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VIRTUAL SCREENING FOR CHALCONE HIT AND IN VITRO BIOLOGICAL SCREENING AS A POTENT ANTI-TUBERCULAR MOLECULE

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Abstract: The fundamental topic in the current exploration work is to discover an intense adversary of tubercular movement for the "chalcone hit" which quell the pharmacological activity created by reactant activity of enoyl-[acyl-transporter protein] reductase (PDB.ID: 2X23) which is a X-ray crystallographic structure, utilized in the silico screening measure, in which the subsidiary of the chalcone were elucidated utilizing PubChem information base. The best hit chalcones were dissected utilizing in vitro bioassay for their objective restricting properties as an adversary of tubercular activity. A "chalcone hit" which is a tentatively bioactive compound has been perceived as compound (E)- 3-(4-hydroxyphenyl)- 1-(2,4,6-trihydroxyphenyl)prop-2-en-1-one.
Keywords: In silico screening, Molecular docking, Chalcone, anti-tubercular agent

摘要：当前勘探工作的基本课题**是**发现“查尔酮**命中”的结核运动的强烈**对手，该“查耳酮**命中”**可抑制**烯酰-[酰**基-转运蛋白]还原酶（PDB.ID：2X23）的反应物活性**产生的药理活性。）这**是一种 X 射线晶体结构，用于硅**筛选**措施，其中使用 PubChem 信息**库阐明查耳酮的附属物。最好的**查尔酮**是利用体外生物**测定法解剖的，因为**它们具有作**为结核活性**对手的客观限制特性。作为暂时生物活性化合物的“查耳酮命中”已被**认为**是化合物**(E)-3-(4-羟基苯基)-1-(2,4,6-三羟基苯基)prop-2-en-1-one。
关键词：计**算机**筛选，**分子**对**接**，查耳酮，**抗**结**核**药物

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INTRODUCTION

Tuberculosis is today among the overall wellbeing dangers. As safe strains of Mycobacterium tuberculosis have gradually arisen, treatment disappointment is time and again a reality, particularly in nations coming up short on the essential medical care association to give the long and expensive therapy adjusted to patients. In view of absence of treatment or absence of adjusted treatment, in any event 2,000,000 individuals will bite the dust of tuberculosis this year. Because of this worry, this irresistible sickness was the focal point of re-established logical interest somewhat recently. Regimens were streamlined, and much was learnt on the instruments of activity of the ant tuberculosis drugs utilized. Besides, the journey for unique medications beating a portion of the issues of ebb and flow regimens additionally turned into the focal point of examination programs and numerous new arrangement of M. tuberculosis development inhibitors were accounted for.

Tuberculosis (TB) is a typical and destructive irresistible infection brought about by different strains of Mycobacterium tuberculosis in people. It is a worldwide medical issue slaughtering around 3 million individuals each year around the world. Patterns in the occurrence of TB along with the improvement of multi drug safe (MDR) and broadly drug safe (XDR) strains of M. tuberculosis and HIV co - contamination raises the need to heighten the quest for more productive medications to battle this infection. With the rising pervasiveness of microorganisms showing obstruction towards current enemy of tubercular medications, there is a critical need to grow new antimicrobial mixtures. Therapeutic plants offer an incredible desire to defeat these requirements in light of their substance variety and their huge job in the medication locating and advancement and furthermore perceived as a helpful wellspring of exceptionally dynamic antimycobacterial metabolites. Prior writing additionally reports the counter mycobacterial action of numerous classes of regular items: like steroids, phenolic compounds, acetogenic quinines, flavonoids and triterpenes.

Based on reported literature, the compounds which consisting chalcone moiety in its basic structure exhibited *in vitro* binding against enoyl-[acyl-carrier protein] reductase (InhA), hence we proposed worthwhile to predict the binding energy of the compounds E)-3-(3-nitrophenyl)-1-(3,4,5-trimethoxyphenyl)-2-propen-1-one (Pub Chem CID 5730821), (E)-3-(3-nitrophenyl)-1-(3,4,5-trimethoxyphenyl)-2-propen-1-one which were synthesized in the present study against InhA protein drug target. However, the role of InhAprotein target has been well established in the pathogenesis of Tuberculosis (TB).

AIM AND OBJECTIVES

The aim of the current research is to determine the virtual screening score of the identified beat chalcone hit and to screen the anitubercular activity of synthesized chalcone.

ENOYL-[ACYL-CARRIER PROTEIN] REDUCTASE (InhA).

Enzymes, InhA have been proposed as targets for INH. The enzyme is a type II dissociated fatty acid biosynthesis pathway (FASII) in *Mycobacterium tuberculosis*, consistent with the observation that INH interferes with the biosynthesis of mycolic acids, very long chain

fatty acid components of the mycobacterial cell wall. InhA, an enoyl reductase that catalyzes the NADH-dependent reduction of long chain *trans*-2-enoyl-acyl carrier proteins (ACPs), was first identified as a target by Jacobs and coworkers. InhA is inhibited by INH. The X-ray crystal structure was obtained from the Brookhaven Protein Data Bank (http://www.rcsb.org/pdb) (PDB.ID: 2X23).

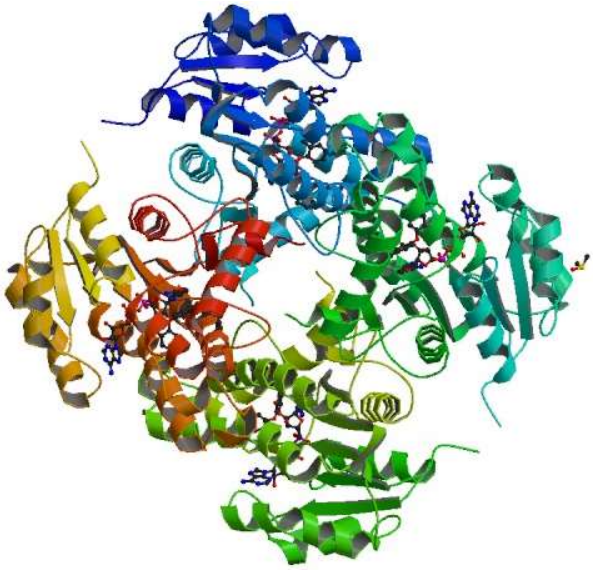

Figure 1 :Xray crystallographic InhA protein-ligand complex (PDB.ID: 2X23)

MATERIALS AND METHODS

COMPUTATIONAL HARDWARE REQUIREMENTS

The minimum central hardware system configuration includes Intel (R) Core (TM) 2Duo Central Processing Unit (CPU), 2.5 GHz, 1 TB hard disk, 2 KV Power Backup, WinXP or higher operating system was used for running selected computer aided drug discovery softwares. All softwares were well compatible with the selected system configuration

COMPUTATIONAL SOFTWARE REQUIREMENTS

Computational drug discovery softwares along with graphical user interface (GUI) were utilized for the study which includes Accelrys Draw for molecular modeling, ArgusLab v 4.0 for energy minimization, and iGemdock v 2.1 molecular docking simulation protocols.

X-RAY CRYSTALLOGRAPHIC STRUCTURES OF VALIDATED DRUG TARGETS

X-ray crystallographic data of validated antitubercular drug targets ligand binding domains (LBD) were obtained from Brookhaven protein data bank (http://www.rcsb.org/pdb). The protein data bank codes were specified in the literature.

GENERAL PROCEDURE FOR THE LIGAND PREPARATION

The chemical structures of the selected ligands were initially modeled as 2D chemical structures using Accelrys Draw software and transformed into 3D chemical structures using Open Babel software and subjected for energy minimization using ArgusLab v 4.0 software. The minimization was executed until the root mean square gradient value reached a value smaller than 0.0001 kcal/mol. Such energy minimized structures were considered for molecular docking studies using iGemdock v 2.1 software. The corresponding docking engine compatible 'MDL MOL' file format has been adapted to ligand by using integral option (save as /MDL MOL).

GENERAL PROCEDURE FOR THE SOFTWARE VALIDATION

iGEMDOCK v 2.1 software validation was performed by using X-ray structure deposited with co-crystallized ligand was obtained from the

Brookhaven Protein Data Bank (http://www.rcsb.org/pdb). The Root Mean Square Deviation (RMSD) between the X-ray co-crystallized ligand and docked conformation was 1.70Ao indicated that the parameters for docking simulation was good in reproducing X-ray crystal structure.

RESULTS& DISCUSSION

ANTITUBERCULAR DRUG TARGET

Chalcones with E)-3-(3-nitrophenyl)-1-(3,4,5-trimethoxyphenyl)-2-propen-1-one (Pub Chem CID 5730821), (E)-3-(3-nitrophenyl)-1-(3,4,5-Trimethoxyphenyl)-2-propen-1-one with their GEMDOCK scores against validated anti-tubercular protein drug target.

Tuberculosis is today amongst the worldwide health threats. As resistant strains of *Mycobacterium tuberculosis* have slowly emerged, treatment failure is too often a fact, especially in countries lacking the necessary health care organization to provide the long and costly treatment adapted to patients. Because of lack of treatment or lack of adapted treatment, at least two million people will die of tuberculosis this year. Due to this concern, this infectious disease was the focus of renewed scientific interest in the last decade. Regimens were optimized, and much was learnt on the mechanisms of action of the antituberculosis drugs used. Moreover, the quest for original drugs overcoming some of the problems of current regimens also became the focus of research programmes and many new series of *M. tuberculosis* growth inhibitors were reported. Based on reported literature, the compounds which consisting chalcone moiety in its basic structure exhibited *in vitro* binding against enoyl-[acyl-carrier protein] reductase (InhA), hence we proposed worthwhile to predict the binding energy of the compounds E)-3-(3-nitrophenyl)-1-(3,4,5-trimethoxyphenyl)-2-propen-1-one (Pub Chem CID 5730821), (E)-3-(3-nitrophenyl)-1-(3,4,5-Trimethoxyphenyl)-2-propen-1-onewhich were synthesized in the present study against InhA protein drug target. However, the role of InhA protein target has been well established in the pathogenesis of Tuberculosis (TB).

ENOYL-[ACYL-CARRIER PROTEIN] REDUCTASE (InhA)

Enzyme InhA, have been proposed as targets for INH. The targeted protein is of type II dissociated fatty acid biosynthesis pathway (FASII) in *Mycobacterium tuberculosis*, consistent with the observation that INH interferes with the biosynthesis of mycolic acids, very long chain fatty acid components of the mycobacterial cell wall. InhA, an enoyl reductase that catalyzes the NADH-dependent reduction of long chain *trans*-2-enoyl-acyl carrier proteins (ACPs), was first identified as a target by Jacobs and coworkers. InhA is inhibited by INH, one of three ketoacyl synthases in the FASII pathway, is a target for INH *in vivo.*The X-ray crystal structure was obtained from the Brookhaven Protein Data Bank (http://www.rcsb.org/pdb) (PDB.ID: 2X23).

Table 1: Compound name E)-3-(3-nitrophenyl)-1-(3,4,5-trimethoxyphenyl)-2-propen-1-one

PubChem CID	Compund Name	Docking Score (kcal/mol)
5730821	E)-3-(3-nitrophenyl)-1-(3,4,5-trimethoxyphenyl)-2-propen-1-one	-107.754

IN-VITRO BIOLOGICAL SCREENING:

Based on the activity showed in *in vitro*InhA enzyme data, The investigation of *in vitro Mycobacterium tuberculosis* E)-3-(3-nitrophenyl)-1-(3,4,5-trimethoxyphenyl)-2-propen-1-one (Pub Chem CID 5730821) demonstrated comparatively the potent inhibitory activity, with IC50 value of 12.11 ± 0.9 µM. which is not so potent as the standard drug Pyrazinamide (3.12 ± 0.4 µM. The activity may be implied to the presence of keto group or trimethoxyphenyl group which may impact the cellular modification of the cell wall major component synthesis i.e Mycolic acid.

CONCLUSION:

In this study the analysis of the best docked ligand against validated antitubercular drug targets revealed the binding mode of compounds involved in this study and confirm the role as potential antitubercular agents with established *in silico* mechanism of binding. Binding energies of the drug–enzyme (receptor) interactions are important to describe how fit the drug binds to the target macromolecule. The obtained results are useful to understand the structural features required to enhance the affinity as well as intrinsic activity. The *in silico* hits identified from the molecular docking simulation pertaining to antitubercular activities are less consistent with the *in vitro* results, the hits will further need to be analyzed using ligand binding assay studies followed by animal studies would be the future implication of this research work.

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