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Original article

Variations of acidic functions at position 2 and substituents at positions 4, 5 and 6 of the indole moiety and their effect on NMDA-glycine site affinity

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Abstract

The synthetic procedures to obtain indole derivatives with different acidic functions at position 2 of the indole are reported. The synthesised and tested derivatives comprise 5-tetrazolyl, 1,3,4-oxadiazol-5-yl-2-one, and indole-2-carboxylic acid amides with 5-aminotetrazole, methanesulphonamide and trifluoromethanesulphonamide moieties. The binding affinity was evaluated using [^{3}H]MDL 105,519 and pig cortical brain membranes. In general, compounds with acidic functions different from a carboxylic acid moiety are less potent than indole-2-carboxylic acid derivatives. Also, the 4,6-dichloro substitution pattern was compared to 5-*tert*-butyl derivatives and compounds not substituted in the benzene moiety of the indole, indicating that the affinity increases from 5-*tert*-butyl over unsubstituted to 4,6-dichloro substituted derivatives.

Keywords: glycine site; NMDA receptor; indole derivatives; variation of the acidic function; structure–activity relationships

1. Introduction

Excitatory amino acid receptors can be divided into two major classes: ionotropic and metabotropic receptors. Metabotropic receptors (mGluRs), which are members of the G-protein coupled receptor family, can be divided into the following groups: group I, which stimulates phospholipase C (mGluR1, mGluR5); group II (mGluR2, mGluR3) and group III (mGluR4, mGluR6-8), which are both coupled to inhibition of adenylate cyclase (for review see [1]). Ionotropic receptors (iGlu receptors) comprise kainate (Kain), (*S*)-2-amino-3-(3-hydroxy-5-methylisoxazol-4-yl)propionic acid (AMPA), and *N*-methyl-D-aspartic acid (NMDA) receptors. These ligand gated ion channels (LGICs) are permeable to Na^+, K^+, and Ca^{2+}-ions. Each class of receptors is named after its selective agonist (Fig. 1). In recent years these glutamate receptors have attracted major scientific interest [2], because they are involved in a number of pathological conditions and diseases: Alzheimer's [3–5], Parkinson's [6,7], and Huntington's disease [8], stroke, trauma [9], dementia [10–12], depressions [13], chronic pain [14], and epilepsy [15,16].

NMDA receptor activation by its agonist glutamate and co-agonist glycine leads to an influx of calcium and sodium ions, whereas potassium ions leave the cell. Other interesting features of the NMDA receptor are its voltage-dependent block by magnesium ions, and its multiple binding and recognition sites (polyamine, PCP, phosphorylation, zinc, pH, redox-site) [17–19]. Because antagonists at the glycine site of the NMDA receptor do not cause the side effects such as vacuolisation and psychotomimetic effects associated with competitive NMDA antagonists and channel blockers, they are of interest for the treatment of diseases, in which excitatory mechanisms are involved [20–22].

The structures developed for glycine antagonism include quinoxalinediones, kynurenic acids and indole-2-carboxylic acids (Fig. 2) [2,23–25].

Besides a high affinity for the glycine site of the NMDA receptor the compounds should have the

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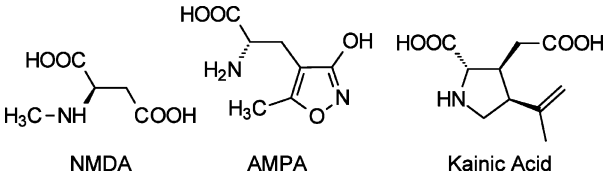


Fig. 1. Selective ionotropic glutamate receptor ligands.

pharmacokinetic and physicochemical properties (brain permeation, solubility) necessary for their development as drug candidates. Poor solubility and brain permeation were a major drawback with early glycine antagonists. One structural component that might contribute to this is the carboxylic acid function. To evaluate if acidic functions other than a carboxylic acid at position 2 of the indole are able to show comparable affinities, different 4,6-dichloro-indoles bearing a variety of acidic functions at position 2 were synthesised. Fig. 3 shows the structures of the reference and target compounds.

In addition, indole-2-carboxylic acid derivatives were synthesised to evaluate the influence of a +I and bulky substituent (5-*tert*-butyl) compared to the well-established −I, +M chloro substituents at positions 4 and 6. Therefore 5-*tert*-butyl-derivatives of indole carboxylic acid and of the hydantoins previously described by our group [25] were evaluated (Fig. 3).

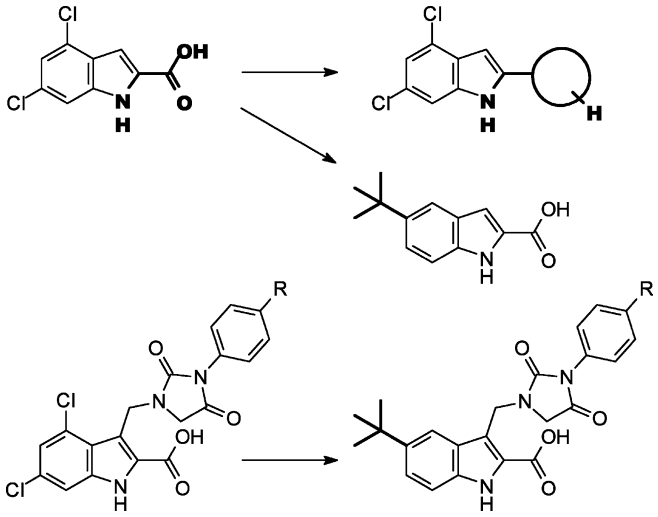


Fig. 3. Structures of reference and target compounds (glycine backbone and modifications are bold).

2. Chemistry

Compound **2** was converted to the acid chloride with phosphor pentachloride and in the same mixture subsequently to the amide **3** by pouring on ammonia solution (Scheme 1). Dehydration of the 4,6-dichloro-indole-2-carboxamide **3** with phosphor oxychloride yielded the cyanide **4** which was cyclised with sodium azide to the tetrazole compound **5**.

Starting from ethyl 4,6-dichloroindole-2-carboxylate **1** the carboxyhydrazide **6** was readily prepared by means of an excess of hydrazine hydrate on the ester. Ring closure of the hydrazide **6** to the 3*H*-1,3,4-oxadiazol-2-one derivative **7** was achieved by reaction with 1,1′-carbonyl diimidazole (CDI) via an intermediate imidazolylcarbonyl hydrazide.

To obtain the 4,6-dichloroindol-2-yl-carboxamides **8–10**, the acid **2** was activated with CDI, and the intermediate imidazolide treated with different amine-compounds (5-aminotetrazole, methanesulphonamide, trifluoromethanesulphonamide).

5-*tert*-Butyl-indole-2-carboxylic acid was synthesised starting from 4-*tert*-butyl-aniline following the same procedures as for the 4,6-dichloro analogue (Scheme 2). First the corresponding hydrazine hydrochloride **11** was produced by diazotisation and subsequent reduction with stannous chloride. Condensation with ethyl pyruvate in ethanol yielded both the *Z*- and *E*-isomer of hydrazone **12**, which were separated by column chromatography. Contrary to the 4,6-dichloro derivative the cyclisation of the hydrazone (*E*/*Z*-mixture) to the indole **13** was not conducted with polyphosphoric acid (PPA)—because this produced many side products—but by saturating the ethanolic solution of the hydrazone with gaseous HCl. Other acidic catalysts such as H_2SO_4 and methanesulphonic acid were shown to be inferior to the HCl method. Basic hydrolysis of the 5-*tert*-butyl-indole-2-carboxylic acid ethyl ester with LiOH in THF–water was used to obtain the test compound **14**.

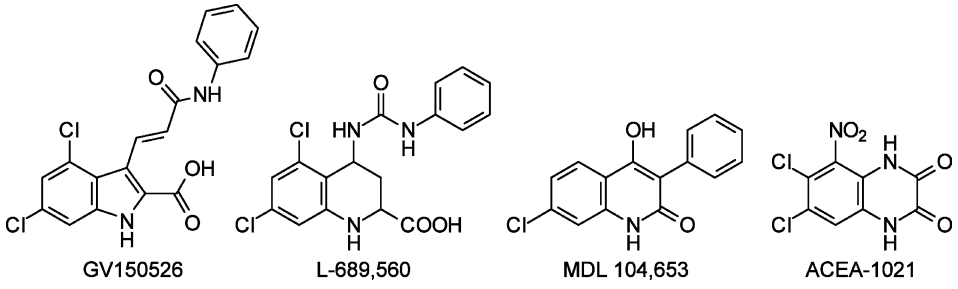
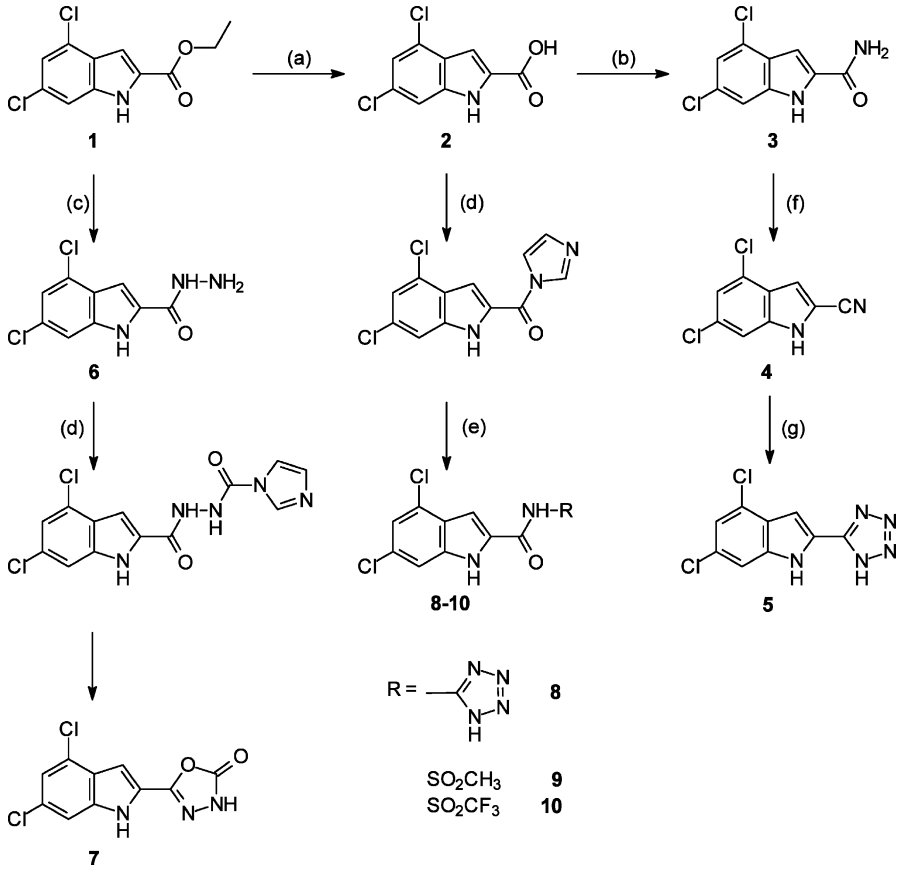
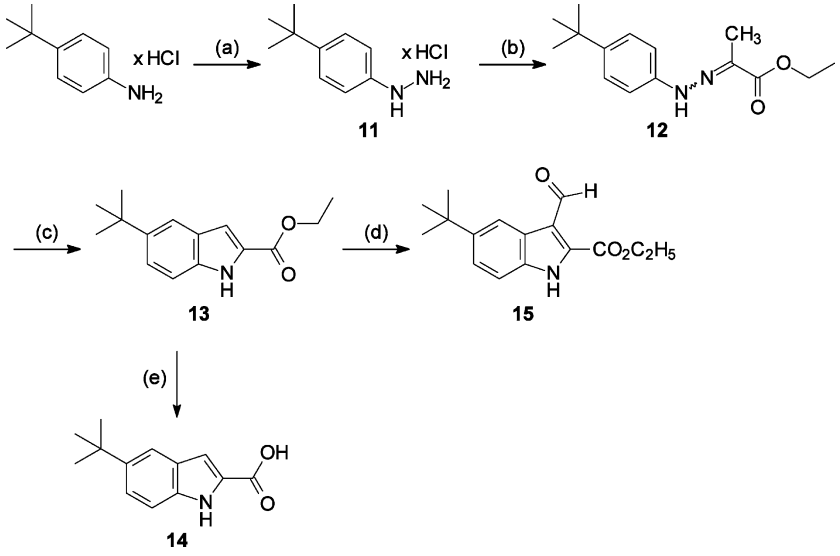


Fig. 2. Structures of NMDA-glycine antagonists.

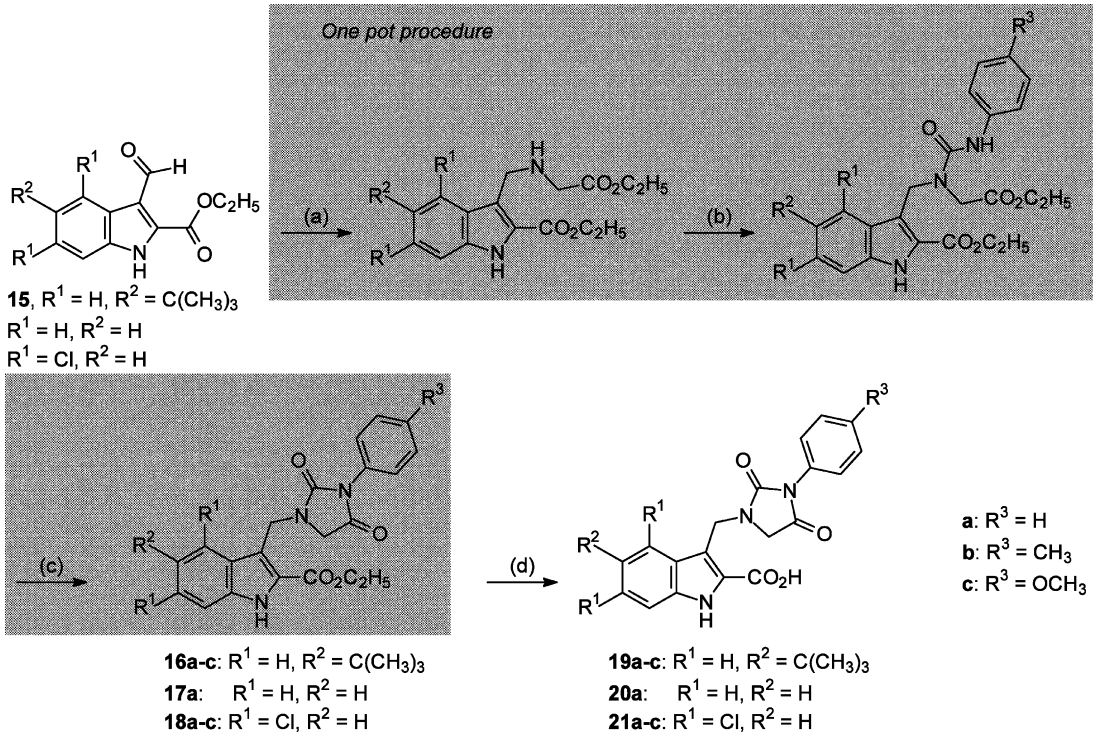


Scheme 1. (a) LiOH; (b) 1. PCl_5 2. NH_3; (c) H_2NNH_2; (d) CDI; (e) H_2NR; (f) $POCl_3$; (g) DPPA.

In order to obtain indole derivatives with higher affinity, compounds with an affinity-enhancing substituent at position 3 of the indole were synthesised as follows. Vilsmeier formylation of **13** produced the 3-indolyl carbaldehyde **15** which was converted to 5-*tert*-butyl-3-[(2,4-dioxo-3-phenyl-1-imidazolidinyl)methyl]-indole-2-carboxylic acid ethyl ester **16a** in a one-pot procedure with ethyl glycinate hydrochloride and phenylisocyanate as described for the corresponding 4,6-dichloro-derivatives in a previous paper [25] (Scheme 3). Reductive amination of the aldehyde **15** with ethyl glycinate and sodium triacetoxyborohydride first leads



Scheme 2. (a) 1. $NaNO_2$ 2. $SnCl_2$; (b) ethyl pyruvate; (c) gaseous HCl; (d) $POCl_3$, *N*-methylformanilide; (e) LiOH.



Scheme 3. (a) Ethyl glycinate–$Na(AcO)_3BH$ (b) R^3-Ph-NCO; (c) Et_3N; reflux; (d) LiOH.

to a secondary amine. This intermediate is transformed to an N-disubstituted-N′-monosubstituted urea by addition of phenylisocyanate and subsequently to the hydantoin derivative **16a** after base-catalysed ring closure. Hydrolysis of the ethyl ester **16a** with LiOH in THF–water yields the test compound **19a**. Starting from commercially available indole-2-carboxylic acid the corresponding 3-formyl ethyl ester derivative was prepared according to the literature [26,27]. This aldehyde was also subjected to hydantoin formation to yield compound **20a**. The same procedure was used to synthesise the hydantoin derivatives **19b–c** and **21a–c**.

3. Results and discussion

Affinity for the glycine site of the NMDA receptor was evaluated using pig brain membranes in a binding assay with [^{3}H]MDL 105,519 (Fig. 4), a ligand of the glycine site of the NMDA receptor with high affinity

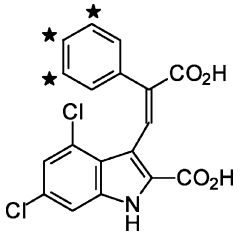


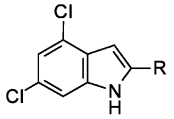
Fig. 4. [^{3}H]MDL 105,519; asterisks indicate tritium atoms.

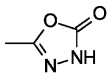
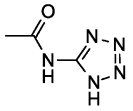
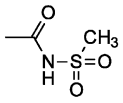
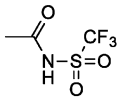
and specificity [41] (see Section 5). The results are reported in Tables 1 and 2. EPS-maps of the deprotonated compounds were calculated with Spartan'02 (semi-empirical, AM1) and are shown in Fig. 5.

The derivatives with the variation of the acidic function show a decrease of affinity in the following order: carboxylic acid (**2**), carbonyl trifluoromethanesulphonamide (**10**), carbonyl-5-aminotetrazolyl amide (**8**), tetrazole (**5**), carbonyl methanesulphonamide (**9**) and 1,3,4-oxadiazol-2-one (**7**). Acidity as indicated by pK_a values increases from 1,3,4-oxadiazol-2-one < tetrazole ≅ amidotetrazole < carboxylic acid < methane sulphonamide < trifluoromethane sulphonamide [28–33]. Consequently the acidity is however not the only influencing factor for receptor affinity. Although carboxylic acid (**2**) and tetrazole (**5**) only show marginal differences in pK_a values, the carboxylic acid (**2**) is 20 times more active. In both cases the negative charge of the deprotonated compound is delocalised. However, in the carboxylate the negative charge is shared equivalently by the two oxygen atoms, whereas the tetrazole-anion—due to its aromatic character—is able to delocalise the charge over all five ring atoms. The advantage of the carboxylate might be its ability for decided bidirectional interactions. This idea is supported by the finding, that the affinity is increased more than 3-fold, when the tetrazole (**5**) is converted to the amidotetrazole (**8**).

The 1,3,4-oxadiazol-2-one (**7**) has by far the lowest acidity and also affinity in this series. The high loss of

Table 1
Binding affinity of the 4,6-dichloroindole-2-carboxylic acid derivatives (4,6-dichlorindole-2-carboxylic acid (**2**) is given as reference)

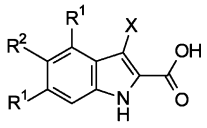


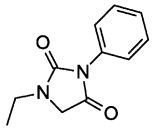
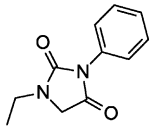
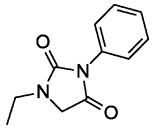
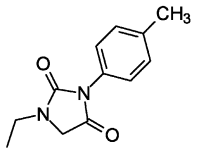
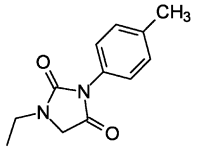
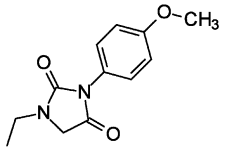
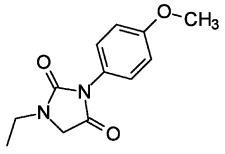
no.	R	[^{3}H]MDL 105,519 $K_i \pm$ SEM [μM]
2	—COOH	2.9 ± 0.8
5		67 ± 5
7		> 200
8		18 ± 10
9		73.4 ± 1.0
10		16.6 ± 1.6

affinity might be a result of the decreased acidity and also of the increased distance between the negative charge of the acidic function and the indole-NH compared to the carboxylic acid derivative. Also the results on sulphonamides **9** and **10** clearly indicate that one contributing factor for affinity is acid strength, since the more acidic trifluoromethanesulphonamide (**10**) is four times more active than the methanesulphonamide derivative (**9**). The indolyl-2-carbonyl compounds **2**, **8**, **9**, and **10** all have one carbonyl-group at the same distance from the indole-N1. The difference in affinity of this series might be attributed to varying acidity (compounds **9** and **10**), but also to the different characters of the delocalised charge (compounds **8** and **9**), as well as to the different distances between the indole-NH and the acidic part.

The 5-*tert*-butyl derivatives **14**, **19a**, **19b**, **19c** (Table 2) are in general accompanied by a strong loss of affinity compared to the corresponding derivatives without this substituent (**2**, **21a**, **20a**, **21b**, **21c**). 5-*tert*-Butylindole-2-carboxylic acid **14** shows a 30-fold reduced affinity compared to 4,6-dichloroindole-2-carboxylic acid **2**. The affinity increases if the imidazolidin-2,4-dione moiety is

Table 2
Binding affinity of the 5-*tert*-butylindole-2-carboxylic acid derivatives (4,6-dichloro-3-[(2,4-dioxo-3-phenyl-1-imidazolidinyl)methyl]-1*H*-indole-2-carboxylic acid (**21a**) [25] is given as reference)



no.	R^1	R^2	X	[^{3}H]MDL 105,519 $K_i \pm$ SEM [μM]
14	H		H	92 ± 5
19a	H			15 ± 3
21a [25]	Cl	H		0.022 ± 0.007
20a	H	H		2.87 ± 0.27
19b	H			62 ± 2
21b [25]	Cl	H		0.036 ± 0.003
19c	H			78 ± 25
21c [25]	Cl	H		0.10 ± 0.06

present at position 3 (**19a**), but the compound is less potent than the 4,6-dichloro reference **21a**. Compounds **19a–c**, **20a**, **21a–c** show that the affinity increases in the

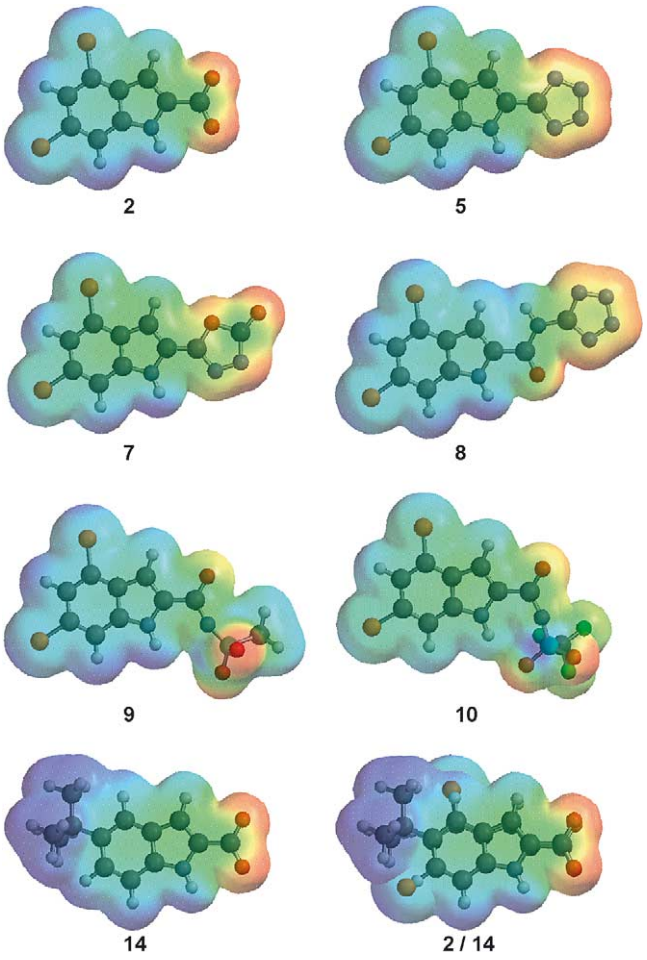


Fig. 5. EPS maps of the test compounds (deprotonated form) presented in energy-minimized conformations calculated with Spartan '02 Windows programme, Wavefunction, Inc. 18401 Von Karman Avenue, Suite 370, Irvine, CA 92612 USA; this surface shows the relative electrostatic potential at the van der Waals surface of the molecule. Regions of negative electrostatic potential are shown in red and positive potentials in purple/blue; **2/14** shows a superposition of the compounds **2** and **14**.

following order: 5-*tert*-butyl < unsubstituted < 4,6-dichloro. The decrease in affinity of the 5-*tert*-butyl-compound (**19a**) is however caused by steric disadvantages, which is in line with known pharmacophore models for the NMDA glycine binding site, that predict size-limited hydrophobic regions at the 4-, 5-, and 6-position [34,35].

4. Conclusion

Varying the acid function of 4,6-dichloroindole carboxylic acid produces less potent compounds. The same is true for a 5-*tert*-butyl substitution with a significant decrease of affinity. However, an additional hydantoin substituent at position 3 increases affinity 7-fold (compounds **14** and **19a**) and up to 100-fold (compounds **2** and **21a**), which is in line with our results previously published [25].

5. Experimental

5.1. Chemistry

Infrared spectra were recorded on a Perkin–Elmer 1310 infrared spectrophotometer. ^{1}H (300 MHz, digital resolution 0.3768 Hz) and ^{13}C (75 MHz, digital resolution 1.1299 Hz) NMR were recorded on a Bruker AC-300 apparatus: the data are reported as follows: chemical shift in parts per million (ppm, δ units) from Me_4Si as external standard, multiplicity, and spin–spin coupling J (Hz). EI-mass spectra were recorded on a Varian MAT 311A (70 eV). Elemental analyses were performed on an Elemental Analyzer Carlo Erba Strumentazione Mod. 1106. Combustion analyses agreed with the calculated data within $\pm 0.4\%$. Melting points were determined on a Büchi apparatus after Dr. Tottoli and are uncorrected. Column chromatography was performed with Merck silica gel 60 (0.063–0.200 mm). Dichloromethane was dried and distilled over CaH_2, whereas THF was used after distillation over K/benzophenone. The progress of the reactions was monitored by thin-layer chromatography (TLC) performed with Merck silica gel 60 F-245 plates. Where necessary reactions were carried out in a nitrogen atmosphere.

4,6-Dichloro-indole-2-carboxylic acid ethyl ester (**1**) and the corresponding acid 4,6-dichloro-indole-2-carboxylic acid (**2**) were synthesised according to procedures previously described [36,37].

5.1.1. 4,6-Dichloro-1H-indole-2-carboxylic acid amide (3) [38]

A mixture of 4,6-dichloro-indole-2-carboxylic acid (**2**) (1 g, 4.35 mmol) and phosphorpentachloride (1.85 g, 8.88 mmol) in dry chloroform was heated under reflux for 30 min. After cooling to ambient temperature, this mixture was added dropwise to concentrated, ice cold, aqueous ammonia solution (15 mL). The resulting light-yellow suspension was filtered to yield the product as light-yellow crystals (0.97 g, 4.23 mmol, 97%), R_f (petroleum ether–ethyl acetate = 2/1): 0.25; melting point (m.p.): 225 °C; IR (KBr) (cm^{-1}): 3400, 3180, 1650, 1550; ^{1}H-NMR (300 MHz) ($CDCl_3$–DMSO-d_6) δ: 6.14 and 7.51 (s each, 1H each, NH_2), 6.76 (d, $J = 1.4$ Hz, 1H, Ind-H7), 6.91 (d, $J = 1.2$ Hz, 1H, Ind-H3), 7.11 (m, 1H, Ind-H5), 11.27 (s, 1H, Ind-NH); MS m/z: 228 [M^+].

5.1.2. 4,6-Dichloro-1H-indole-2-carbonitrile (4) [39]

Compound **3** (0.97 g, 4.23 mmol) and phosphorus oxychloride (10 mL) were heated under reflux for 5 min.

After cooling to ambient temperature, this mixture was poured on a mixture of crushed ice (80 g) and aqueous ammonia solution (20 mL). Afterwards the pH was adjusted to alkaline reaction by adding further aqueous ammonia solution. The formed solid was filtered, suspended in water and extracted with ethyl acetate to remove inorganic impurities. After drying over $MgSO_4$, the solvent was evaporated under vacuum to obtain beige crystals (0.72 g, 3.43 mmol, 81%), R_f (petroleum ether–ethyl acetate = 2/1): 0.68; m.p.: 212 °C; IR (KBr) (cm^{-1}): 3180, 2180, 1700, 1570; ^{1}H-NMR (300 MHz) (DMSO-d_6) δ: 7.34 (d, $J = 1.9$ Hz, 1H, Ind-H7), 7.43 (m, 1H, Ind-H3), 7.54 (m, 1H, Ind-H5), 12.92 (s, 1H, NH); ^{13}C-NMR (75 MHz) (DMSO-d_6) δ: 108.3 (Cq), 111.56 (CH), 111.60 (CH), 113.9 (Cq), 121.1 (CH), 124.0 (Cq), 126.8 (Cq), 130.5 (Cq), 137.6 (Cq); MS *m/z*: 210 [M^+].

5.1.3. 4,6-Dichloro-2-(1H-tetrazol-5-yl)-1H-indole (5)

A mixture of compound **4** (0.3 g, 1.42 mmol), sodium azide (0.11 mg, 1.69 mmol) and acetic acid (0.1 mL, 1.78 mmol) was refluxed in *n*-butanol until the starting material disappeared from TLC. After hydrolysation the precipitated product (light yellow crystals) is collected by filtration (0.25 mg, 0.97 mmol, 68%), R_f (petroleum ether–ethyl acetate–acetic acid = 2/1/0.03): 0.24; m.p.: 301 °C; IR (KBr) (cm^{-1}): 3540, 3400, 3320, 1580, 1540; ^{1}H-NMR (300 MHz) (DMSO-d_6) δ: 6.73 (m, 1H, Ind-H3), 7.09 (d, $J = 1.67$ Hz, 1H, Ind-H7), 7.46 (dd, $J = 1.67$ Hz, $J = 0.95$ Hz, 1H, Ind-H5), 12.24 (s, 1H, NH); ^{13}C-NMR (75 MHz) (DMSO-d_6) δ: 95.9 (CH), 110.7 (CH), 118.60 (CH), 124.5 (Cq), 125.1 (Cq), 126.3 (Cq), 134.2 (Cq), 137.4 (Cq), 155.2 (Cq); MS *m/z*: 253 [M^+]; Anal. $C_9H_5Cl_2N_2 \times 3\ H_2O$ (C, H, N).

5.1.4. 4,6-Dichloro-1H-indole-2-carboxylic acid hydrazide (6) [40]

A solution of compound **1** (0.67 g, 2.61 mmol) in methanol (60 mL) was treated with hydrazine hydrate (0.63 mL, 13 mmol). After heating under reflux for 24 h and cooling to ambient temperature the precipitated product was collected by filtration and washed with methanol. White crystals were obtained (0.52 g, 2.14 mmol, 82%), M.p.: > 280 °C; IR (KBr) (cm^{-1}): 3220, 1590, 1520; ^{1}H-NMR (300 MHz) (DMSO-d_6) δ: 4.55 (s, 2H, NH_2), 7.20 (m, 2H, Ind-H3,7), 7.40 (m, 1H, Ind-H5), 9.97 (s, 1H, CONH), 12.12 (s, 1H, Ind-NH); MS *m/z*: 243 [M^+].

5.1.5. 5-(4,6-Dichloro-1H-indol-2-yl)-3H-[1,3,4]-oxadiazol-2-one (7)

Compound **6** was suspended in a mixture of dry THF (18 mL) and dry DMF (1.8 mL). After adding 1,1′-carbonyl-diimidazole (CDI) (0.46 g, 2.75 mmol) and triethylamine (0.5 L, 3.66 mmol) the mixture became clear and slightly yellow. The solution was heated under reflux for 20 h. Then it was cooled in an icebath and the product precipitated by acidification with 1N HCl. The obtained crystals were separated by filtration and washed with methanol (0.4 g, 1.5 mmol, 82%), R_f (petroleum ether–ethyl acetate = 1/1): 0.46; m.p.: 287 °C; IR (KBr) (cm^{-1}): 3400–2600, 3240, 1750, 1620; ^{1}H-NMR (300 MHz) (DMSO-d_6) δ: 6.95 (d, $J = 1.51$ Hz, 1H, Ind-H3), 7.24 (d, $J = 1.51$ Hz, 1H, Ind-H7), 7.39 (m, 1H, Ind-H5), 12.55 (s, 1H, NH), 12.76 (s, 1H, NH); MS *m/z*: 269 [M^+]; Anal. $C_{10}H_5Cl_2N_3O_2 \times 1/2\ H_2O$ (C, H, N).

5.1.6. 4,6-Dichloro-1H-indole-2-carboxylic acid (1H-tetrazol-5-yl)-amide (8)

To a solution of compound **2** (0.59 g, 2.56 mmol) in dry THF (40 mL) CDI (0.46 g, 2.81 mmol) was added. After heating under reflux for 1 h the mixture was cooled to room temperature and 5-aminotetrazole (0.26 g, 2.56 mmol) was added. The mixture was refluxed for 3 h. Then it was cooled in an icebath and 1N HCl (170 mL) was added to obtain a crystalline solid, which was purified by crystallisation in methanol/chloroform (0.39 g, 1.32 mmol, 51%), m.p.: > 290 °C; IR (KBr) (cm^{-1}): 3280, 3080, 1630, 1580; ^{1}H-NMR (300 MHz) (DMSO-d_6) δ: 7.29 (d, $J = 1.88$ Hz, 1H, Ind-H7), 7.47 (m, 1H, Ind-H3), 7.81 (d, $J = 1.51$ Hz, 1H, Ind-H5), 12.44 (s, 1H, NH), 12.64 (s, 1H, NH), 16.07 (s, 1H, NH); MS *m/z*: 297 [M^+]; Anal. $C_{10}H_6Cl_2N_6O \times 1\ H_2O$ (C, H, N); Calc.: C, 38.12; H, 2.56; N, 26.67. Found: C, 38.57; H, 2.69; N, 26.13%.

5.1.7. General procedure for the synthesis of the indole-2-carbonylmethanesulphonamides

The carboxylic acid **2** (0.4 g, 1.74 mmol) was dissolved in dry THF (10 mL). Then a solution of CDI (0.3 g, 1.85 mmol) in dry THF (10 mL) was added dropwise. The resulting solution was heated under reflux for 1 h. After only a few min of heating a precipitate was formed. The mixture was cooled to ambient temperature and the sulphonamide (1.85 mmol) was added. The reaction mixture was stirred for 10 min and a solution of 1,8-diazabicyclo[5.4.0]undec-7-en (DBU) (0.28 mL, 1.85 mmol) in dry THF (10 mL) was added. After stirring at room temperature overnight the solvent was removed under vacuum. The residue was treated with ethyl acetate and washed consecutively with 1N HCl and H_2O. The organic phase was dried ($MgSO_4$) and evaporated to dryness. Subsequent crystallisation in ethyl acetate afforded the pure products.

5.1.7.1. N-(4,6-Dichloro-1H-indole-2-carbonyl)-methanesulphonamide (9).

Slight yellow crystals (0.34 g, 1.11 mmol, 64%), m.p.: > 310 °C; IR (KBr) (cm^{-1}): 3270, 3230, 1910, 1650; ^{1}H-NMR (300 MHz) (DMSO-d_6) δ: 3.40 (s, 3H, CH_3), 7.28 (d, $J = 1.5$ Hz, 1H, Ind-H7), 7.44 (m, 1H, Ind-H3), 7.67 (d, $J = 1.5$ Hz, 1H, Ind-

H5), 12.38 (s, 2H, NH); MS *m/z*: 305 [M^+]; Anal. $C_{10}H_8Cl_2N_2O_3S$ (C, H, N).

*5.1.7.2. N-(4,6-Dichloro-1H-indole-2-carbonyl)-trifluoromethanesulphonamide (**10**).* Slight beige crystals (0.26 g, 0.73 mmol, 42%), m.p.: 265 °C; IR (KBr) (cm^{-1}): 3310, 3250, 1675; ^{1}H-NMR (300 MHz) (DMSO-d_6) δ: 6.80 (m, 1H, Ind-H3), 7.16 (d, $J = 1.4$ Hz, 1H, Ind-H7), 7.39 (m, 1H, Ind-H5), 11.78 (s, 2H, NH); MS *m/z*: 359 [M^+]; Anal. $C_{10}H_5Cl_2F_3N_2O_3S \times 2H_2O$ (C, H, N); Calc.: C, 30.24; H, 2.28; N, 7.05. Found: C, 30.14; H, 2.55; N, 6.40%.

*5.1.8. 4-tert-Butylphenylhydrazine hydrochloride (**11**)*

A suspension of *tert*-butylaniline (10 g, 67 mmol) in acetic acid (55 mL) and concentrated HCl (160 mL) was treated with an ice cold solution of $NaNO_2$ (4.62 g, 67 mmol) in H_2O (26 mL). The temperature was maintained below −10 °C. At the same time stannous dichloride dihydrate (45.4 g, 201 mmol) was dissolved in concentrated HCl (33 mL) and the solution cooled to a temperature below −15 °C under a nitrogen atmosphere. To this mixture the diazonium mixture is slowly added over a period of 2 h and the temperature always kept below −15 °C. The reaction was completed in an icebath overnight. The product which was obtained after filtration was purified by dissolving the stannous salts with boiling HCl. The solid hydrazine hydrochloride **11** was collected and dried under vacuum (10.8 g, 54 mmol, 81%). An analytical sample was obtained by treating the hydrochloride with aqueous NaOH, extracting the free base into ether, and precipitating the hydrochloride from the dried ethereal phase with freshly prepared ethereal HCl solution. M.p.: 242 °C; ^{1}H-NMR (300 MHz) (DMSO-d_6) δ: 1.22 (s, 9H, $C(CH_3)_3$), 6.93 (d, $J = 8.7$ Hz, 2H, Ph-H2,6), 7.27 (d, $J = 8.7$ Hz, 2H, Ph-H3,5), 10.24 (s, 3H, NH); MS *m/z*: 164 [M^+ free base].

*5.1.9. 2-[(4-tert-Butylphenyl)-hydrazono]-propionic acid ethyl ester (**12**)*

Compound **11** was dissolved in ethanol (150 mL), treated with a solution of ethyl pyruvate (5.7 g, 49 mmol) in ethanol (40 mL) and stirred at ambient temperature overnight. Part of this mixture (10 mL) was evaporated to dryness and chromatographed on silica gel to obtain two analytical samples, *Z*- and *E*-isomer (2/3). The remaining crude solution was used for the next step without further treatments. *Z*-isomer: yellow crystals; R_f (petroleum ether–ethyl acetate = 3/1): 0.64; m.p.: 63 °C; IR (KBr) (cm^{-1}): 3200, 2910, 1650, 1530, 1500; ^{1}H-NMR (300 MHz) (DMSO-d_6) δ: 1.24 (m, 12H, $C(CH_3)_3$, CH_2CH_3), 2.07 (s, 3H, N=CCH_3), 4.22 (q, $J = 7.2$ Hz, 2H, CH_2CH_3), 7.11 (d, $J = 8.8$ Hz, 2H, Ph-H2,6), 7.28 (d, $J = 8.8$ Hz, 2H, Ph-H3,5), 11.90 (s, 1H, NH); MS *m/z*: 262 [M^+]. *E*-isomer: orange crystals; R_f (petroleum ether–ethyl acetate = 3/1): 0.36; m.p.: 101 °C; IR (KBr) (cm^{-1}): 3240, 2920, 1680, 1500; ^{1}H-NMR (300 MHz) (DMSO-d_6) δ: 1.24 (m, 12H, $C(CH_3)_3$, CH_2CH_3), 2.03 (s, 3H, N=CCH_3), 4.17 (q, $J = 7.2$ Hz, 2H, CH_2CH_3), 7.18 (d, $J = 8.8$ Hz, 2H, Ph-H2,6), 7.28 (d, $J = 8.8$ Hz, 2H, Ph-H3,5), 9.75 (s, 1H, NH); MS *m/z*: 262 [M^+].

*5.1.10. 5-tert-Butyl-1H-indole-2-carboxylic acid ethyl ester (**13**)*

The crude mixture obtained for compound **12** is saturated with gaseous HCl and left at room temperature overnight. After evaporating most of the ethanol under reduced pressure the residue is hydrolysed and extracted several times with dichloromethane. The combined organic extracts were dried ($MgSO_4$) and evaporated to dryness. The residue was chromatographed on silica gel (petroleum ether–ethyl acetate = 10/1) to obtain a white solid (4.35 g, 18 mmol, 46%), R_f (petroleum ether–ethyl acetate = 10/1): 0.42; m.p.: 137 °C; IR (KBr) (cm^{-1}): 3290, 2920, 1670, 1500; ^{1}H-NMR (300 MHz) (DMSO-d_6) δ: 1.32 (m, 12H, $C(CH_3)_3$, CH_2CH_3), 4.31 (q, $J = 7.2$ Hz, 2H, CH_2CH_3), 7.08 (d, $J = 1.9$ Hz, 1H, Ind-H3), 7.36 (m, 2H, Ind-H6,7), 7.36 (m, 1H, Ind-H4), 11.72 (s, 1H, NH); MS *m/z*: 245 [M^+].

*5.1.11. 5-tert-Butyl-1H-indole-2-carboxylic acid (**14**)*

The ester **13** was dissolved in THF (22 mL) and a solution of LiOH monohydrate (0.11 g, 2.68 mmol) in water (11 mL) was added. The reaction was kept at room temperature overnight. Then it was cooled in an icebath and acidified with 10% HCl. The mixture was extracted several times with ethyl acetate. The combined organic extracts were dried ($MgSO_4$) and evaporated to dryness. Crystallisation from ethyl acetate yielded the pure product as slight beige crystals (0.43 g, 2.0 mmol, 82%), m.p.: 220 °C; IR (KBr) (cm^{-1}): 3300, 2920, 1640, 1500; ^{1}H-NMR (300 MHz) (DMSO-d_6) δ: 1.30 (s, 9H, $C(CH_3)_3$), 7.03 (d, $J = 1.5$ Hz, 1H, Ind-H3), 7.32 (m, 2H, Ind-H6,7), 7.56 (m, 1H, Ind-H4), 11.59 (s, 1H, NH), 12.84 (s, 1H, COOH); ^{13}C-NMR (75 MHz) (DMSO-d_6) δ: 31.9 (CH_3), 34.6 (Cq), 107.8 (CH), 115.2 (CH), 112.3 (CH), 117.4 (CH), 123.2 (CH), 127.0 (Cq), 128.6 (Cq), 135.8 (Cq), 142.5 (Cq), 163.2 (C=O); MS *m/z*: 217 [M^+]; Anal. $C_{13}H_{15}NO_2$ (C, H, N).

*5.1.12. 5-tert-Butyl-3-formyl-1H-indole-2-carboxylic acid ethyl ester (**15**)*

A mixture of *N*-methylformanilide (4 mL, 30 mmol) and compound **13** (5 g, 20.4 mmol) in 1,2-dichloroethane (20 mL) was stirred under a nitrogen atmosphere at room temperature. After 10 min phosphorus oxychloride (3 mL, 30 mmol) and 1,2-dichloroethane (40 mL) were added and the reaction heated under reflux for 8 h. The reaction mixture is hydrolysed by pouring it on

a mixture of sodium acetate trihydrate (9 g) and ice (12 g) and stirring at 4 °C overnight. The product separated as a solid and is collected by filtration. Light beige crystals were obtained (4.4 g, 16.1 mmol, 79%), R_f (*n*-hexane–ethyl acetate = 3/2): 0.57; m.p.: 206 °C; IR (KBr) (cm^{-1}): 3140, 2940, 1710, 1620; ^{1}H-NMR (300 MHz) (DMSO-d_6) δ: 1.32 (m, 12H, $C(CH_3)_3$, CH_2CH_3), 4.31 (q, $J = 6.8$ Hz, 2H, CH_2CH_3), 7.49 (m, 2H, Ind-H6,7), 8.23 (m, 1H, Ind-H4), 10.60 (s, 1H, CHO), 12.71 (s, 1H, NH); MS *m/z*: 273 [M^+].

5.1.13. General procedure for the synthesis of the hydantoin derivatives **16–18**

The glycine ethyl ester hydrochloride (1.5 equiv.) was suspended in dry dichloromethane (10 mL) and triethylamine (1.8 equiv.) was added. The indole-3-carbaldehyde derivative (2.0 equiv.) was added and the mixture left at room temperature for 20 min. After the addition of sodium triacetoxyborohydride (2.3 equiv.) the mixture was stirred at room temperature for 24 h. Phenylisocyanate (2.0 equiv) was added, and after another hour triethylamine (1.8 equiv.). Subsequently the reaction mixture was refluxed for 12 h. After dissolving in ethyl acetate the organic layer was washed with 5% HCl, saturated $NaHCO_3$ solution and water, dried ($MgSO_4$) and concentrated under reduced pressure.

5.1.13.1. 5-tert-Butyl-3-[(2,4-dioxo-3-phenyl-1-imidazolidinyl)methyl]-1H-indole-2-carboxylic acid ethyl ester (**16a**).

2.0 mmol carbaldehyde **15** and phenylisocyanate were used. Column chromatography (*n*-hexane–ethyl acetate = 3/1) and crystallisation from dichloromethane–*n*-hexane afforded the pure product (0.4 g, 0.93 mmol, 62%), R_f (*n*-hexane–ethyl acetate = 3/2): 0.08; m.p.: 178 °C; IR (KBr) (cm^{-1}): 3250, 2930, 1750, 1680, 1540; ^{1}H-NMR (300 MHz) (DMSO-d_6) δ: 1.29 (s, 9H, $C(CH_3)_3$), 1.38 (t, $J = 7.15$ Hz, 3H, CH_2CH_3), 3.95 (s, 2H, NCH_2CO), 4.38 (q, $J = 7.15$ Hz, 2H, CH_2CH_3), 5.05 (s, 2H, $ArCH_2N$), 7.30–7.83 (m, 8H, Ar-H), 11.75 (s, 1H, NH); MS *m/z*: 434 [M^+].

5.1.13.2. 5-tert-Butyl-3-{[3-(4-methylphenyl)-2,4-dioxo-1-imidazolidinyl]methyl}-1H-indole-2-carboxylic acid ethyl ester (**16b**).

1.17 mmol **15** and 4-methylphenylisocyanate were used. Column chromatography (*n*-hexane–ethyl acetate = 3/1) and crystallisation from dichloromethane–*n*-hexane afforded the pure product (0.35 g, 0.78 mmol, 78%): m.p.: 189 °C; IR (KBr) (cm^{-1}): 3300, 2960, 1710; ^{1}H-NMR (300 MHz) (DMSO-d_6) δ: 1.29 (s, 9H, $C(CH_3)_3$), 1.38 (t, $J = 7.15$ Hz, 3H, CH_2CH_3), 2.32 (s, 3H, Ph-CH_3); 3.93 (s, 2H, NCH_2CO), 4.38 (q, $J = 7.15$ Hz, 2H, CH_2CH_3), 5.05 (s, 2H, $ArCH_2N$), 7.19 (d, $J = 8.35$ Hz, 2H, Ph-H3,5), 7.27 (d, $J = 8.35$ Hz, 2H, Ph-H2,6), 7.39 (m, 2H, Ind-H6,7), 7.83 (m, 1H, Ind-H4), 11.73 (s, 1H, NH); MS *m/z*: 448 [M^+].

5.1.13.3. 5-tert-Butyl-3-{[3-(4-methoxyphenyl)-2,4-dioxo-1-imidazolidinyl]methyl}-1H-indole-2-carboxylic acid ethyl ester (**16c**).

1.17 mmol **15** and 4-methoxyphenylisocyanate were used. Column chromatography (*n*-hexane–ethyl acetate = 3/1) and crystallisation from dichloromethane–*n*-hexane afforded the pure product (0.34 g, 0.73 mmol, 73%): m.p.: 205 °C; IR (KBr) (cm^{-1}): 3220, 2920, 1740, 1680, 1490; ^{1}H-NMR (300 MHz) (DMSO-d_6) δ: 1.29 (s, 9H, $C(CH_3)_3$), 1.38 (t, $J = 7.2$ Hz, 3H, CH_2CH_3), 3.77 (s, 3H, OCH_3), 3.92 (s, 2H, NCH_2CO), 4.38 (q, $J = 7,2$ Hz, 2H, CO_2CH_2), 5.04 (s, 2H, $ArCH_2N$), 7.01 (d, $J = 8.82$ Hz, 2H, Ph-H3,5), 7.21 (d, $J = 8.82$ Hz, 2H, Ph-H2,6), 7.39 (m, 2H, Ind-H6,7), 7.82 (m, 1H, Ind-H4), 11.73 (s, 1H, NH); MS *m/z*: 462 [M^+].

5.1.13.4. 3-[(2,4-Dioxo-3-phenyl-1-imidazolidinyl)methyl]-1H-indole-2-carboxylic acid ethyl ester (**17a**).

2.0 mmol 3-formyl-indole-2-carboxylic acid ethyl ester and phenylisocyanate were used. Column chromatography (*n*-hexane–ethyl acetate = 3/1) and crystallisation from methanol afforded the pure product (0.37 g, 0.98 mmol, 65%); m.p.: 192 °C; IR (KBr) (cm^{-1}): 3200, 1750, 1690; ^{1}H-NMR (300 MHz) (DMSO-d_6) δ: 1.39 (t, $J = 7.2$ Hz, 3H, CH_3), 3.96 (s, 2H, NCH_2CO), 4.40 (q, $J = 7.2$ Hz, 2H, CH_2CH_3), 5.05 (s, 2H, $IndCH_2N$), 7.07–7.84 (m, 9H, Ph-H, Ind-H), 11.90 (s, 1H, NH); MS *m/z*: 376 [M^+].

5.1.13.5. 4,6-Dichloro-3-[(2,4-dioxo-3-phenyl-1-imidazolidinyl)methyl]-1H-indole-2-carboxylic acid ethyl ester (**18a**) and the derivatives **18b** and **18c**.

Were prepared as described previously [25].

5.1.14. General procedure for the hydrolysis of the esters to yield acids **19–21**

The ethyl ester (1 equiv.) was dissolved in THF (15 mL) and a solution of LiOH monohydrate (1.2 equiv.) in water (7 mL) was added. Subsequently the reaction mixture was stirred at room temperature until the ester disappeared (TLC). Then the solution was cooled in an icebath and acidified with 10% HCl and extracted with several portions of ethyl acetate. The combined organic extracts were dried ($MgSO_4$) and evaporated to dryness.

5.1.14.1. 5-tert-Butyl-3-[(2,4-dioxo-3-phenyl-1-imidazolidinyl)methyl]-1H-indole-2-carboxylic acid (**19a**).

Ethyl ester **16a** (0.35 g, 0.81 mmol) was hydrolysed according to the general procedure. Crystallisation from dichloromethane–*n*-hexane afforded the pure product as white crystals (0.28 g, 0.7 mmol, 86%), m.p.: 237 °C; IR (KBr) (cm^{-1}): 3400–2300, 3180, 2920, 1750, 1685, 1640, 1530; ^{1}H-NMR (300 MHz) (DMSO-d_6) δ: 1.28 (s, 9H, $C(CH_3)_3$), 3.93 (s, 2H, NCH_2CO), 5.06 (s, 2H, $ArCH_2N$), 7.30–7.80 (m, 8H, Ar-H), 11.63 (s, 1H, NH), 13.24 (bs, 1H, COOH); ^{13}C-NMR (75

MHz) (DMSO-d_6) δ: 31.9 (3 CH_3), 34.7 (Cq), 36.3 (CH_2), 49.5 (CH_2), 112.4 (CH), 115.6 (Cq), 116.0 (CH), 123.8 (CH), 126.3 (Cq), 126.8 (2 CH), 128.1 (CH), 129.1 (2 CH), 132.6 (Cq), 134.7 (Cq), 142.7 (Cq), 155.3 (C=O), 163.4 (C=O), 169.5 (C=O); MS *m/z*: 404 [M^+]; Anal. $C_{23}H_{23}N_3O_4$ (C, H, N).

*5.1.14.2. 5-tert-Butyl-3-{[3-(4-methylphenyl)-2,4-dioxo-1-imidazolidinyl]methyl}-1H-indole-2-carboxylic acid (**19b**).* Ethyl ester **16b** (0.30 g, 0.67 mmol) was hydrolysed according to the general procedure. Crystallisation from dichloromethane/n-hexane afforded the pure product as white crystals (0.11 g, 0.27 mmol, 40%), m.p.: >190 °C; IR (KBr) (cm^{-1}): 3600–2500, 3320, 2910, 1740, 1675; ^{1}H-NMR (300 MHz) (DMSO-d_6) δ: 1.28 (s, 9H, $C(CH_3)_3$), 2.32 (s, 3H, CH_3), 3.90 (s, 2H, NCH_2CO), 5.05 (s, 2H, $ArCH_2N$), 7.17 (d, $J = 8.11$ Hz, 2H, Ph-H2,6), 7.26 (d, $J = 8.11$ Hz, 2H, Ph-H3,5), 7.36 (m, 2H, Ind-H6,7), 7.79 (m, 1H, Ind-H4), 11.62 (s, 1H, NH), 13.00 (bs, 1H, COOH); MS *m/z*: 420 [M^+]; Anal. $C_{24}H_{25}N_3O_4 \times 1/3\ H_2O$ (C, H, N).

*5.1.14.3. 5-tert-Butyl-3-{[3-(4-methoxyphenyl)-2,4-dioxo-1-imidazolidinyl]methyl}-1H-indole-2-carboxylic acid (**19c**).* Ethyl ester **16c** (0.29 g, 0.60 mmol) was hydrolysed according to the general procedure. Crystallisation from dichloromethane–*n*-hexane afforded the pure product as white crystals (0.23 g, 0.53 mmol, 88%), m.p.: 240 °C; IR (KBr) (cm^{-1}): 3600–2500, 3320, 2920, 1740, 1680, 1490; ^{1}H-NMR (300 MHz) (DMSO-d_6) δ: 1.28 (s, 9H, $C(CH_3)_3$), 3.77 (s, 3H, OCH_3), 3.89 (s, 2H, NCH_2CO), 5.05 (s, 2H, $ArCH_2N$), 7.01 (d, $J = 8.67$ Hz, 2H, Ph-H2,6), 7.21 (d, $J = 8.67$ Hz, 2H, Ph-H2,6), 7.36 (m, 2H, Ind-H6,7), 7.79 (m, 1H, Ind-H4), 11.62 (s, 1H, NH), 13.25 (bs, 1H, COOH); ^{13}C-NMR (75 MHz) (DMSO-d_6) δ: 31.9 (3 CH_3), 34.7 (Cq), 36.3 (CH_2), 49.4 (CH_2), 55.7 (CH_3), 112.4 (CH), 114.4 (2 CH), 115.7 (Cq), 116.0 (CH), 123.7 (CH), 125.2 (Cq), 126.3 (Cq), 126.8 (Cq), 128.2 (2 CH), 134.7 (Cq), 142.7 (Cq), 155.6 (C=O), 158.9 (Cq), 163.4 (C=O), 169.7 (C=O); MS *m/z*: 437 [M^+]; Anal. $C_{24}H_{25}N_3O_5$ (C, H, N).

*5.1.14.4. 3-[(2,4-Dioxo-3-phenyl-1-imidazolidinyl)methyl]-1H-indole-2-carboxylic acid (**20a**).* Ethyl ester **17a** (0.30 g, 0.79 mmol) was hydrolysed according to the general procedure. Crystallisation from dichloromethane–*n*-hexane afforded the pure product as white crystals (0.17 g, 0.5 mmol, 63%); m.p.: 264 °C; IR (KBr) (cm^{-1}): 3550–2400, 3320, 1740, 1680; ^{1}H-NMR (300 MHz) (DMSO-d_6) δ: 3.94 (s, 2H, NCH_2CO), 5.05 (s, 2H, $ArCH_2N$), 7.07–7.80 (m, 9H, Ar-H), 11.79 (s, 1H, NH), 13.37 (bs, 1H, COOH); ^{13}C-NMR (75 MHz) (DMSO-d_6) δ: 36.2 (CH_2), 49.6 (CH_2), 112.9 (CH), 115.4 (Cq), 120.4 (CH), 120.7 (CH), 125.1 (CH), 126.4 (Cq), 126.9 (2 CH), 127.2 (Cq), 128.0 (CH), 129.0 (2 CH), 132.6 (Cq), 136.4 (Cq), 155.4 (C=O), 163.3 (C=O), 169.5 (C=O); Anal. $C_{19}H_{15}N_3O_4 \times H_2O$ (C, H, N).

*5.1.15. 4,6-Dichloro-3-[(2,4-dioxo-3-phenyl-1-imidazolidinyl)methyl]-1H-indole-2-carboxylic acid (**21a**) and derivatives **21b** and **21 c***

These were prepared starting from **18** as described previously [25].

5.2. Binding assay

The final compounds were dissolved in DMSO–H_2O (v% = 50:50); when necessary one fifth of the water was replaced with 0.1N NaOH. The substances were tested at ten different concentrations in duplicate in displacing [^{3}H]MDL 105,519 (2 nM) from pig cortical brain membranes as described by Baron et al. [41]. A reference compound was always included as an internal control. Pig brain membranes were prepared according to procedures described by Hoefner et al. [42,43], Baron et al. [41,44] and Marvizón et al. [45]. The protein content was measured using a procedure described by Bradford [46] and adjusted to contain 50 μg protein in 0.5 mL final volume during the displacement experiment which was performed in 50 mM Tris buffer (pH 8.0). IC_{50} values were measured from at least ten-point inhibition curves. Data were analysed using the curve-fitting software GraFit, Erithacus Software Ltd. K_i values were calculated from IC_{50} values using a method described by Cheng and Prusoff [47]. All the experiments were repeated at least twice and the K_i values stated represent the geometrical mean of all the values obtained for a specific substance.

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References

[1] T. Knöpfel, R. Kuhn, H. Allgeier, J. Med. Chem. 38 (1995) 1417–1426.
[2] H. Bräuner-Osborne, J. Egebjerg, E.Ø. Nielsen, U. Madsen, P. Krogsgaard-Larsen, J. Med. Chem. 43 (2000) 2609–2645.
[3] M.P. Mattson, B. Cheng, D. Davis, K. Bryant, I. Lieberburg, R.E. Rydel, J. Neurosci. 12 (1992) 376–389.
[4] C. Arias, I. Arrieta, R. Tapia, J. Neurosci. Res. 41 (1995) 561–566.
[5] M.E. Harris, Y.N. Wang, N.W. Pedigo, K. Hensley, D.A. Butterfield, J.M. Carney, J. Neurochem. 67 (1996) 277–286.
[6] P.A. Löschmann, K.W. Lange, M. Kunow, K.J. Rettig, P. Jähnig, T. Honoré, L. Turski, H. Wachtel, P. Jenner, C.D. Marsden, J.

Neural Transm.: Parkinson's Dis. Dementia Sect. 3 (1973) 203–213.
[7] J.L. Wright, T.F. Gregory, S.R. Kesten, P.A. Boxer, K.A. Serpa, L.T. Meltzer, L.D. Wise, J. Med. Chem. 43 (2000) 3408–3419.
[8] S.E. Purdon, E. Mohr, V. Ilivitsky, B.D. Jones, J. Psychiatry Neurosci. 19 (1994) 359–367.
[9] W. Löscher, Progr. Neurobiol. 54 (1998) 721–741.
[10] L.J. Rogers, J.W. Hambley, Behav. Brain Res. 4 (1982) 1–18.
[11] R.G. Morris, E. Anderson, G.S. Lynch, M. Baudry, Nature 319 (1986) 774–776.
[12] G. Collingridge, Nature 330 (1987) 604–605.
[13] P. Skolnick, Amino Acids 23 (2002) 153–159.
[14] R. Di Fabio, N. Conti, M. Corsi, D. Donati, P. Gastaldi, G. Gaviraghi, S. Giacobbe, G. Pentassuglia, M. Quartaroli, E. Ratti, D.G. Trist, A.R. Ugolini, Drugs Future 25 (2000) 137–145.
[15] M.J. Croucher, J.F. Collins, B.S. Meldrum, Science 216 (1982) 899–901.
[16] C.E. Herron, R.A. Lester, E.J. Coan, G.L. Collingridge, Nature 322 (1986) 265–268.
[17] J.C. Marvizón, M. Baudry, Eur. J. Pharmacol. 269 (1994) 165–175.
[18] C.M. Low, F. Zheng, P. Lyuboslavsky, S.F. Traynelis, Proc. Natl. Acad. Sci. USA 97 (2000) 11062–11067.
[19] C.J. McBain, M.L. Mayer, Physiol. Rev. 74 (1994) 723–760.
[20] R.G.M. Morris, E. Anderson, G.S. Lynch, M. Baudry, Nature 319 (1986) 774–776.
[21] D.W. Choi, S.M. Rothman, Annu. Rev. Neurosci. 13 (1990) 171–182.
[22] W. Koek, F.C. Colpaert, J. Pharmacol. Exp. Ther. 252 (1990) 349–357.
[23] (a) F.G. Salituro, B.L. Harrison, B.M. Baron, P.L. Nyce, K.T. Stewart, I.A. McDonald, J. Med. Chem. 33 (1990) 2944–2946;
(b) N.M. Gray, M.S. Dappen, B.K. Cheng, A.A. Cordi, J.P. Biesterfeldt, W.F. Hood, J.B. Monahan, J. Med. Chem. 34 (1991) 1283–1292;
(c) P.D. Leeson, R.W. Carling, K.W. Moore, A.M. Moseley, J.D. Smith, G. Stevenson, T. Chan, R. Baker, A.C. Foster, S. Grimwood, J.A. Kemp, G.R. Marshall, K. Hoogsteen, J. Med. Chem. 35 (1992) 1954–1968;
(d) V. Colotta, D. Catarzi, F. Varano, L. Cecchi, G. Filacchioni, A. Galli, C. Costaldi, Arch. Pharm. Pharm. Med. Chem. 330 (1997) 129–134;
(e) R.W. Carling, P.D. Leeson, K.W. Moore, C.R. Moyes, M. Duncton, M.L. Hudson, R. Baker, A.C. Foster, S. Grimwood, J.A. Kemp, G.R. Marshall, M.D. Tricklebank, K.L. Saywell, J. Med. Chem. 40 (1997) 754–765;
(f) G. Dannhardt, B.K. Kohl, Curr. Med. Chem. 5 (1998) 253–263;
(g) H. Poschenrieder, G. Höfner, H.D. Stachel, Arch. Pharm. Pharm. Med. Chem. 331 (1998) 389–394;
(h) C. Balsamini, A. Bedini, G. Diamantini, G. Spadoni, A. Tontini, G. Tarzia, R. Di Fabio, A. Feriani, A. Reggiani, G. Tedesco, R. Valigi, J. Med. Chem. 41 (1998) 808–820;
(i) R. Di Fabio, N. Conti, E. De Magistris, A. Feriani, S. Provera, F.M. Sabbatini, A. Reggiani, L. Rovatti, R.J. Barnaby, J. Med. Chem. 42 (1999) 3486–3493;
(j) H. Poschenrieder, G. Höfner, H.D. Stachel, Arch. Pharm. Pharm. Med. Chem. 333 (2000) 211–216;
(k) G. Dannhardt, B.K. Kohl, Arch. Pharm. Pharm. Med. Chem. 333 (2000) 123–129;
(l) S. Katayama, N. Ae, R. Nagata, J. Org. Chem. 66 (2001) 3474–3483.
[24] R. Di Fabio, A.M. Capelli, N. Conti, A. Cugola, D. Donati, A. Feriani, P. Gastaldi, G. Gaviraghi, C.T. Hewkin, F. Micheli, A. Missio, M. Mugnaini, A. Pecunioso, A.M. Quaglia, E. Ratti, L. Rossi, G. Tedesco, D.G. Trist, A. Reffiani, J. Med. Chem. 40 (1997) 841–850.
[25] M. Jansen, H. Potschka, C. Brandt, W. Löscher, G. Dannhardt, J. Med. Chem. 46 (2003) 64–73.
[26] C. Gränacher, A. Mahal, M. Gerö, Helv. Chim. Acta 7 (1924) 579–586.
[27] Z.L. Hickman, C.F. Sturino, N. Lachance, Tetrahedron Lett. 21 (2000) 8217–8220.
[28] J.S. Mihina, R.M. Herbst, J. Org. Chem. 15 (1950) 1082–1092.
[29] R.M. Herbst, W.L. Garbrecht, J. Org. Chem. 18 (1953) 1283–1291.
[30] D.C. Bhattacharyya, A.K. Basu, S. Aditya, J. Indian Chem. Soc. 61 (1984) 956–959.
[31] A.D. Headley, S.D. Starnes, L.Y. Wilson, G.R. Famini, J. Org. Chem. 59 (1996) 8040–8046.
[32] K. Chamberlain, A.A. Evans, R.H. Bromilow, Pestic. Sci. 47 (1996) 265–272.
[33] V.S. Talanov, G.G. Talanova, M.G. Gorbunova, R.A. Bartsch, J. Chem. Soc. Perkin Trans. 2 2 (2002) 209–215.
[34] P.D. Leeson, L.L. Iversen, J. Med. Chem. 37 (1994) 4053–4067.
[35] I.G. Tikhonova, I.I. Baskin, V.A. Palyulin, N.S. Zefirov, J. Med. Chem. 46 (2003) 1609–1616.
[36] G. Pappalardo, T. Vitali, Gazz. Chim. Ital. 88 (1958) 1147–1169.
[37] F.D. Chattaway, O.C. Ellington, J. Chem. Soc. 109 (1916) 587–593.
[38] P. Hill, R. Robinson, J. Chem. Soc. (1933) 486–488.
[39] F.P. Doyle, W. Ferrier, D.O. Holland, M.D. Mehta, J.H.C. Nayler, J. Chem. Soc. (1956) 2853–2857.
[40] P. Desos, J.M. Lepagnol, P. Morain, P. Lestage, A.A. Cordi, J. Med. Chem. 39 (1996) 197–206.
[41] B.M. Baron, B.W. Siegel, B.L. Harrison, R.S. Gross, C. Hawes, P. Towers, J. Pharm. Exp. Therap. 279 (1996) 62–68.
[42] G. Höfner, K.Th. Wanner, J. Receptor Signal Transduction Res. 16 (1996) 297–313.
[43] G. Höfner, K.Th. Wanner, Neurosci. Lett. 226 (1997) 79–82.
[44] B.M. Baron, B.W. Siegel, A.L. Slone, B.L. Harrison, M.G. Palfreyman, S.D. Hurt, Eur. J. Pharmacol.—Mol. Pharm. Sect. 206 (1991) 149–154.
[45] J.C. Marvizón, M. Baudry, J. Neurochem. 63 (1994) 963–971.
[46] M.M. Bradford, Anal. Biochem. 72 (1976) 248–254.
[47] Y. Cheng, W.J. Prusoff, Biochem. Pharmacol. 22 (1973) 3099–3108.