Available online at www.pelagiaresearchlibrary.com

Pelagia Research Library

Der Pharmacia Sinica, 2013, 4(1):88-92

Der Pharmacia Sinica
ISSN: 0976-8688
CODEN (USA): PSHIBD

Docking analysis of substituted pyrazoles as *M. tuberculosis* Enoyl ACP reductase inhibitors

Purnima. S.[1], Subashini. S.[2], Subhashini. N.[3] and Solairaj. P[4]

[1]*Dept.of.Pharmaceutical Chemistry, Grace College of Pharmacy, Kodunthirapully, Palakkad, Kerala, India*
[2]*Department of Plant Molecular Biology and Bioinformatics, Centre for Plant Molecular Biology and Biotechnology, Tamil Nadu Agricultural University, Coimbatore,Tamil Nadu, India*
[3]*Dept. of Pharmacology, S.B College of Pharmacy, Anaikuttam, Sivakasi, Tamil Nadu, India*
[4]*Dept. of Pharmaceutical Chemistry, S. B. College of Pharmacy, Anaikuttam, Sivakasi, Tamil Nadu, India*

ABSTRACT

Tuberculosis continues to be a major cause of morbidity and mortality all over the world. No new drug has been developed in the past 30 yr. Consequently, there is an urgent need to identify new drug targets in mycobacteria and eventually, develop new drugs. The enoyl acyl carrier protein reductase (ENR) from Mycobacterium tuberculosis is one of the key enzymes involved in the type II fatty acid biosynthesis pathway of M. tuberculosis. A series of pyrazoles linked with imidazoles were computationally designed and energy minimized. These ligands were investigated for drug like properties by calculating Lipinski's rule of five using molinspiration. These compounds were docked into the active site of ENR (PDB code, 2H7I) using Argus lab docking software which showed good affinity for the enzyme, when compared with the binging energies of standard drug isoniazid (-8.39kcal/mol.) Among all the designed ligands, the ligand 4 showed more binding energy values (-10.17kcal/mol). Further we planned to synthesise these derivatives and to screen for their anti mycobacterial activity.

Key words: Tuberculosis, ENR, Arguslab, Lipinski's rule of five

---

INTRODUCTION

Tuberculosis (TB) is a chronic infectious disease caused by mycobacteria of the ''tuberculosis complex'', which includes primarily *Mycobacterium tuberculosis*. Tuberculosis continues to be a major cause of morbidity and mortality throughout the world. Five decades of tuberculosis control programmes using potentially efficacious drugs and the availability of BCG vaccine, have failed to reduce the prevalence of infection in most parts of the world. WHO report says that India alone accounts for more than 25% of the world's incident cases [1].It has been reported that more than 3 billion people have been vaccinated with BCG, but still TB kills more than 50,000 people every week. It has been estimated that TB accounts for around 32 per cent deaths in HIV infected individuals [2].In spite of this, no new drug has been developed till now. Hence, it is an essential need to identify new drug targets in mycobacteria and in order to, develop new drugs.

The NADH-dependent enoyl- ACP reductase of the *Mycobacterium tuberculosis* is one of the important molecular targets. The target for the first line anti tubercular drug isoniazid (INH) is also the same [3]. Enoy l-ACP reductase (ENR) is a key regulatory enzyme in fatty acid elongation, and it catalyses the NADH-dependent stereo specific reduction o f α, β -unsaturated fatty acids bound to the acyl carrier protein [4]. Inhibition of ENR disrupts the biosynthesis of the mycolic acids that are central constituents of the mycobacterial cell wall [5].

Thus, we are claiming for a new class of anti tubercular agent with a different mode of action and it has led medicinal chemists to explore a wide variety of chemical structures. Whereas many reports [6-10] were proved the emergence of pyrazoles as potent antitubercular agents. Imidazole derivatives are known to possess antitubercular, antifungal, anti-neoplastic activities [11-14].

This initiated the construction of our compounds containing both the pyrazole and imidazole ring systems in the same matrix to serve as a new scaffold towards the development of novel antimycobacterial agents. Accordingly we planned to link pyrazoles and imidazoles to produce better anti tubercular agents and to evaluate the interactions with the target by using ARGUS LAB docking software.

MATERIALS AND METHODS

In the present study biological databases like PDB [15] (Protein Data Bank) and bioinformatics software like Argus lab [16], ACD ChemSketch [17], Online servers like Molnspiration[18] were been used. The Protein Data Bank (PDB) is a repository for the 3-D structural data of large biological molecules, such as proteins and nucleic acids. The data, typically obtained by X-ray crystallography or NMR spectroscopy and submitted by biologists and biochemists from around the world, are freely accessible .The structure of enoyl acyl carrier protein synthase was retrived from Brookheaven Protein Data Bank with the PDB Id : 2H7I.

Using Chemsketch the structures of these ligands were sketched. These Ligands were taken for analysing its druglikeness properties. Drug likeliness index leads to the ideal procedure of rejection nonviable lead compounds before they are even synthesized. One of the traditional rules is Lipinski's rule of five. For calculating Lipinski's rule of five we have used Molinspiration server. Molinspiration is an online server used to calculate the molecular properties of the ligands. Molinspiration tools are written in Java, therefore can be used practically on any computer platform.

Molecular docking is a method to predict the preferred orientation of one molecule to a second when bound to each other to form a stable complex. Computers and programs (software) are often used to predict or simulate the possible reaction and/or interactions between two molecules based on their three dimensional structures. This method can be used to predict possible binders or inhibitors and as well to predict the association between the molecules [19]. ArgusLab is a molecular modeling, graphics, and drug design program for Windows operating systems. Argus Lab is the electronic structure program that is based on the quantum mechanics, it predicts the potential energies, molecular structures; geometry optimization of structure, vibration frequencies of coordinates of atoms, bond length, bond angle and reactions pathway [20].

RESULTS AND DISCUSSION

Druglikeness properties of the ligands, the binding energies of the ligands and the Enoy l-ACP reductase (2H7I) are reportedin table 1& 2.

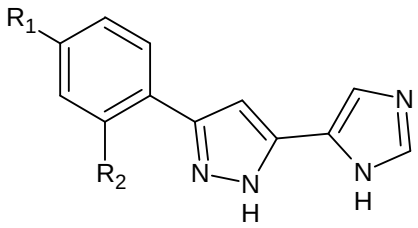


Table1: Druglike properties of the designed ligands

S.No	Compound code	R_1	R_2	Log P	M.Wt	Hydrogen acceptors	Hydrogen donors	No.of violations
1	Ligand 1	Cl	H	2.209	244.685	4	2	0
2	Ligand 2	Br	H	2.341	289.136	4	2	0
3	Ligand 3	F	H	1.695	228.23	4	2	0
4	Ligand 4	Cl	Cl	2.815	279.13	4	2	0
5	Ligand 5	NH_2	H	0.607	225.255	5	4	0
6	Ligand 6	NO_2	H	1.49	255.37	7	2	0
7	Ligand 7	OH	H	1.052	226.239	5	3	0
8	Ligand 8	OH	OH	0.761	242.238	6	4	0

In vivo absorption capabilities of the designed molecules were tentatively assessed by means of Lipinski's rule of five, which predicts that a compound administered orally, will more likely have a good absorption or permeation. All of them satisfies the rule; which indicates all the ligands1-8 have good absorption. So, the ligands 1-8 can be taken for the docking study.

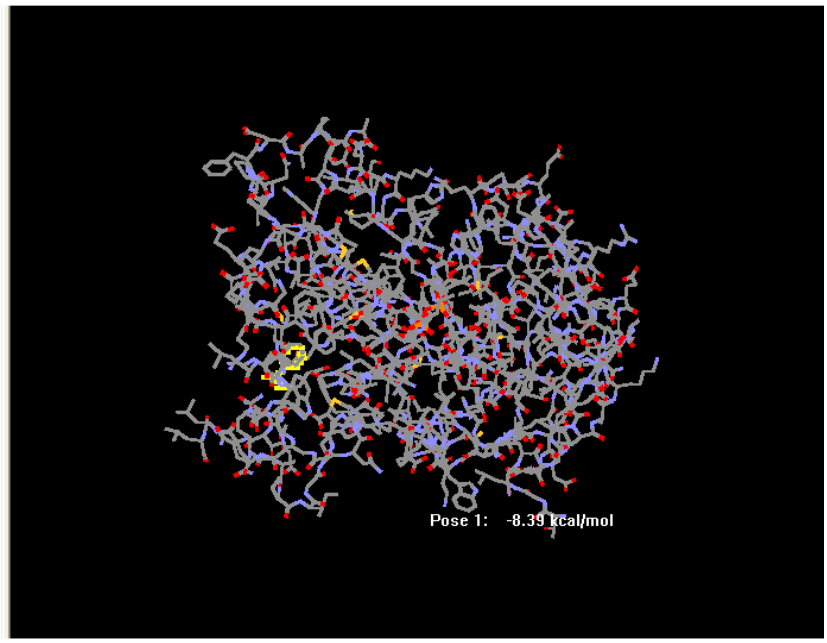
Docking results between ENR (2H7I) enzyme and designed ligands are reported in Table 2

Table 2: Docking results of 2H7I with the ligands1-8

S. No	Compound code	Binding energy (Kcal/mol)
1	Ligand1	-9.67
2	Ligand2	-9.52
3	Ligand3	-8.94
4	Ligand4	-10.17
5	Ligand5	-8.61
6	Ligand6	-8.47
7	Ligand7	-7.90
8	Ligand8	- 8.01
9	ISONIAZID	-8.39

.

Fig 1: Interaction and binding energy of Isoniazid with 2H7I

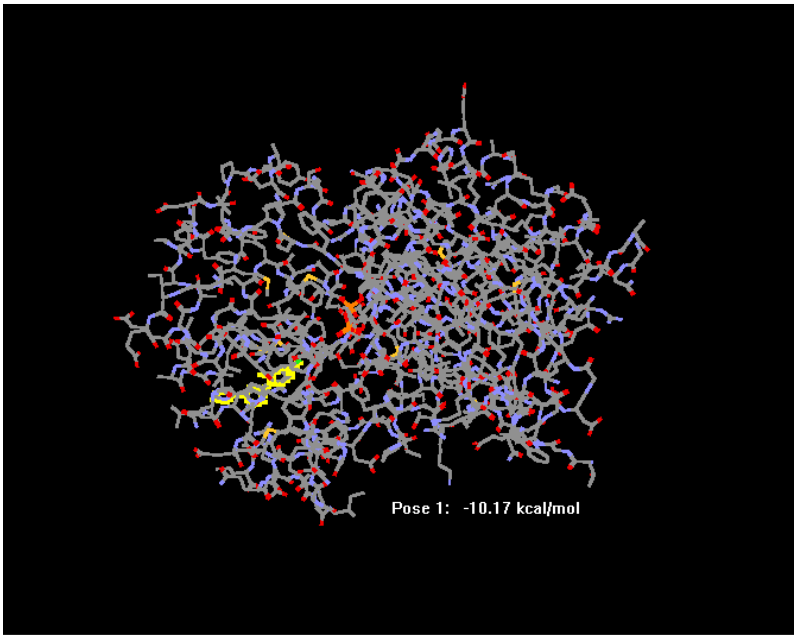


Based on the literature, it has been shown clearly that pyrazoles and imidazole can be used to target enoyl acyl carrier protein reductase. The energy values were calculated using Argus lab.

Grid box size X: 18.75A^0, Y:20 A^0, Z:20 A^{0}Grid resolution: 0.4 A^{0}Flexible docking mode was chosen for docking. Binding sites Gly96, Phe97, Ile102, Met103, Phe149, Met155, Pro156, Ala157, Tyr158, Met161, Pro193, Met199, Val203, Ile215, Leu218 and Trp222 were chosen. The standard anti-tubercular agent isoniazid on docking with enoyl acyl carrier protein reductase produced the energy value of -8.39 kcal (Figure 1).

Ligand 4 showed a decreased in energy values (-10.17 kcal) (Figure 2) which means it was more compatible with the receptor than the standard and other designed ligands. Ligand 4 containing chorine at the 4th position of aromatic ring showed better binding nature, which resulted in decrease in binding energy

Fig 2: Interaction and binding energy of Ligand 4 with 2H7I



CONCLUSION

The enoyl acyl carrier protein reductase (ENR) from *Mycobacterium tuberculosis* is one of the key enzymes involved in the type II fatty acid biosynthesis pathway of *M. tuberculosis*. Pyraole linked imidazoles were designed and their druglikeliness scores showed that it can be docked. These ligands were docked with ENR using Arguslab. With the standard isoniazid binding energy was -8.39kcal/mol. While ligand 4 showed a drastic decrease in the binding energy, i.e., -10.17 kcal/mol. So it can be concluded that the designed compounds can be potent anti mycobacterial agent. In future research work we planned to synthesis these pyrazole derivatives and screen for their *in-vitro* anti mycobacterial activity

REFERENCES

[1] www.searo.who.int/linkfiles/tuberculosis who-tb-report-**2012**.pdf
[2] Puneet Chopra, L.S. Meena, Yogendra Singh, *Indian J Med Res*, **2003**, 117, 1.
[3] A. Banerjee, E. Dubnau, A. Quemard, V. Balasubramanian, *Science*,**1994**, 263, 227.
[4] Suvarna. G. Kini, Anilchandra.R.Bhat, Byron Bryant, John.S.Williamson, Franck.E. Dayan, *European Journal of Medicinal Chemistry*, **2009**,44, 492.
[5] Xin He, Akram Alian, Paul R Ortiz de Montellano, *Bioorganic & Medicinal Chemistry*, **2007**, 15, 6649.
[6] Mack.R.Kuo, Hector.R.Morbidoni, David Alland, Scott.F.Sneddon, Brian.B.Gourlie, Mark.M. Stavesk, *The Journal Of Biological Chemistry*, **2003,** 278, 23, 20851.
[7] Joseph.A.Maddry, Subramanian Ananthan, Robert.C.Goldman, *Tuberculosis*, **2009**, 1.
[8] D.H.Vyas, M.F. Dhakuk, S.Tala, J.D. Akbari, H.S. Joshi, *Indian Journal of Heterocyclic Chemistry*, **2007**, 16**,** 169.

[9] H.M.Kanjariya, T.V. Radhakrishnan, Ramachandran, P.Hansa, *Indian Journal of Chemistry*, **2004**, 43B, 1569.
[10] Marco radi, Vincenz O Brenardo, Beat Rice, Bechi Daniele, *Tetrahydron letters*, **2009**, 50, 6572.
[11] Preeti Gupta, Shahul Hameed, Rahul Jain, *European Journal of Medicinal chemistry*, **2004**, 39, 805.
[12] Jyoti Pandey, Vinod. K. Tiwari, Shyam.S.Verma, S.Vinita Chaturvedi, *European Journal of Medicinal Chemistry*, **2009**, 44, 3350.
[13] Afshin Fassihi, Zahra Azadpour, Neda Delbari , Lotfollah Saghaie *et al*, *European Journal of Medicinal Chemistry*, **2009**, 44, 3253.
[14] Sang-Ho Lee; Suhyun Kim; Min-Han Yun; Yong Sup Lee *et al*, *Bioorganic & Medicinal Chemistry Letters*, **2011**, 21, 1515.
[15] Research Collaboratory for Structural Bioinformatics. Protein Data Bank. [Internet]. **2011** [updated 2011 Mar 22; cited **2011** Mar 26]. Available from: http://www.pdb.org/
[16] ArgusLab 4.0.1, Mark A. THOMPSON, Planaria Software LLC, Seattle, WA, Downloaded from http://www.arguslab.com
[17] Downloaded from http://www.acdlabs.com/resources/freeware/chemsketch/
[18] Molinspiration Cheminformatics. Molinspiration. [Internet]. **2010** [cited 2011 Mar 26]. Available from: http://www.molinspiration.com/cgi-bin/properties.
[19] http://www.scribd.com/doc/8931374/Introduction-to-Molecular-Docking
[20] C. Peng, P.Y. Ayali, H.B. Schlegel, M.J.Frisch, *J. Comp. Chem*, **1995**, 16,49.