

IN-SILICO PROTEIN LIGAND INTERACTION STUDY OF TYPICAL ANTIPSYCHOTIC DRUGS AGAINST DOPAMINERGIC D2 RECEPTOR

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Received: 24 Dec 2012, Revised and Accepted: 02 Feb 2013

ABSTRACT

Objective: Psychotropic drugs or antipsychotics are those which exert primary effect on psyche and are used to treat psychosis. Among these, typical antipsychotics are found to show potent D2 receptor blocking activity. Present study deals with interaction of D2 receptor protein (2HLB) with some typical antipsychotics (ligands).

Methods: The nature of interaction between D2 receptor and the typical antipsychotics (under study) was investigated by molecular modelling using docking protocol.

Results: The docking results emphasizing on the hydrogen bonds between the receptor and the ligands (drugs under study) along with their different binding energies were analysed. The binding energies were found to be within the range of -6.55 to -8.56 Kcal/mol with reference drugs according to Table 1.

Conclusion: The least binding energy was found to be -8.56 Kcal/mol corresponding to the drug Prochlorperazine which establishes its maximum potency amongst the drugs under study.

Keywords: Exome Horizon, Molecular Docking, Autodock 4.2, D2 receptor

INTRODUCTION

An **antipsychotic** (or **neuroleptic**) is a psychiatric medication primarily used to manage psychosis (including delusions or hallucinations, as well as disordered thought), particularly in schizophrenia and bipolar disorder, and is increasingly being used in the management of non-psychotic disorders. A first generation of antipsychotics, known as typical antipsychotics, was discovered in the 1950s. Most of the drugs in the second generation, known as atypical antipsychotics, have been developed more recently, although the first atypical antipsychotic, clozapine, was discovered in the 1950s and introduced clinically in the 1970s. Both generations of medication tend to block receptors in the brain's dopamine pathways, but antipsychotic drugs encompass a wide range of receptor targets. The discovery of chlorpromazine's psychoactive effects in 1952 led to greatly reduced use of restraint, seclusion, and sedation in the management of agitated patients [1] and also led to further research that resulted in the development of antidepressants, anxiolytics, and the majority of other drugs now used in the management of psychiatric conditions. Until the 1970s there was considerable debate within psychiatry on the most appropriate term to use to describe the new drugs [2]. In the late 1950s the most widely used term was "neuroleptic", followed by "major tranquilizer" and then "ataraxic" [2]. The first recorded use of the term tranquilizer dates from the early nineteenth century [2]. Antipsychotics are broadly divided into two groups, the typical or first-generation antipsychotics and the atypical or second-generation antipsychotics. The typical antipsychotics are classified according to their chemical structure while the atypical antipsychotics are classified according to their pharmacological properties. These include serotonin-dopamine antagonists (see dopamine antagonist and serotonin antagonist), multi-acting receptor-targeted antipsychotics (MARTA, those targeting several systems), and dopamine partial agonists, which are often categorized as atypical [3]. In particular, antipsychotic occupancy of dopamine D2-receptors has been the focus of extensive research. Blockade of cortical and limbic dopamine D2-receptors is thought to mediate both clinical response to antipsychotics and the occurrence of adverse events. D2-receptor-related adverse events are mediated

via blockade of striatal and tuberoinfundibular D2-receptors, which are associated with extrapyramidal symptoms (EPS) [4] and prolactin elevation [5], respectively. Studies have demonstrated that the atypical antipsychotics generally have a much lower affinity for D2-receptors than the older, conventional antipsychotic agents [6-8]. The antipsychotic effects of neuroleptic drugs are mediated by dopamine DA-2 receptors, and dopamine DA-1 receptors, linked to cAMP (Cyclic Adenosine Mono Phosphate) formation, are not involved [9]. In addition, "antipsychotics" are increasingly used to treat non-psychotic disorders. For example, they are sometimes used off-label to manage aspects of Tourette syndrome or autism spectrum disorders. They have multiple off-label uses as an augmentation agent (i.e. in addition to another medication), for example in "treatment-resistant" depression [10] or OCD (Obsessive Compulsive Disorder) [11]. Despite the name, the off-label use of "antipsychotics" is said to involve deploying them as antidepressants, anti-anxiety drugs, mood stabilizers, cognitive enhancers, anti-aggressive, anti-impulsive, anti-suicidal and hypnotic (sleep) medications [12].

MATERIAL AND METHODS

Retrieval of 3D Structure

The 3D structure of the protein was downloaded from RCSB (Research Collaboratory for Structural Bioinformatics), Protein Databank (PDB, <http://www.pdb.org>). The PDB ID of the selected protein was found to be 2HLB. The Water molecules and ligands attached to the protein were removed by using Swiss PDB Viewer. The Protein was having 359 no. of groups, 2905 no. of atoms and 2954 no. of bonds.

Structural Assessment of the Protein

The protein was sent for structural assessment to Exome Horizon. The Ramchandran Plot for all residue types was given in **Fig.1**, Chi1-Chi2 plots, Main-chain parameters, Side-chain parameters, Residue properties, Main-chain bond length, Main-chain bond angles, RMS distances from planarity and distorted geometry were analyzed for input atom only [13].

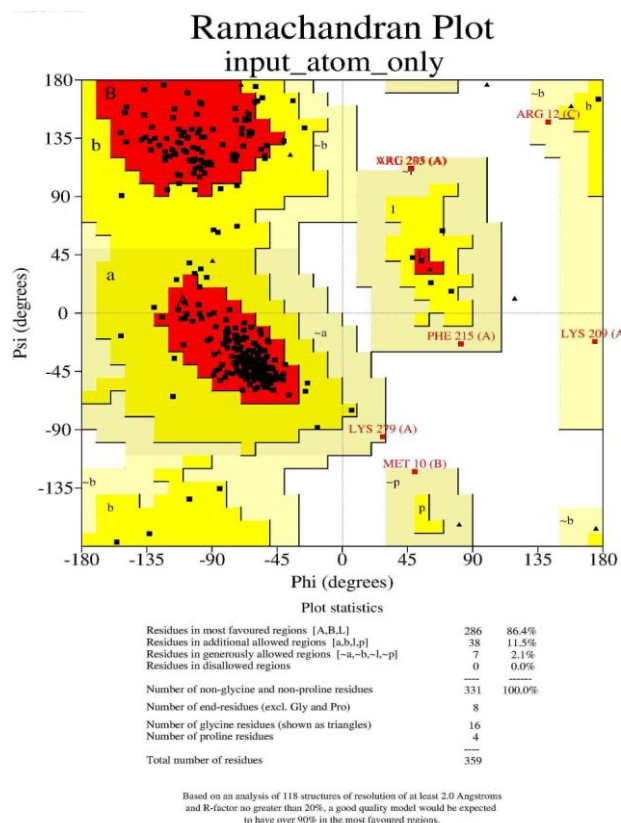


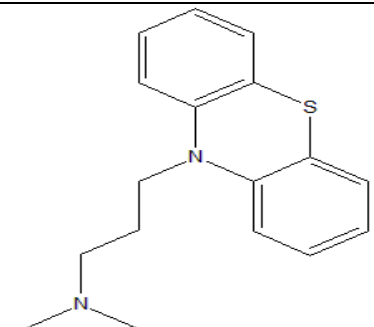
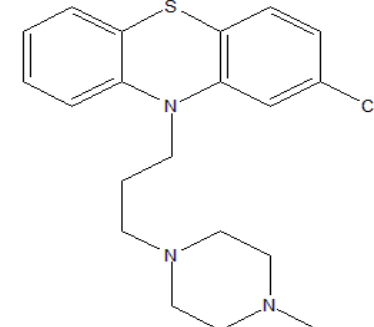
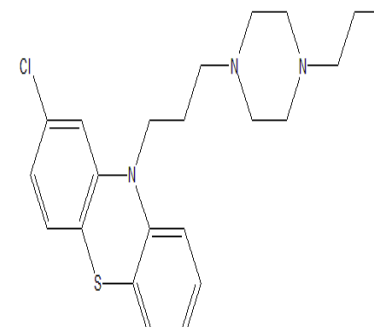
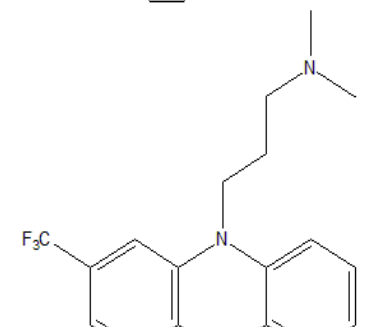
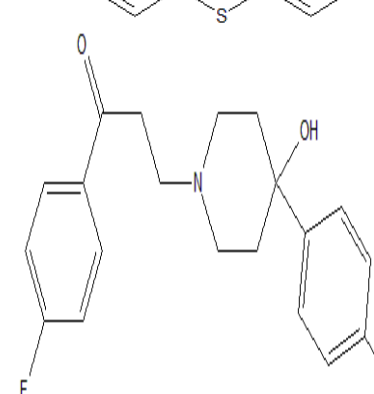
Fig. 1: It shows Ramachandran plot analysis of D2 Receptor

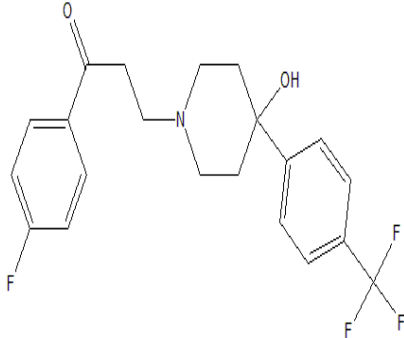
Ligand Preparation

The ligands were drawn using Moldraw tool of Exome™ Horizon in 2D and were converted into 3D before submission for docking. The ligands and its properties were given in **Table 1**.

Table 1: It shows typical antipsychotics with their respective chemical properties

S. No.	Ligand name	IUPAC name	Mol. Formulae	Log P	2D structure
1	Chlorpromazine	3-(2-chloro-10H-phenothiazin-10-yl)-N,N-dimethylpropan-1-amine	C ₁₇ H ₁₉ ClN ₂ S	5.23	
2	Thioridazine	10-(2-(1-methylpiperidin-2-yl)ethyl)-2-(methylthio)-10H-phenothiazine	C ₁₈ H ₂₀ ClNO	3.58	

3	Promazine	N,N-dimethyl-3-(10H-phenothiazin-10-yl)propan-1-amine	C ₁₇ H ₂₀ N ₂ S	3.68	
4	Prochlorperazine	2-chloro-10-(3-(4-methylpiperazin-1-yl)propyl)-10H-phenothiazine	C ₂₀ H ₂₄ ClN ₃ S	4.0	
5	Perfenazine	2-(4-(3-(2-chloro-10H-phenothiazin-10-yl)propyl)piperazin-1-yl)ethanol	C ₂₂ H ₂₆ ClN ₃ OS	3.48	
6	Triflupromazine	N,N-dimethyl-3-(2-(trifluoromethyl)-10H-phenothiazin-10-yl)propan-1-amine	C ₁₈ H ₁₉ F ₃ N ₂ S	4.62	
7	Haloperidol	3-(4-(4-chlorophenyl)-4-hydroxypiperidin-1-yl)-1-(4-fluorophenyl)propan-1-one	C ₂₀ H ₂₁ ClFNO ₂	3.21	

8	Trifluoperidol	1-(4-fluorophenyl)-3-(4-hydroxy-4-(4-(trifluoromethyl)phenyl)piperidin-1-yl)propan-1-one	C ₂₁ H ₂₁ F ₄ NO ₂	3.58	
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Protein-Ligand Docking Studies

Protein-ligand docking is used to check the structure, position and orientation of a protein when it interacts with small molecules like ligands. Protein-ligand docking aims to predict and rank the structures arising from the association between a given ligand and a target protein of known 3D structure. Protein-Ligand Docking module is further divided into different parts for user convenience like Receptor Preparation, Ligand Preparation, Binding Site Analysis, Dock and Analysis [14]. The protein-ligand docking was performed using Lamarckian genetic algorithm with default parameter [15].

Binding Site Analysis

Binding Site analysis is a fast detection program for 'the identification and visualization of possible binding sites and 'the distribution of surrounding residues in the active sites'. The centre of active site was chosen as grid map values for preparation of the grids. The spacing of grid was set to 1.00 Å and the no. of grid point were taken as 60 x 60 x 60 Å and protein-ligand docking was performed using Lamarckian genetic algorithm using default parameter [16]. The active sites were given in Table 2.

Table 2: It shows active sites and the centre of active sites of the receptor

S. No.	Name of active sites	Residues in active sites	Centre of active sites
1.	H1	ESGKSTDSLRTNRKAT	-17.124, -38.356, 2.487
2.	H2	GAVESCVLDYNRMHESM	-22.689, -48.087, 5.161
3.	H3	TFTDTSITKEIYVNN	-41.724, -44.176, -8.785
4.	H4	FRSREYQLNDSDDK	-10.448, -47.327, 6.608
5.	H5	GAVESGKSTRVTDVGG	-23.754, -37.533, 7.530
6.	H6	KGVTADTSNNLKDCG	-44.647, -36.860, -5.088

RESULTS

The analogues were successfully docked into the binding pocket. The binding energy was observed in the range of -6.55 to -8.56 Kcal/mol. The key result in a docking log file (DLG) are the docked structure or conformation found at the end of each run, the energies of these docked structures and their similarities to each other. The DLG file provides docked conformations, orientations and the binding energies. The similarity of docked structures is measured by computing the root-mean-square deviation (RMSD) between the coordinates of selected molecular conformation with the molecular

conformation having lowest interaction energy which is ranked on top. Clusters are created based on the comparison of conformations using RMSD values. The docking results consist of the PDBQT of the transformed 3D Cartesian coordinates of the ligand atoms as docked to the receptor molecule [13].

The binding energy of the selected ligands were plotted in the graph and from the graph (Fig. 2) the binding energy of all the active sites were observed among which the best ligand which shows better activity in all the active site was found to be Prochlorperazine. The aminoacids and the drug interactions were given in the Fig. 3 (a-h).

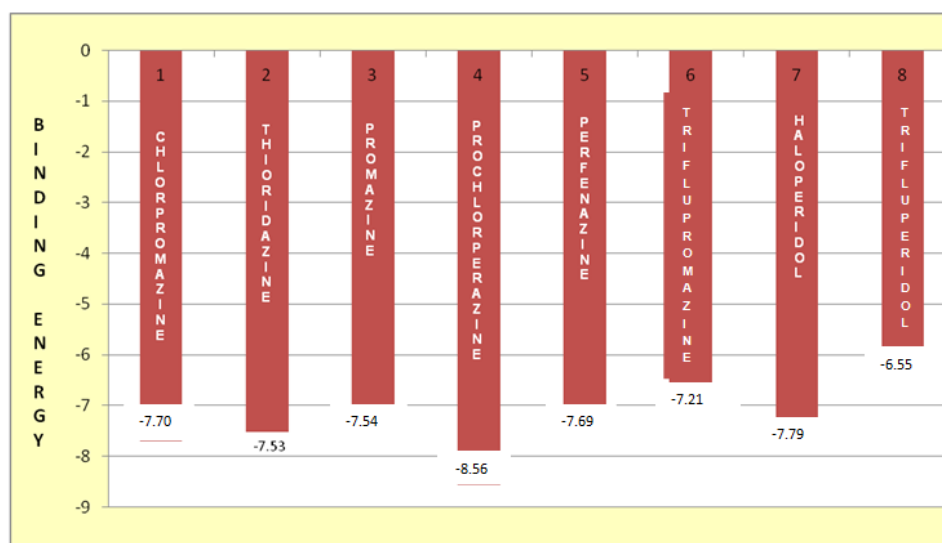
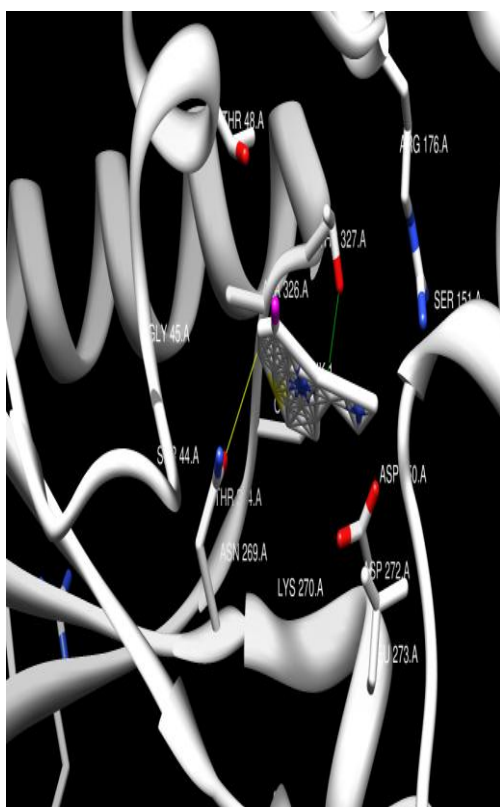
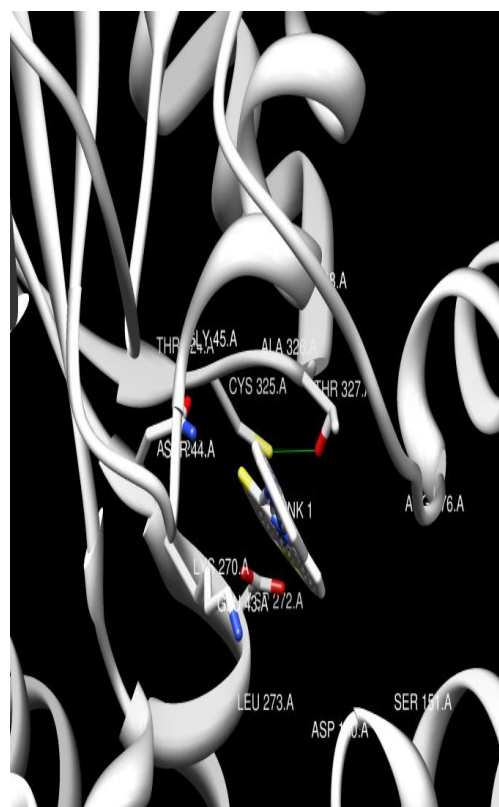


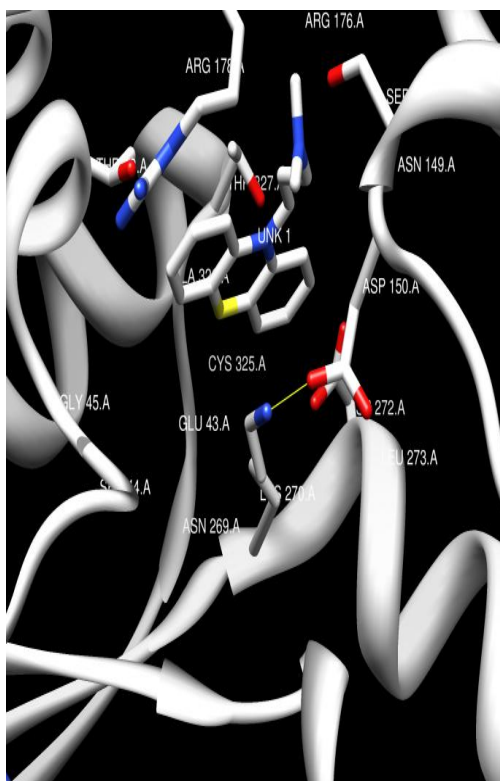
Fig. 2: It shows representation of Binding Energies of the drugs against D2 Receptor



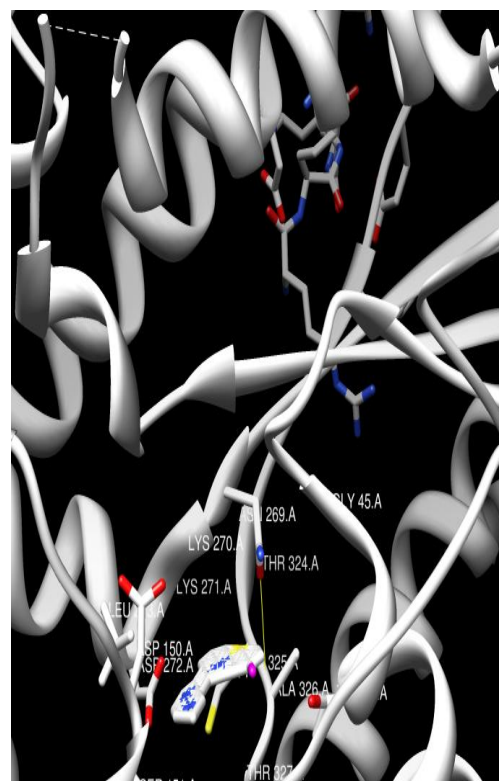
a) CHLORPROMAZINEeE



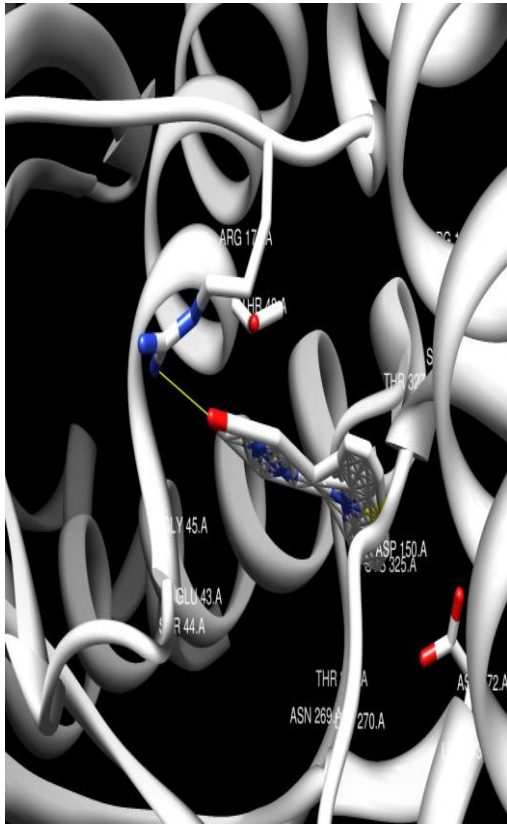
b) THIORIDAZINE



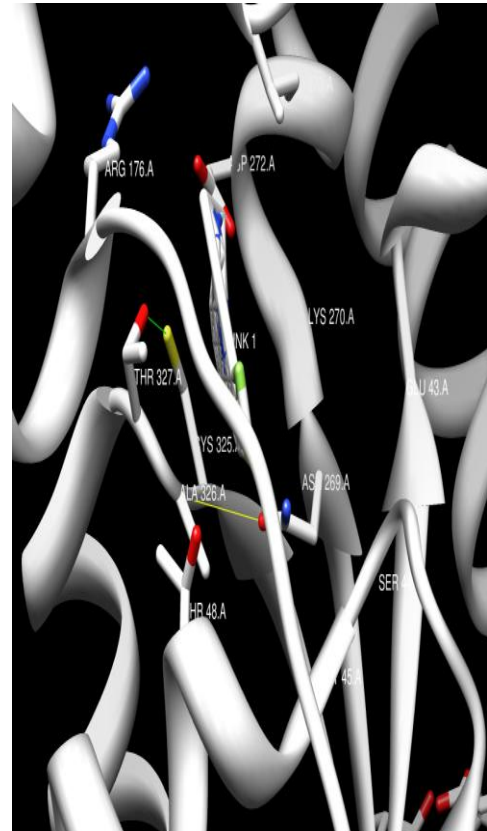
c) PROMAZINE



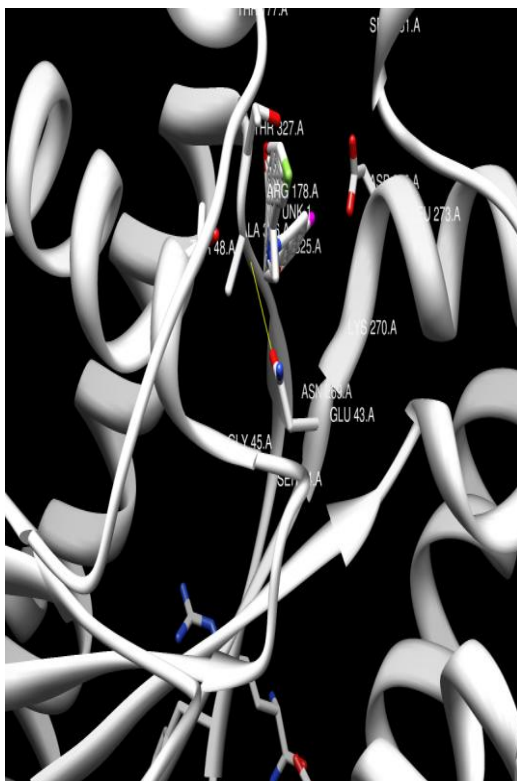
d) PROCHLORPERAZINE



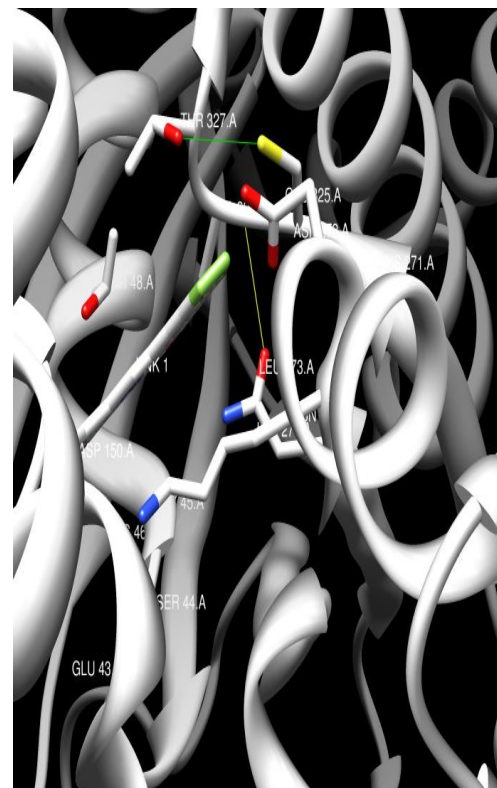
e) PERFENAZINE





f) TRIFLUPROMAZINE



g) HALOPERIDOL



h) TRIFLUPERIDOL

Fig. 3: It shows interaction of drugs against the protein 2HLB. The thin lines with colours   represent interacting hydrogen bonds between the protein and the drugs

DISCUSSION

Both generations of medication tend to block receptors in the brain's dopamine pathways, but compared to the typicals, the atypicals are less likely to cause extrapyramidal motor control disabilities in patient, which include unsteady Parkinson's disease-type movements, body rigidity and involuntary tremors[17]. Side effects vary among the various agents in this class of medications, but common side effects include: dry mouth, muscle stiffness, muscle cramping, tremors, EPS and weight-gain. EPS is a cluster of symptoms consisting of akathisia, parkinsonism, dystonias. Anticholinergics such as benztropine and diphenhydramine are commonly prescribed to treat the symptoms of EPS. 4% of patients develop the Rabbit syndrome while on typical antipsychotics [18]. The role of typical antipsychotics has come into question recently as studies have suggested that atypical antipsychotics may increase the risk of death in elderly patients. A retrospective cohort study from the *New England Journal of Medicine* on Dec. 1, 2005 showed an increase in risk of death with the use of typical antipsychotics that was on par with the increase shown with atypical antipsychotics [19]. A measure of "chlorpromazine equivalence" is used to compare the relative effectiveness of antipsychotics [20-21]. The measure specifies the amount (mass) in milligrams of a given drug that must be administered in order to achieve desired effects equivalent to those of 100 milligrams of chlorpromazine. Agents with a chlorpromazine equivalence ranging from 5 to 10 milligrams would be considered "medium potency", and agents with 2 milligrams would be considered "high potency"[22]. Prochlorperazine (Compazine, Buccastem, Stemetil) and Pimozide (Orap) are less commonly used to treat psychotic states, and so are sometimes excluded from this classification [23]. The current research paper is focussed on docking study of typical antipsychotic drugs with D2 receptor protein (PDB ID: 2HLB). On the basis of binding energy value, Prochlorperazine is found to be the most potent drug (Fig. 2). Prochlorperazine (Compazine, Stemetil, Buccastem, Phenotil) is a dopamine (D2) receptor antagonist that belongs to the phenothiazine class of antipsychotic agents that are used for the antiemetic treatment of nausea and vertigo. It is also a highly potent typical antipsychotic, 10-20 times more potent than chlorpromazine. It is also used to treat migraine headache. Hypersensitivities to Prochlorperazine can occur and there is cross-reactivity with other drugs in the phenothiazine class. Symptoms of a reaction include dyskinesia (unusual, uncontrollable body or face movements, including abnormal movements of the tongue, also known as tardive dyskinesia), seizures and seizure-like symptoms in individuals who have never had a seizure before. Long-term delay of medical treatment can lead to long-term effects. In extreme cases, it has been known to produce permanent damage to the lower jaw and the jaw joint due to extended seizure symptoms. Due to various side effect prochlorperazine is not clinically used as antipsychotic agent. In order to obtain an active neuroleptic derivatives, the hydrogen atoms attached to carbon C-2 and nitrogen N-10 atoms were substituted by different chemical groups, and structures of various Phenothiazines given in the literature contained at the N-10 position: piperazine, piperidine, or aliphatic side chain [10]. Depending on the structure of substituents in the side chain, the intensity of neuroleptic action of Phenothiazines could be ranked as follows: piperazine group > piperidine group > aliphatic chain [24]. The piperazine phenothiazines demonstrate the strongest antipsychotic action.

CONCLUSIONS

The least binding energy was found to be -8.56 Kcal/mol corresponding to the drug Prochlorperazine which establishes its maximum potency amongst the drugs under study. Keeping the above study under consideration, further modifications can be carried out taking Prochlorperazine as the reference of choice for better antipsychotic activity.

ACKNOWLEDGEMENTS

The authors gratefully acknowledge Bioinformatics Infrastructure Facility (BIF) funded by Department of Biotechnology, Govt. of India, at Centre for Studies in Biotechnology, Dibrugarh University.

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