Available online at www.derpharmachemica.com

Scholars Research Library

Der Pharma Chemica, 2015, 7(12):33-43
(http://derpharmachemica.com/archive.html)

ISSN 0975-413X
CODEN (USA): PCHHAX

Computer aided docking studies of indole derivatives as Hepatitis C NS5B polymerase inhibitor

Thayaillany Rajandran[1, 2], Radha Prabhu[1*] and M. Prabhu[1]

[1]Faculty of Pharmacy, Asia Metropolitan University, Selangor Darul Ehsan, Malaysia
[2]Faculty of Science, Technology, and Engineering La Trobe University, Bendigo, Australia

---

ABSTRACT

Hepatitis C is a liver disease caused by the Hepatitis C virus (HCV) and the infection has affected approximately180 million people around the world. Nonstructural protein 5B (NS5B) polymerase is a viral protein found in HCV and it plays a major role in the replication of the virus. Over the decade, it has been found that inhibition of the enzyme prevents the replication of the virus and thus treats the disease. In this study, molecular docking was performed on a series of 2-phenylindole derivatives by Autodock 4.2 into active sites of NS5B polymerase enzyme.

Keywords: Hepatitis C, NS5B polymerase, 2-phenylindole derivatives, Molecular docking.

INTRODUCTION

Previous studies over the decade have demonstrated that compounds with indole nucleus possess many therapeutic properties. This includes antimicrobial, anti-viral, antitubercular, anti-inflammatory, anticancer, anti-diabetic, anticonvulsant, antimicrobial, antioxidant, antidepressant activities [1, 2]. Nonstructural protein 5B (NS5B) polymerase is a viral protein found in HCV. NS5B polymerase plays a critical role in the replication of the virus. NS5B polymerase contains subunits that have additional roles during the infection process that are independent of RNA synthesis [3]. Therefore, this protein has become an attractive target in drug designing as inhibition of the protein prevents the virus from affecting normal cellular processes as well as inhibiting HCV RNA synthesis. Great variability is possible with the inhibitors of HCV as multiple allosteric binding sites are present on NS5B polymerase [4]. The enzyme has four binding sites which are non-nucleoside inhibitor (NNI) site I, II, III and IV. NNI site I and II are located in the thumb domain, while III and IV are closer to the active site in the palm domain. The upper section of the thumb domain, approximately 30Å from the active site at the juncture of the thumb and finger loop is the target of indole derivatives. This will interfere with conformational changes required for the formation of productive RNA enzyme complex thus inhibiting the elongation process [5].

MATERIALS AND METHODS

2.1 Preparation of protein molecule

Crystal structure of target protein was retrieved from Protein Data Bank (PDB ID: 3UPI). The protein was bounded with inhibitor 4,5-dihydrofurano indole. The ligand 4,5-dihydrofurano indole was removed from the target protein using protein data bank data base. The energy of the target protein was minimized using USCF Chimera. The protocol of USCF Chimera prepares the proteins by inserting missing atoms in incomplete residues, modeling

missing loop regions, deleting alternate conformations, removing water molecules, standardizing atom names, protonating titratable residues using predicted pKs.

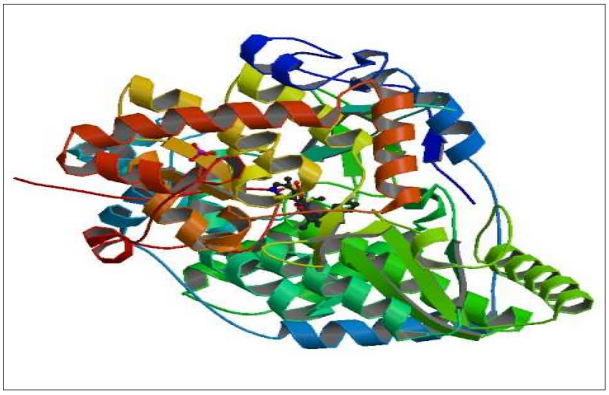


Figure 1: Ribbon structure of target protein (PDB ID: 3UPI)

2.2 Preparation of ligands

The structures of 2-phenylindole derivatives were selected from the literature S.Y.Liao et al., 2009. In this literature, 3D quantitative structure-activity relationship (QSAR) and docking studies have been carried out for 2-phenylindole derivatives with anticancer activity against human breast cancer cell line. These compounds were found to possess anticancer activity by preventing the polymerization of the α/β tubulin dimers to functional microtubules. The 2D structures of the selected ligands were drawn by ChemSketch. The 2D structure of the ligands was converted into 3D structure using Marvin Sketch. The 3D structure of the compounds will be saved in protein databank (pdb) format. The pdb format of the ligands will be viewed in USCF Chimera and the energy of the ligands will be minimized using USCF Chimera.

2.3 Validation of software

Software method validation was performed in Autodock 4.2 using the protein NS5B polymerase (PDB ID: 3UPI). The x-ray crystal structure of 3UPI was recovered from protein data bank. The co-crystallized ligand 4,5-dihydrofurano indole was redocked and the docked position was compared to the crystal structure position by calculating RMSD value.

2.4 Molecular docking

The ligand-flexible docking studies were performed using Lamarckian Genetic Algorithm of the Autodock 4.2 program [7]. This program was designed to predict how small molecules, such as substrates or drug candidates, bind to a receptor of known 3D structure. This was achieved through rapid grid based energy evaluation and efficient search of torsional freedom

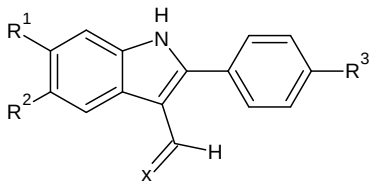
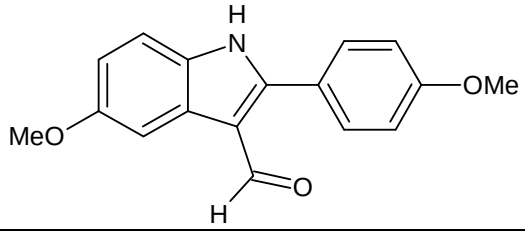
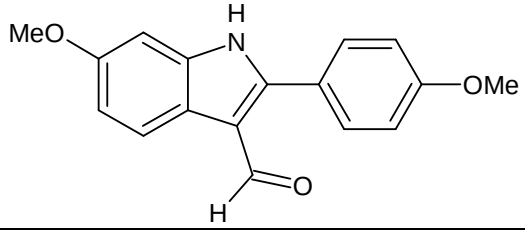
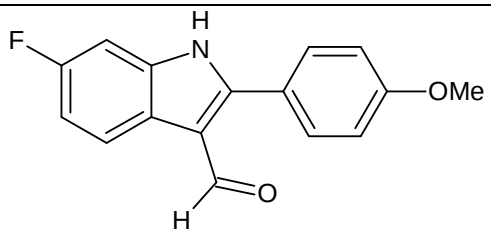
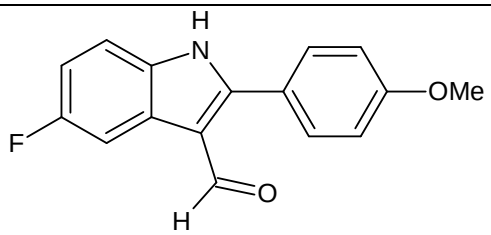
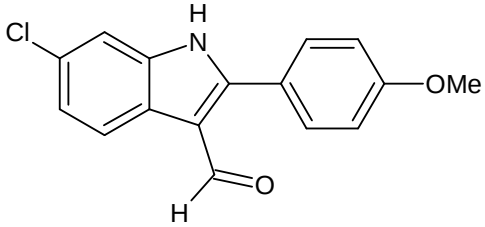
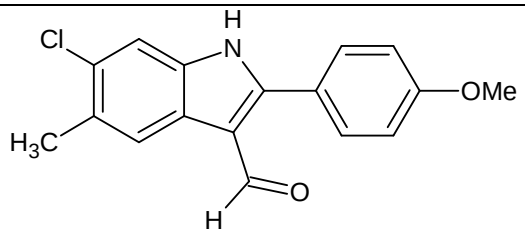
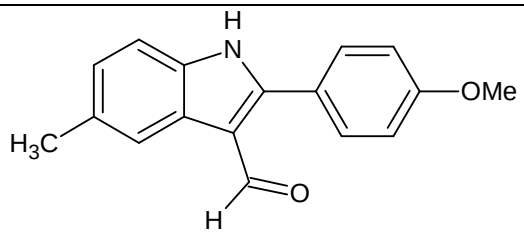
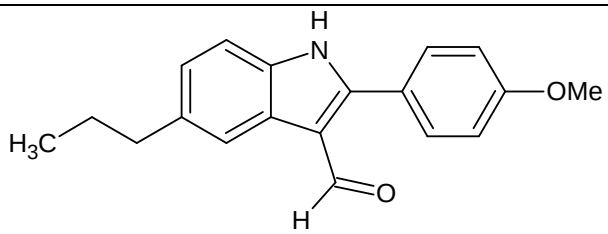
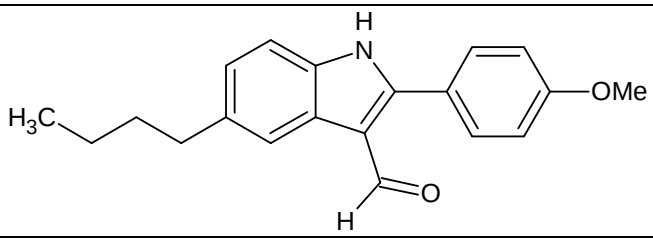
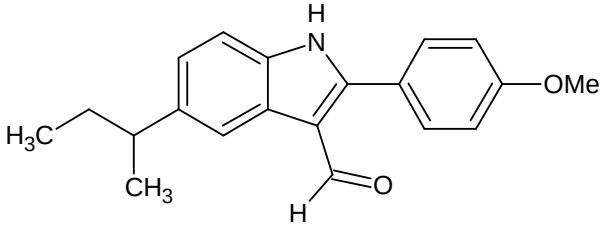
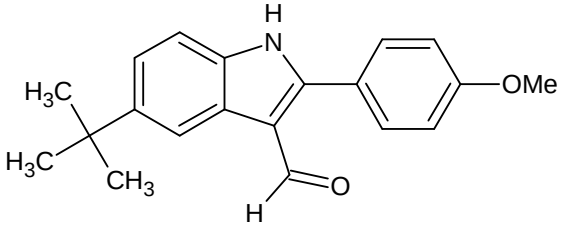
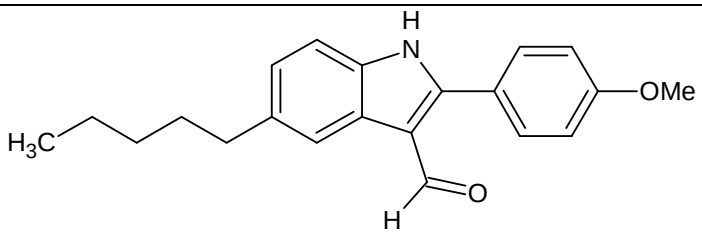
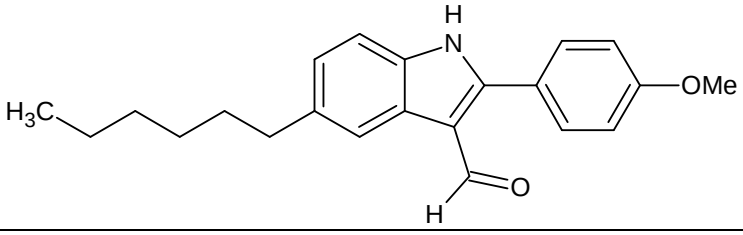
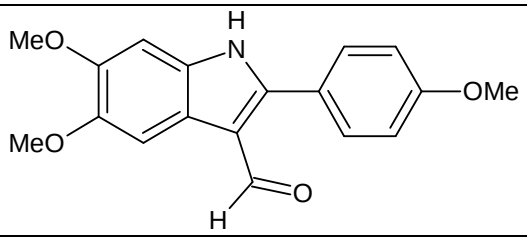
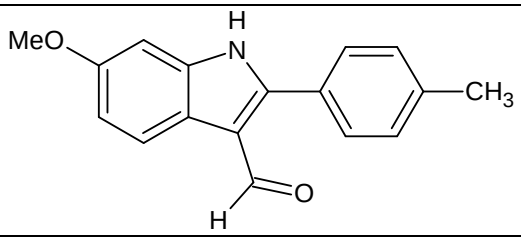


Figure 2: Molecular structure of 2-phenylindole

Table 1: List of Docked Compounds

No.	Compound ID	Structure of Compound	IUPAC Name of Compound
1.	1a		5-methoxy-2-(4-methoxyphenyl)-1H-indole-3-carbaldehyde
2.	2a		6-methoxy-2-(4-methoxyphenyl)-1H-indole-3-carbaldehyde
3.	3a		6-fluoro-2-(4-methoxyphenyl)-1H-indole-3-carbaldehyde
4.	4a		5-fluoro-2-(4-methoxyphenyl)-1H-indole-3-carbaldehyde
5.	5a		6-chloro-2-(4-methoxyphenyl)-1H-indole-3-carbaldehyde
6.	6a		6-chloro-2-(4-methoxyphenyl)-5-methyl-1H-indole-3-carbaldehyde

7.	7a		2-(4-methoxyphenyl)-5-methyl-1H-indole-3-carbaldehyde
8.	8a		2-(4-methoxyphenyl)-5-propyl-1H-indole-3-carbaldehyde
9.	9a		5-butyl-2-(4-methoxyphenyl)-1H-indole-3-carbaldehyde
10.	10a		5-(butan-2-yl)-2-(4-methoxyphenyl)-1H-indole-3-carbaldehyde
11.	11a		5-tert-butyl-2-(4-methoxyphenyl)-1H-indole-3-carbaldehyde
12.	12a		2-(4-methoxyphenyl)-5-pentyl-1H-indole-3-carbaldehyde
13.	13a		5-hexyl-2-(4-methoxyphenyl)-1H-indole-3-carbaldehyde

14.	14a		5,6-dimethoxy-2-(4-methoxyphenyl)-1H-indole-3-carbaldehyde
15.	15a		6-methoxy-2-(4-methylphenyl)-1H-indole-3-carbaldehyde

RESULTS

Table 2: Docking Results of Ligands

No.	Ligands	Ligand efficiency	Inhibition constant (uM)	Intermolecular energy	Van der Waals dissolution energy	Electrostatic energy	Total internal	Unbound energy
1.	1A	-0.42	1.65	-8.19	-8.22	-0.04	-0.12	-0.12
2.	2A	-0.36	8.28	-7.23	-7.27	-0.03	-0.08	-0.08
3.	3A	-0.36	11.02	-7.06	-7.09	-0.02	-0.11	-0.11
4.	4A	-0.37	6.32	-7.39	-7.43	-0.04	-0.12	-0.12
5.	5A	-0.36	8.7	-7.2	-7.11	-0.99	-0.12	-0.12
6.	6A	-0.37	4.26	-7.62	-7.54	-0.08	-0.12	-0.12
7.	7A	-0.42	1.61	-8.2	-8.24	-0.04	-0.12	-0.12
8.	8A	-0.41	10.94	-9.43	-9.45	-0.03	-0.23	-0.23
9	9A	-0.31	10.2	-8.0	-7.95	-0.05	-0.31	-0.31
10.	10A	-0.34	3.31	-8.37	-8.28	-0.09	-0.32	-0.32
11.	11A	-0.34	2.9	-8.15	-8.07	-0.08	-0.32	-0.32
12.	12A	-0.29	11.93	-8.21	-8.13	-0.08	-0.42	-0.42
13.	13A	-0.31	3.42	-9.25	-9.19	-0.06	-0.61	-0.61
14.	14A	-0.38	3.1	-7.81	-7.86	-0.05	-0.1	-0.1
15.	15A	-0.36	11.31	-7.05	-6.98	-0.07	-0.12	-0.12

Table 3: Binding energy, Interaction Residues, Hydrogen Bonds and Hydrogen Bond Distance of Ligands

No.	Ligands	Binding energy (kJ/mol)	Interacting residues	Hydrogen Bond	Hydrogen bond distance (Å)
1.	1A	-7.89	LEU 384, MET 414, PRO 197, PRO 417, TYR 383	NS5B:A:MET414:O	2.120
2.	2A	-6.93	ARG 200, LEU 384, MET 414, PRO 197, PRO 417, TYR 383, TYR 415	2A:UNKO:N1	2.732
3.	3A	-6.76	ARG 200, HIS 467, LEU 384, MET 414, PRO 197, PRO 417, SER 368, TYR 383, TYR 415	3A:UNKO:N1	2.647
4.	4A	-7.09	ARG 200, CYS 366, HIS 467, LEU 384, MET 414, PRO 197, PRO 417, SER 368, TYR 415	NS5B:A:MET 414:O	2.679
5.	5A	-6.9	ARG 200, ASN 411, CYS 366, GLY 410, MET 414, SER 368, SER 407, TYR 415, TYR 448	NS5B:A:TYR 415:HH1 NS5B:A:TYR 448:HN1	2.193 2.127
6.	6A	-7.33	ARG 200, ASN 411, CYS 366, GLY 410, MET 414, SER 368, SER 407, TYR 415, TYR 448	NS5B:A:TYR 415:HH1 NS5B:A:TYR 448:HN1	2.193 2.105
7.	7A	-7.9	ARG 200, HIS 467, LEU 384, MET 414, PRO 197, PRO 417, SER 368, TYR 383, TYR 415, VAL 201	NS5B:A:MET 414:O	2.63
8.	8A	-8.53	ARG 200, HIS 467, LEU 384, LYS 198, LYS 366, MET 414, PRO 197, SER 368, TYR 383, TYR 415, VAL 201	8A:UNKO:N1	2.532
9	9A	-6.81	ARG 200, ASN 316, ASN 411, CYS 366, GLN 446, GLY	NSB8:A:TYR 448:HN	2.029

			410, ILE 405, MET 414, SER 407, TYR 415, TYR 448		
10.	10A	-7.48	ARG 200, GLN 446, GLY 410, LEU 384, MET 414, PRO 197, SER 407, TYR 415, TYR 448	NS5B:A:TYR 448:HN	1.652
11.	11A	-7.56	ARG 200, GLY 410, LEU 384, MET 414, SER 407, TYR 415, TYR 448	NS5B:A:TYR 448:HN	1.725
12.	12A	-6.72	ARG 200, ARG 386, ARG 394, ASN 411, CYS 366, GLY 410, LEU 384, MET 414, SER 368, TYR 415	NS5B:A:ARG 386:HH11	2.225
13.	13A	-7.46	ARG 200, CYS 366, GLN 446, ILE 447, LEU 384, MET 414, PRO 197, SER 407, TYR 415, TYR 448	NS5B:A:TYR 448:HN	1.794
14.	14A	-7.51	ARG 200, LEU 384, MET 414, PRO 197, PRO 417, TYR 383, TYR 415, VAL 201	NO HYDROGEN BOND	-
15.	15A	-6.75	ARG 200, ASN 411, CYS 366, GLY 410, MET 414, SER 368, SER 407, TYR 415, TYR 448	NS5B:A:TYR 415:HH NS5B:A:TYR 448:HN	2.181 2.199

Figure 3: Interacting residues of ligand 6A with NS5B

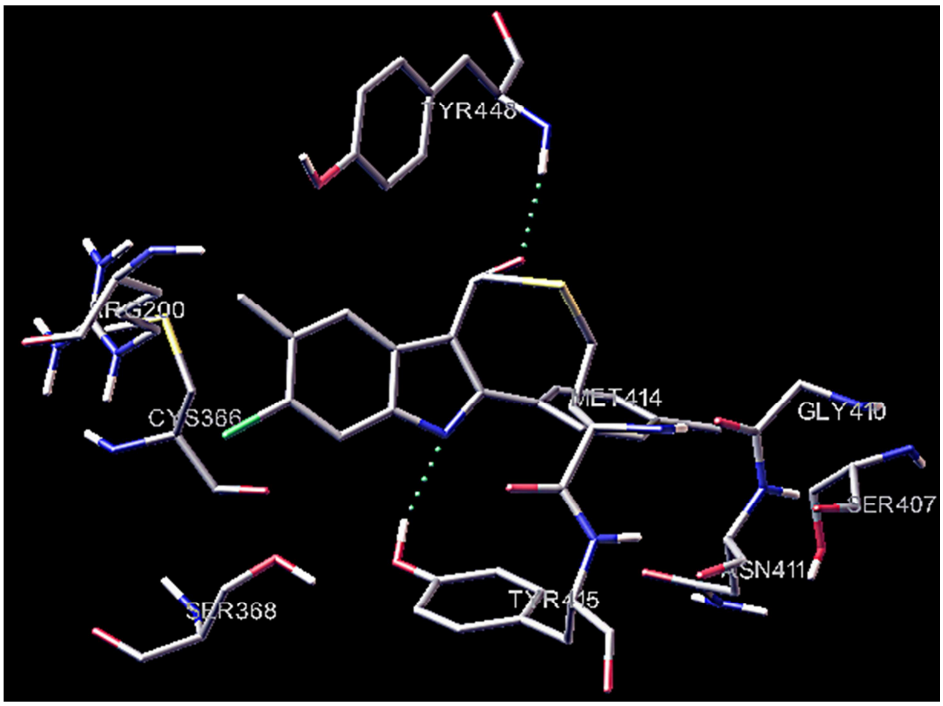


Figure 4: Surface view of ligand 6A with NS5B

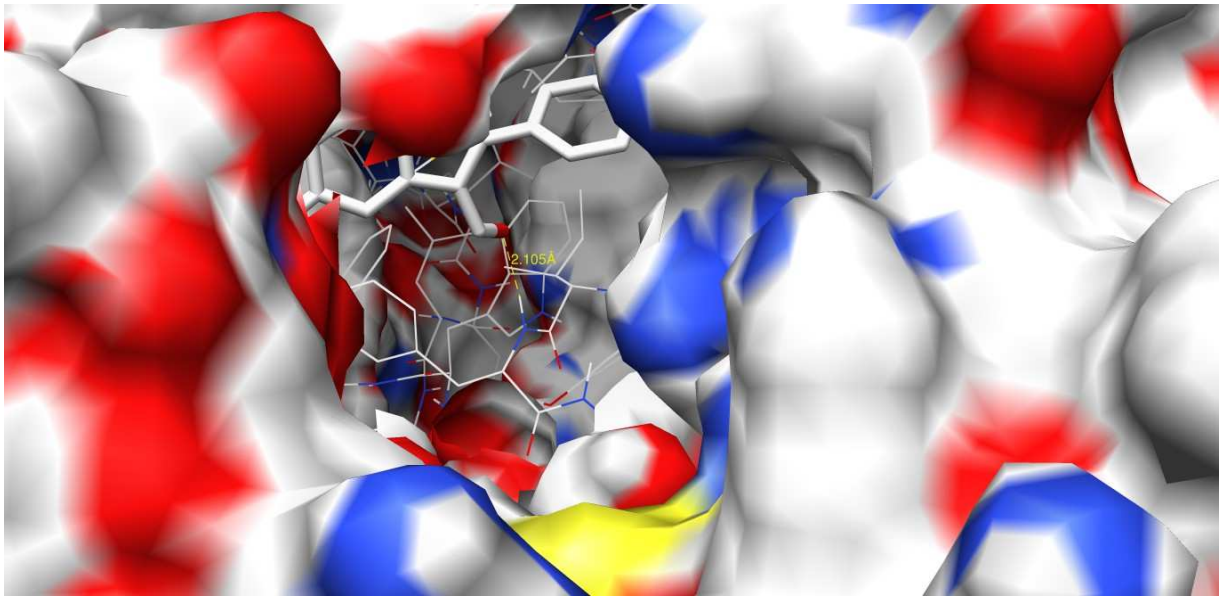


Figure 5: Interacting residues of ligand 7A with NS5B

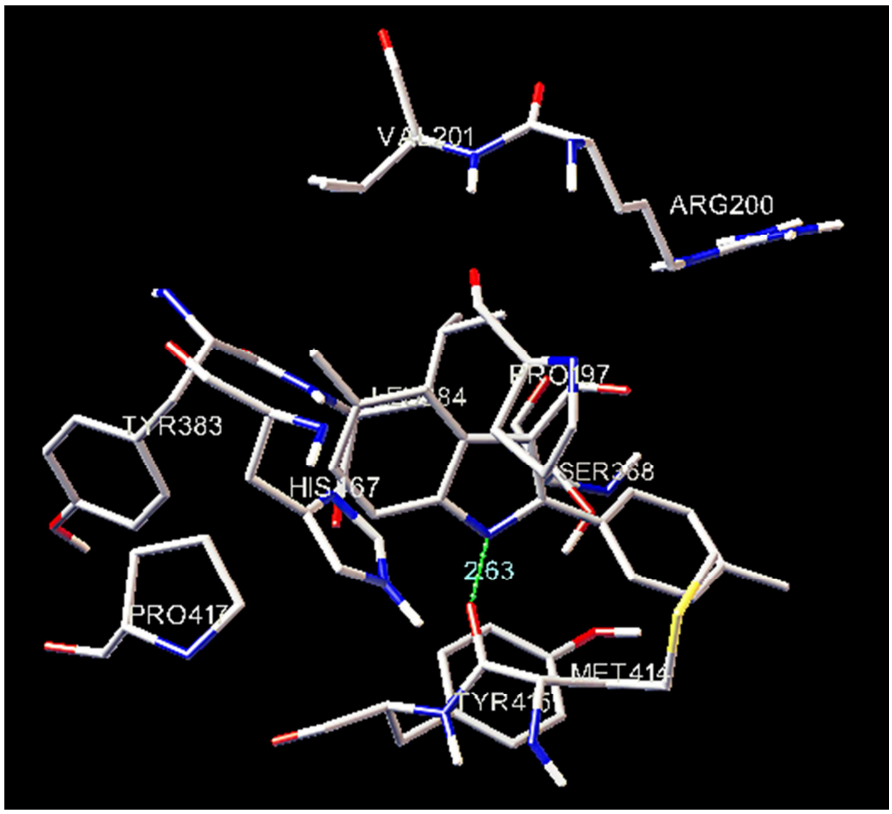


Figure 6: Surface view of ligand 7A complexed with NS5B

The binding energy of ligand 6A with NS5B polymerase enzyme is -7.33kJ/mol. The ligand efficiency of 6A is found to be -0.37 and the Van der Waals dissolution energy is -7.54. This ligand has an intermolecular energy of -7.62 with a total internal of -0.12. The inhibition constant is 4.26 µM. The docking of the ligand 6A with NS5B has 9 Van der Waals interacting residues at binding site which are ARG 200, ASN 411, CYS 366, GLY 410, MET 414,

SER 368, SER 407, TYR 415and TYR 448. Ligand 6A exhibits 2 hydrogen bond interactions with amino acids, TYR 415 and TYR 448 in the active site. The hydroxyl group of benzene ring in TYR 415 forms hydrogen bond with amino group of indole in ligand 6A with a hydrogen bond distance of 2.193Å. The second hydrogen bond is formed between oxygen at the 16th position of the ligand and the amino group of TYR 448 with a hydrogen bond distance of 2.105Å.

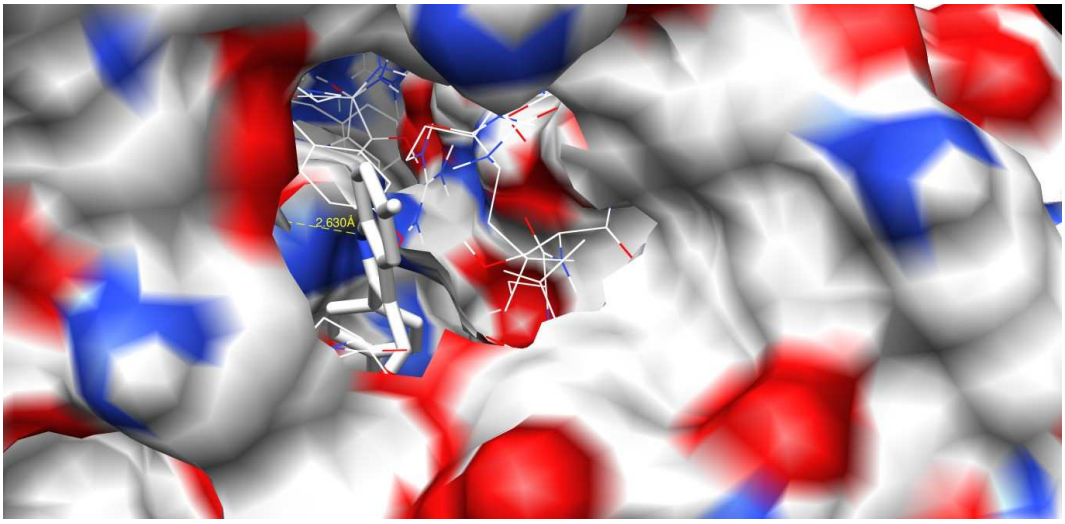
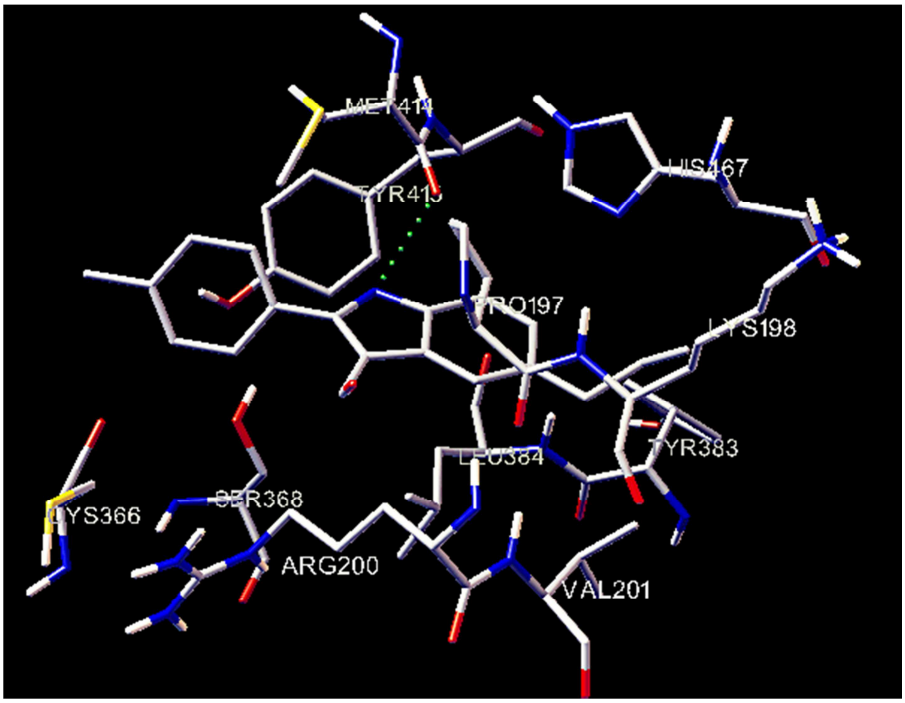


Figure 7: Interacting residues of ligand 8A with NS5B



The binding energy of ligand 7A with NS5B polymerase enzyme is -7.9kJ/mol. The ligand efficiency of 7A is -0.42 and the Van der Waals dissolution energy is found to be -8.24. This ligand has an intermolecular energy of -8.2 with a total internal energy of -0.12. The inhibition constant is 1.61 µM. The docking of the ligand with NS5B has 10 Van der Waals interacting residues which are ARG 200, HIS 467, LEU 384, MET 414, PRO 197, PRO 417, SER 368, TYR 383, TYR 415 and VAL 201. Ligand 7A exhibits a hydrogen bond interaction with amino acid, MET 414

in the active site. The hydrogen bond is formed between amino group of indole ring in the ligand and oxygen in the carboxyl group of amino acid with a hydrogen bond distance of 1.725Å.

Figure 8: Surface view of ligand 8A complexed with NS5B

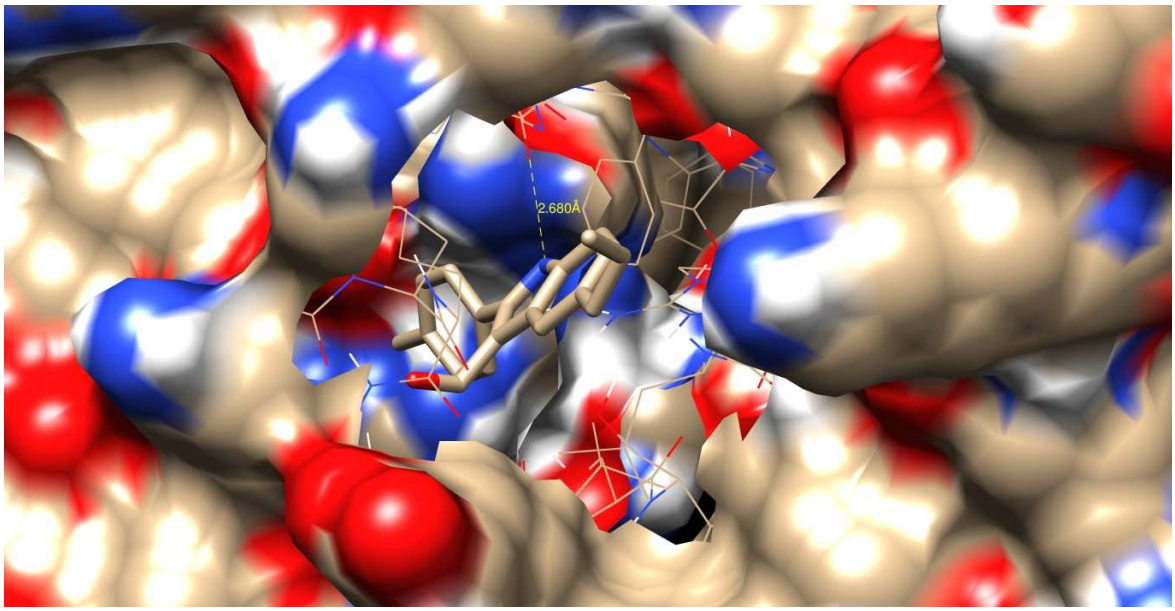


Figure 9: Interacting residues of Ligand 11 A with NS5B

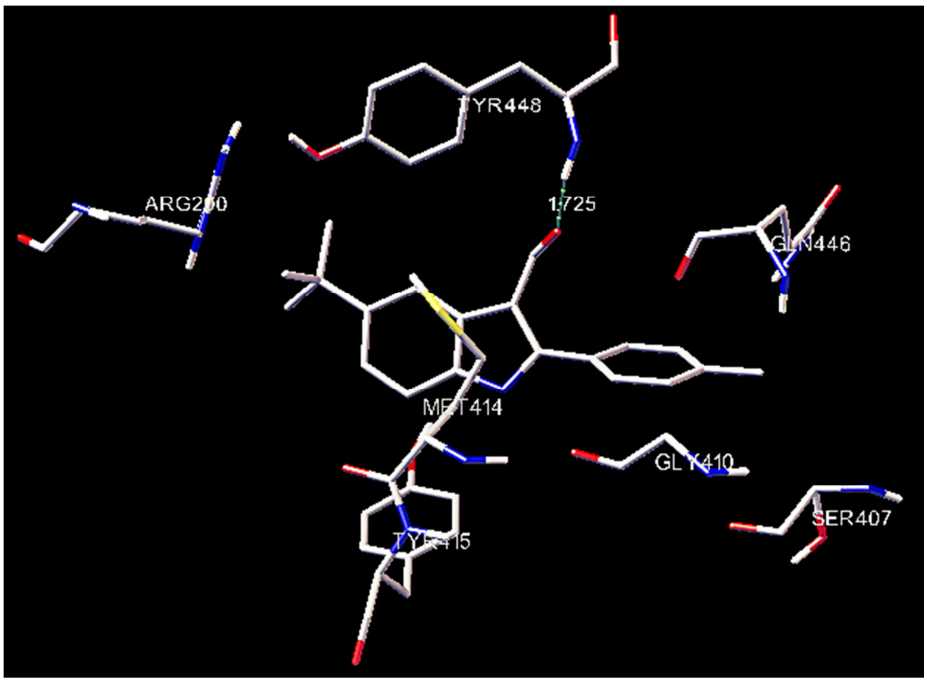
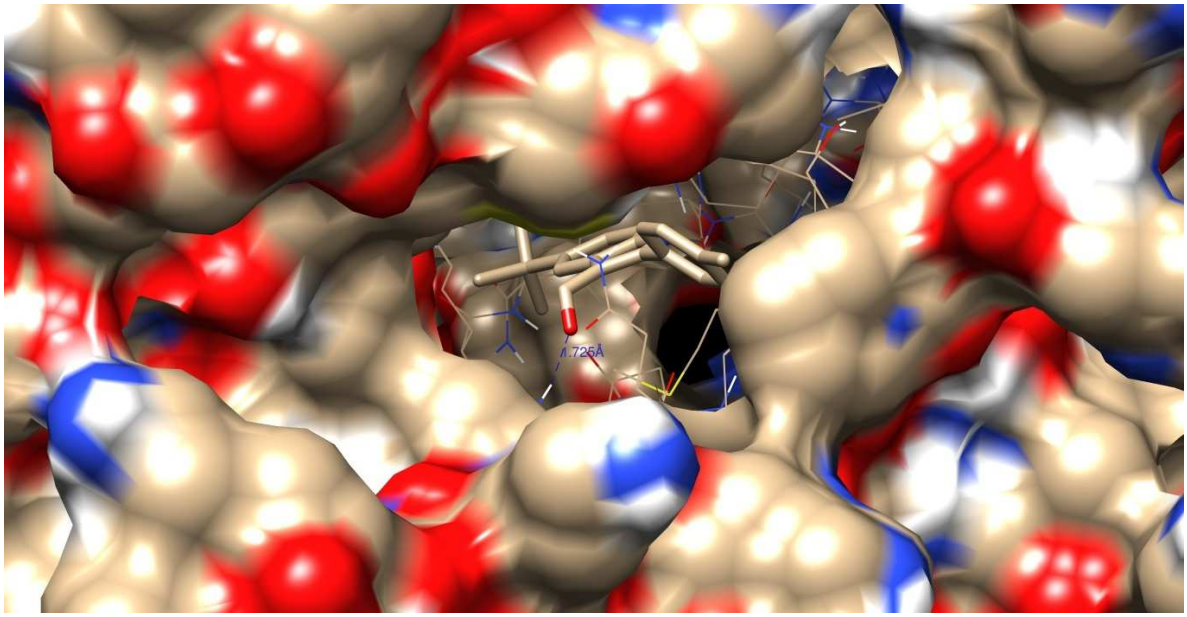


Figure 10: Surface view of Ligand 11 A with NS5B

The binding energy of ligand 8A with NS5B polymerase enzyme is -.8.53kJ/mol. The ligand efficiency of 8A is -0.41 and the Van der Waals dissolution energy is found to be -9.45. This ligand has an intermolecular energy of -9.43 with a total internal energy of -0.23. The inhibition constant is 10.94 µM. The docking of the ligand with NS5B has 11 Van der Waals interacting residues at binding site which are ARG 200, HIS 467, LEU 384, LYS 198, LYS

366, MET 414, PRO 197, SER 368, TYR 383, TYR 415, and VAL 201. Ligand 8A exhibits hydrogen bond interaction with amino acid MET 414 in the active site. The hydrogen bond is formed between amino group of indole ring in the ligand and oxygen in the carboxyl group of amino acid with a hydrogen bond distance of 2.680 Å.



The binding energy of ligand 11A with NS5B polymerase enzyme is -7.56kJ/mol. The ligand efficiency of 7A is -0.34 and the Van der Waals dissolution energy is found to be -8.07. This ligand has an intermol energy of -8.15 with a total internal energy of -0.32. The electrostatic energy of this ligand is -0.08 with the inhibition constant of 2.9 µM. The docking of the ligand with NS5B has 7 Van der Waals interacting residues at binding site which are ARG 200, GLY 410, LEU 384, MET 414, SER 407, TYR 415 and TYR 448. Ligand 11A exhibits hydrogen bond interaction with amino acid, TYR 448 in the active site. The hydrogen bond is formed between oxygen at the 16th position of the ligand and the amino group of TYR 448 with a hydrogen bond distance of 1.725Å.

DISCUSSION

Computational techniques are routinely used in modern drug design to help in understanding the drug-receptor interaction. It has been shown in many literatures that molecular docking can strongly support and help the design of novel, more potent inhibitors by revealing the mechanism of drug-receptor interaction. The present docking study is carried out for fifteen compounds against target protein NS5B polymerase enzyme (3UPI) within the active site which is MET 414. Table 2 and 3 show the binding energy and the inhibition constant of the selected ligands respectively. This docking study revealed that maximum number of the selected compounds has good binding energy with the target protein which range from -6.72 to -8.53 kJ/mol. The ligands 8A and 7A showed the highest binding energy of -8.53 kJ/mol and -7.9 kJ/mol, with ligand efficiency of 0.41 and 0.42 respectively. These molecules are completely surrounded by the amino acid residues in the active site pocket as shown in Figure 3 and Figure 4. However compound 14A did not form a hydrogen bond with the target protein although it has a high binding energy of 7.51Å. The residues in the active site which are involved in the hydrogen bonding of most of the ligands are MET 414, TYR 415 and TYR 448. Ligand 6A formed two hydrogen bonds with 2 residues in the active site which are TYR 415 and TYR 448 with hydrogen bond distance of 2.193Å and 2.105Å respectively and it has the highest binding energy compared to other ligands that formed 2 hydrogen bonds with the target protein.

CONCLUSION

Molecular docking study was performed in a series of 2-phenylindole derivatives as inhibitors of NS5B polymerase enzyme (3UPI). Almost all of the selected ligands showed good binding energy together with good inhibition constant. Of all the ligands docked, compounds 8A and 7A exhibit the highest binding energy which is -8.53kJ/mol

and 7.9kJ/mol respectively. Meanwhile compound 6A exhibits 2 hydrogen bonds with a high binding energy which is 7.33 kJ/mol. This study has provided a theoretical framework to drug design of 2-phenylindole derivatives as novel inhibitors of NS5B polymerase enzyme of Hepatitis C virus.

REFERENCES

[1] K.X. Chen, C.A. Lesburg, B. Vibulbhan, W. Yang, T.Y. Chan, S. Venkatraman et al., *J. Med. Chem*.,. **2012**, 55, 2089−2101.
[2] M.H. Powdrill, J.A. Bernatchez, M.Götte. *Viruses*, **2010**, 2, 2169–2195.
[3] B.K. Biswal, M.M. Cherney, M. Wang, L. Chan, C.G. Yannopoulos, D. Bilimoria, et.al., *J Biol Chem*., **2005**, 280, 18202-10.
[4] A.C. Anderson, *Chem. Biol*., **2003**, 10, 787–797.
[5] G. Varun, M. Lokesh, M. Sandeep, SajadShahbazi, G.Deepak Reddy. *Bangladesh J Pharmacol*, **2014**, 9, 290-297.
[6] S. Bressaneli, L. Tomei, A.Roussel, I. Incitti, R.L.Vitale, M. Mathieu, et.al., *Proc Natl Acad Sci USA*, **1999**, 96, 13034-39.
[7] S. Di Marco, C. Volpari, L. Tomei, S. Altamura, S. Harper, F. Narjes, *J. Biol. Chem*. **2005**, 280, 29765–29770.
[8] F. Pauwels, W. Mostmans, L. M. Quirynen, L. Van Der Helm, C.W. Boutton, et.al., *J Virol*, **2007,** 81, 6909–6919.
[9] G. Jin, S. Lee, M. Choi, S.Son, G.W. Kim, J.W. Oh, et.al., Eur. J. Med. Chem., **2014**, 75, 413-425.
[10] G.M. Morris, D.S. Goodsell, R.S. Halliday, R. Huey, W.E. Hart, R.K. Belew, et.al., *J Comput Chem*, **1998**, 19, 1639-1662
[11] Nagendra Kumar Kaushik, Neha Kaushik, Pankaj Attri, Naresh Kumar, Chung Hyeok Kim, Akhilesh Kumar Verma, and Eun Ha Choi. *Molecules*, **2013**, 20, 19907–19913.
[12] R. Dhani, A. Avinash, S. K. Salenaagina, M. V. Saicharan Teja, P. Masthanaiah, P. Raja Rathnam, V.Chandana Silpa, *J. Chem. Pharm. Res*., **2011**, 3, 519-523
[13] S.Y. Liao, L. Quan, T.F. Miao, H.L. Lu, K.C. Zheng. *Eur J Med Chem*, **2009**, 44, 2822-2827.