Section I

Chapter 5: Experimental

5. Experimental

5.1 Chemistry

Melting points were measured using VEEGO multi-programmable melting point apparatus and are uncorrected. IR spectra were recorded on FT-IR system-2000 Bruker spectrometer on KBr pellets or neat samples. ^{1}H-NMR spectra were recorded on Bruker Avance II 400 MHz FT-NMR spectrometer in $CDCl_3$ solvent unless stated. Chemical shifts are expressed in δ units relative to tetramethylsilane (TMS) signal as internal reference. The following abbreviations are used in reporting NMR data: s, singlet; bs, broad singlet; b, broad; d, doublet; t, triplet; q, quartet; dq, doublet of quartet; dd, doublet of doublet and m, mutiplet. Mass spectra were recorded on Thermo Scientific DSQ-II Mass analyzer. Elemental analyses were performed on ThermoFisher FLASH 2000 organic elemental analyzer.

The chemical work carried out has been discussed under the following heads:

5.1.1 Synthesis of 3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one derivatives

5.1.2 Synthesis of 3-ethyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one derivatives

5.1.3 Synthesis of 1-imino derivatives of 3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one

5.1.1 Synthesis of 3-Methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one derivatives (4-27)

5.1.1.1 *N*-methyl-2-phenylethanamine (1)

Methylamine (120 ml, 1.435 mol, 40% aqueous solution) and THF (45ml) were taken in a reaction vessel and 2-phenylethylbromide (10.0 gm, 54.04 mmol) in THF (75 ml) in portions was added to it at room temperature. The reaction mixture was stirred for 12 hr and completion of the reaction was monitored by TLC. The intermediate (**1**) was extracted in chloroform and concentrated to afford yellow oil (6.28 ml, 86 %). The intermediate was used in form of chloroform solution for further reaction.

Anal.:

TLC : R_f 0.45 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 3422, 3064, 2933, 1564, 1491, 747 and 699.

5.1.1.2 2-Chloro-*N*-methyl-*N*-phenethylacetamide (2)

N-Methyl-2-phenylethylamine (10.0 gm, 74.07 mmol) in chloroform (15 ml) was taken in a reaction vessel and aqueous solution of K_2CO_3 (30.67 gm, 222.22 mmol) was added to it. The reaction mixture was stirred at 0 °C for 15 min. 2-Chloroacetyl chloride (8.84 ml, 111.11 mmol) diluted in $CHCl_3$ was added in to the reaction mixture drop wise over a period of 30 min, while maintaining the temperature at 0 ^oC. Completion of the reaction was monitored by TLC. After completion, the organic layer was separated, dried over sodium sulphate and the solvent distilled out to get a product (**2**) (15.1 gm, 97 %) as yellow oil.

TLC : R_f 0.40 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 3061, 2937, 1652, 1492, 749 and 703.

5.1.1.3 3-Methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (3)

A solution of 2-chloro-*N*-methyl-*N*-phenethylacetamide (5.4 gm, 25.38 mmol) in 1,2-DCB was taken in a reaction vessel and anhydrous $AlCl_3$ powder (5.0 gm, 38.06 mmol) was added to it in portions. After stirring at rt for 15 minutes the reaction mixture was heated at 165 °C, while monitoring the reaction with TLC for completion of the reaction. After completion, the reaction mixture was cooled at 0^oC and quenched with dil. HCl. The mixture was extracted with DCM (10ml x 3) and washed with sodium bicarbonate solution. From the combined organic extracts, solvent was removed to afford a crude product which was purified by column chromatography using silica gel as stationary phase and hexane: ethyl acetate as eluent to yield a white solid product (2.4 gm, 55%). m.p.: 105 - 107 °C

Anal.:

TLC : R_f 0.25 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 3010, 1645 and 744.

^{1}H-NMR : 7.26 (d,1H, Ar-*H*), 7.19-7.12 (m, 2H, Ar-*H*), 7.10 (d, 1H, Ar-*H*), 3.90

(s, 2H, Ar-*CH*$_2$-CO), 3.72-3.69 (m, 2H, Ar-CH$_2$-*CH*$_2$-N), 3.15-3.12 (m, 2H, Ar-*CH*$_2$-CH$_2$-N) and 3.02 (s, 3H, N-*CH*$_3$).

MS (m/z) : 175 $(M)^+$

5.1.1.4 3-Methyl-4,5-dihydro-3*H*-benzo[*d*]azepine-1,2-dione (4)

An oven-dried flask was charged with 3-methyltetrahydro-3*H*-benzazepin-2-one (1.0 gm, 5.71 mmol), evacuated and backfilled with nitrogen. Dioxane (10 ml) was added to dissolve the solid and selenium oxide (SeO_2) (1.127 gm, 11.42 mmol) was added to the solution. The reaction mixture was refluxed for 6-8 hr with stirring and completion of the reaction was monitored by TLC. When the reaction got completed, the volatiles were evaporated. To the resulting mixture, ether (20 ml) and H_2O (30 ml) were added. The aqueous layer was separated and additionally extracted with ether (10x2 ml). Organic fractions were combined, dried over $MgSO_4$, volatiles removed and the residue so obtained was purified by column chromatography on silica gel using hexane/ether (2:1) solvent system, to obtain the compound (**4**), (1.0 gm, 88 %), as a colourless solid. m. p.: 118-120 °C

Anal.:

TLC : R_f 0.20 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 1690, 1657, 1483, 1418, 1398, 1228 and 749

^{1}H-NMR : 7.76 (d, 1H, Ar-*H*), 7.52-7.48 (m, 1H, Ar-*H*), 7.36-7.33 (m, 1H, Ar-*H*), 7.27 (d, 1H, Ar-*H*), 3.74-3.72 (m, 2H, $C_6H_4CH_2C\mathit{H}_2N$), 3.26-3.24 (m, 2H, $C_6H_4C\mathit{H}_2CH_2N$) and 3.13 (s, 3H, $NC\mathit{H}_3$)

MS (m/z) : 189.44 (M^+)

5.1.1.5 1-Hydroxy-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (5)

Methanolic (15 ml) solution of 3-methyl-4,5-dihydro-3*H*-benzo[*d*]azepine-1,2-dione (**4**) (1.0 gm, 5.29 mmol) was taken in a reaction vessel and was added sodium borohydride (0.4 gm, 10.58 mmol) in portions with continuous stirring at room temperature. After completion of the reaction, excess solvent from the reaction mixture was removed and the slurry was poured into the ice-cold water. The aqueous layer was

extracted with diethyl ether (10x2 ml). The organic fractions were combined, dried over $MgSO_4$, volatiles removed and the residue so obtained was purified by column chromatography on silica gel using hexane/ether (2:1) solvent system, to obtain compound (**5**), (0.7 gm, 67 %), as a white solid m. p.: 97-99 °C

Anal.:

TLC : R_f 0.47 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 3424, 1664, 1483, 1418, 1398, 1228 and 749

^{1}H-NMR : 7.77 (d, 1H, Ar-*H*), 7.28-7.18 (m, 2H, Ar-*H*), 7.10 (d, 1H, Ar-*H*), 5.71 (d, 1H, CH-O*H*, *J* = 5.1 Hz), 4.55 (d, 1H, C*H*-OH, *J* = 5.1 Hz), 4.05-3.99 (m, 1H, $C_6H_4CH_2CH_2N$), 3.33-3.06 (m, 3H, $C_6H_4CH_2CH_2N$, $C_6H_4CH_2CH_2N$) and 3.04 (s, 3H, NCH_3)

MS (m/z) : 191.31 (M^+).

5.1.1.6 3-Methyl-2-oxo-2,3,4,5-tetrahydro-1*H*-benzo[*d*]azepin-1-yl methanesulfonate (6)

An oven-dried flask was charged with 1-hydroxy-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**5**) (1.0 gm, 5.23 mmol) in dichloromethane (DCM) (20 ml), and the solution was flushed with nitrogen. To this solution, triethylamine (1.05 gm, 10.46 mmol) and methanesulfonyl chloride (0.9 gm, 7.85 mmol) was added drop-wise at 0 °C. The reaction mixture was refluxed for 6-8 hr and completion of the reaction was monitored by TLC. After completion of the reaction, the reaction mixture was washed with water (20x3 ml) and the combined aqueous layer was extracted with 20 ml methylene dichloride (MDC). The organic fractions were combined, dried over $MgSO_4$, volatiles removed and the residue so obtained was purified by column chromatography on silica gel using hexane/Ether (2:1) solvent system, to obtain compound (**6**), (1.1 gm, 74 %) as a colourless solid. m. p.: 81-83 °C

Anal.:

TLC : R_f 0.47 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 1657, 1488, 1440, 1397, 1235, 1112 and 752

MS (m/z) : 269.31 (M^+).

5.1.1.7 Synthesis of 1-amino substituted 3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one derivatives (7-24)

3-Methyl-1-propylamino-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (7)

A THF (10 ml) solution of 3-methyl-2-oxo-2,3,4,5-tetrahydro-1*H*-benzo[*d*]azepin-1-yl methanesulfonate (**6**) (0.5 gm, 1.86 mmol) was taken under nitrogen environment to the reaction vessel. Anhydrous K_2CO_3 (0.385 gm, 2.79 mmol) and *n*-propylamine (0.165 gm, 2.79 mmol) were added and the reaction mixture was stirred for 6-8 hr at room temperature. Completion of the reaction was monitored by TLC. After completion, the reaction mixture was poured into ice-cold water and extracted successively with ethyl acetate (10x3 ml). The organic fractions were combined, dried over $MgSO_4$, volatiles removed and the residue so obtained was purified by column chromatography on silica gel using hexane/ether (2:1) solvent system, to obtain compound (**7**)**,** (0.3 gm, 70 %) as white solid. m. p.: 55-57 °C

Anal.:

TLC : R_f 0.49 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 3129, 1657, 1480, 1399, 1228 and 748

^{1}H-NMR : 7.68 (d, 1H, Ar-*H*), 7.23-7.15 (m, 2H, Ar-*H*), 7.09 (d, 1H, Ar-*H*), 4.88 (s, 1H, C*H*NHCH$_2$), 4.09-4.02 (m, 1H, $C_6H_4CH_2CH_2N$), 3.59-3.53 (m, 1H, $C_6H_4CH_2CH_2N$), 3.28-3.06 (m, 2H, $C_6H_4CH_2CH_2N$), 3.01 (s, 3H, NCH_3), 2.73-2.67 (m, 1H, NHCH_2CH$_2$CH$_3$), 2.56-2.50 (m, 1H, NHCH_2CH$_2$CH$_3$), 1.97 (bs, 1H, N*H*CH$_2$CH$_2$CH$_3$), 1.64-1.55 (m, 2H, NHCH$_2$CH_2CH$_3$) and 0.97 (t, 3H, NHCH$_2$CH$_2$CH_3)

MS (m/z) : 231.22 (M^+-1)

1-Isobutylamino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (8)

The compound 1-isobutylamino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**8**) was synthesized as per the method described for compound (**7**) by replacing *n*-propylamine with isobutylamine (0.204 gm, 2.79 mmol) to obtain compound (**8**), (0.3 gm, 66 %) as white solid. m. p.: 52-54 °C

Anal.:

TLC : R_f 0.48 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 3130, 1658, 1466, 1399, 1172 and 746

^{1}H-NMR : 7.71 (d, 1H, Ar-*H*), 7.23-7.15 (m, 2H, Ar-*H*), 7.09 (d, 1H, Ar-*H*), 4.84 (s, 1H, C*H*NHCH$_2$), 4.06-3.99 (m, 1H, $C_6H_4CH_2CH_2N$), 3.61-3.58 (m, 1H, $C_6H_4CH_2CH_2N$), 3.23-3.06 (m, 2H, $C_6H_4CH_2CH_2N$), 3.01 (s, 3H, NCH_3), 2.58-2.54 (m, 1H, NHCH_2CH(CH$_3$)$_2$), 2.38-2.28 (m, 1H, NHCH_2CH(CH$_3$)$_2$), 2.17 (bs, 1H, N*H*CH$_2$CH(CH$_3$)$_2$), 1.85-1.79 (m, 1H, NHCH$_2$C*H*(CH$_3$)$_2$) and 0.98 (dd, 6H, NHCH$_2$CH(CH_3)$_2$)

MS (m/z) : 246.4 (M^+)

1-Butylamino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (9)

The compound 1-butylamino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**9**) was synthesized as per the method described for compound (**7**) by replacing *n*-propylamine with *n*-butylamine (0.204 gm, 2.79 mmol) to obtain compound (**9**), (0.3 gm, 66 %) as yellow oil.

Anal.:

TLC : R_f 0.50 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 3416, 1658, 1478, 1399, 1172 and 746

^{1}H-NMR : 7.67 (d, 1H, Ar-*H*), 7.26-7.15 (m, 2H, Ar-*H*), 7.09 (d, 1H, Ar-*H*), 4.87 (s, 1H, C*H*NHCH$_2$), 4.09-4.01 (m, 1H, $C_6H_4CH_2CH_2N$), 3.60-3.53 (m, 1H, $C_6H_4CH_2CH_2N$), 3.28-3.06 (m, 2H, $C_6H_4CH_2CH_2N$), 3.01 (s, 3H, NCH_3), 2.75-2.69 (m, 1H, NHCH_2CH$_2$CH$_2$CH$_3$), 2.57-2.53 (m, 1H, NHCH_2CH$_2$CH$_2$CH$_3$), 2.03 (bs, 1H, N*H*CH$_2$CH$_2$CH$_2$CH$_3$), 1.60-1.55 (m, 2H, NHCH$_2$CH_2CH$_2$CH$_3$), 1.46-1.39 (m, 2H, NHCH$_2$CH$_2$CH_2CH$_3$) and 0.93 (t, 3H, NHCH$_2$CH$_2$CH$_2$CH_3)

MS (m/z) : 246.55 (M^+)

1-Cyclopropylamino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (10)

The compound 1-cyclopropylamino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**10**) was synthesized as per the method described for compound (**7**) by replacing *n*-propylamine with cyclopropylamine (0.159 gm, 2.79 mmol) to obtain compound (**10**), (0.4 gm, 84 %) as white solid. m. p.: 113-115 °C

Anal.:

TLC : R_f 0.43 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 3298, 1648, 1472, 1439, 1397, 1175 and 754

^{1}H-NMR : 7.59 (dd, 1H, Ar-*H*), 7.19-7.13 (m, 2H, Ar-*H*), 7.09 (d, 1H, Ar-*H*), 5.05 (s, 1H, C*H*NHCH$_2$), 4.20-4.12 (m, 1H, $C_6H_4CH_2CH_2N$), 3.51-3.45 (m, 1H, $C_6H_4CH_2CH_2N$), 3.22-3.15 (m, 2H, $C_6H_4CH_2CH_2N$), 3.03 (s, 3H, NCH_3), 2.86 (bs, 1H, CHN*H*CyPr), 2.31-2.27 (m, 1H, NHC*H*(CH$_2$)$_2$) 0.57-0.47 (m, 2H, NHCH(CH_2)$_2$) and 0.45-0.30 (m, 2H, NHCH(CH_2)$_2$)

MS (m/z) : 230.7 (M^+)

3-Methyl-1-phenylamino-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (11)

The compound 3-methyl-1-phenylamino-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**11**) was synthesized as per the method described for compound (**7**) by replacing *n*-propylamine with aniline (0.26 gm, 2.79 mmol) to obtain compound (**11**) (0.3 gm, 53 %) as white solid. m. p.: 206-208 °C

Anal.:

TLC : R_f 0.44 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 3362, 1660, 1443, 1400, 1176, 751 and 693

^{1}H-NMR : 7.51 (d, 1H, Ar-*H*), 7.25-7.11 (m, 5H, Ar-*H*), 6.69 (t, 1H, Ar-*H*), 6.53

(d, 2H, Ar-*H*), 5.67 (bs, 1H, CHN*H*C_6H_5), 5.55 (d, 1H, C*H*NHC$_6H_5$), 4.24-4.18 (m, 1H, $C_6H_4CH_2CH_2$N), 3.43-3.36 (m, 2H, $C_6H_4CH_2CH_2$N, $C_6H_4CH_2CH_2$N), 3.23-3.17 (m, 1H, $C_6H_4CH_2CH_2$N) and 3.05 (s, 3H, NCH_3)

MS (m/z) : 266.41 (M^+)

3-Methyl-1-(pyridin-2-ylamino)-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (12)

The compound 3-methyl-1-(pyridin-2-ylamino)-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**12**) was synthesized as per the method described for compound (**7**) by replacing *n*-propylamine with 2-aminopyridine (0.26 gm, 2.79 mmol) to obtain compound (**12**) (0.2 gm, 34 %) as yellow oil.

Anal.:

TLC : R_f 0.42 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 3486, 2930, 1640, 1601 and 747

^{1}H-NMR : 8.55-8.53 (d, 1H, N*H*), 7.78-7.09 (m, 8H, Ar-*H*), 5.29 (d, 1H, C*H*NHPy), 4.18-4.14 (m, 1H, $C_6H_4CH_2CH_2$N), 4.01-3.92 (m, 1H, $C_6H_4CH_2CH_2$N), 3.19-3.18 (m, 2H, $C_6H_4CH_2CH_2$N) and 3.01 (s, 3H, NCH_3)

MS (m/z) : 267.3 (M^+)

1-Benzylamino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (13)

The compound 1-benzylamino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**13**) was synthesized as per the method described for compound (**7**) by replacing *n*-propylamine with benzylamine (0.3 gm, 2.79 mmol) to obtain compound (**13**) (0.3 gm, 63 %) as white solid. m. p.: 68-70 °C

Anal.:

TLC : R_f 0.45 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 3305, 1666, 1490, 1451, 1399, 1172, 749 and 700

^{1}H-NMR : 7.80 (d, 1H, Ar-*H*), 7.41 (d, 2H, Ar-*H*), 7.33-7.15 (m, 5H, Ar-*H*),

7.08 (d, 1H, Ar-*H*), 4.93 (s, 1H, C*H*NHCH$_2$), 4.04-3.96 (m, 2H, C$_6$H$_4$CH$_2$C*H*$_2$N, NHC*H*$_2$Ar), 3.77 (d, 1H, NHC*H*$_2$Ar), 3.51-3.48 (m, 1H, C$_6$H$_4$CH$_2$C*H*$_2$N), 3.17-3.12 (m, 2H, C$_6$H$_4$C*H*$_2$CH$_2$N), 3.03 (s, 3H, NC*H*$_3$) and 2.65 (bs, 1H, N*H*CH$_2$Ar)

MS (m/z) : 278.81 (M^+-1)

3-Methyl-1-(4-methylbenzyl)amino-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (14)

The compound 3-methyl-1-(4-methylbenzyl)amino-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**14**) was synthesized as per the method described for compound (**7**) by replacing *n*-propylamine with 4-methylbenzylamine (0.338 gm, 2.79 mmol) to obtain compound (**14**) (0.3 gm, 59 %) as white solid. m. p.: 101-103 °C

Anal.:

TLC : R_f 0.41 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 3305, 1657, 1487, 1448, 1396, 1175 and 755

^{1}H-NMR : 7.68 (d, 1H, Ar-*H*), 7.22 (d, 2H, Ar-*H*, *J* = 7.8 Hz), 7.18-7.06 (m, 2H, Ar-*H*), 7.05 (d, 2H, Ar-*H*, *J* = 7.8 Hz), 7.00 (d, 1H, Ar-*H*), 4.88 (s, 1H, C*H*NHCH$_2$Ar), 3.95-3.89 (m, 1H, C$_6$H$_4$CH$_2$C*H*$_2$N), 3.86 (d, 1H, CHNHC*H*$_2$Ar, *J* = 13.0 Hz), 3.67 (d, 1H, CHNHC*H*$_2$Ar, *J* = 13.0 Hz), 3.46-3.39 (m, 1H, C$_6$H$_4$CH$_2$C*H*$_2$N), 3.24 (bs, 1H, CHN*H*CH$_2$Ar), 3.13-3.04 (m, 2H, C$_6$H$_4$C*H*$_2$CH$_2$N), 2.94 (s, 3H, NC*H*$_3$) and 2.25 (s, 3H, ArC*H*$_3$)

MS (m/z) : 293.42 (M^+-1)

1-(4-Methoxybenzyl)amino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (15)

The compound 1-(4-methoxybenzyl)amino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**15**) was synthesized as per the method described for compound (**7**) by replacing *n*-propylamine with 4-methoxybenzylamine (0.382 gm, 2.79 mmol) to obtain compound (**15**) (0.3 gm, 59 %) as white solid. m. p.: 63-65 °C

Anal.:

TLC : R_f 0.41 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 3415, 1655, 1400, 1245 and 753

^{1}H-NMR : 7.77 (d, 1H, Ar-*H*), 7.33 (d, 2H, Ar-*H*, *J* = 8.6 Hz), 7.25-7.15 (m, 2H, Ar-*H*), 7.08 (d, 1H, Ar-*H*), 6.86 (d, 2H, Ar-*H*, *J* = 8.6 Hz), 4.92 (s, 1H, C*H*NHCH$_2$), 4.04-3.96 (m, 1H, $C_6H_4CH_2CH_2N$), 3.90 (d, 1H, NHC*H*$_2$Ar, *J* = 12.8 Hz), 3.79 (s, 3H, OC*H*$_3$), 3.70 (d, 1H, NHC*H*$_2$Ar, *J* = 12.8 Hz), 3.52-3.48 (m, 1H, $C_6H_4CH_2CH_2N$), 3.17-3.12 (m, 2H, $C_6H_4CH_2CH_2N$), 3.02 (s, 3H, NC*H*$_3$) and 2.20 (bs, 1H, N*H*CH$_2$Ar)

MS (m/z) : 307.65 (M^+-2)

3-Methyl-1-(4-trifluoromethylbenzyl)amino-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (16)

The compound 3-methyl-1-(4-trifluoromethylbenzyl)amino-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**16**) was synthesized as per the method described for compound (**7**) by replacing *n*-propylamine with 4-trifluoromethylbenzylamine (0.49 gm, 2.79 mmol) to obtain compound (**16**) (0.4 gm, 62 %) as yellow oil.

Anal.:

TLC : R_f 0.39 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 3197, 1653, 1489, 1449, 1401, 1124 and 748

^{1}H-NMR : 7.51 (d, 1H, Ar-*H*), 7.43-7.39 (m, 3H, Ar-*H*), 7.27-7.18 (m, 3H, Ar-*H*), 7.14 (d, 1H, Ar-*H*), 5.04 (s, 1H, C*H*NHCH$_2$), 4.37-3.30 (m, 1H, $C_6H_4CH_2CH_2N$), 3.53-3.47 (m, 1H, $C_6H_4CH_2CH_2N$), 3.50 (s, 2H, CHNHC*H*$_2$Ar), 3.17-3.12 (m, 2H, $C_6H_4CH_2CH_2N$) and 3.05 (s, 3H, NC*H*$_3$)

MS (m/z) : 349.64 (M^++1)

1-(4-Fluorobenzyl)amino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (17)

The compound 1-(4-fluorobenzyl)amino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**17**) was synthesized as per the method described for compound (**7**) by replacing *n*-propylamine with 4-Fluorobenzylamine (0.35 gm, 2.79 mmol) to obtain compound (**17**) (0.4 gm, 69 %) as white solid. m. p.: 103-105 °C

Anal.:

TLC : R_f 0.37 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 3302, 1658, 1483, 1448, 1399, 1151, 1091, 748 and 686

^{1}H-NMR : 7.78 (d, 1H, Ar-*H*), 7.39-7.36 (m, 2H, Ar-*H*), 7.24-7.16 (m, 2H, Ar-*H*), 7.09 (d, 1H, Ar-*H*), 7.02-6.97 (m, 2H, Ar-*H*), 4.91 (s, 1H, C*H*NHCH$_2$), 4.05-3.98 (m, 1H, $C_6H_4CH_2CH_2N$), 3.94 (d, 1H, NHC*H*$_2$Ar, *J* = 13.1 Hz), 3.72 (d, 1H, NHC*H*$_2$Ar, *J* = 13.1 Hz), 3.50-3.46 (m, 1H, $C_6H_4CH_2CH_2N$), 3.18-3.13 (m, 2H, $C_6H_4CH_2CH_2N$), 3.03 (s, 3H, NC*H*$_3$) and 2.61 (bs, 1H, N*H*CH$_2$Ar)

MS (m/z) : 299.83 (M^+-2)

1-(4-Chlorobenzyl)amino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (18)

The compound 1-(4-chlorobenzyl)amino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**18**) was synthesized as per the method described for compound (**7**) by replacing *n*-propylamine with 4-chlorobenzylamine (0.39 gm, 2.79 mmol) to obtain compound (**18**) (0.4 gm, 62 %) as white solid. m. p.: 106-108 °C

Anal.:

TLC : R_f 0.39 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 3302, 1659, 1487, 1448, 1399, 1172 and 746

^{1}H-NMR : 7.78 (d, 1H, Ar-*H*), 7.35 (d, 2H, Ar-*H*, *J* = 8.4 Hz), 7.28 (d, 2H, Ar-*H*, *J* = 8.4 Hz), 7.23-7.16 (m, 2H, Ar-*H*), 7.09 (d, 2H, Ar-*H*), 4.89 (s, 1H, C*H*NHCH$_2$), 4.04-3.88 (m, 1H, $C_6H_4CH_2CH_2N$), 3.96 (d, 1H, NHC*H*$_2$Ar, *J* = 13.5 Hz), 3.72 (d, 1H, NHC*H*$_2$Ar, *J* = 13.5

Hz), 3.49-3.45 (m, 1H, $C_6H_4CH_2CH_2N$), 3.18-3.13 (m, 2H, $C_6H_4CH_2CH_2N$), 3.03 (s, 3H, NCH_3) and 1.59 (bs, 1H, $NHCH_2Ar$)

MS (m/z) : 314.28 (M^+)

1-(3-Chlorobenzyl)amino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (19)

The compound 1-(3-chlorobenzyl)amino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**19**) was synthesized as per the method described for compound (**7**) by replacing *n*-propylamine with 3-chlorobenzylamine (0.39 gm, 2.79 mmol) to obtain compound (**19**) (0.4 gm, 60 %) as yellow oil.

Anal.:

TLC : R_f 0.40 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 3129, 1659, 1481, 1427, 1400, 1172, 747 and 684

^{1}H-NMR : 7.79 (d, 1H, Ar-*H*), 7.42 (s, 1H, Ar-*H*), 7.31-7.17 (m, 5H, Ar-*H*), 7.09

(d, 1H, Ar-*H*), 4.91 (s, 1H, $CHNHCH_2$), 4.06-4.00 (m, 1H, $C_6H_4CH_2CH_2N$), 3.98 (d, 1H, $NHCH_2Ar$, J = 13.5 Hz), 3.74 (d, 1H, $NHCH_2Ar$, J = 13.5 Hz), 3.51-3.44 (m, 1H, $C_6H_4CH_2CH_2N$), 3.19-3.11 (m, 2H, $C_6H_4CH_2CH_2N$) and 3.06 (s, 3H, NCH_3)

MS (m/z) : 315.67 (M^++1)

1-(2-Chlorobenzyl)amino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (20)

The compound 1-(2-chlorobenzyl)amino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**20**) was synthesized as per the method described for compound (**7**) by replacing *n*-propylamine with 2-chlorobenzylamine (0.39 gm, 2.79 mmol) to obtain compound (**20**) (0.3 gm, 53 %) as white solid. m. p.: 102-104 °C

Anal.:

TLC : R_f 0.41 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 3129, 1657, 1479, 1426, 1400, 1199 and 746

^{1}H-NMR : 7.79 (d, 1H, Ar-*H*), 7.42-7.17 (m, 6H, Ar-*H*), 7.09 (d, 1H, Ar-*H*),

4.91 (s, 1H, C*H*NHCH$_2$), 4.06-4.00 (m, 1H, C$_6$H$_4$CH$_2$C*H*$_2$N), 3.97 (d, 1H, NHC*H*$_2$Ar, *J* = 13.5 Hz), 3.73 (d, 1H, NHC*H*$_2$Ar, *J* = 13.5 Hz), 3.50-3.44 (m, 1H, C$_6$H$_4$CH$_2$C*H*$_2$N), 3.18-3.11 (m, 2H, C$_6$H$_4$C*H*$_2$CH$_2$N) and 3.06 (s, 3H, NC*H*$_3$)

MS (m/z) : 314.89 (M^+)

1-(3,4-Dimethoxybenzyl)amino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (21)

The compound 1-(3,4-dimethoxybenzyl)amino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**21**) was synthesized as per the method described for compound (**7**) by replacing *n*-propylamine with 3,4-dimethoxybenzylamine (0.47 gm, 2.79 mmol) to obtain compound (**21**) (0.4 gm, 57 %) as yellow oil.

Anal.:

TLC : R_f 0.34 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 3128, 1657, 1461, 1399, 1138 and 761

^{1}H-NMR : 7.80 (d, 1H, Ar-*H*), 7.25-7.16 (m, 2H, Ar-*H*), 7.09 (d, 1H, Ar-*H*), 7.20 (s, 1H, Ar-*H*), 6.92 (d, 1H, Ar-*H*, *J* = 8.1 Hz), 6.81 (d, 1H, Ar-*H*, *J* = 8.1 Hz), 4.94 (s, 1H, C*H*NHCH$_2$), 4.04-3.98 (m, 1H, C$_6$H$_4$CH$_2$C*H*$_2$N), 3.95 (d, 1H, NHC*H*$_2$Ar, *J* = 13.0 Hz), 3.86 (s, 6H, OC*H*$_3$), 3.76 (d, 1H, NHC*H*$_2$Ar, *J* = 13.0 Hz), 3.48-3.44 (m, 1H, C$_6$H$_4$CH$_2$C*H*$_2$N), 3.17-3.12 (m, 2H, C$_6$H$_4$C*H*$_2$CH$_2$N) and 3.02 (s, 3H, NC*H*$_3$)

MS (m/z) : 340.18 (M^+)

3-Methyl-1-(pyridin-2-yl)methylamino-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (22)

The compound 3-methyl-1(pyridin-2-yl)-methylamino-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**22**) was synthesized as per the method described for compound (**7**) by replacing *n*-propylamine with 2-amino methyl pyridine (0.3 gm, 2.79 mmol) to obtain compound (**22**) (0.3 gm, 58 %) as yellow oil.

Anal.:

TLC : R_f 0.45 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 3309, 2924, 1654, 1433, 1399 and 750

^{1}H-NMR : 8.46-8.45 (m, 1H, Ar-*H*), 7.69 (d, 1H, Ar-*H*), 7.58-7.54 (m, 1H, Ar-*H*), 7.42 (d, 1H, Ar-*H*), 7.19-6.99 (m, 4H, Ar-*H*), 4.89 (s, 1H, C*H*NHCH$_2$), 4.04 (d, 1H, NHC*H*$_2$Ar, *J* = 13.0 Hz), 3.93-3.87 (m, 1H, $C_6H_4CH_2CH_2N$), 3.84 (d, 1H, NHC*H*$_2$Ar, *J* = 13.0 Hz), 3.70 (bs, 1H, N*H*), 3.52-3.46 (m, 1H, $C_6H_4CH_2CH_2N$), 3.14-2.98 (m, 2H, $C_6H_4CH_2CH_2N$) and 2.93 (s, 3H, NC*H*$_3$)

MS (m/z) : 281.8 (M^+)

3-Methyl-1-phenethylamino-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (23)

The compound 3-methyl-1-phenethylamino-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**23**) was synthesized as per the method described for compound (**7**) by replacing *n*-propylamine with 2-phenethylamine (0.34 gm, 2.79 mmol) to obtain compound (**23**) (0.3 gm, 60 %) as yellow oil.

Anal.:

TLC : R_f 0.47 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 3416, 1656, 1480, 1453, 1393, 1138, 754 and 699

^{1}H-NMR : 7.63-7.61 (d, 1H, Ar-*H*), 7.30-7.13 (m, 7H, Ar-*H*), 7.09-7.07 (m, 1H, Ar-*H*), 4.86 (s, 1H, C*H*NHCH$_2$), 3.24-3.05 (m, 4H, $C_6H_4CH_2CH_2N$, $C_6H_4CH_2CH_2N$), 3.02 (s, 3H, NC*H*$_3$), 2.99.2.79 (m, 4H, NHC*H*$_2$C*H*$_2$Ar) and 1.85 (bs, 1H, N*H*CH$_2$CH$_2$Ar)

MS (m/z) : 290.61 (M^+-2)

1-(4-Chlorophenethyl)amino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (24)

The compound 1-(4-chlorophenethyl)amino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**24**) was synthesized as per the method described for compound (**7**) by replacing *n*-propylamine with 2-(4-chlorophenyl)ethylamine (0.435 gm, 2.79 mmol) to obtain compound (**24**) (0.3 gm, 56 %) as yellow oil.

Anal.:

TLC : R_f 0.45 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 3302, 1661, 1486, 1425, 1398, 1171 and 759

^{1}H-NMR : 7.61 (d, 1H, Ar-*H*), 7.25-7.15 (m, 6H, Ar-*H*), 7.08 (d, 1H, Ar-*H*), 4.88 (s, 1H, C*H*NHCH$_2$CH$_2$), 4.00-3.94 (m, 1H, $C_6H_4CH_2CH_2N$), 3.51-3.47 (m, 1H, $C_6H_4CH_2CH_2N$), 3.21-3.10 (m, 2H, $C_6H_4CH_2CH_2N$), 2.99 (s, 3H, NCH_3) and 2.91-2.80 (m, 4H, NHCH_2CH_2Ar)

MS (m/z) : 330.87 (M^++2)

5.1.1.8 Synthesis of 1-oxy substituted 3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one derivatives (25-27)

3-Methyl-1-phenoxy-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (25)

A methanolic solution (10 ml) of 3-methyl-2-oxo-2,3,4,5-tetrahydro-1*H*-benzo[*d*]azepin-1-yl methanesulfonate (**6**) (0.5 gm, 1.86 mmol) was taken in a reaction vessel under nitrogen environment and was added with sodium methoxide (0.15 gm, 2.79 mmol). The reaction mixture was stirred for 6-8 hr at room temperature and the progress of the reaction was monitored by TLC. After completion, the reaction mixture was poured into ice-cold water and extracted successively with ethyl acetate (10x3 ml). The organic fractions were combined, dried over $MgSO_4$, volatiles removed and the residue so obtained was purified by column chromatography on silica gel using hexane/ether (2:1) solvent system, to obtain compound (**25**)**,** (0.2 gm, 40 %) as a semisolid mass.

Anal.:

TLC : R_f 0.33 (Ethyl acetate: Hexane) (1:1)

IR (cm^{-1}) : 2929, 1654, 1489, 1083 and 749

^{1}H-NMR : 7.42 (d, 1H, Ar*H*), 7.43-7.13 (m, 2H, Ar-*H*), 7.14 (d, 1H, Ar*H*), 5.01

(s, 1H, C*H*), 4.37-4.30 (m, 1H, NCH_2CH$_2$), 3.53-3.47 (m, 1H, NCH_2CH$_2$), 3.50 (s, 3H, OCH_3), 3.17-3.12 (m, 2H, NCH$_2$CH_2) and 3.05 (s, 3H, NCH_3)

MS (m/z) : 205.9 (M^+)

1-Ethoxy-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (26)

The compound 1-ethoxy-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**26**) was synthesized as per the method described for compound (**25**) by taking 3-methyl-2-oxo-2,3,4,5-tetrahydro-1*H*-benzo[*d*]azepin-1-yl methanesulfonate (**6**) (0.5 gm, 1.86 mmol) in ethanol and replcing sodium methoxide with sodium ethoxide (0.19 gm, 2.79 mmol) to obtain compound (**26**) (0.2 gm, 50 %) as a semisolid mass.

Anal.:

TLC : R_f 0.30 (Ethyl acetate: Hexane) (1:1)

IR (cm^{-1}) : 1657, 1398, 1076 and 749

^{1}H-NMR : 7.75 (d, 1H, Ar-*H*), 7.40 (d, 1H, Ar-*H*), 7.20-7.08 (m, 2H, Ar-*H*), 5.07 (s, 1H, Ar-C*H*), 4.56-4.44 (m, 1H, NCH_2CH$_2$), 4.05-3.57 (m, 2H, OCH_2), 3.46-3.40 (m, 1H, NCH_2CH$_2$), 3.33-3.07 (m, 2H, NCH$_2$CH_2), 3.04 (s, 3H, NCH_3) and 1.26 (t, 3H, CH$_2$CH_3)

MS (m/z) : 241.8 ($M+Na^+$)

3-Methyl-1-phenoxy-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (27)

The compound 3-methyl-1-phenoxy-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**27**) was synthesized as per the method described for compound (**25**) by replcing sodium methoxide with sodium phenoxide (0.32 gm, 2.79 mmol) to obtain compound (**27**) (0.2 gm, 45 %) as a semisolid mass.

Anal.:

TLC : R_f 0.28 (Ethyl acetate: Hexane) (1:1)

IR (cm^{-1}) : 3060, 2924, 1655, 1493, 1400, 1239, 753 and 691

^{1}H-NMR : 7.38 (d, 1H, Ar-*H*), 7.22-6.88 (m, 8H, Ar-*H*), 5.16 (s, 1H, Ar-C*H*), 4.38-4.31 (m, 1H, NCH_2CH_2), 3.44-3.37 (m, 1H, NCH_2CH_2), 3.15-3.08 (m, 2H, NCH_2CH_2) and 2.90 (s, 3H, NCH_3)

MS (m/z) : 267.8 (M^+)

5.1.2 Synthesis of 3-ethyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one derivatives (31-48)

Synthesis of 3-ethyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one derivatives **(31-48)** was carried out by a similar procedure as used for the synthesis of 3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one derivatives **(4-27)** by taking ethylamine instead of methylamine in the synthesis of *N*-ethyl-2-phenylethylamine.

5.1.2.1 *N*-Ethyl-2-phenylethanamine (28)

Synthesis of *N*-ethyl-2-phenylethanamine was carried out by the same method used for the synthesis of *N*-methyl-2-phenylethanamine by replacing methylamine with ethylamine (35 ml, 54.04mM, 70% aqueous solution). After removal of solvent, **28** obtained as yellow oil (6.28 ml, 86 %). The intermediate was used in form of chloroform solution for further reaction..

Anal.:

TLC : R_f 0.45 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 3443, 2956, 1450, 750 and 697

5.1.2.2 2-Chloro-*N*-ethyl-*N*-phenethylacetamide (29)

N-Ethyl-2-phenylethylamine was reacted with 2-chloroacetyl chloride (8.0 ml, 10.05 mmol) in the similar way as described for the synthesis of intermediate (**2**) to afford 2-chloro-*N*-ethyl-*N*-phenethylacetamide (**29**) (15.1 gm, 97 %) as yellow oil.

Anal.:

TLC : R_f 0.64 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 3027, 2977, 1650, 1455, 752 and 703

5.1.2.3 3-Ethyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (30)

Synthesis of the cyclized intermediate was obtained by the method similar to the synthesis of compound **(3)** and yielded the intermediate 3-ethyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one **(30)** (0.4 gm, 54 %) as a white solid. m.p.: 106-108 ^{o}C

Anal.:

TLC : R_f 0.54 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 1639, 1456 and 760

^{1}H-NMR : 7.77-7.75 (m, 1H, Ar-*H*), 7.52-7.48 (m, 1H, Ar-*H*), 7.35 (t, 1H, Ar-*H*), 7.27-7.25 (m, 1H, Ar-*H*), 3.89 (s, 2H, COCH_2), 3.73- 3.70 (m, 2H, Ar-CH_2C*H_2*N), 3.48 (q, 2H, NCH_2CH_3), 3.12-3.15 (m, 2H, NCH_2C*H_2*) and 1.26 (t, 3H, NCH_2C*H_3*)

5.1.2.4 3-Ethyl-4,5-dihydro-3*H*-benzo[*d*]azepine-1,2-dione (31)

The 3-ethyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**30**) was oxidized by selenium oxide (0.88 gm, 7.9 mmol) as done for the synthesis of compound (**4**) to afford a white solid compound (**31**) (0.6 gm, 56 %). m.p.: 102-104 ^{o}C

Anal.:

TLC : R_f 0.27 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 1729, 1658, 1451 and 741

^{1}H-NMR : 7.76 (d, 1H, Ar*H*), 7.50 (t, 1H, Ar*H*), 7.35 (t, 1H, Ar*H*), 7.26 (d, 1H, Ar*H*), 3.72-3.70 (m, 2H, NCH_2CH_2), 3.58 (q, 2H, NCH_2CH_3, *J*=7.2 Hz), 3.26-3.24 (m, 2H, NCH_2C*H_2*) and 1.26 (t, 3H, NCH_2C*H_3*, *J*=7.2 Hz)

MS (m/z) : 204.1 (M+1)

5.1.2.5 3-Ethyl-1-hydroxy-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (32)

Synthesis of 3-ethyl-1-hydroxy-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**32**) was carried out by a similar method used for the synthesis of compound (**5**) using sodium borohydride (0.693 gm, 7.4 mmol). The residue so obtained was purified by column chromatography on silica gel using hexane/ether (2:1) solvent system, to obtain compound (**32**), (0.8 gm, 77 %) as a white solid. m.p.: 115-117 ^{o}C

Anal.:

TLC : R_f 0.42 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 3419, 1655, 1560, 1454 and 746

^{1}H-NMR : 7.80 (d, 1H, Ar*H*), 7.28-7.19 (m, 2H, Ar*H*), 7.09 (d, 1H, Ar*H*), 5.73 (d, 1H, CHO*H*, *J*=5.0 Hz), 4.53 (d, 1H, C*H*OH, *J*=5.0 Hz), 4.09-4.02 (m, 1H ,NCH_2CH$_2$), 3.67-3.58 (m, 1H ,NCH_2CH$_2$), 3.49-3.24 (m, 2H, NCH_2CH$_3$), 3.17-3.08 (m, 2H, NCH$_2$CH_2) and 1.17 (t, 3H, NCH$_2$CH_3)

MS (m/z) : 206.1(M+1)

5.1.2.6 3-Ethyl-2-oxo-2,3,4,5-tetrahydro-1*H*-benzo[*d*]azepin-1-yl methanesulfonate (33)

Synthesis of 3-ethyl-2-oxo-2,3,4,5-tetrahydro-1*H*-benzo[*d*]azepin-1-yl methanesulfonate (**33**) was carried out by a similar method used for the synthesis of compound (**6**) using methanesulfonyl chloride (0.738 gm, 7.3 mmol) to obtain compound (**33**), (0.9 gm, 68 %) as a semisolid.

Anal.:

TLC : R_f 0.72 (Ethyl acetate: Hexane) (2:8)

IR (cm^{-1}) : 2973, 1655, 1485, 1203 and 762

5.1.2.7 Synthesis of 1-amino substituted 3-ethyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one derivatives (34-48)

3-Ethyl-1-propylamino-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (34)

A THF (10 ml) solution of 3-ethyl-2-oxo-2,3,4,5-tetrahydro-1*H*-benzo[*d*]azepin-1-yl-methanesulfonate (**33**) (0.5 gm, 1.8 mmol) was taken under nitrogen environment to the reaction vessel. Anhydrous K_2CO_3 (0.37 gm, 2.6 mmol) and *n*-propylamine (0.16 gm, 2.6 mmol) were added and the reaction mixture was stirred for 6-8 hr at room temperature. Completion of the reaction was monitored by TLC. After completion, the reaction mixture was poured into ice-cold water and extracted successively with ethyl acetate (10x3 ml). The organic fractions were combined, dried over $MgSO_4$, volatiles removed and the residue so obtained was purified by column chromatography on silica gel using hexane/ether (2:1) solvent system, to obtain compound (**34**) (0.3 gm, 63 %) as white solid. m. p. : 72-74 °C

Anal.:

TLC : R_f 0.54 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 3292, 3059, 2964, 1648, 1424 and 743

^{1}H-NMR : 7.60 (d, 1H, Ar*H*), 7.14-7.07 (m, 2H, Ar*H*), 7.00 (d, 1H, Ar*H*), 4.78 (s, 1H, C*H*NH), 4.00-3.92 (m, 1H, NCH_2CH$_2$), 3.59- 3.53 (m, 1H, NCH_2CH$_2$), 3.51-3.30 (m, 2H, NCH_2CH$_3$), 3.19-3.01 (m, 2H, NCH$_2$CH_2), 2.65-2.59 (m, 1H, NHCH_2CH$_2$), 2.49-2.43 (m, 1H, NHCH_2CH$_2$), 1.59-1.47 (m, 2H, NHCH$_2$CH_2), 1.05 (t, 3H, NCH$_2$CH_3) and 0.90 (t, 3H, CH$_2$CH$_2$CH_3)

MS (m/z) : 246.04(M)

3-Ethyl-1-(isobutylamino)-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (35)

The compound 3-ethyl-1-(isobutylamino)-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**35**) was synthesized as per the method described for compound (**34**) by replacing *n*-propylamine with isobutylamine (0.193 gm, 2.6 mmol) to obtain compound (**35**) (0.3 gm, 63 %) as a semisolid product.

Anal.:

TLC : R_f 0.69 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 3292, 2956, 2927, 1655, 1471 and 753

^{1}H-NMR : 7.63 (d, 1H, Ar*H*), 7.15-7.07 (m, 2H, Ar*H*), 7.00 (d, 1H, Ar*H*), 4.75 (s, 1H, C*H*NH), 3.97-3.90 (m, 1H, NCH_2CH$_2$), 3.64- 3.58 (m, 1H, NCH_2CH$_2$), 3.49-3.32 (m, 2H, NHCH_2CH$_3$), 3.19-3.04 (m, 2H, NCH$_2$CH_2), 2.51-2.47 (m, 1H, NHCH_2CH), 2.33-2.29 (m, 1H, NHCH_2CH), 1.80-1.73 (m, 1H, NHCH$_2$C*H*), 1.06 (t, 3H, NCH$_2$CH_3) and 0.94-0.90 (m, 6H, CH_3CHCH_3)

MS (m/z) : 259.86(M)

1-Butylamino-3-ethyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (36)

The compound 1-butylamino-3-ethyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**36**) was synthesized as per the method described for compound (**34**) by replacing *n*-propylamine with *n*-butylamine (0.193 gm, 2.6 mmol) to obtain compound (**36**) (0.3 gm, 63 %) as a semisolid product.

Anal.:

TLC : R_f 0.50 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 3305, 3061, 2953, 1657, 1477 and 755

^{1}H-NMR : 7.59 (d, 1H, Ar*H*), 7.15-7.07 (m, 2H, Ar*H*), 7.00 (d, 1H, Ar*H*), 4.78 (s, 1H, C*H*NH), 4.00-3.92 (m, 1H, NCH_2CH$_2$), 3.60- 3.32 (m, 3H, NCH_2CH$_2$, NCH_2CH$_3$), 3.18-3.01 (m, 2H, NCH$_2$CH_2), 2.68-2.62 (m, 1H, NHCH_2CH$_2$), 2.53-2.47 (m, 1H, NHCH_2CH$_2$), 1.54-1.46 (m, 2H, NHCH$_2$CH_2), 1.39-1.30 (m, 2H, NHCH$_2$CH$_2$CH_2),1.05 (t, 3H, NCH$_2$CH_3) and 0.86 (t, 3H, CH$_2$CH$_2$CH_3)

MS (m/z) : 260.11(M)

3-Ethyl-1-phenylamino-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (37)

The compound 3-ethyl-1-phenylamino-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**37**) was synthesized as per the method described for compound (**34**) by replacing *n*-propylamine with aniline (0.257 gm, 2.6 mmol) to obtain compound (**37**) (0.3 gm, 63 %) as a white solid. m.p.: 186-188 °C

Anal.:

TLC : R_f 0.70 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 3361, 2990, 2933, 1656 1499, 759 and 692

^{1}H-NMR : 7.55 (d, 1H, Ar*H*), 7.28-7.15 (m, 5H, Ar*H*), 6.72 (m, 2H, Ar*H*), 6.58 (d, 1H, ArH), 5.70 (d, 1H, CHN*H*), 5.58 (d, 1H, C*H*NH), 4.28-4.20 (m, 1H, NCH_2CH_2), 3.70-3.62 (m, 1H, NCH_2CH_3), 3.51- 3.36 (m, 3H, NCH_2CH_3, NCH_2CH_2), 3.28-3.20 (m, 1H, NCH_2CH_2) and 1.17 (t, 3H, NCH_2CH_3)

MS (m/z) : 279.11(M^+)

3-Ethy-1-(2-furanylmethylamino)-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (38)

The compound 3-ethy-1-(2-furanylmethylamino)-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**38**) was synthesized as per the method described for compound (**34**) by replacing *n*-propylamine with (furan-2-yl)methylamine (0.257 gm, 2.6 mmol) to obtain compound (**38**) (0.4 gm, 78 %) as semisolid product.

Anal.:

TLC : R_f 0.50 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 3407, 2972, 2929, 1654, 1490 and 745

^{1}H-NMR : 7.73 (d, 1H, Ar*H*), 7.38 (m, 1H, Ar*H*), 7.24-7.17 (m, 2H, Ar*H*), 7.09 (d, 1H, Ar*H*), 6.33-6.32 (m, 1H, Ar*H*), 6.24-6.23 (m, 1H, Ar*H*), 4.97 (s, 1H, C*H*NH), 4.06-3.98 (m, 1H, NCH_2CH_2), 3.95 (d, 1H, NHCH_2), 3.84 (d, 1H, NHCH_2),3.63- 3.52 (m, 2H, N-CH_2CH_3), 3.46-3.39 (m, 1H, NCH_2CH_2), 3.25-3.14 (m, 2H, NCH_2CH_2) and 1.14 (t, 3H, NCH_2CH_3)

MS (m/z) : 284.73(M)

1-Benzylamino-3-ethyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (39)

The compound 1-benzylamino-3-ethyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**39**) was synthesized as per the method described for compound (**34**) by replacing *n*-propylamine with benzylamine (0.284 gm, 2.6 mmol) to obtain compound (**39**) (0.5 gm, 89 %) as white solid. m.p.: 92-94 °C

Anal.:

TLC : R_f 0.45 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 3439, 3060, 2971, 1653, 1425, 769 and 698

^{1}H-NMR : 7.79 (d, 1H, Ar*H*), 7.43-7.41 (m, 2H, Ar*H*), 7.33-7.30 (m, 2H, Ar*H*), 7.29-7.27 (m, 1H, Ar*H*), 7.25-7.15 (m, 2H, Ar*H*), 7.06 (d, 1H, Ar*H*), 4.92 (s, 1H, C*H*NH), 4.02-3.95 (m, 2H, NHCH_2, NCH_2CH_2), 3.78 (d, 1H, NHCH_2), 3.60-3.51 (m, 2H, NCH_2CH_3), 3.47-3.38 (m, 1H, NCH_2CH_2), 3.21-3.08 (m, 2H, NCH_2CH_2) and 1.13 (t, 3H, NCH_2CH_3)

MS (m/z) : 293.74 (M^+)

3-Ethyl-1-(4-methylbenzylamino)-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (40)

The compound 3-ethyl-1-(4-methylbenzylamino)-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**40**) was synthesized as per the method described for compound (**34**) by replacing *n*-propylamine with 4-methylbenzylamine (0.312 gm, 2.6 mmol) to obtain compound (**40**) (0.5 gm, 94 %) as white solid. m.p.: 94-95 °C

Anal.:

TLC : R_f 0.40 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 3283, 3057, 2922, 1654, 1441, 744 and 683

^{1}H-NMR : 7.77 (d, 1H, Ar*H*), 7.30 (d, 2H, Ar*H*), 7.25-7.11 (m, 4H, Ar*H*), 7.06 (d, 1H, Ar*H*), 4.91 (s, 1H, C*H*NH), 4.01-3.96 (m, 1H, NCH_2CH_2), 3.92 (d, 1H, NHCH_2Ar), 3.73 (d, 1H, NHCH_2Ar), 3.61-3.51 (m,

2H, NC*H*$_2$CH$_3$), 3.47-3.40 (m, 1H, NC*H*$_2$CH$_2$), 3.17-3.12 (m, 2H, NCH$_2$C*H*$_2$), 2.33 (s, 3H, ArC*H*$_3$) and 1.13 (t, 3H, NCH$_2$C*H*$_3$)

MS (m/z) : 307.44 (M^+)

3-Ethyl-1-(4-methoxybenzylamino)-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (41)

The compound 3-ethyl-1-(4-methoxybenzylamino)-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**41**) was synthesized as per the method described for compound (**34**) by replacing *n*-propylamine with 4-methoxybenzylamine (0.364 gm, 2.6 mmol) to obtain compound (**41**) (0.6 gm, 97 %) as semisolid product.

Anal.:

TLC : R_f 0.45 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 3283, 3010, 2965, 1647, 1442, 744 and 683

^{1}H-NMR : 7.76 (d, 1H, Ar*H*), 7.34-7.32 (m, 2H, Ar*H*), 7.20-7.14 (m, 2H, Ar*H*),
7.07-7.05 (m, 1H, Ar*H*), 6.87-6.85 (m, 2H, Ar*H*), 4.91 (s, 1H,C*H*NH), 3.98-3.95 (m, 1H, NC*H*$_2$CH$_2$), 3.90 (d, 1H, NHC*H*$_2$Ar), 3.79 (s, 3H, OC*H*$_3$), 3.71 (d, 1H, NHC*H*$_2$Ar), 3.59-3.50 (m, 2H, NC*H*$_2$CH$_3$), 3.45-3.40 (m, 1H, NC*H*$_2$CH$_2$), 3.17-3.12 (m, 2H, NCH$_2$C*H*$_2$) and 1.13 (t, 3H, NCH$_2$C*H*$_3$)

MS (m/z) : 323.70(M^+)

3-Ethyl-1-(4-fluorobenzylamino)-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (42)

The compound 3-ethyl-1-(4-fluorobenzylamino)-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**42**) was synthesized as per the method described for compound (**34**) by replacing *n*-propylamine with 4-fluorobenzylamine (0.332 gm, 2.6 mmol) to obtain compound (**42**) (0.3 gm, 63 %) as semisolid product.

Anal.:

TLC : R_f 0.58 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 3307, 3062, 2930, 1654, 1508 and 750

^{1}H-NMR : 7.69 (d, 1H, Ar*H*), 7.32-7.29 (m, 2H, Ar*H*), 7.15-7.07 (m, 2H, Ar*H*),
6.99 (d, 1H, Ar*H*), 6.95-6.89 (m, 2H, Ar*H*), 4.82 (s, 1H, C*H*NH), 3.95-3.88 (m, 1H, NCH_2CH_2), 3.86 (d, 1H, NHCH_2), 3.65 (d, 1H, NHCH_2), 3.52- 3.43 (m, 2H, N-CH_2CH_3), 3.39-3.30 (m, 1H, NCH_2CH_2), 3.13-3.05 (m, 2H, NCH_2CH_2) and 1.05 (t, 3H, NCH_2CH_3)

MS (m/z) : 311.82(M^+)

3-Ethyl-1-(3-fluorobenzylamino)-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (43)

The compound 3-ethyl-1-(3-fluorobenzylamino)-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**43**) was synthesized as per the method described for compound (**34**) by replacing *n*-propylamine with 3-fluorobenzylamine (0.332 gm, 2.6 mmol) to obtain compound (**43**) (0.4 gm, 67 %) as white solid. m.p.: 95-96 °C

Anal.:

TLC : R_f 0.45 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 3437, 3063, 2972, 1652, 1482 and 771

^{1}H-NMR : 7.71 (d, 1H, Ar*H*), 7.21-7.07 (m, 5H, Ar*H*), 6.98 (d, 1H, Ar*H*), 6.87-
6.82 (m, 1H, Ar*H*), 4.81 (s, 1H, C*H*NH), 3.94-3.86 (m, 2H, NHCH_2Ar, NCH_2CH_2), 3.67 (d, 1H, NHCH_2Ar), 3.52-3.42 (m, 2H, NCH_2CH_3), 3.38-3.30 (m, 1H, NCH_2CH_2), 3.12-3.04 (m, 2H, NCH_2CH_2) and 1.05 (t, 3H, NCH_2CH_3)

MS (m/z) : 311.27(M^+)

1-(3-Chlorobenzylamino)-3-ethyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (44)

The compound 1-(3-chlorobenzylamino)-3-ethyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**44**) was synthesized as per the method described for compound (**34**) by replacing *n*-propylamine with 3-chlorobenzylamine (0.376 gm, 2.6 mmol) to obtain compound (**44**) (0.5 gm, 83 %) as semisolid compound.

Anal.:

TLC : R_f 0.54 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 3324, 3061, 2973, 1654, 1477 and 762

^{1}H-NMR : 7.78 (d, 1H, Ar*H*), 7.42 (s, 1H, Ar*H*), 7.31-7.15 (m, 5H, Ar*H*), 7.07 (d, 1H, Ar*H*), 4.89 (s, 1H, C*H*NH), 4.03-3.94 (m, 2H, NHCH_2, NCH_2CH_2), 3.73 (d, 1H, NHCH_2), 3.60-3.51 (m, 2H, NCH_2CH_3), 3.46-3.37 (m, 1H, NCH_2CH_2), 3.21-3.12 (m, 2H, NCH_2CH_2) and 1.13 (t, 3H, NCH_2CH_3)

MS (m/z) : 328.51(M^+)

3-Ethyl-1-(2-methylbenzylamino)-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (45)

The compound 3-ethyl-1-(2-methylbenzylamino)-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**45**) was synthesized as per the method described for compound (**34**) by replacing *n*-propylamine with 2-methylbenzylamine (0.321 gm, 2.6 mmol) to obtain compound (**45**) (0.3 gm, 57 %) as a white solid. m.p.: 96-97 °C

Anal.:

TLC : R_f 0.50 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 3436, 3061, 2972, 1654, 1481 and 761

^{1}H-NMR : 7.70 (d, 1H, Ar*H*), 7.33-7.31 (m, 1H, Ar*H*), 7.17-7.07 (m, 5H, Ar*H*), 6.99 (d, 1H, Ar*H*), 4.85 (s, 1H, C*H*NH), 3.95-3.86 (m, 2H, NHCH_2Ar, NCH_2CH_2), 3.67 (d, 1H, NHCH_2Ar), 3.55-3.44 (m, 2H, NCH_2CH_3), 3.41-3.34 (m, 1H, NCH_2CH_2), 3.10-3.05 (m, 2H, NCH_2CH_2), 2.31 (s, 3H, ArCH_3) and 1.06 (t, 3H, NCH_2CH_3)

MS (m/z) : 307.61(M^+)

3-Ethyl-1-phenethylamino-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (46)

The compound 3-Ethyl-1-phenethylamino-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**46**) was synthesized as per the method described for compound (**34**) by replacing *n*-propylamine with 2-phenethylamine (0.322 gm, 2.6 mmol) to obtain compound (**46**) (0.6 gm, 97 %) as a white solid. m.p.: 86-87 °C

Anal.:

TLC : R_f 0.63 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 3302, 3054, 2975, 1650, 1484, 747 and 702

^{1}H-NMR : 7.62-7.59 (m, 1H, Ar*H*), 7.29-7.23 (m, 4H, Ar*H*), 7.21-7.13 (m, 3H, Ar*H*), 7.06-7.04 (m, 1H, Ar*H*), 4.85 (s, 1H, C*H*NH), 3.96-3.88 (m, 1H, NCH_2CH_2) 3.60- 3.45 (m, 3H, NHCH_2CH_2, NCH_2CH_2), 3.20-2.80 (m, 6H, NHCH_2CH_2, NCH_2CH_2, NCH_2CH_3) and 1.10 (t, 3H, NCH_2CH_3)

MS (m/z) : 308.09(M^+)

1-(4-Chlorophenethylamino)-3-ethyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (47)

The compound 1-(4-chlorophenethylamino)-3-ethyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**47**) was synthesized as per the method described for compound (**34**) by replacing *n*-propylamine with 2-(4-chloro)phenylethylamine (0.412 gm, 2.6 mmol) to obtain compound (**47**) (0.5 gm, 78 %) as white solid. m.p.: 84-86 °C

Anal.:

TLC : R_f 0.50 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 3460, 3060, 2975, 1652, 1496 and 757

^{1}H-NMR : 7.53-7.51 (m, 1H, Ar*H*), 7.18-7.16 (m, 2H, Ar*H*), 7.12-7.05 (m, 4H, Ar*H*), 7.00-6.97 (m, 1H, Ar*H*), 4.77 (s, 1H, C*H*NH), 3.90-3.84 (m, 1H, NCH_2CH_2), 3.52- 3.39 (m, 4H, NHCH_2CH_2), 3.37-3.30 (m, 1H, NCH_2CH_2), 3.14-3.03 (m, 2H, NCH_2CH_3), 2.81-2.70 (m, 2H, NCH_2CH_2) and 1.03 (t, 3H, NCH_2CH_3)

MS (m/z) : 342.75(M^+)

3-Ethyl-1-(4-methoxyphenethylamino)-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (48)

The compound 3-ethyl-1-(4-methoxyphenethylamino)-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**48**) was synthesized as per the method described for compound (**34**) by replacing *n*-propylamine with 2-(4-methoxy)phenylethylamine (0.402 gm, 2.6 mmol) to obtain compound (**48**) (0.4 gm, 63 %) as white solid. m.p.: 86-87 °C

Anal.:

TLC : R_f 0.36 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 3439, 3061, 2972, 1651, 1425 and 766

^{1}H-NMR : 7.53-7.51 (m, 1H, Ar*H*), 7.20-7.05 (m, 4H, Ar*H*), 6.99-6.97 (m, 1H, Ar*H*), 6.77-6.73 (m, 2H, Ar*H*), 4.90 (s, 1H, C*H*NH), 4.33-4.26 (m, 1H, NCH_2CH_2), 3.70 (s, 3H. ArCH_3), 3.53-3.29 (m, 4H, NHCH_2CH_2Ar), 3.14-3.02 (m, 2H, NCH_2CH_3), 2.93-2.89 (m, 1H, NCH_2CH_2), 2.80-2.71 (m, 2H, NCH_2CH_2) and 1.01 (t, 3H, NCH_2CH_3)

MS (m/z) : 337.77(M^+)

5.1.3 Synthesis of 1-imino derivatives of 3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (49-63)

3-Methyl-4,5-dihydro-1-propylimino-1*H*-benzo[*d*]azepin-2(3*H*)-one (49)

Ethanolic solution (dry) of 3-methyl-4,5-dihydro-3*H*-benzo[*d*]azepine-1,2-dione (**4**) (1 gm, 5.29 mmol) was taken in a reaction vessel to which tosic acid (1.36 gm, 7.9 mmol) and *n*-propyl amine (0.87 ml, 10.58 mmol) were added. The mixture was refluxed for at least 48 hrs and completion of the reaction was monitored by TLC. After the completion, the reaction mixture was poured into ice cold water and extracted with DCM (3x10ml). The combined organic solvent was dried and the solvent recovered to get a crude compound. The crude product so obtained was purified by column chromatography

using silica gel as stationary phase and hexane: ethyl acetate as eluent to afford compound (**49**) (0.7 gm, 54 %) as yellowish solid. m.p.: 72-76 °C

Anal.:

TLC : R_f 0.55 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 2954, 1643, 1483 and 745.

^{1}H-NMR : 7.56 (d, 1H, Ar*H*), 7.34-7.23 (m, 2H, Ar*H*), 7.13 (d, 1H, Ar*H*), 3.70 (bs, 2H, NCH_2CH_2), 3.61 (t, 2H, NCH_2CH_2CH_3), 3.14-3.12 (m, 2H, NCH_2CH_2), 3.09 (s, 3H, NCH_3), 1.85-1.76 (m, 2H, CH_2CH_2CH_3) and 1.00 (t, 3H, CH_2CH_2CH_3)

MS (m/z) : 231.3 (M+1)

1-Isobutylimino- 3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (50)

The compound 1-(isobutylimino)-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**50**) was synthesized as per the method described for compound (**49**) by replacing *n*-propylamine with isobutylamine (0.87 ml, 10.58 mmol) to obtain compound (**50**) (0.7 gm, 54 %) as yellowish solid. m.p.: 72-76 °C

Anal.:

TLC : R_f 0.55 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 2954, 1643, 1483 and 745.

^{1}H-NMR : 7.56 (d, 1H, Ar*H*), 7.33-7.23 (m, 2H, Ar*H*), 7.13 (d, 1H, Ar*H*), 3.70 (bs, 2H, NCH_2CH_2), 3.45 (d, 2H, NCH_2CH), 3.14-3.12 (m, 2H, NCH_2CH_2), 3.08 (s, 3H, NCH_3), 2.17-2.10 (m, 1H, NCH_2C*H*) and 1.00 (d, 6H, CH_2CH(CH_3)$_2$)

MS (m/z) : 245.3 (M+1)

1-Butylimino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (51)

The compound 1-butylimino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**51**) was synthesized as per the method described for compound (**49**) by replacing *n*-propylamine with *n*-butylamine (1.04 ml, 10.58 mmol) to obtain compound (**51**) (0.6 gm, 47%) as semisolid product.

Anal.:

TLC : R_f 0.52 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 2957, 1654, 1564 and 742.

^{1}H-NMR : 7.48 (d, 1H, Ar*H*), 7.26-7.15 (m, 2H, Ar*H*), 7.05 (d,1H, Ar*H*), 3.62-3.55 (m, 2H, $NCH_2CH_2CH_2CH_3$), 3.57 (t, 2H, $ArCH_2CH_2N$), 3.06 (t, 2H, $ArCH_2CH_2N$), 3.01 (s, 3H, NCH_3), 1.72-1.65 (m, 2H, $NCH_2CH_2CH_2CH_3$), 1.39-1.32 (m, 2H, $NCH_2CH_2CH_2CH_3$) and 0.88 (t, 3H, $NCH_2CH_2CH_2CH_3$)

MS (m/z) : 245.2 (M+1)

1-Cyclopropylimino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (52)

The compound 1-cyclopropylimino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**52**) was synthesized as per the method described for compound (**49**) by replacing *n*-propylamine with cyclopropylamine (0.73 ml, 10.58 mmol) to obtain compound (**52**) (0.7 gm, 58 %) as yellowish solid. m.p.: 147-148 °C

Anal.:

TLC : R_f 0.55 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 2933, 1653, 1482 and 745.

^{1}H-NMR : 7.51 (d, 1H, Ar*H*), 7.31-7.21 (m, 2H, Ar*H*), 7.13 (d, 1H, Ar*H*), 3.74 (m, 2H, NCH_2CH_2), 3.35-3.31 (m, 1H, N*CH*), 3.15-3.11 (m, 5H, NCH_2CH_2, NCH_3), 1.07-1.03 (m, 2H, $NCH(CH_2)_2$) and 1.00-0.98 (m, 2H, $NCH(CH_2)_2$)

MS (m/z) : 229.3 (M+1)

1-Cyclohexylimino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (53)

The compound 1-cyclohexylimino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**53**) was synthesized as per the method described for compound (**49**) by replacing *n*-propylamine with cyclohexylamine (1.210 ml, 10.58 mmol) to obtain compound (**53**) (0.9 gm, 63 %) as yellowish solid. m.p.: 152-154 °C

Anal.:

TLC : R_f 0.55 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 2922, 1641, 1485 and 751.

^{1}H-NMR : 7.53 (d, 1H, Ar*H*), 7.32-7.22 (m, 2H, Ar*H*), 7.12 (d, 1H, Ar*H*), 3.71 (bs, 2H, NCH_2CH_2), 3.62-3.5 (m, 1H, NC*H*), 3.13 (bs, 2H, NCH_2CH_2), 3.08 (s, 3H, NCH_3), 1.81-1.78 (m, 4H, CH_2), 1.68-1.55 (m, 4H, CH_2) and 1.42-1.23 (m, 2H, CH_2)

MS (m/z) : 271.4 (M+1)

3-Methyl-1-phenylimino-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (54)

The compound 3-methyl-1-phenylimino-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**54**) was synthesized as per the method described for compound (**49**) by replacing *n*-propylamine with aniline (0.96 ml, 10.58 mmol) to obtain compound (**54**) (0.8 gm, 59 %) as yellowish solid. m.p.: 185-187 °C

Anal.:

TLC : R_f 0.42 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 3056, 1655, 1590 and 755.

^{1}H-NMR : 7.85 (d, 1H, Ar*H*), 7.41-7.11 (m, 7H, Ar*H*), 7.01 (d, 1H, Ar*H*), 3.88-3.86 (m, 2H, ArCH$_2$CH_2N), 3.23-3.22 (m, 2H, ArCH_2CH$_2$N) and 3.08 (s, 3H, NCH_3)

MS (m/z) : 265.2 (M+1)

1-Benzylimino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (55)

The compound 1-benzylimino-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**55**) was synthesized as per the method described for compound (**49**) by replacing *n*-propylamine with benzylamine (1.16 ml, 10.58 mmol) to obtain compound (**55**) (0.7 gm, 48 %) as yellowish solid. m.p.: 93-95 °C

Anal.:

TLC : R_f 0.56 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 2924, 1653, 1595 and 745.

^{1}H-NMR : 7.61 (d, 1H, Ar*H*), 7.43-7.23 (m, 6H, Ar*H*), 7.14 (d, 1H, Ar*H*), 4.88 (s, 2H, NCH_2Ar), 3.64-3.63 (m, 2H, ArCH_2CH_2N), 3.13-3.12 (m, 2H, ArCH_2CH_2N) and 3.10 (s, 3H, NCH_3)

MS (m/z) : 279.2 (M+1)

1-(4-Methylbenzylimino)-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (56)

The compound 1-(4-methylbenzylimino)-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**56**) was synthesized as per the method described for compound (**49**) by replacing *n*-propylamine with 4-methylbenzylamine (0.134 ml, 1.058 mmol) to obtain compound (**56**) (0.7 gm, 45 %) as yellowish solid. m.p.: 102-104 °C

Anal.:

TLC : R_f 0.54 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 3053, 1656, 1598 and 743.

^{1}H-NMR : 7.53 (d, 1H, Ar*H*), 7.29-7.05 (m, 7H, Ar*H*), 4.76 (s, 2H, NCH_2Ar), 3.56-3.58 (m, 2H, ArCH_2CH_2N), 3.07-3.05 (m, 2H, ArCH_2CH_2N), 3.03 (s, 3H, NCH_3) and 2.26 (s, 3H, PhCH_3)

MS (m/z) : 293.2 (M+1)

1-(4-Methoxybenzylimino)-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (57)

The compound 1-(4-methoxybenzylimino)-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin- 2(3*H*)-one (**57**) was synthesized as per the method described for compound (**49**) by replacing *n*-propylamine with 4-methoxybenzylamine (0.138 ml, 1.058 mmol) to obtain compound (**57**) (0.8 gm, 49 %) as yellowish solid. m.p.: 257-259 °C

Anal.:

TLC : R_f 0.40 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 3063, 1659, 1608 and 743.

^{1}H-NMR : 7.51 (d, 1H, Ar*H*), 7.28-7.14 (m, 4H, Ar*H*), 7.05 (d, 1H, Ar*H*), 6.82-6.79 (m, 2H, Ar*H*), 4.74 (s, 2H, NCH_2Ar), 3.71 (s, 3H, PhOCH_3), 3.58-3.55 (m, 2H, ArCH_2CH_2N), 3.06-3.03 (m, 2H, ArCH_2CH_2N) and 3.025 (s, 3H, NCH_3)

MS (m/z) : 309.2 (M+1)

1-(4-Fluorobenzylimino)-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (58)

The compound 1-(4-fluorobenzylimino)-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**58**) was synthesized as per the method described for compound (**49**) by replacing *n*-propylamine with 4-fluorobenzylamine (1.20 ml, 10.58 mmol) to obtain compound (**58**) (0.7 gm, 45 %) as a semisolid product.

Anal.:

TLC : R_f 0.46 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 2963, 1676, 1597 and 747.

^{1}H-NMR : 7.52 (d, 1H, Ar*H*), 7.34-7.17 (m, 6H, Ar*H*), 7.08 (d, 1H, Ar*H*), 4.76

(s, 2H, NCH_2Ar), 3.59-3.58 (m, 2H, ArCH_2CH_2N), 3.09-3.07 (m, 2H, ArCH_2CH_2N) and 3.04 (s, 3H, NCH_3)

MS (m/z) : 297.3 (M+1)

1-(4-Chlorobenzylimino)-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (59)

The compound 1-(4-chlorobenzylimino)-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**59**) was synthesized as per the method described for compound (**49**) by replacing *n*-propylamine with 4-chlorobenzylamine (1.27 ml, 10.58 mmol) to obtain compound (**59**) (0.8 gm, 49 %) as yellowish solid. m.p.: 136-138 °C

Anal.:

TLC : R_f 0.43 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 3000, 1651, 1549 and 751.

^{1}H-NMR : 7.59 (d, 1H, Ar*H*), 7.38-7.24 (m, 6H, Ar*H*), 7.15 (d, 1H, Ar*H*), 4.83 (s, 2H, NCH_2Ar), 3.66-3.65 (m, 2H, ArCH_2CH_2N), 3.16-3.15 (m, 2H, ArCH_2CH_2N) and 3.11 (s, 3H, NCH_3)

MS (m/z) : 313.2 (M+1)

1-(4-Bromobenzylimino)-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (60)

The compound 1-(4-bromobenzylimino)-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**60**) was synthesized as per the method described for compound (**49**) by replacing *n*-propylamine with 4-bromobenzylamine (1.44 ml, 10.58 mmol) to obtain compound (**60**) (0.9 gm, 48 %) as yellowish solid. m.p.: 157-160 °C

Anal.:

TLC : R_f 0.46 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 2927, 1649, 1485 and 747

^{1}H-NMR : 7.58 (d, 1H, Ar*H*), 7.47-7.24 (m, 6H, Ar*H*), 7.15 (d, 1H, Ar*H*), 4.81 (s, 2H, NCH_2Ar), 3.66-3.64 (m, 2H, ArCH_2CH_2N), 3.16-3.13 (m, 2H, ArCH_2CH_2N) and 3.10 (s, 3H, NCH_3)

MS (m/z) : 357.1 $(M)^+$

1-(3-Chlorobenzylimino)-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (61)

The compound 1-(3-chlorobenzylimino)-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**61**) was synthesized as per the method described for compound (**49**) by replacing *n*-propylamine with 3-chlorobenzylamine (0.129 ml, 1.058 mmol) to obtain compound (**61**) (0.8 gm, 46 %) as a semisolid product.

Anal.:

TLC : R_f 0.59 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 2926, 1654, 1573 and 742.

^{1}H-NMR : 7.61 (d, 1H, Ar*H*), 7.43 (s, 1H, Ar*H*), 7.37-7.21 (m, 5H, Ar*H*), 7.16 (d, 1H, Ar*H*), 4.84 (s, 2H, NCH_2Ar), 3.66-3.65 (m, 2H, CH_2CH_2N), 3.15-3.14 (m, 2H, ArCH_2CH_2N) and 3.11 (s, 3H, NCH_3)

MS (m/z) : 313.3 (M+1)

3-Methyl-1-phenethylimino-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (62)

The compound 3-methyl-1-phenethylimino-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**62**) was synthesized as per the method described for compound (**49**) by replacing *n*-propylamine with 2-phenethylamine (1.36 ml, 10.58 mmol) to obtain compound (**62**) (0.7 gm, 45 %) as yellowish solid. m.p.: 120-122 °C

Anal.:

TLC : R_f 0.45 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 3056, 1644, 1598 and 745.

^{1}H-NMR : 7.47 (d, 1H, Ar*H*), 7.25-7.11 (m, 7H, Ar*H*), 7.02 (d, 1H, Ar*H*), 3.91 (t, 2H, NCH_2CH_2Ph), 3.08-3.05 (m, 2H, ArCH_2CH_2N), 3.05 (t, 2H, NCH_2CH_2Ph), 2.94-2.92 (m, 2H, ArCH_2CH_2N) and 2.91 (s, 1H, NCH_3)

MS (m/z) : 293.2 (M+1)

1-(4-Methoxyphenethylimino)-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (63)

The compound 1-(4-methoxyphenethylimino)-3-methyl-4,5-dihydro-1*H*-benzo[*d*]azepin-2(3*H*)-one (**63**) was synthesized as per the method described for compound (**49**) by replacing *n*-propylamine with 4-methoxyphenethylamine (1.551 ml, 10.58 mmol) to obtain compound (**63**) (0.8 gm, 47 %) as yellowish solid. m.p.: 121-124 °C

Anal.:

TLC : R_f 0.55 (Ethyl acetate: Hexane) (3:7)

IR (cm^{-1}) : 2953, 1659, 1511 and 751.

^{1}H-NMR : 7.54 (d, 1H, Ar*H*), 7.33-7.29 (m, 1H, Ar*H*), 7.26-7.23 (m, 1H, Ar*H*), 7.18-7.14 (m, 2H, Ar*H*), 7.10 (d, 1H, Ar*H*), 6.83-6.79 (m, 2H, Ar*H*), 3.94 (t, 2H, NCH_2CH_2Ar, *J* = 7.0 Hz), 3.78 (s, 3H, OCH_3), 3.21-3.20 (m, 2H, NCH_2CH_2), 3.06 (t, 2H, NCH_2CH_2Ar, *J* = 7.0 Hz), 3.02-3.00 (m, 2H, NCH_2CH_2) and 2.98 (s, 3H, NCH_3)

MS (m/z) : 323.2 (M+1)

5.2 Biological activity

The biological activity of the synthesized compounds has been carried out by the researchers of the pharmacology section and not by the candidate himself.

5.2.1 Cell culturing and MTT assay

The SH-SY5Y cells (National Centre for Cell Science, Pune) were cultured in Dulbecco's Modified Eagle Medium (DMEM) supplemented with (10 % v/v) fetal bovine serum (FBS), 100 U/ml penicillin and 100 U/ml streptomycin at 37 ºC and 5 % CO_2. The cells, cultured in 75 cm^2 flasks, were seeded in 96 well plates and incubated for 24 hr. Subsequently, the growth media was replaced with fresh normal media (control cultures) or with media supplemented with NMDA (5 mM) (Sigma) for another 24 hr to induce excitotoxicity. For antagonism studies, SH-SY5Y cells were exposed to a series of benzazepine derivatives (at 10 µM) for 2 hr followed by NMDA treatment for 24 hr. After

incubation period, cell viability was assessed using 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay. In brief, 20 µl of MTT (Sigma) solution (5 mg/ml stock solution) was added into each well and incubated for 4 hr at 37 °C. Later, the culture media was replaced by 200 µl DMSO to dissolve the formazan crystals. The optical density was measured at 570 nm with 630 nm reference wavelength using a micro plate reader 680 XR (BIO-RAD, India). The absorbance of the control cells was considered as 100 % of the cell viability.[212] Percentage protection against NMDA-induced excitotoxic damage was calculated for individual compounds.

5.2.2 Thioflavin T (ThT) assay for determining $A\beta_{1-42}$ aggregation inhibitory activity of the test compounds

In order to identify multi-target-directed ligands, $A\beta_{1-42}$ aggregation inhibitory activity was determined using the thioflavin T (ThT) fluorescence assay for the most promising test compounds obtained from the *in vitro* MTT assay, as per the earlier described method.[213] $A\beta_{1-42}$ (Sigma) dissolved in phosphate buffer saline (PBS) was further diluted with 0.215 M sodium phosphate buffer (*p*H 8) to get the final concentration of 20 µM. The test compounds were dissolved in DMSO and diluted with 0.215 M sodium phosphate buffer (*p*H 8). To determine $A\beta_{1-42}$ aggregation, the solution containing 20 µM of $A\beta_{1-42}$ or $A\beta_{1-42}$ plus the test compound (10 µM) in 0.215 M sodium phosphate buffer was incubated at room temperature for 24 hr. After incubation, 20 µM ThT (prepared in 50 mM glycine-NaOH buffer; *p*H 8.5) was added to the above solution. Finally, the fluorescence intensity was read at 442 nm excitation and 490 nm emission wavelengths using spectrofluorometer (RF-5301 PC, Shimadzu). The percentage inhibition of $A\beta_{1-42}$ aggregation was calculated using the formula: 100-(IFi/IFo×100), where IFi and IFo are fluorescence intensities in the presence and absence of the test compound respectively. Each assay was run in triplicate.

5.2.3 Congo red (CR) binding assay for determining $A\beta_{1-42}$ aggregation inhibitory activity of the test compounds

$A\beta_{1-42}$ Aggregation inhibitory potential of the potent test compounds was assessed using Congo red (CR) binding assay. CR (Hi-Media) solution prepared in PBS (*p*H 7.4) was diluted to get a final concentration of 5 µM. $A\beta_{1-42}$ (Sigma) prepared in PBS was diluted to get a final concentration of 20 µM. Briefly, 20 µM of $A\beta_{1-42}$ was incubated with or without the test compound (10 µM) for 6 hr at 37 °C. Later on, the mixture was

incubated with 5 μM of CR for 30 min at room temperature. Following incubation, CR spectra were measured using a UVspectrophotometer (UV-1700, Shimadzu) at 480 nm and 540 nm wavelengths. CR binding was calculated using the following formula: CB (M) = (OD at 540 nm/25,295) - (OD at 480 nm/46,306); where, CB (M) is the amount of CR bound with β sheets of $A\beta_{1-42}$ and OD is the optical density.[214,215]

5.2.4 Assessment of the effect of test compounds on cell viability challenged by $A\beta_{1-42}$–induced excitotoxicity

Cell viability of hippocampal neurons was determined using MTT assay as described earlier.[216] Hippocampal neuronal cells, cultured in 96 well plates were exposed to different concentrations of the test and the standard compounds (10-40 μM) for 24 hr to assess their cytotoxicity. Another set of experiments was performed to determine the neuroprotective role of potent NMDAR antagonist test compounds against $A\beta_{1-42}$ toxicity. Cultured neurons were exposed to the test and the standard compounds (10-40 μM) for 2 hr followed by $A\beta_{1-42}$ (10 μM) treatment for 24 hr. MTT assay was performed in both the experiments to determine the cell viability.

5.2.5 Morris Water Maze test for assessing spatial learning ability

The spatial learning ability of the animals was assessed using Morris Water Maze (MWM) test. The test was performed during the last five days of the treatment period. Time required to reach the hidden platform (i.e. escape latency time-ELT) and number of crossings over the platform area were recorded during 2 min of training session to determine spatial learning ability.[217]

5.2.6 Y maze test for assessing immediate working memory

Immediate working memory was evaluated using Y-maze test.[218] The Y-maze test was also carried out during the last five days of the treatment period. Each animal was placed at the end of any one arm of the maze and allowed to explore all the three arms. The sequence and the number of arm entries were recorded visually over a period of 5 min. An actual “alteration” was defined as entries in all three arms in consecutive choices (i.e. ABC, BCA or CAB but not BAB). Repeat arm entry was considered as a sign of memory impairment. The number of arm entries indicated locomotor activity. The “alteration score” for each rat was calculated using the equation:

% Alternation = [(Number of alternations) / (Total arm entries−2)] × 100.

5.2.7 ROS scavenging activity of test compounds

The ROS scavenging activity of the potent test compounds was determined using 2′,7′-dichlorofluorescin diacetate (DCFH-DA) assay.[219] Briefly, primary rat hippocampal neuronal cells were seeded in 96 well plates. Cells were exposed to the test compounds (10-40 μM) for 2 hr, followed by $A\beta_{1-42}$ (10 μM) insult for 24 hr. After the incubation period, the hippocampal cells were loaded with 10 μM DCFH-DA (Sigma) at 37 °C for 30 min. Fluorescence intensity was determined using the Synergy HTX multi-mode microplate reader with excitation wavelength of 492 nm and emission of 495 nm. The fluorescence intensities in the presence and absence of the inhibitors were compared using appropriate control and the percentage inhibition of ROS was calculated for individual inhibitors.

5.2.8 Statistical analysis

The observed data were analysed using GraphPad Prism (version 5) software. The data are expressed as mean ± SEM. Significant difference between the experimental groups was determined using two way ANOVA (MWM and Y maze test) and one way ANOVA followed by Bonferroni test. Images were visualized and captured using a Carl-Zeiss confocal microscope. While capturing images, all parameters such as gain, contrast, brightness, the positions, and types of filters were set to standard parameters, such that the signals were not saturated and all images could be quantified and compared to one another. Captured images were analysed using ZEN 2012 imaging software. For statistical analysis, a minimum of 30 randomly chosen cells per condition were analysed (n=4 independent experiments with 3-4 replicates). A value of $p<0.05$ was considered significant. * and # indicated the level of significance.