Available online at www.derpharmachemica.com

Scholars Research Library

Der Pharma Chemica, 2012,4 (3):909-914
(http://derpharmachemica.com/archive.html)

ISSN 0975-413X
CODEN (USA): PCHHAX

Synthesis and biological screening of 3-chloro-2-piperazinylmethyl-N-aryl sulfonamide indole derivatives as 5-HT$_6$ receptor ligands: Part III

Ramakrishna V. S. Nirogi [1], Amol D. Deshpande [1, 2], Anil K. Chindhe [1], Ramasastri Kambhampati [1], Anil K. Shinde[1*] and P. K. Dubey [2]

[1]Discovery Research - Medicinal Chemistry, Suven Life Sciences Ltd., Hyderabad 500 034, India
[2]Department of Chemistry, Jawaharlal Nehru Technological University Hyderabad, Kukatpally, Hyderabad 500 085, India

---

ABSTRACT

In continuation to our earlier work and to improve upon the potentency of 2-piperazinylmethyl-N-aryl sulfonamide indole derivatives, a new series of compounds were taken up for synthesis by inserting halo group at 3rd position of indole. The critical intermediate 3-halo-2-formyl indole was achieved through the Vilsmerier reaction, which on further reductive amination followed by sulfonylation gave 3-chloro-2-piperazinylmethyl-N-aryl sulfonamide indole derivatives as a targeted series of compounds. The design, synthesis and their in-vitro affinity towards the 5-HT$_6$ receptor is discussed in this communication.

Key words: 3-halo-2-piperazinylmethyl N_1-arylsulfonyl indole derivatives, Vilsmerier reaction, 5-HT$_6$ receptor, *in-vitro* affinity.

INTRODUCTION

5-HT$_6$ receptor mainly found in limbic, cortical and hippocampus region of the brain [1]. Due to exclusive distribution of this receptor in the brain, makes it a promising and novel target for central nervous system (CNS) mediated diseases such as Alzheimer's disease, schizophrenia, anxiety and obesity [2 - 4].

In our earlier communication, we reported 2-(4-methylpiperazin-1-yl methyl)-1-(arylsulfonyl)-*1H*-indole derivatives as the potential 5-HT$_6$ receptor ligands. In continuation to this effort and to make these molecules more potent we developed a new series of compounds by insertion of halo group at 3rd position of indole nucleus. Thus, the main aim of the study is to see the effect of insertion of halogen on the binding affinity towards the 5-HT$_6$ receptor.

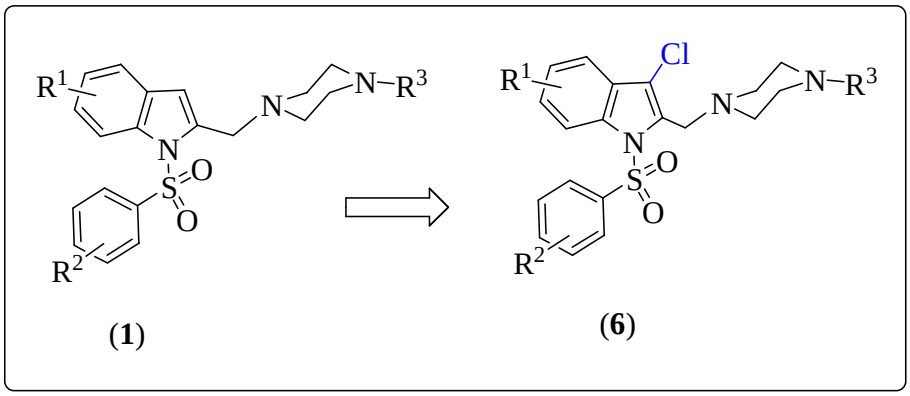


Figure 1: Genesis of ligands

MATERIALS ANDS METHODS

2.1 Chemistry

Melting points of synthesized compounds were determined using Electro Derman open capillary apparatus and are uncorrected. Infra red spectra were recorded on KBr disc and in solid state using Perkin-Elmer model 1600 FT-IR spectrophotometer (Perkin-Elmer, Norwalk, CT, USA). Electrospray ionization mass spectra were recorded on a API 4000 triple quadrupole instrument (MDS-SCIEX, Concord, Ontario, Canada). ^{1}H-NMR spectra were obtained on a Bruker proton NMR spectrometer (Fallanden, Switzerland) at 400 MHz. Deuterated reagents were used as solvents and were commercially procured. Tetramethylsilane (TMS) was used as an internal standard. Chemical shift values are expressed in parts per million (δ) and coupling constants are expressed in Hz. Thin layer chromatography (TLC) was performed on Merck silica gel 60 F_{254} plates. Chromatography refers to column chromatography performed using 60-120 mesh silica gel and executed under nitrogen pressure (flash chromatography) conditions. All the reagents and chemicals used were of 'reagent grade'.

2.2 General procedure for 2-(Carboxymethyl-amino)-benzoic acid derivatives (3)

To a stirred solution of **2** (0.03 mol) in water (30 mL) was added NaOH (0.09 mol) dissolved in water (35 mL). Chloroacetic acid (0.036 mol) was added to the reaction mixture and the reaction mixture was heated to 90 °C. After completion of reaction, the reaction mixture was cooled to RT and the separated solids were filtered. ML was discarded and the solid mass was taken in chilled water (50 mL). pH was adjusted to 3 using 10 % aqueous HCl solution. The resulting mass was extracted with ethyl acetate (2 x 50 mL). Combined organic layer was washed with brine solution (1 x 25 mL) and dried over sodium sulfate. The solvent was removed under reduced pressure to obtain the crude compound, which was purified by column chromatography using ethyl acetate to obtain **3.**

5-Bromo-2-(carboxymethyl amino)benzoic acid (3a, R^1 = Br): MR (^{o}C): 205 - 206; IR (cm^{-1}): 3351, 1720 (acid carbonyl stretching), 1569, 1507, 1227; ^{1}H-NMR ($CDCl_3$): δ 3.98 (2H, s, CH_2), 7.12 - 7.13 (1H, d, aromatic), 7.60 - 7.63 (1H, dd, aromatic), 7.84 - 7.84 (1H, d, aromatic); Mass (m/z): 272.0, 273.9 (M-H)$^-$.

2-(carboxymethyl amino) benzoic acid (3b, R^1 = H): IR (cm^{-1}): 3373, 1726 (acid carbonyl stretching), 1665, 1574, 1398, 1223; ^{1}H-NMR (DMSO-d_6): δ 3.97 (2H, S, CH_2), 6.56 - 6.60 (2H, m), 7.32 - 7.37 (1H, m), 7.77 - 7.81 (1H, m), 8.10 (1H, bs, -NH), 12.72 (2H, bs, -COOH); Mass (m/z): 193.9 (M-H)$^-$.

5-Chloro - 2-(carboxymethyl amino) benzoic acid (3c, R^1 = Cl): IR (cm^{-1}): 3352, 1724 (acid carbonyl stretching), 1562, 1524, 1230; ^{1}H-NMR (DMSO-d_6): 4.01 - 4.04 (2H, d, CH_2), 6.59 - 6.64 (2H, m), 7.75 - 7.80 (1H, d), 8.23 (1H, bs, -NH), 12.82 (2H, bs, -COOH); Mass (m/z): 228.1, 230.4 (M-H)$^-$.

2.3. General procedure for 3-Chloro-2-formyl indole derivatives (4)

To a stirred solution of **3** (0.01 mol) in DMF (25 mL) was added $POCl_3$ (0.06 mol) at -10 °C. The above mixture was stirred for 30 minutes maintaining the temperature below -5 °C and then heated at 90 °C for 7 h. After completion of reaction, the mixture was quenched into ice water (100 mL) and pH made neutral with saturated sodium acetate solution (50 mL). The aq. layer was extracted with ethyl acetate (2 x 50 mL) and washed with brine

solution (1 x 25 mL). The combined organic layer was dried over sodium sulphate and evaporated under reduced pressure. The residual crude product was purified by flash chromatography using 5 % ethyl acetate in hexane to afford **4** in moderate yield.

5-Bromo-3-chloro-2-formyl indole (4a, R^1 = 5-Br): MR (^oC): 255.7 - 258.6; IR (cm^{-1}): 3273 (indole NH stretching), 1653 (aldehydic carbonyl stretching); ^{1}H-NMR (DMSO-d_6): δ 7.38 - 7.44 (2H, m), 7.81 - 7.82 (1H, m), 10.04 (1H, aldehydic), 12.02 (1H, indole NH); Mass (m/z): 256.1, 258.1 $(M+H)^+$.

3-Chloro-2-formyl indole (4b, R^1 = H): MR (^oC): 169 - 172; IR (cm^{-1}): 3288 (indole NH stretching), 1655 (aldehydic carbonyl stretching); ^{1}H-NMR ($CDCl_3$): δ 7.24 - 7.26 (1H, m), 7.44 - 7.47 (2H, m), 7.75 - 7.77 (1H, m), 9.17 (1H, indole NH), 10.06 (1H, aldehydic); Mass (m/z): 178.3, 180.2 $(M-H)^-$.

3,6 -Dichloro-2-formyl indole (4c, R^1 = 6-Cl): MR (^oC): 219 - 220; IR (cm^{-1}): 3277 (indole NH stretching), 1651 (aldehydic carbonyl stretching); ^{1}H-NMR ($CDCl_3$): δ 7.20 - 7.23 (1H, dd, J= 8.6, 1.48 Hz), 7.43 - 7.44 (1H, d, J=1.1 Hz), 7.67 - 7.69 (1H, d, J= 8.6 Hz), 9.04 (1H, indole NH), 10.03 (1H, aldehydic); Mass (m/z): 211.9, 213.8 $(M-H)^-$.

2.4 General procedure for 3-Chloro-2-(4-methylpiperazin-1-ylmethyl)-1H-indole derivatives (5)

To a stirred solution of **4** (0.0038 mol) in EDC (10 vol) was added N-methylpiperazine (0.46g, 0.0046 mol) followed by acetic acid (2-3 drops) and the resulting mixture was stirred at RT for 2 h. Sodium triacetoxyborohydride was added to the reaction mixture and the reaction was stirred at RT for 4 h. After completion of reaction on TLC, the mass was diluted with DCM (50 mL) and washed with water (50 mL). Collective organic layer was dried over Na_2SO_4 and the organic volatiles were evaporated under reduced pressure to obtain a crude mass, which was further purified by column chromatography using 1% TEA in ethyl acetate to obtain **5** in good yields.

5-Bromo-3-chloro-2-(4-methyl-piperazin-1-ylmethyl)-1H-indole (5a, R^1 = 5-Br) : MR (^oC): 179.2 - 183.5; IR (cm^{-1}): 3109 (indole NH stretching), 1451, 1290, 1155, 789; ^{1}H-NMR ($CDCl_3$): δ 2.29 (3H, s, N-CH_3), 2.46 - 2.54 (8H, bs, piperazinyl) 3.69 (2H, s, -CH_2), 7.15 - 7.19 (1H, d, C-7, J = 8.6Hz), 7.26 - 7.29 (1H, dd, C-6, J = 2 Hz), 7.69 - 7.70 (1H, d, C-4, J = 1.6 Hz); Mass (m/z): 342.2.3, 344.2 $(M+H)^+$.

3-Chloro-2-(4-methyl-piperazin-1-ylmethyl)-1H-indole (5b, R^1 = H): MR (^oC): 179.2 - 183.5; IR (cm^{-1}): 3114 (indole NH stretching), 2838, 1480, 1280; ^{1}H-NMR (CDCl3): δ 2.24 (3H, s, N-CH_3), 2.44 - 2.52 (8H, bs, piperazinyl) 3.72 (2H, s, -CH_2), 7.10 - 7.23 (4H, m); Mass (m/z): 364.2, 366.1 $(M+H)^+$.

3,6-Dichloro-2-(4-methyl-piperazin-1-ylmethyl)-1H-indole (5c, R^1 = 6-Cl): MR (^oC): 176.1 - 178.2; IR (cm^{-1}): 3128 (indole NH stretching), 2819, 1455, 1329; ^{1}H-NMR (CD_3OD): δ 2.27 (3H, s, N-CH_3), 2.43 - 2.64 (8H, bs, piperazinyl) 3.72 (2H, s, -CH_2), 7.04 - 7.06 (1H, dd, C-5, J = 8.4, 1.84Hz), 7.34 - 7.35 (1H, d, C-7, J = 1.8 Hz), 7.39 - 7.41 (1H, dd, C-4, J = 8.4 Hz); Mass (m/z): 296.1, 298.1 $(M-H)^-$.

2.5. General procedure for 1-arylsulfonyl-3-chloro-2-(4-methyl-piperazin-1-ylmethyl)-*1H*-indole derivatives (6)

To a stirred solution of sodium hydride (0.65 mol, 50 % suspension in mineral oil) in 10 mL THF was added a solution of **5** (0.5 mol) dissolved in 5 mL of THF under nitrogen atmosphere at 25 - 30 ^oC. The mass was further stirred for 45 min. A solution of aryl sulfonyl chloride (0.65 mol) in 5 ml THF was added to the above reaction mass and the mass was further stirred for 2 hr at 25 - 30 ^oC. The progress of the reaction was monitored by TLC. After completion of the reaction, the mass was quenched on to water and the product was extracted with ethyl acetate (2 x 25 mL). The combined organic layer was washed with brine solution (1 x 100 mL) and dried over anhydrous magnesium sulfate. Organic volatiles were removed under vacuum to obtain the title compound. The product was purified by flash chromatography using 1% TEA in ethyl acetate.

5-Bromo-3-chloro-2-(4-methyl-piperazin-1-ylmethyl)-1-(toluene-4-sulfonyl)-1H-indole (6a, R^1 = 5-Br, R^2 = 4'-CH_3) : MR (^oC): 142.4 - 151; IR (KBr, cm^{-1}): 2935, 2792, 1595, 1442, 1379 (SO_2 stretching), 1177 (SO_2 stretching), 1011; ^{1}H-NMR ($CDCl_3$): δ 2.27 (3H, s, N-CH_3), 2.37 (3H, s, -CH_3), 2.59 - 2.61 (8H, bs, piperazinyl), 3.96 (2H, s, benzylic), 7.21- 7.23 (2H, d, J= 8.2 Hz), 7.42 - 7.45 (1H, dd, J= 8.2, 1.84 Hz), 7.65 - 7.66 (1H, d, J = 1.76 Hz), 7.96 - 7.98 (1H, d, J = 8.2 Hz), 8.11 – 8.13 (2H, d, J= 8.2 Hz); Mass (m/z): 496.2, 498.1 $[M+H]^+$.

5-Bromo-1-(2-bromobenzenesulfonyl)-3-chloro-2-(4-methyl-piperazin-1-ylmethyl)-1H-indole (6b, R^1 = 5-Br, R^2 = 2'- Br) : MR (^oC): 209.2 - 211; IR (KBr, cm^{-1}): 2941, 1446, 1372 (SO_2 stretching), 1178 (SO_2 stretching), 1011; ^{1}H-NMR ($CDCl_3$): δ 2.04 (3H, s, N-CH_3), 2.14 – 2.35 (8H, bs, piperazinyl), 3.86 (2H, s, benzylic), 7.29- 7.34 (1H, m), 7.43 - 7.52 (3H, m), 7.73 - 7.74 (1H, d, J = 1.93 Hz), 7.96 - 7.98 (1H, d, J = 8.98 Hz); Mass (m/z): 562, 564.1 $[M+H]^+$.

5-Bromo-1-(2-chlorobenzenesulfonyl)-3-chloro-2-(4-methyl-piperazin-1-ylmethyl)-1H-indole (6c, R^1 = 5-Br, R^2 = 2'- Cl) : MR (^oC): 186.8 - 187.8; IR (KBr, cm^{-1}): 2945, 2796, 1452, 1372 (SO_2 stretching), 1179 (SO_2 stretching), 1008; ^{1}H-NMR ($CDCl_3$):δ 2.03 (3H, s, N-CH_3), 2.06 - 2.35 (8H, bs, piperazinyl), 3.85 (2H, s, benzylic), 7.29- 7.42 (3H, m), 7.43 - 7.46 (1H, dd), 7.73 - 7.75 (1H, m), 7.95 - 7.98 (1H, d, J = 8.96 Hz); Mass (m/z): 516.1, 518.1 $[M+H]^+$.

3,6-Dichloro-1-(4-isopropylbenzenesulfonyl)-2-(4-methyl-piperazin-1-ylmethyl)-1H-indole (6d, R^1 = 6-Cl, R^2 = 4'- i-Pr) : MR (^oC): 156.6 - 161; IR (KBr, cm^{-1}): 2945, 2796, 1452, 1372 (SO_2 stretching), 1179 (SO_2 stretching), 1008; ^{1}H-NMR (CD_3OD):δ 1.21 - 1.23 (6H, d), 2.00 (3H, s, N-CH_3), 2.40 (8H, bs, piperazinyl), 2.91 -2.99 (1H, sept), 3.98 (2H, s, benzylic), 7.35 - 7.38 (1H, dd, J= 1.74, 8.46 Hz), 7.40 - 7.43 (2H, dd), 7.52 - 7.54 (1H, d, J= 8.42 Hz), 7.89 - 7.91 (2H, dd), 8.16 - 8.17 (1H, d, J= 1.42 Hz); Mass (m/z): 480.2, 482.1 $[M+H]^+$.

3,6-Dichloro-1-(2-bromobenzenesulfonyl)-2-(4-methyl-piperazin-1-ylmethyl)-1H-indole (6e, R^1 = 6-Cl, R^2 = 2'-Br) : MR (^oC): 169.8 - 172.1; IR (KBr, cm^{-1}): 2934, 2794, 1452, 1366 (SO_2 stretching), 1174 (SO_2 stretching), 1007; ^{1}H-NMR (CD_3OD):δ 1.97 - 2.40 (11H, N-CH_3 and piperazinyl), 2.91 - 2.99 (1H, sept), 3.88 (2H, s, benzylic), 7.37 - 7.40 (1H, dd, J= 1.68, 8.43 Hz), 7.47 - 7.66 (4H, m), 7.83 - 7.85 (1H, d, J= 7.86 Hz), 8.03 - 8.04 (1H, d, J= 1.6 Hz); Mass (m/z): 516.2, 518.2 $[M+H]^+$.

3,6-Dichloro-1-(4-fluorobenzenesulfonyl)-2-(4-methyl-piperazin-1-ylmethyl)-1H-indole (6f R^1 = 6-Cl, R^2 = 4'- F) : MR (^oC): 226.4 - 228.1; IR (KBr, cm^{-1}): 2944, 2794, 1592, 1494, 1358 (SO_2 stretching), 1178 (SO_2 stretching), 1009; ^{1}H-NMR (CD_3OD): δ2.79 - 2.97 (11H, N-CH_3 and piperazinyl), 4.08 (2H, s, benzylic), 7.30 - 7.40 (3H, m), 7.53 - 7.55 (1H, d, J= 8.42 Hz), 8.05 – 8.10 (2H, m), 8.14 - 8.15 (1H, d, J = 1.42 Hz); Mass (m/z): 456, 458.1 $[M+H]^+$.

1-Benzenesulfonyl-3-chloro-2-(4-methyl-piperazin-1-ylmethyl)-1H-indole (6g, R^1 = 6-Cl, R^2 = H) : MR (^oC): 203.1 - 206.7; IR (KBr, cm^{-1}): 2960, 2787, 1592, 1366 (SO_2 stretching), 1171 (SO_2 stretching), 1098; ^{1}H-NMR (CD_3OD):δ 2.56 - 3.00 (11H, N-CH_3 and piperazinyl), 4.11 (2H, s, benzylic), 7.54 - 7.58 (3H, m), 7.81 - 7.87 (4H, m), 8.17 (1H, d); Mass (m/z): 438, 440.2 $[M+H]^+$.

3,*6-Dichloro-2-(4-methyl-piperazin-1-ylmethyl)-1-(toluene-4-sulfonyl)-1H-indole (6h, R^1 = 6-Cl, R^2 = 4'- CH_3) :* MR (^oC): 179.3 - 181.3; IR (KBr, cm^{-1}): 2939, 2797, 1599, 1452, 1382 (SO_2 stretching), 1179 (SO_2 stretching), 1006; ^{1}H-NMR (CD_3OD): δ 2.21 (3H, s, N-CH_3), 2.20 - 2.26 (4H, bs, piperazinyl), 2.37 (3H, s, -CH_3), 2.56 (4H, piperazinyl), 3.95 (2H, s, benzylic), 7.32 - 7.35 (3H, m), 7.49 - 7.51 (1H, d, J= 8.39 Hz), 7.92 - 7.94 (2H, dd, J= 8.42 Hz), 8.13 - 8.14 (1H, d, J= 1.71 Hz); Mass (m/z): 452.1, 454 $[M+H]^+$.

3-Chloro-1-(4-fluoro-benzenesulfonyl)-2-(4-methyl-piperazin-1-ylmethyl)-1H-indole (6i, R^1 = H, R^2 = 4'- F) : Oily mass; IR (KBr, cm^{-1}): 2930, 2791, 1582, 1484, 1356 (SO_2 stretching), 1179 (SO_2 stretching); ^{1}H-NMR (CD_3OD): 2.71 - 2.99 (11H, N-CH_3 and piperazinyl), 4.06 (2H, s, benzylic), 7.30 - 7.40 (6H, m), 7.91 – 8.10 (2H, m); Mass (m/z): 422, 424.1 $[M+H]^+$.

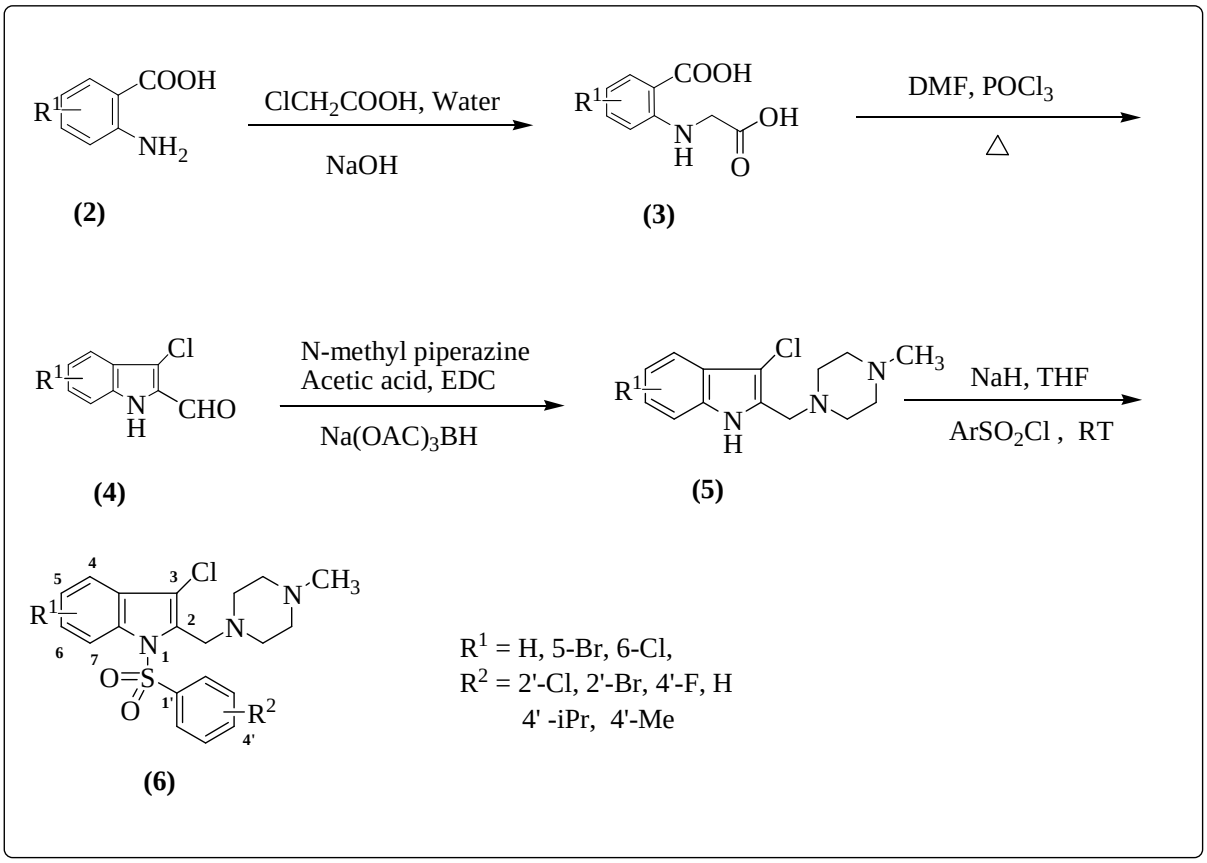
3-Chloro-2-(4-methyl-piperazin-1-ylmethyl)-1-(toluene-4-sulfonyl)-1H-indole (6j, R^1 = H, R^2 = 4'- CH_3) : Oily mass; IR (KBr, cm^{-1}): 2934, 2783, 1570, 1474, 1366 (SO_2 stretching), 1170 (SO_2 stretching); ^{1}H-NMR (CD_3OD): δ 2.23 (3H, s, N-CH_3), 2.25 - 2.30 (4H, bs, piperazinyl), 2.35 (3H, s, -CH_3), 2.58 (4H, piperazinyl), 3.93 (2H, s, benzylic), 7.29 - 7.38 (6H, m), 7.91 – 8.15 (2H, m); Mass (m/z): 418, 420.2 $[M+H]^+$.

RESULTS AND DISCUSSION

The general synthetic strategy used for the preparation of title **Compounds 6a-j** has been summarized in **Scheme 1**.

The commercially available substituted anthranilic acids were treated with chloro acetic acid in presence of aqueous sodium hydroxide at reflux temperature [5] to obtain the dicarboxylicacid derivatives **3**. These derivatives showed a broad absorption band at 1700 - 1720 cm^{-1} due to acid carbonyl group stretching and peak at 3351 cm^{-1} for the -OH stretching. The dicarboxylicacid derivatives **(3)** were further subjected to cyclization in presence of phosphorus oxychloride ($POCl_3$) and dimethyl formamide (DMF) at 90 ^{0}C [6].

The IR spectra of these derivatives show broad peak at ~1653 cm^{-1} due to the aldehyde carbonyl stretching, while peak at ~3273 cm^{-1} for indole NH confirmed the ring closer.



Scheme -1

The aldehyde derivatives **(4)** were further reacted with N-methyl piperazine under reductive amination condition to obtain intermediate **5**, which was confirmed by analytical data. The sulfonylation was achieved in presence of strong base, NaH and substituted arylsulfonyl chlorides in DMF solvent. All final compounds were characterized by IR, NMR and Mass spectral data.

5-HT_6 Receptor binding studies

The *in-vitro* 5-HT_6 receptor binding assay [7] was carried out on Human recombinant receptor expressed in HEK-293 cells; Radioligand used was [3H]LSD (60-80 Ci/mmol). Final ligand concentration was 1.5 nM, Non-specific Determinant was Methiothepin mesylate - [0.1 μM]; Reference Compound was Methiothepin mesylate, Positive Control was Methiothepin mesylate.

Table -1: 5-HT_6 receptor binding data

Compound No.	R^1	R^2	% Inhibition at 1 µM
6a	5- Br	4'-CH_3	46.21 %
6b	5- Br	2'- Br	48.30 %
6c	5- Br	2'- Cl	40.81 %
6d	6-Cl	4'-iPr	45.81 %
6e	6-Cl	2'- Br	39.80 %
6f	6-Cl	4'-F	61.15 %
6g	6-Cl	H	85.86 %
6h	6-Cl	4'-CH_3	79.12 %
6i	H	4'-F	62.30 %
6j	H	4'-CH_3	46.67 %

All the synthesized compounds were screened for their % inhibition towards the human 5-HT_6 receptor at 1µM concentration. All derivatives have shown improvement in the binding affinity towards the 5-HT_6 receptor as compared to the parent series of compounds. Compounds **6g** and **6h** showed the highest inhibition of 85.86 % and 79.12 % respectively.

CONCLUSION

In summary, we successfully synthesized a new series of 3-Chloro-2-piperazinylmethyl-N-aryl sulfonamide indole derivatives using the Vilsmerier reaction. By comparing the *in-vitro* data with the parent series, we came to the conclusion that insertion of halo groups at 3rd position of indole nucleus is not only advantageous but also improves the binding affinity towards the 5-HT_6 receptor. The detailed profiling of these derivatives is under progress.

Acknowledgments
The authors wish to acknowledge the support received from Mr. Venkateswarlu Jasti, CEO, Suven Life Sciences Ltd., Hyderabad, India.

REFERENCES

[1] W. J.Geldenhuys, C.V. Schyf, *Exper. Rev. Neurother.*, **2009**, 9, 1073.
[2] J. Holenz, P. J. Pauwels, J. L.Diaz, R. Merce, X. Codony, *Drug Disc. Today*, **2006,** 11, 283.
[3] S. L. Davies, J. S. Silvestre, X. Guitart, *Drugs of Future*, **2005**, 30, 479.
[4] J. G. Werner, J. V. Cornelis, *Curr. Top. in Med. Chem.*, **2008**, 8, 1035.
[5] Su, H.C.F. Tsou, K.C. *J. Am. Chem. Soc.*, **1960,** 82, 1187.
[6] P. T. Perumal, V. J. Majo, *J. Org. Chem.*, **1996**, 61, 6523.
[7] Assays performed at Novascreen Biosciences Corporation, Hanover, Maryland, U.S.A.