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3-Aminobenzoic Acid Analogues as a Novel GABA-at Inhibitors: Synthesis, Pharmacological Evaluation and Molecular Docking Studies

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Abstract: In present investigation we attempt to synthesized and evaluated rigid GABA analogues as potential anticonvulsant. Substituted 3-aminobenzoic acid derivatives (**A to H**) were obtained is good yields by the reaction of substituted aniline with amino protected (phthalimide) 3-aminobenzoic acid. Among 8 tested molecule five molecules found active in scPTZ model of anticonvulsant screening. The compounds A; 3-(1, 3-dioxo-2, 3-dihydro-1H-isoindol-2yl)-N-(2-nitrophenyl) benzamide and D; (N-(3-chlorophenyl)-3-(1, 3-dioxo-2, 3-dihydro-1H-isoindol-2yl) benzamide) were found active at the dose level of 100 mg/kg, the compound F;N-(2-chloro-4-nitrophenyl)-3-(1, 3-dioxo-2, 3-dihydro-1H-isoindol-2yl) benzamide and G;N-(4-bromophenyl)-3-(1, 3-dioxo-2, 3-dihydro-1H-isoindol-2yl) benzamide found to possess moderate anticonvulsant potential. Further compound B; N-(2-bromo-2, 4-dinitrophenyl)-3-(1, 3-dioxo-2, 3-dihydro-1H-isoindol-2yl) benzamidewas found also safe in long spam of time i.e. 4hr.,this result clearly indicated compound B(-5.69) are most and equivalent to diazepam(-5.68), Inhibition constant (Ki) values of compound B and diazepam, 67.79 μm and 68.93 μm respectively. however compound A and D were found protective at the dose level of 100 mg/kg in long duration of action. But the compounds F and G filed in long term treatment.Moreover the animal recovery was more than 90 %.

Key words: 3-aminobenzoic acid, epilepsy, GABA-AT (γ-aminobutyric acidaminotransferase), scPTZ (subcutaneously pentylinetetrazole), Docking.

Introduction

Epilepsy is a disorder of CNS (central nervous system). Epileptic seizure is widely used to describe an abnormal spasm or convulsion, generated by excessive electrical activity in the brain. Epilepsy is among the least understood of major chronic medical conditions [16]. It has been estimated that worldwide there are at least 50 million people who have epilepsy, affecting 0.5-1.0 % population in India [23,35]. Worldwide is the third most common neurological disorder in the United States after Alzheimer's disease and stroke [12]. Its prevalence is greater than that of cerebral palsy, multiple sclerosis and Parkinson's disease combined [24]. Based on the prevalence of epilepsy in different studies and accounting for incomplete case identification the estimated number of children and adolescents in Europe with active epilepsy is 0.9 million (prevalence 4.5-5.0 per 1000), 1.9 million in ages 20–64 years (prevalence six per 1000) and 0.6 million in ages 65 years and older (prevalence seven per 1000).

Approximately 20-30 % of the epilepsy populations have more than one seizure per month. Based on the age specific incidence rates in European studies, the estimated number of new cases per year amongst European children and adolescents is 130 000 (incidence rate 70 per 100 000), 96 000 in adults 20-64 years (incidence rate 30 per 100 000) and 85 000 in the elderly 65 years

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and older (incidence 100 per 100 000) [22]. The incidence rates are higher in the children and the elderly,Epilepsy strikes most often among the very young and the very old, although anyone can develop it at any age. In the U.S., it currently affects more than 326,000 children under the age of fifteen, more than 90,000 of whom have severe seizures that cannot be adequately treated [4].

Currently, a plethora of anti-epileptic drugs is available with distinct mechanism of actions. They act predominantly by targeting voltage gated sodium channels (phenytoin, carbamazepine, topiramate, valproic acid); gamma amino butyric acid A (GABAA) channels (benzodiazepines, tiagabine&vigabatrine); calcium channels (ethosuximide) and synaptic vesicle protein SV2A (levetiracetam, brivarace-tam) [27]. Despite such an armamentarium of antiepileptics being available, complete seizure control is not achieved in many patients as they fail to respond completely to the pharmacotherapy while others suffer from intolerable side effects of these drugs which many a times leads to discontinuation of the treatment. Hence clinical research continues in this field to come up with novel antiepileptic with new targets and mechanisms of actions which have improved activity and/or reduced side effects [29]. A mechanistic approach to the pharmacological management of each epilepsy syndrome has the potential to optimize the chance of perfect seizure control and help more people achieve safer and more fulfilled lives, GABA (γ-amino butyric acid) is a endogenous ligand and a major neuro-transmitter producing inhibitory effect by neurotransmitter release in mammalian central nerves system that has been regarded as central inhibitory neuroharmonal modulator [17]. If imbalance exists between the release of excitatory amino acid (Glutamate) and inhibitory amino acid (GABA) then this might causes epileptic seizures [11]. GABA does not penetrate the blood-brain barrier; it synthesized in the brain from glutamate using the enzymeL-glutamic acid decarbo-xylase(GAD) and pyridoxal phosphate (which is the active form of vitamin B_6) as a cofactor via a metabolic pathway called the GABA shunt [25]. This process converts glutamate, the principal excitatory neurotransmitter into the principal inhibitory neurotransmitter (GABA). GABA is found in high concentration in Cerebellum, its play vital role in regulating neuronal excitability by acting at inhibitory synopses in the brain by binding specific receptor in plasma membrane of both presynaptic and postsynaptic neuronal process.

There are two type of GABA receptor $GABA_A$ (ligand-gated ion channel or ionotropic receptor) and $GABA_B$ (G protein-coupled receptor or metabotropic). GABA metabolism depends on the removal of GABA from extracellular space, neuron and glia cell uptake GABA via specific GABA transports GATs (GAT1-GAT4) each with characteristic distribution in CNS. With in the cell widely distributed mitochondrial enzyme GABA-AT (γ-amino butyric acid transferase) that catalyzes the GABA to succinic semialdehyde (SSA) which is subsequently oxidized to succinic acid by SSA dehydrogenase then inters into Krebs's-cycle to become α-ketoglutarate and again GABA-AT regenerate glutamate from α-ketoglutarate [8]. Transaminase and thus inhibits degradation of GABA. Two important neurotransmitters involved in the regulation of brain neuronal activity are GABA, one of the most widely distributed inhibitory neurotransmitters, and L-glutamic acid, an excitatory neurotransmitter [26]. In mammals, GABA-AT plays an important role in the regulation of GABA concentration in the brain [2,20].

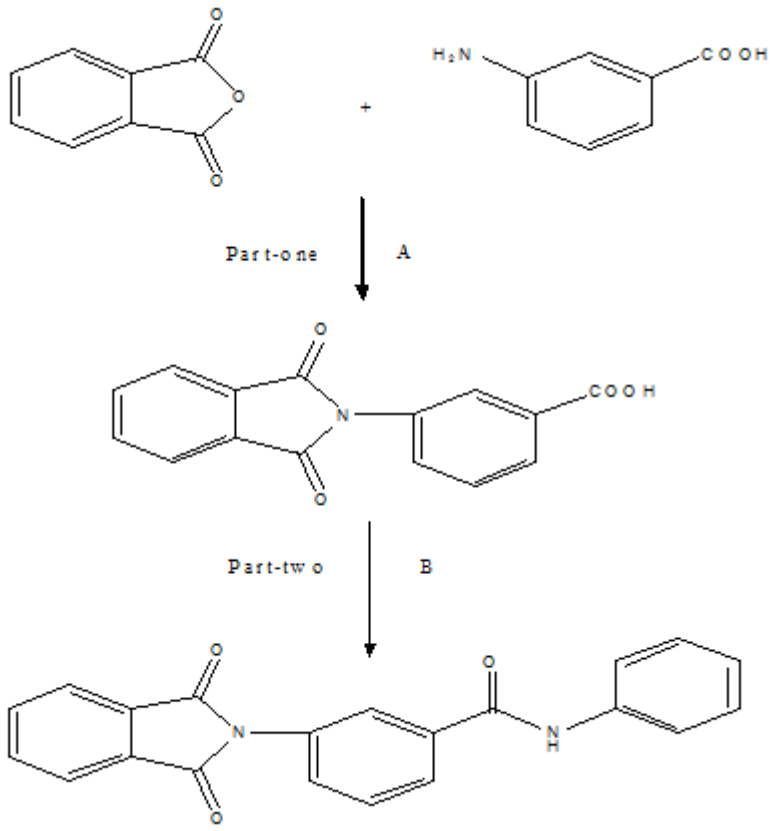
Amino transferases are the enzyme that catalyzes the amino group to oxacids [1]. It is similar vitamin B_6-dependent mechanism [32]. The concentration of GABA is regulated by two pyridoxal 52 - phosphate (PLP)-dependent enzymes, L-glutamic acid decarboxylase, which catalyzes the conversion of L-glutamate to GABA, and GABA aminotransferase (GABA-AT, EC 2.6.1.19), which degrades GABA to succinic semialdehyde [2]. GABA-AT is another subgroup-II of aminotransferase; At least 10 enzymes with approximately 30 known sequences belong to this subgroup. The coordinates for the structure of one of these enzymes, dialkylglycine decarboxylase, are available [33,34].

Phosphate covalently bound to Lys-329 via a Schiff base. The primary sequence of GABA-AT

has been deduced from the cDNA of pig brain [21] and from peptide fragments of the pig liver enzyme [9]. Which is bearing α-amino group an enzyme whose substrate is major inhibitory neuro-transmitter in brain [15,19,28]. Administration of GABA peripherally is not effective, because GABA, under normal conditions, cannot cross the blood-brain barrier (BBB); however, several other approaches have been taken to increase the brain concentrations of GABA. One approach is the use of a compound that crosses the blood-brain barrier and then selectively inhibits GABA-AT, thereby causing a build-up of GABA. Numerous competitive inhibitors of GABA-AT, Particularly compounds having a backbone structure similar to GABA [5,6,13,14,20,31] and a variety of mechanism-based in activators [34] of GABA-AT [30] show anticonvulsant activity.

Material and method

All the chemicals and reagents used were of analytical or pharmaceutical grade, the pure compounds 2, 4, dinitro aniline, 2-Bromo-4, 6-dinitroaniline, 4-Bromo aniline, 4-Nitro aniline, 2-Chloro-2-methyl aniline,2-Chloro-4-nitro aniline, 3-aminobenzoic acid (sigma Aldrich). Solvent were used in reaction are analytical grade, Ethanol, Dichloromethane (DCM). Analyses of compounds were done by using standard analytical instrument. Determinations of Melting point ranges of newly synthesized compounds were determined by open capillary method using the melting point apparatus (theils tube). The IR was carried out from KBr by Perkin Elmer Spectrum RXI- FTIR system. 1H-NMR spectra were measured on a Bruker Advance 500 MHz using TMS as a reference. Designing of synthesized compound were arranged by using different designing software Marvin Sketch 5.0.6.1, Chem Draw Ultra 6.1, AutoDock Tools 1.4.6, AutoDock 4.0, AutoGrid 4.0.



A: Fusion at 185-230°C in oil bath, stirred first 10min, leave for 30 min.
B: Aniline (0.016 mole), DCC (N, N'-dicyclohexylcarbodimide), DCM (dichloromethane), stirred ice cold condition (0-3°C) for 8-10hr.

Synthesis protocol of compounds (a-h)

General procedure for synthesis of the compounds part one

Synthesis of 3-(1, 3-dioxo-2, 3-dihydro-1H-isoindol-2-yl) benzoic acid [36]

Phthalic anhydride (1 mole) was taken in test tube and heated to 185-230°C in oil bath until it converted into a solution. 3-aminobenzoic acid (2 moles) was added simultaneously in solution. The mixture stirred occasionally for first 10min and removed after 30 minute from oil bath which result into the formation of 3-(1,3-dioxo-2,3-dihydro-1H-isoindol-2-yl) benzoic acid. The resulting precipitate was filtered, dried and recrystallised from aqueous ethanol to get pure precipitate.

Compound A; 3-(1, 3-dioxo-2, 3-dihydro-1H-isoindol-2yl)-N-(2-nitrophenyl) benzamide

IR(KBr,Vmax,cm^{-1}): CH (aromatic): 2998. 69,C=C(str):1627.36,C-C(str): 1222. 52, C=O (lactone):1718.83,C-N(str):1378.75, C=O (amide):1627.36, NH(str):3328.17, NH(bend): 1575.60 N=O(asymmetric):1525.03 N=O (symmetric):1378.75, 1H-NMR(500MHz, CDCl3)δ: 7.793-7.971(d, 2H,CH isoindol; t, 2H, CH, isoindol), 7.982-7.992(d, 2H,CH, benzamide; t, 1H, CH, benzamide), 7.999-8.246 (s, 1H,CH benzamide; s, 1H, NH, benzamide; d, 2H, CH, nitrophenyl; t,2H, CH, nitrophenyl), m.p.-144-147°C, yield-38.83 %.

Compound B; N-(2-bromo-2, 4-dinitrophenyl)-3-(1, 3-dioxo-2, 3-dihydro-1H-isoindol-2yl) benzamide

IR(KBr,Vmax,cm^{-1}): CH(aromatic):3008.36, C=C(str):1575.53, C-C(str):1211.95, C=O (lactone): 1736.66, C-N(str):1379.79, C=O (amide):1627.97, NH(str):3326.33, NH(bend): 1575.53, N=O(asymmetric):1504.82 N=O (symmetric):1449.75 C-Br:729.28, 1H-NMR (500MHz, CDCl3)ä: 8.108(d, 2H,CH isoindol; t, 2H, CH, isoindol), 8.583-8.588 (s, 1H, CH, benzamide; d, 2H,CH, benzamide; t, 1H, CH, benzamide), 8.821-8.826 (s, 1H,NH benzamide; s, 2H, CH, dinitrophenyl), m.p.-160-162°C, yield-45.5 %.

Compound C; N-(4-chlorophenyl)-3-(1,3-dioxo-2,3-dihydro-1H-isoindol-2yl) benzamide

IR(KBr,Vmax,cm^{-1}):CH(aromatic):3066.65, C=C(str):1466.75, C-C(str):1171.54, C=O (lactone): 1747.56, C-N(str):1126.56, C=O (amide):1627.36, NH(str):3495.73, NH(bend): 1581.12, C-Cl:714.65, 1H-NMR(500MHz, CDCl3)δ: 7.552-7.617(d, 2H,CH isoindol; t, 2H, CH, isoindol), 7.624-7.801(s, 1H, CH, benzamide; d, 2H,CH, benzamidet, 1H, CH, benzamide), 7.806-7.967(s, 1H,NH benzamide; d, 4H, CH, chlorophenyl), m.p.-119-121^{0}C, yield-49.16 %.

Compound D; N-(3-chlorophenyl)-3-(1,3-dioxo-2, 3-dihydro-1H-isoindol-2yl) benzamide

IR(KBr,Vmax,cm^{-1}):CH(aromatic):3038.84, C=C(str):1585.48, C-C(str):1217.73, C=O (lactone): 1716.92, C-N(str):1106.04, C=O (amide):1633.92, NH(str):3344.53, NH (bend):1522.34, C-Cl:716.13,1H-NMR(500MHz, CDCl3)δ:7.4755-7.5647(d, 2H,CH isoindol; t, 2H, CH, isoindol), 7.5797-7.8304 (s, 1H, CH, benzamide; d, 2H,CH, benzamide; t, 1H, CH, benzamide), 7.8354-8.8304 (s, 1H,NH benzamide; s, 1H, CH, chlorophenyl; d, 2H, CH, chlorophenyl; t, 1H, 2H, chlorophenyl),m.p.-86-88°C, yield-40.81%.

Compound E: 3-(1, 3-dioxo-2, 3-dihydro-1H-isoindol-2yl)-N-(3-nitrophenyl) benzamid

IR(KBr,Vmax,cm^{-1}):CH(aromatic):3001.64, C=C(str):1575.57, C-C(str):1234.14, C=O (lactone): 1731.12, C-N(str):1374.49, C=O (amide):1674.35, NH(str):3334.50, NH(bend): 1628.08, N=O(asymmetric):1525.53, N=O (symmetric):1437.55,1H-NMR(500MHz, CDCl3)ä:7.509-7.599(d, 2H,CH isoindol; t, 2H, CH, isoindol), 8.583-8.588(s, 1H, CH, benzamide; d, 2H,CH, benzamide; t, 1H, CH, benzamide), 8.821-8.826(s, 1H,NH benzamide; s, 1H, CH, nitrophenyl; d, 2H, CH, nitrophenyl; t, 1H, CH, nitrophenyl). m.p.-140-142°C, yield-47.30 %.

Compound F: N-(2-chloro-4-nitrophenyl)-3-(1, 3-dioxo-2,3-dihydro-1H-isoindol-2yl) benzamide

IR(KBr,Vmax,cm^{-1}):CH(aromatic):3048.03, C=C(str):1586.36, C-C(str):1235.31, C=O (lactone): 1730.64, C-N(str):1112.05, C=O

(amide):1690.05, NH(str):3331.63, NH(bend): 1625.37, N=O(asymmetric):1524.95, N=O (symmetric):1373.79, C-Cl:745.49,1H-NMR (500MHz, CDCl3)δ:7.883-7.983(d, 2H,CH isoindol; t, 2H, CH, isoindol), 7.993-8.021(d, 2H,CH, benzamide; s, 1H, CH, benzamide; t, 1H, CH, benzamide), 8.024-8.314(s, 1H,NH benzamide; s, 1H, CH, nitrophenyl; d, 2H, CH, nitrophenyl), m.p.-144-146°C, yield-42.10 %.

Compound G: N-(4-bromophenyl)-3-(1,3-dioxo-2, 3-dihydro-1H-isoindol-2yl) benzamid

IR(KBr,Vmax,cm^{-1}):CH(aromatic):3035.05, C=C(str):1627.36, C-C(str):1220.92, C=O (lactone): 1745.45, C-N(str):1126.51, C=O (amide):1627.36, NH(str):3328.17, NH (bend):1575.60, C-Br:1011.14,1H-NMR (500MHz, CDCl3)δ: 6.619-6.891(d, 2H,CH isoindol; t, 2H, CH, isoindol), 6.934-7.282(s, 1H, CH, benzamide; d, 2H, CH, benzamide; t, 1H, CH benzamide), 7.386-7.986(s, 1H,NH benzamide; d, 4H,CH, bromopheny),m.p.-147^{0}C, yield-40.5 %.

Compound H: N-(3-chloro-2-methylphenyl)-3-(1,3-dioxo-2,3-dihydro-1H-isoindol-2yl) benzamid

IR(KBr,Vmax,cm^{-1}):CH(aromatic):3025.21, C=C(str):1627.36, C-C(str):1234.37, C=O (lactone): 1759.44, C-N(str):1089.48, C=O (amide):1627.36, NH(str):3333.33, NH(bend): 1573.86, CH_3(asymmetric):2931.89, CH_3 (symmetric):2853.44 ,1H-NMR(500MHz, CDCl3)ä:7.420-7.604(d, 2H,CH isoindol; t, 2H, CH, isoindol), 7.611-7.784(s, 1H, CH, benzamided, 2H, CH, benzamide; t, 1H, CH benzamide) 7.948-8.092(s, 1H,NH benzamide; d, H, CH, methylphenyl; t, 1H, CH, methyl-phenyl),m.p.-148-149°C, yield-42.5 %.

Anticonvulsant activity

All the successfully synthesized were subjected to *in vivo* pharmacological screening for anticonvulsant activity. All the tests have been performed in accordance with the guidelines laid out by the Institutional Animal Ethics Committee.The anticonvulsant activity of the selected compounds was carried out according to the protocol of the Antiepileptic Drug Development Program of the National Institute of Neurological Disorder and Strokes [18]. Anticonvulsant test, chemical induced seizures (scPTZ)] was employed for screening procedures. Diazepam was used as reference compound. All the tested compounds were administered intraperitoneally (i.p.) at the dose level of 30, 100, 300 mg/kg in accordance with ASP of NINDS, NIH Bethesda, USA.

Molecular designing

With the aim of getting insights into the structural basis for its activity, 3-aminobenzoic acid analogs (compounds **A–H**) were docked into the active site of GABA-AT enzyme (PDB code: 1OHV). The Autodock docking program was employed for each docking experiment; the lowest energy docked conformation was selected from 100 runs. The central processing unit for a single docking experiment took 10-15 min, on a 2.19 GHz Intel (R) core2 Duo machine with 2.96 GB of RAM and Red Hat Enterprise Linux 5.0 operating system.

Result and discussion

A series of anti-convulsant compounds possessing 3-aminobenzoic acid were designed based on 'analogue-based' design strategy. The selection of 3-aminobenzoic acid pharmacophore was motivated by its good compliance with both geometrical and surface properties necessary for GABA-AT inhibition. In search of structural similarity, the design foresaw replacement of the 'central methyl chain' with a aromatic structure phenyl, and followed by introduction of phthalimide group to NH_2 moiety.

A set of N-protected 3-aminobenzoic acid that possess a nitro, chloro, bromo or methyl substituted phenyl ring at its carboxylic end were designed and synthesized for anticonvulsant potential. The synthetic strategy adopted for the synthesis of title compounds has been depicted in **Scheme.** Eight analogues of 3-aminobenzoic acid were synthesized. Among 8 tested molecule five molecules found active in scPTZ model of anticonvulsant screening. The results are clearly indicating the **compound B** as most and equipotent to diazepam at dose level of 30 mg/kg. The compounds **A and D** were found active at the dose

level of 100mg/kg, the compound **F and G** found to possess moderate anticonvulsant potential. Further compound **B** was found also safe in long spam of time i.e. 4hr. however compound **A and D** were found protective at the dose level of 100 mg/kg in long duration of action. But the compounds **F and G** filed in long term treatment.The results are summarized in Table: 1, Moreover the animal recovery was more than 90%.

In order to evaluate accuracy of docking, binding energy and numbers in cluster were used. Ki values (μM) were recorded for the lowest binding energy mode. Few of these molecules showed better inhibition potential than *Diazepam*, the result of molecular docking studies are tabulated in Table 2 similarly binding poses are shown in **Figure 1.** The docking analysis at the (PDB ID: 1OHV) has shown binding interaction with amino acid identified in **table 3** All molecules screened, some of them show the hydrogen bonding of with ARG404, SER403, SER74, RES1, and LYS329 are illustrated in **Figure 1**, **Table 2 and Table 3.** Considering free energy of binding and inhibition constant as criteria of evaluation a total of 8

Table 1. anticonvulsant activity of synthesized compounds Intraperitoneally injection in rat (dose 30, 100 and 300 mg/kg were administered)

Compound code	scPTZ screening 0.5 hr	scPTZ screening 4 hr	Death / Recovery
A	100	300	2/16
B	30	300	1/17
C	-	-	4/14
D	100	300	1/17
E	-	-	5/13
F	300	-	2/16
G	300	-	2/16
H	-	-	3/15
Control	-	-	4/14
Diazepam	30	100	0/18

*The figures in table indicate the minimum dose where bioactivity was demonstrated in half or more of the rats. The dash (-) indicates an absence of activity at the maximum dose administered (300 mg/kg)

Table 2. Detail of binding interaction (ligand-protein)

No.	Commpound Code	Binding Energy	RMSD	H-Bond	Inhibition Constant (Ki- μm)
1	A	-4.59	27.565	-	432.84 μm
2	B	-5.69	23.640	LYS330, PLP600	67.79 μm
3	C	-5.15	24.885	SER403,ARG404	169.25 μm
4	D	-4.74	24.840	-	332.93 μm
5	F	-4.71	32.137	SER74	300.77 μm
6	G	-4.86	28.924	RES1	273.28 μm
7	H	-4.61	21.146	-	420.96 μm
8	I	-5.23	27.035	SER74, LYS329	146.99 μm
9	Diazepam	-5.68	23.600	SER137, ARG192	68.93 μm

Table 3. binding interaction of titled compound

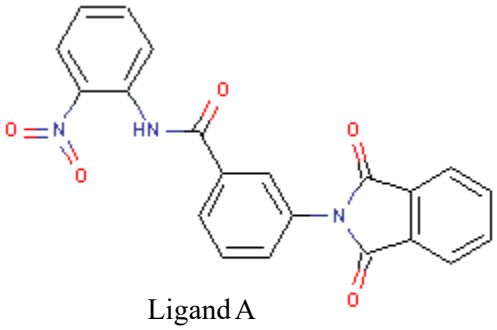
No.	Code	Interaction with amino acid
1	A	ARG192, ACT500, PLP600, ILE72, VAL300, GLN301, LYS329, SER74, SER73, LYS330
2	B	TYR69, ACT500, LYS329, SER74, PLP600, CYS135, ARG192, ILE72
3	C	GLN71, PHE414, ILE72, VAL300, SER328, GLN301, SER403, SER74, SER73, THR193 SER137, LYS330, LYS329, PLP600
4	D	SER74, LYS329, SER73, GLN301, PLP600, SER137, ARG192, THR193
5	E	MET332, LYS129, SER74, CYS136, PLP600, FES400, SER137, ILE72, CYS136, PLP600, FES400, SER137, ARG192, THR193
6	F	LYS330, SER74, SER73, CYS329, GLN201, VAL300, PLP600, ILE72, ACT500, ARG192, HIS206
7	G	LEU51, ASP67, HIS44, ALA42, ARG102, ILE72, TYR69, GLN71, MET435
8	H	MET332, SER71, LYS329, ILE72, PLP600, SLR137, ACT500, ARG192
9	Diazepam	PLP600, SER73, SER74, LYS329, SER329, LYS330, GLN301, ILE72, VAL300

compounds was predicted to be potent. The structure of the all the target molecule are shown in **figure 2**

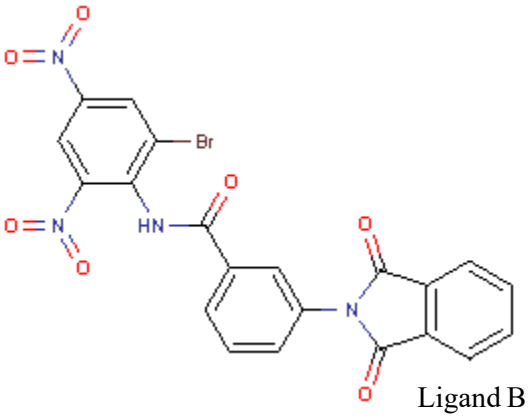
The presence of aromatic ring was found to play a major role in determining inhibitory activity for GABA-AT. Amino acids LYS329 and SER74 seemed to be in key interactions with ligands. Introduction of rigidity (decreased number of conformation of methyl chain between amino and carboxyl group) produces novel GABA-AT inhibitors. Reproducibility of the experimental conformations of the bound ligands such as **B**, and **Diazepam** flurbiprofen indicates the better performance of AutoDock method. Good correlation between the binding score and the anticonvulsant activity was observed. Rigid docking of eighty GABA-AT inhibitors have been successfully carried out. Some false positives and false negatives were observed but considering the limitations of the available docking programs, the results are encouraging. The detailed analysis of the resulted COX-2-l igand complexes may improve our knowledge in understanding the binding interactions in detail. Thus, this study will be useful for the design of new novel GABA-AT inhibitors based on docking.

Conclusion

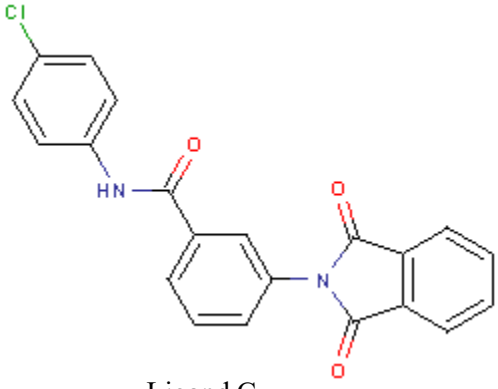
GABA is an important inhibitory neuro-transmitter containing NH_2 at one end and COOH at another end. The Beauty of GABA is that it forms Schiff's bases with PLP of GABA-AT and being metabolized.Therefore GABA-AT is valuated target for epileptic treatment because its



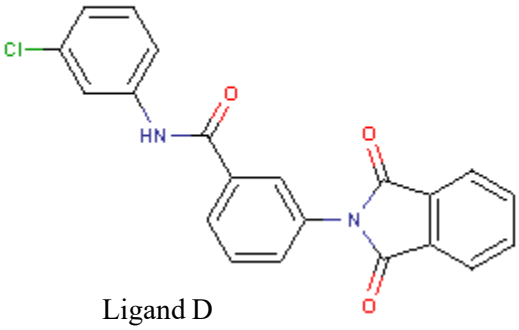
Ligand A



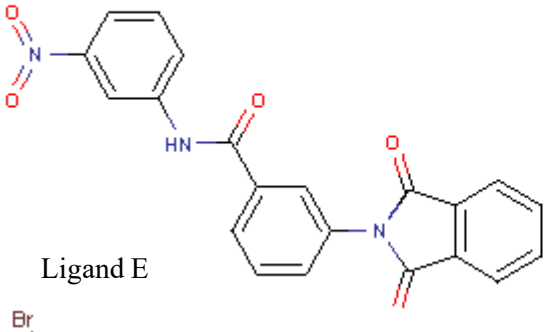
Ligand B



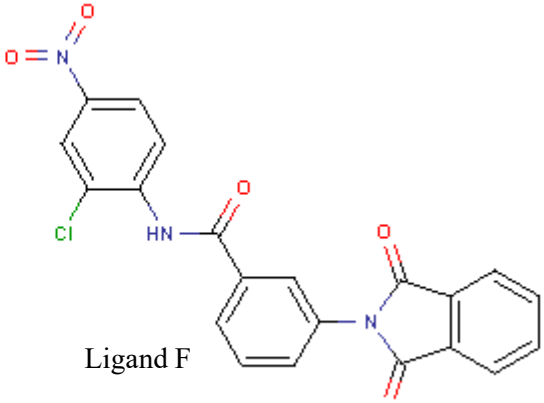
Ligand C



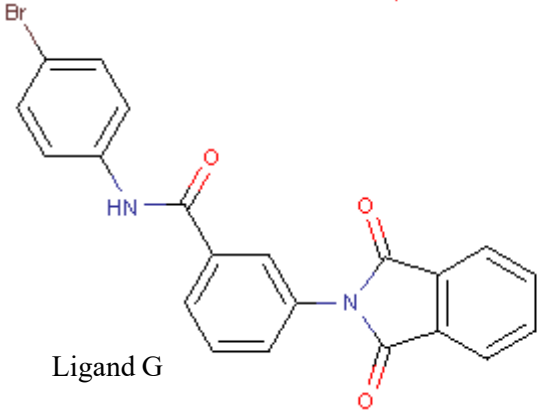
Ligand D



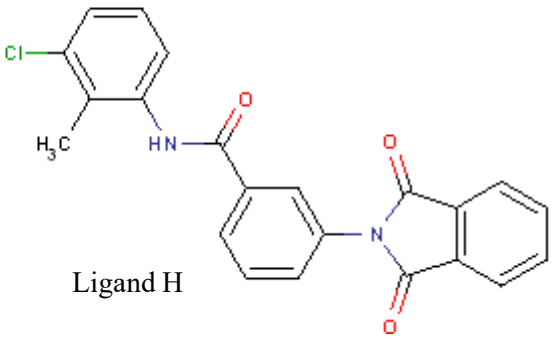
Ligand E



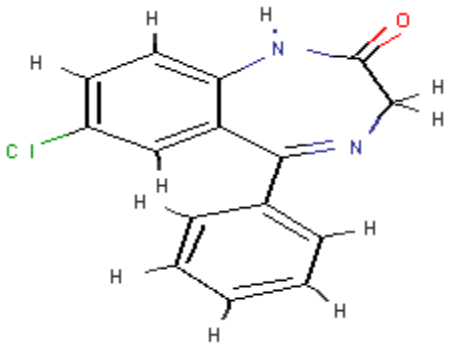
Ligand F



Ligand G



Ligand H



Reference ligand Diazepam

Figure 2. Structure of targeted molecules

mechanistic inhibition raises the level of GABA in brain and controls the seizure. In present investigation we attempt to synthesized and evaluated rigid GABA analogues as potential anticonvulsant.

Substituted 3-aminobenzoic acid derivatives (**A, B, C, D, E, F, G, H**) were obtained is good yields by the reaction of substituted aniline with amino protected (phthalimide) 3-aminobenzoic acid. The structure if all synthesized compounds were characterized by using physiochemical (m.p., TLC, solubility, % N estimation) and spectral parameter (IR, ^{1}H-NMR). Successfully synthesized compounds were subjected to biological evaluation as per guidelines given by ASP program of NINDS, NIH Bethesda USA.

Among 8 tested molecule five molecules found active in scPTZ model of anticonvulsant screening. The compounds **A and D** were found active at the dose level of 100 mg/kg, the compound **F and G** found to possess moderate anticonvulsant potential. Further compound **B** was found also safe in long spam of time i.e. 4hr. however compound **A and D** were found protective at the dose level of 100mg/kg in long duration of action. But the compounds **F and G** filed in long term treatment.

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