-induced contractions in rat tail artery by LE 300 and **19**. Only the mean ± *SEM* of second curves is shown. *Upper panel:* Concentration-effect curve for 5-HT in the absence (●, N = 7) and presence of LE 300 at 10 nM (❍, N = 5), 30 nM (Δ, N = 5), 100 nM (∇, N = 5), and 1000 nM (❑, N = 7). Schild plot for LE 300 (inset): pA_2 = 8.32 ± 0.02 (95% confidence limits 8.27-8.37), slope not significantly different from unity (1.04 ± 0.03, $P > 0.05$, N = 22). *Lower panel:* Concentration-effect curve for 5-HT in the absence (●, N = 6) and presence of **19** at 10 nM (❍, N = 6), 100 nM (∇, N = 6), and 1000 nM (❑, N = 8). Schild plot for **19** (inset): pA_2 = 8.01 ± 0.03 (95% confidence limits 7.95-8.07), slope not significantly different from unity (1.00 ± 0.03, $P > 0.50$, N = 20).

Experimental

Chemistry

Melting points were determined in open glass capillaries on a Gallenkamp melting point apparatus and are uncorrected. The infrared (IR) spectra were determined on a Perkin-Elmer FTIR "Paragor 1000" spectrophotometer using the KBr disc technique unless otherwise indicated. The ^{1}H NMR spectra were recorded, unless otherwise stated, on a Bruker WH 90 (90 MHz) spectrometer using tetramethylsilane as the internal standard with coupling constants J in Hz. Mass spectra were recorded on an A.E.I. 50 mass spectrometer. The elemental analyses were performed at the Microanalytical Unit, Institut of Pharmacy, University of Bonn, Germany, and are within ±0.4% of the theoretical values, unless otherwise indicated. Follow up of the reactions and checking the homogeneity of the compounds were made by TLC on silica gel-coated aluminium sheets (Type 60 F254, Merck) and the spots were detected by exposure to a UV-lamp at 254 nm for few seconds.

*4-(2-Nitroethenyl)-1-phenyl-1H-pyrazole (**1**)*

To a stirred solution of 1-phenyl-1*H*-pyrazole-4-carboxaldehyde[7] (12.1 g, 70 mmol) in absolute ethanol (100 mL) was added nitromethane (4.3 g, 70 mmol), glacial acetic acid (3 mL) and piperidine (3 mL), respectively. The reaction mixture was allowed to stand overnight at room temperature and the precipitated yellow product was filtered, washed with petroleum ether (bp 40–60 °C), and dried. It crystallized from ethanol as bright yellow needles, mp 138–140 °C; yield 14.0 g (93%). IR: ν (cm^{-1}) 1598 (C-NO_2), 1530 (C=C). ^{1}H NMR ([D_6]DMSO): δ (ppm) 9.11 (d, J = 15 Hz, 1H, =CH-NO_2), 8.35 (d, J = 15 Hz, 1H, pyrazole-CH=), 8.06 (s, 1H, C_3-H), 7.88 (s, 1H, C_5-H), 7.77–7.38 (m, 5H, Ar-H). Anal. ($C_{11}H_9N_3O_2$) C, H, N.

*2-(1-Phenyl-1H-pyrazol-4-yl)-ethylamine (**2**)*

Under strictly anhydrous conditions, a solution of **1** (10.8 g, 50 mmol) in freshly distilled THF (50 mL) was added dropwise to a cooled, stirred suspension of lithium aluminium hydride (9.5 g, 250 mmol) in freshly distilled THF (100 mL) over a period of 45 min. Subsequently, the reaction mixture was refluxed with stirring for 48 h. After chilling in an ice-salt bath, the reaction mixture was diluted with ether (100 mL) and excess lithium aluminium hydride was deactivated by addition of methanol (20 mL) and sodium hydroxide solution (w = 20%, 30 mL). The organic layer was separated and the granular precipitate was extracted thoroughly with ether (4 × 150 mL). The combined organic extracts were washed with water, dried over anhydrous $MgSO_4$ and filtered. Evaporation of the ether under reduced pressure afforded a deep yellow oil, yield 7.1 g (76%). It was used directly in the following reaction without further purification. IR (NaCl): ν (cm^{-1}) 3300–2900 (NH and CH). ^{1}H NMR ($CDCl_3$): δ (ppm) 7.77–7.23 (m, 7H, C_3-H, C_5-H, and Ar-H) 2.88 (t, J = 7 Hz, 2H, CH_2-NH_2), 2.73 (t, J = 7 Hz, 2H, pyrazole-CH_2) and 1.35 (s, 2H, NH_2).

*3-(2-Hydroxy-ethyl)-1H-indole-2-carboxylic Acid [2-(1-Phenyl-1H-pyrazol-4-yl)-ethyl]-amide (**4**)*

Compound **3** (3.7 g, 20 mmol) and the amine **2** (5.6 g, 30 mmol) were refluxed in toluene (50 mL) for 48 h. After cooling to room temperature, ethanol (40 mL) was added and the precipitated product was filtered, washed with cold ethanol and dried. It was crystallized from ethanol as colourless microcrystals, mp 232–234 °C; yield 3.4 g (55%). IR: ν (cm^{1}) 3237–2910 (NH and OH), 1600 (C=O, amide I band), a band split at 1560 and 1470 (amide II band, C=N, and aromatics). ^{1}H NMR ($CDCl_3$): δ (ppm) 11.27 (s, 1H, indole-NH), 8.85 (s, 1H, amide-NH), 8.6 (s, 1H, pyrazole-C_5-H), 8.38 (s, 1H, pyrazole-C_3-H), 7.8–7.0 (m, 9H, Ar-H), 5.38 (s, 1H, OH), 3.6 (t, *J* = 5.3 Hz, 2H, C*H*$_2$-OH), 3.3 (t, *J* = 7 Hz, 2H, C*H*$_2$-NH), 3.08 (t, *J* = 5.3 Hz, 2H, indole-CH_2), and 2.8 (t, *J* = 7 Hz, 2H, pyrazole-CH_2). Anal. ($C_{22}H_{22}N_4O_2$) C, H, N.

*1-Methyl-2-(2-nitroethenyl)-1H-pyrrole (**7**)*

A mixture of **6** (10.9 g, 100 mmol), ammonium acetate (4 g, 52 mmol) and nitromethane (25 mL) was heated under reflux for 30 min and then cooled to room temperature. Excess nitromethane was removed under reduced pressure. The remaining solid residue was crystallized from ethanol, affording shiny yellow needles, mp 104–106 °C; yield 9.2 g (61%). IR: ν (cm^{1}) 1614 (C-NO_2), 1560 (C=C). ^{1}H NMR ($CDCl_3$): δ (ppm) 8.1 (d, *J* = 15 Hz, 1H, =CH-NO_2), 7.5 (d, *J* = 15 Hz, 1H, pyrrole-CH=), 6.9 (d, *J* = 3.5 Hz, 1H, C_5-H), 6.77 (d, *J* = 3.5 Hz, 1H, C_3-H), 6.26 (t, *J* = 3.5 Hz, 1H, C_4-H), and 3.77 (s, 3H, CH_3). Anal. ($C_7H_8N_2O_2$) C, H, N.

*2-(1-Methyl-1H-pyrrol-2-yl)-ethylamine (**8**)*

Under strictly anhydrous conditions, a solution of **7** (9.1 g, 60 mmol) in freshly distilled THF (50 mL) was added dropwise to a cooled stirred suspension of lithium aluminium hydride (11.4 g, 300 mmol) in freshly distilled THF (100 mL) over a period of 45 min. Working up as described for compound **2** afforded a deep yellow oil (bp[21] 66–68 °C/1.4 Torr), yield 5.8 g (78%). It was used directly in the next reaction step without further purification. IR (NaCl): ν (cm^{1}) 3290–2890 (NH and CH). ^{1}H NMR ($CDCl_3$): δ (ppm) 6.6 (d, *J* = 3.5 Hz, 1H, C_5-H), 6.0–5.8 (m, 2H, C_3-H, C_4-H), 3.5 (s, 3H, CH_3), 2.9 (t, *J* = 7 Hz, 2H, C*H*$_2$-NH_2), 2.65 (t, *J* = 7 Hz, 2H, pyrrole -CH_2) and 1.77 (s, 2H, NH_2).

*3-(2-Hydroxyethyl)-1H-indole-2-carboxylic Acid [2-(1-Methyl-1H-pyrrol-2-yl)-ethyl]-amide (**9**)*

Pyranoindole **3**[7] (3.7 g, 20 mmol) and the amine **8** were refluxed in toluene (50 mL) for 48 h. Working up as described for compound **4** gave rise to **9**. It was crystallized from ethanol as white microcrystals, mp 232–234 °C; yield 3.4 g (55%). IR: ν (cm^{-1}) 3448 (NH and OH), 1665 (C=O, amide I band), a band split at 1600 and 1530 (amide II band and aromatics). ^{1}H NMR ($CDCl_3$): δ (ppm) 11.23 (s, 1H, indole-NH), 8.77 (t, *J* = 7 Hz, 1H, amide-NH), 7.62–6.92 (m, 4H, Ar-H), 6.6 (d, *J* = 3.5 Hz, 1H, pyrrole-C_5-H), 6.1–5.84 (m, 2H, pyrrole-C_3-H, -C_4-H), 5.3 (s, 1H, OH), 3.62 (t, *J* = 5.3 Hz, 2H, C*H*$_2$-OH), 3.5 (s, 3H, CH_3), 3.38 (t, *J* = 7 Hz, 2H, C*H*$_2$-NH), 3.07 (t, *J* = 5.3 Hz, 2H, indole-CH_2), and 2.8 (t, *J* = 7 Hz, 2H, pyrrole-CH_2). Anal. ($C_{18}H_{21}N_3O_2$) C, H, N.

3-(2-Bromo-ethyl)-1H-indole-2-carboxylic Acid Ethyl Ester **10**

A stirred solution of compound **3** (11.2 g, 60 mmol) in absolute ethanol (150 mL) was saturated with dry hydrogen bromide gas at 0 °C. Cooling and stirring were maintained for 18 h. The precipitated product was filtered, washed several times with cold ethanol and crystallized twice from ethanol. Creamy white shiny needles, mp 153–155 °C; yield 13 g (73%). IR: ν (cm^{1}) 3400 (NH), 1705 (C=O ester), 1275 (C-O-C). ^{1}H NMR ($CDCl_3$): δ (ppm) 9.0 (s, 1H, NH), 7.77–7.08 (m, 4H, Ar-H), 4.42 (q, *J* = 5.3 Hz, 2H, COO-CH_2), 3.77–3.42 (m, 4H, CH_2-CH_2), and 1.46 (t, *J* = 5.3 Hz, 3H, COO-CH_2-C*H*$_3$). Anal. ($C_{13}H_{14}BrNO_2$) C, H, N.

*2-[2-(1-Methyl-1H-pyrrol-2-yl)-ethyl]-2,3,4,9-tetrahydro-pyrido[3,4-b]-indol-1-one (**11**)*

Under nitrogen atmosphere, a mixture of **10** (5.9 g, 20 mmol), the amine **8** (3.7 g, 30 mmol), anhydrous potassium carbonate (4.1 g, 30 mmol) and few crystals of potassium iodide was refluxed in absolute ethanol (120 mL) for 72 h. After removing the solvent to dryness under reduced pressure, the residue was treated with excess water and extracted several times with methylene chloride. The organic extract was washed twice with a saturated solution of sodium chloride, then with water, dried over anhydrous magnesium sulphate and filtered. Rotary evaporation of the methylene chloride afforded a light brown semisolid residue which was crystallized from toluene/petroleum ether (3:1) as creamy white crystals, slightly hygroscopic, mp 196–198 °C; yield 1.6 g (27%). IR: ν (cm^{-1}) 3250 (NH), 1630 (C=O amide I band), a band split at 1600 and 1530 (amide II band and aromatics). ^{1}H NMR ($CDCl_3$): δ (ppm) 9.11 (s, 1H, indole-NH), 7.6–7.07 (m, 4H, Ar-H), 6.5 (d, *J* = 3.5 Hz, 1H, at pyrrole-C_5-H), 6.15–5.65 (m, 2H, pyrrole-C_3-H, -C_4-H), 3.54 and 3.38 (two t, *J* = 7 Hz, 4H, CH_2-N-CH_2), 3.4 (s, 3H, CH_3), 3.07 (t, *J* = 7 Hz, 2H, indole-CH_2), and 2.8 (t, *J* = 7 Hz, 2H, pyrrole -CH_2). Anal. ($C_{18}H_{19}N_3O$ x 1/3 H_2O) C, H, N.

*3-Methyl-4,5,6,7,12,12b-hexahydro-3H-indolo[2,3-a]pyrrolo[2,3-h]quinolizine (**12**)*

Method A: Under anhydrous conditions, the amide **9** (3.5 g, 11 mmol) was heated under reflux with phosphorus oxychloride (10 mL) for 3 h. After cooling to room temperature, excess ether was added, and a dark brown oily residue was separated. The organic layer was decanted, the residue was treated with ether (5 × 50 mL) and excess ether was evaporated under reduced pressure. The remaining residue was dissolved in methanol (100 mL) and then treated with sodium borohydride (8.0 g) portionwise over a period of 30 min while cooling to 0 °C. Stirring was kept for 18 h at room temperature, then methanol was evaporated to dryness under reduced pressure. The remaining light yellow residue was treated with excess water (150 mL) and extracted thoroughly with ether. The ethereal extract was washed twice with a saturated solution of sodium chloride then with water, dried over anhydrous magnesium sulphate, filtered, and evaporated to dryness under reduced pressure. Crystallization of the residue from ethanol/petroleum ether (4:1) afforded deep yellow microcrystals, mp 202–204 °C; yield 1.0 g (33%).

Method B: Under anhydrous conditions, the lactam **11** (2.9 g, 10 mmol) was refluxed with phosphorus oxychloride (10 mL) in toluene (60 mL) for 5 h. The reaction mixture was worked up as described under method A; yield 0.6 g (20%). IR: ν (cm^{1}) 3390 (NH), 1560 (aromatics). ^{1}H NMR ($CDCl_3$): δ (ppm) 7.98 (s, 1H, indole-NH), 7.5–7.0 (m, 4H, Ar-H), 6.57 (d, *J* = 3.5 Hz, 1H, C_2-H), 6.2 (d, *J* = 3.5 Hz, 1H, C_1-H), 5.13 (s, 1H, C_{12b}-H), 3.46 (s, 3H, CH_3), and 3.7–2.6 (m, 8H, overlapped signals of two CH_2-CH_2 moieties). Anal. ($C_{18}H_{19}N_3$ x C_2H_5OH) C, H, N.

*3,5a-Dimethyl-4,5,6,7,12,12b-hexahydro-3H-indolo[2,3-a]pyrrolo[2,3-h] quinolizinium Iodide (**13**)*

Under anhydrous conditions, methyl iodide (7.1 g, 50 mmol) was added to a stirred solution of **12** (2.7 g, 10 mmol) in dry acetone (30 mL). Stirring was maintained at room temperature for 18 h and the precipitated deep yellow solid was separated by centrifugation. It was used directly in the next reaction without further purification.

*3,6-Dimethyl-3,4,5,6,7,8,13,13,14-octahydro-indolo[3,2-d]pyrrolo[3,2-g]-azecine (**14**)*

To a suspension of the quaternary ammonium compound **13** in absolute ethanol (5 mL) in a three-necked flask immersed in a liquefied nitrogen bath ammonia was condensed (100 mL). To the mixture (maintained at –40 °C with methanol-dry ice) sodium (1 g, 4.35 mmol) was added and the resulting dark blue solution was stirred for 45 min. The blue colour was quenched by adding few crystals of ammonium chloride. Ammonia was allowed to evaporate and the remaining residue was treated with water (20 mL) and extracted thoroughly with methylene chloride. The organic layer was washed twice each with 10 mL of sodium hydroxide solution (*w* = 5%) and then with water, dried over anhydrous magnesium sulphate, filtered and evaporated to dryness under reduced pressure. The resulting dark orange oil (300 mg) was purified by column chromatography on alumina (Fluka, type 507C neutral),

using ethyl acetate:methanol (8:2) as the eluent, yielding 130 mg of pure light orange oily product. IR (NaCl): ν (cm^1) a band split at 3400 and 2925 (NH, CH), 1460 (aromatics). ^{1}H NMR (Bruker AC 200 (200 MHz)) ($CDCl_3$): δ (ppm) 7.9 (s, 1H, NH), 7.5–7.0 (m, 4H, Ar-H), 6.5 (d, *J* = 3.5 Hz, 1H, C_2-H), 6.0 (d, *J* = 3.5 Hz, 1H, C_1-H), 4.1 (s, 2H, C_{14}-H), 3.46 (s, 3H, 3-CH_3), 2.75–2.25 (m, 8H, overlapped signals of two CH_2-CH_2 moieties), and 2.2 (s, 3H, 6-CH_3). MS: *m/z* (%) 293 (18.5) ([M^+]), 279 (13.5), 235 (10), 167 (37.1), 149 (100), 107 (52.8), 71 (20), 57 (30). Anal. ($C_{19}H_{23}N_3$) C, H, N.

*2-(2-Hydroxy-ethyl)-N-[2-(1-methyl-1H-pyrrol-2-yl)-ethyl]-benzamide (**16**)*

A mixture of isochromanone **15** (3.7 g, 25 mmol) and the amine **8** (3.7 g, 30 mmol), in toluene (30 mL) was heated under reflux for 48 h. After cooling to room temperature, all the toluene was evaporated under reduced pressure and the remaining dark sticky mass was triturated with ethanol (5 mL) and allowed to stay 2 d in the refrigerator. The formed crystals were filtered, washed with cold ethanol and finally crystallized twice from ethanol as creamy white needles, mp 106–108 °C; yield 2.4 g (35%). IR: ν (cm^{-1}) a band split at 3229, 3100, and 2852 (NH, OH, CH), 1618 (C=O, amide I band), a band split at 1565 and 1442 . ^{1}H NMR ($CDCl_3$): δ (ppm) 7.5–7.0 (m, 4H, Ar-H), 6.84 (s, 1H, amide-NH), 6.5 (d, *J* = 3.5 Hz, 1H, pyrrole-C_5-H), 6.15–5.84 (m, 2H, pyrrole-C_3-H, -C_4-H), 4.27 (s, 1H, OH), 3.8 (t, *J* = 5.3 Hz, 2H, C*H*$_2$-OH), 3.7 (t, *J* = 5.3 Hz, 2H, phenyl-CH_2), 3.55 (s, 3H, CH_3), and 3.07–2.7 (m, 4H, (C*H*$_2$)$_2$-NH). Anal. ($C_{16}H_{20}N_2O_2$) C, H, N.

*3-Methyl-3,4,5,6,7,11b-hexahydro-benzo[a]pyrrolo[2,3-h]quinolizine (**17**)*

Under anhydrous conditions, the amide **16** (3 g, 11 mmol) was heated under reflux with phosphorus oxychloride (10 mL) for 3 h. Work up as described for compound **12** except that the remaining dark brown oil was purified by column chromatography on alumina (Fluka, type 507C neutral) using ethyl acetate:methanol (8:2) as the eluent to give the title compound; yield 0.75 g (29%). IR (NaCl): ν (cm^1) 1620–1580 (aromatics). ^{1}H NMR ($CDCl_3$): δ (ppm) 7.5–7.0 (m, 4H, Ar-H), 6.5 (d, *J* = 3.5 Hz, 1H, C_2-H), 6.0 (d, *J* = 3.5 Hz, 1H, C_1-H), 5.07 (s, 1H, C_{11b}-H), 3.46 (s, 3H, CH_3), and 2.75–2.25 (m, 8H, overlapped signals of two CH_2-CH_2 moieties). MS: *m/z* (%) 238 (100) ([M^+]), 223 (10), 167 (10), 149 (20), 107 (50), 72 (44), 58 (67). The oily compound contains some amounts of solvents and does not give a satisfactory elemental analysis. See next reaction step.

*3,5a-Dimethyl-3,4,5,6,7,11b-hexahydrobenzo[a]pyrrolo[2,3-h]quinolizinium Iodide (**18**)*

Under anhydrous conditions, methyl iodide (1.7 g, 12 mmol) was added to a stirred solution of **17** (1 g, 4 mmol) in dry acetone (30 mL). Stirring was maintained at room temperature for 18 h and the precipitated light yellow product was filtered, washed with cold acetone and dried. It was crystallized from ethanol as light yellow microcrystals, mp 237–239 °C; yield 0.55 g (35%). IR: ν (cm^1) a band split at 3436 and 2917 (N-C, C-H). ^{1}H NMR ($CDCl_3$): δ (ppm) 7.42–7.15 (m, 4H, Ar-H), 6.53 (d, *J* = 3.5 Hz, 1H, C_2-H), 5.75 (d, *J* = 3.5 Hz, 1H, C_1-H), 5.65 (s, 1H, C_{11b}-H), 3.6 (s, 3H, 3-CH_3), 3.5 (s, 3H, 5a-CH_3), and 3.38–2.6 (m, 8H, overlapped signals of two CH_2-CH_2 moieties). Anal. ($C_{17}H_{21}IN_2 \times H_2O$) C, H, N.

*3,6-Dimethyl-4,5,6,7,8,13-hexahydro-3H-benzo[d]pyrrolo[3,2-g]azecine (**19**)*

Prepared from the quaternary ammonium compound **18** (0.6 g, 1.0 mmol) following the same procedure described for compound **14**. The yield of the yellow oily product was 0.2 g (50%). IR (NaCl): ν (cm^1) a band split at 3350 and 2900 (N-C, C-H), 1460 (aromatics). ^{1}H NMR (Bruker AC 200 (200 MHz)) ($CDCl_3$): δ (ppm) 7.3–7.0 (m, 4H, Ar-H), 6.5 (d, *J* = 3.5 Hz, 1H, C_2-H), 6.0 (d, *J* = 3.5 Hz, 1H, C_1-H), 3.9 (s, 2H, CH_2), 3.6 (s, 3H, 3-CH_3), 3.0–2.6 (m, 8H, overlapped signals of two CH_2-CH_2 moieties), and 2.2 (s, 3H, 6-CH_3). MS: *m/z* (%) 254 (92.5) ([M^+]), 239 (10), 210 (57), 196 (78.5), 182 (100), 167 (28), 149 (28), 115 (20), 107 (54.3), 94 (30), 72 (45), 58 (70). Anal. ($C_{17}H_{22}N_2$) C, H, N.

In Vitro Pharmacology

Rat Tail Artery (Ring Segments): 5-HT$_{2A}$ Receptors

In brief, cylindrical segments of 3–4 mm length were mounted isometrically (initial tension 5 mN) by means of two stainless L-shaped steel hooks (diameter 0.15 mm) in a modified Krebs-Henseleit solution (37 °C) of composition (mM): NaCl 118.1, KCl 4.7, $CaCl_2$ 2.5, $MgSO_4$ 1.2, KH_2PO_4 1.2, $NaHCO_3$ 25.0, and D-glucose 10.0. The solution was aerated with 95% O_2 / 5% CO_2 and contained prazosin (30 nM) to block α_1 adrenoceptors, and cocaine (6 μM) to block neuronal uptake of amines. During an equilibration period of 120 min the preparations were primed once (after 60 min) with 5-HT (1 and 10 μM) to monitor tissue viability. Two cumulative concentration-effect curves for 5-HT (0.01–30 μM for the first curve) were determined in the absence and presence of potential antagonists which were usually incubated for 30 min or 120 min (LE 300, **19**, ketanserin at 0.47 and 1 nM, respectively). Control experiments in the absence of antagonist revealed that for the 30-min protocol, two successive concentration-effect curves for 5-HT were superimposable (data not shown). When the 120-min protocol was applied, a leftward shift of the control curves of approximately 0.1–0.3 logarithmic units was usually observed. The daily mean sensitization was used to correct the dextral shift measured for treated organs.

Guinea-Pig Ileum (Whole Segments): H$_1$ Receptors

Whole segments of ileum, 1.5–2.0 cm in length, were mounted isotonically (preload 0.5 g) in Tyrode solution (37 °C) of composition (mM): NaCl 136.9, KCl 2.7, $CaCl_2$ 1.8, $MgCl_2$ 1.0, NaH_2PO_4 0.4, $NaHCO_3$ 11.9, and D-glucose 5.1. The solution was aerated with 95% O_2 / 5% CO_2 and contained atropine (0.1 μM) to block ileal M_3 receptors. During an equilibration period of 80 min the organs were primed three times with histamine (1 and 10 μM every time). Up to four cumulative concentration-effect curves for histamine (0.01–30 μM for the first curve) were determined in the absence and presence of potential antagonists which were incubated for 10 min. Control experiments in the absence of antagonist revealed that four successive concentration-effect curves for histamine were superimposable (data not shown).

Pharmacological Parameters

Results are expressed as mean ± standard error (*SEM* or *SE*) unless otherwise indicated. Antagonist affinity was calculated as apparent pA_2 value according to equation (1) when only one or two antagonist concentrations were used[22]. ([*c*] = mol/L, *r* is the ratio of agonist concentrations in the presence and absence of antagonist, that elicit 50% of the respective maximum effect).

$$pA_2 = -\log_{10} c(\text{antagonist}) + \log_{10} (r - 1) \quad (1)$$

Full pA_2 values were calculated according to the method of Schild[23] when a set of different antagonist concentrations over at least 1.5 logarithmic units was studied. Single organ preparations were from at least two (H_1) or three (5-HT_{2A}) animals.

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