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The application of advanced drug discovery approaches in the fight against Tuberculosis

Shivkant Patel*[1], Surabhi Jain [2], Kinjal Patel[1],Phool Singh Yaduwanshi[3], Anubha Jain[4]

[1]Department of Pharmacy, Sumandeep Vidyapeeth Deemed to be University, Piparia, Vadodara, Gujarat, India.

[2]Faculty of Pharmacy, B. Pharmacy College Rampura-kakanpur,(Gujarat Technological University), Panchmahals, Gujarat, India

.[3]IITM, Department of Pharmacy, IES University, Bhopal (M.P.). [4] Sri Aurobindo College of Pharmacy, Indore (M.P.)

***Correspondence to Author**

Shivkant Patel

Assistant Professor,

Department of Pharmacy,

Sumandeep Vidyapeeth Deemed to be University, Vadodara.

E-mail: shivapatel2609@gmail.com

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Abstract:

The application of advanced drug discovery approaches in the fight against tuberculosis (TB) has significantly accelerated the identification and development of novel therapeutics. This review explores various contemporary strategies, including high-throughput screening, structure-based drug design, and computational modelling, that have been employed to discover new anti-TB drugs. Emphasis is placed on the integration of omics technologies, such as genomics, proteomics, and metabolomics, which provide comprehensive insights into Mycobacterium tuberculosis biology and pathogenesis. Additionally, the role of drug repurposing and combination therapy in enhancing treatment efficacy and overcoming drug resistance is discussed. This multifaceted approach aims to address the urgent need for effective TB therapies in the face of rising multidrug-resistant TB strains.

Keywords*: Tuberculosis, Drug Discovery, High-Throughput Screening, Structure-Based Drug Design, Computational Modelling*

1. Introduction

The practice through which drugs are identified or created in the realm of medicine is known as drug discovery. A drug is any substance (other than food or a device) that is used to diagnose, cure, relieve, treat, or prevent disease, or that is designed to influence the structure or function of the body. Traditionally, most medications were found either by finding the active component in traditional remedies or by chance. The drug development process includes steps such as candidate discovery, preparation, characterization, screening, and therapeutic effectiveness studies. In a stance when molecule has proven its worth in these assays, it will enter the development of new drugs prior to clinical trials. With respect to treatment of Tuberculosis, different analogues with different mode of action are shown in

above Tables 1.1-1.3. With the increase in number of tuberculosis infected population, it has taken the shape of most influential disease in the world. Since conventional drug design approach is having long design cycle and high cost whereas rational drug design approach has taken step forward to former. Rational approach includes structure-based drug design, molecular modeling studies, rapid DNA sequencing, automated high throughput screening (HTS), docking studies, QSAR and many more. The bioinformatics knowledge and computational approaches have speeded up the drug discovery process in very effective manner [1].

2. Rational Drug Design Approach

This approach made the way half easy to discover the candidate as drug. This technique is classified in two categories: Indirect (Ligand based) e.g. QSAR and Direct (Structure based) drug design. The former method relies on binding knowledge of molecules on pharmacological proven target whereas latter method based on biological target protein information.

2.1. Quantitative Structure Activity Relationship

Within Rational based approach, QSAR is an unbelievable method established by scientist Hansch and Fujita [2]. The essential premise in this method is that differences in the structural, physical, and chemical features of a sequence of congeneric molecules with a shared mode of action are connected with their biological activity [3]. The QSAR is a mathematical link that exists amongst biological activity and physicochemical qualities of molecules such as hydrophobicity, electronic properties, and so on. A QSAR equation is represented as:

$$\text{Biological activity} = (P_1*C_1) + (P_2*C_2) + \ldots\ldots + (P_n*C_n) + \text{constant}$$

Where, the parameters P1 to Pn are determined to every molecule in the array, whereas the coefficient C1 to Cn are obtained by matching variations in the respective parameters and bioactivities. Depending upon type of properties available in making a mathematical relationship with biological activity, QSARs are formalized into different dimensions i.e. 2D-, 3D-, 4D- etc. 0D-2D QSAR is also referred as classical approach which uses physico-chemical and structural parameters without referring to their conformations/ geometric preferences. 3D QSAR based approaches can be alignment-dependent or alignment-independent[4].

2.2. Structure Based Drug Design

Through tremendous evolution in structure or direct designing approaches, developments such as obtaining proteins with high purity, emergence of improved protein purification methods such as chromatography, increased sensitivity of NMR instruments, crystallized protein database, progression of cryo-cooling technique, and so on. The emergence of direct drug design as a new technology is nevertheless a attractive development with worldwide importance. This approach was used for the virtual (database) screening (VS) of new lead compounds. This VS has been accomplished by 2 approaches first is ‘VS by docking’ and second is ‘similarity-based VS’. Using information about the target's 3D structure, the first approach prioritizes compounds based on their propensity to bind to the protein. The ‘similarity-based VS’ is based on the idea that one or more chemicals known to bind to the protein are utilized as structural queries. The screening technique retrieves chemicals from the database using a similarity criterion that is appropriate for the situation. These criteria should consider compounds that bind firmly to the same proteins as comparable in order for the screening approach to be effective [5].

2.2.1. Docking

The Docking based structure drug design approach is similar to lock and key hypothesis. This hypothesis states that there is only one key for a lock. Similarly, only one orientation of molecule fitted to protein can produce a desired effect. It predicts ideal orientation of a

molecule (such as ligand or protein) to another (such as a protein or receptor) when they are bonded together to create a stable complex [6]. In relation to obtain different conformations of ligand, various docking programs are available. The computer enabled docking programs generates possess of small molecules in target structure in different positions, variation in conformations along with diverse orientations. These 3 collectively makes the different poses of ligand. Each pose generated is scored in comparison with different other poses in the grounds of energy and RMSD to ligand, to bind with protein. A predicted pose with good scores for submitted molecules indicates its potentiality as effective binders. This technique is replicated for all compounds, which will then be ranked according to docking or affinity scores. The selected top scorer active molecules were either purchased or synthesized then biological investigation was done. Taking into assumption that poses and its associated predicted affinity scores are in accordance with reasonable accuracy, this procedure reduces large proportion of time and cost then compared to random selection method. The technique is divided into 2 categories on basis of protein flexibility i.e. rigid and flexible docking.

Procedure employed for Docking:

a. **Receptor structure –** The receptor structure is important part of docking studies. The receptor provides the cavity or space for the binding of molecule to protein. The structure of the receptor or protein is most generally characterized either by X−ray crystallography or NMR. If 3D structure isn't known, protein structure prediction techniques can be used. Threading and homology modeling are two extensively used approaches for predicting protein structure. The former technique evaluates if a particular array of amino acids is consistent with desired structures. Homology, also referred as comparative modelling, is based on comprehensive connection or homology between the desired protein's sequence and at least one established structure. After determination of structure if a condition appears where function of protein is unknown, then putative binding sites should be evaluated. These binding sites can then be investigated for particular drug binding or correlated to other known binding sites. An examination of putative binding features and/or associations with a specific molecule can give critical findings for the development of new ligands or the docking of prospective leads.

b. **Prediction of Pose** - By doing a number of trials and keeping the positions with the least energy, the proper binding mode of ligand may be discovered.Since most ligand molecules are flexible, it entails determining the precise orientation as well as the suitable conformation of the docked molecule. It suggests ligands overall degrees of freedom like translational, rotational along with rotatable bonds. The study is ended after a certain number of trials have been completed or an acceptable collection of poses has been discovered. Algorithms that maintain record of earlier identified lowest energy and lead the exploration into new space. The judgment to preserve a trial pose is dependent on the pose's affinity score to target. Numerous programs create a "dock" score in order to rank them from high to low energy scores along with its orientations in an acceptable amount of time [7].

c. **Scoring of Predicted Pose -** The scoring functions are designed to molecules to respective receptor binding sites to segregate them as seeming inactive and active ligand molecules. There are 3 kinds of existing estimation methods: force field based, empirical, and knowledge based. The force field scoring functions estimate the binding free energy as a sum of individual molecular mechanics force field potentials. Free energy scoring methodologies in terms of Molecular Mechanics – Poisson-Boltzmann Surface Area/ Generalized Born Surface Area (MM-PBSA/ GBSA) might be advantageous in boosting binding affinity prediction and grading the actives. Applying the scoring function to experimental binding affinity data results in an empirical approximation of the binding free energy as a weighted combination of protein and ligand interaction components. The knowledge-based scoring method were

developed by statistical analysis of atom pair frequencies in protein ligand complexes with known 3D structure [8].

d. Principal Algorithms and Computer-Enabled Docking Platforms- Several docking programs and search algorithms are available to carry out desired docking process. There are different criterions to classify docking programs, the first criterion for classifying the underlying algorithms is the way the ligands are treated during docking. In some of these algorithm's ligand is divided into fragments that are docked separately in the receptor site. After the fragments are docked the parts are fused together. This fragmentation allows the algorithm to consider ligand flexibility. Rigid fragments that are docked initially work like "anchors" that are united secondarily by flexible parts of ligand which have rotatable bonds. In this way the ligand is gradually "constructed" inside the binding site of receptor. This approach is known as Incremental Construction. Programs that follow this approach include Hammerhead [9], DOCK [10], and FlexX [11]. The second algorithm is Monte Carlo simulations were firstly introduced as a minimization procedure in molecular dynamics applications. This search algorithm tries to dock the ligand inside the receptor site through many random positions and rotations, which decreases the chances of being trapped in the local minima. Programs used are MCDOCK [12] and ICM[13]. In Simulated Annealing (SA) a biomolecular system is simulated by a specific kind of dynamic simulation. Every docking conformation is carried into a simulation where temperature is decreased gradually during regular intervals of time in each cycle of simulation. In Genetic Algorithm (GA) programming, crossover, which is a genetic operator that combines (mates) two chromosomes (parents) to produce a new chromosome (offspring), is applied in order to generate new chromosome that may be better than both of the parents if it takes the best characteristics from each of the parents. This process that swaps large regions of the "parents," is permitted in genetic algorithms. In this process many complex scoring functions are used, taking into account a set of parameters, such as mutation rates, crossover rates and number of evolutionary rounds. The GA adopted in GOLD docking program. In addition to ligand flexibility, it may be desirable to keep at least part of the receptor flexible in order to allow for conformational changes that are necessary to accommodate the ligand, a phenomenon referred to as 'induced fit.' Because it is computationally expensive, few docking programs allow protein flexibility. The way flexibility is handled differs from program to program. For example, FlexE [14] uses multiple receptor conformations. A list of popular docking programs is given in Table 1 and 2 along with their description and algorithm is presented.

Program	Flexible Protein	Flexible Ligands	Algorithm	Description	Ref.
Hammerhead	No	Yes	Genetic Algorithm	Tail and Anchor Fragments Linked from Genetic Algorithms	9
DOCK	No	Yes	Shape Matching (Sphere Images)	Docks Small Molecules or Fragments	10
Flexx	No	Yes	Incremental Construction	Incremental Construction	11
MCDOCK(Monti Carlo Dock)	No	Yes	Monti Carlo (Stochastic Algorithm)	Stochastic Algorithm	12
ICM	Yes	Yes	Stochastic Algorithm	Stochastic Algorithm	13
AUTODOCK	Yes	Yes	Genetic Algorithm	simulated annealing	14
Flexe	Yes	Yes	Incremental Construction	Incremental Construction	15
FRED (Fast rigid exhaustive docking)	No	Yes	Exhaustive Search Algorithm	Shape Matching (Gaussian Functions)	16
Glide	No	Yes	Exhaustive Search Algorithm	Search for orientation Ligand.	17
Prodock	Yes	Yes	Monti Carlo (Stochastic Algorithm)	Stochastic Algorithm for Flexible Ligand and receptor	18
ADAM	No	Yes	Incremental Construction	Aligned Based Fragments based on Hydrogen Bond	19

Table 1. DE NO VO Generation of Ligands

Program	Flexible Protein	Flexible Ligands	Description	Ref.
LUDI	No	Yes	Incremental Construction	20
GRID	No	Yes	Energy Calculation	21
Multi copy simultaneous search methodology	No	Yes	Monte Carlo Growth Algorithm	22
Smog (Small Molecule Growth)	No	Yes	Monte Carlo Growth Algorithm	23, 24
CONCERTS	No	Yes	Stochastic search	25
Legend	No	Yes	Raises structure Atom by Atom	26
DLD	No	Yes	Monte Carlo Growth Algorithm	27

Growmol	No	Yes	Incremental Construction	27
Genstar	No	Yes	Forms molecules by Sp^3 Carbons	27
GROW	No	Yes	amino acid Residue Addition	27
Groupbuild	No	Yes	Forms molecules by defined Collection of Fragments	27
HOOK	No	Yes	Exploration of chemical Database	27

Table 2. Virtual screening methods

3. Computational Drug Designing's Role in Tuberculosis Drug Research

The well recognized fact that most of current available anti-tuberculosis treatments were discovered either by coincidence or by chemical changes of previously available medicines. Despite the fact that existing frontline medicines have improved the prognosis of Tb patients, there is an urgent need to apply creative tactics for the identification of viable therapeutic options. Furthermore, the loss of efficacy of currently available active drugs, as well as a scarcity of distinct chemical substances in healthcare situations, has caused serious damage to current Tb eradication efforts. As a result, unique reasoning tactics are critical for the discovery of new medications and therapeutic targets to treat tuberculosis. For more than three decades, computer-aided techniques have dominated this quest. This image may be shown by the fact that several researches have been conducted in order to find *M. tuberculosis* druggable targets. *Cui et al.* conducted an investigation of protein-protein interaction networks using homogeneous protein mapping in this approach. The study discovered that molecular chaperones, ribosomal proteins, and ABC transporters are all intricately linked proteins [28]. Computational approaches have enabled the discovery of fresh molecules with desirable characteristics intended for the effective management of various types of tuberculosis, including resistant to drug and persistent forms of Tuberculosis, as well as HIV co-infected patients. Several QSAR, pharmacophore modeling, docking investigations, and molecular dynamics simulations (MDS) studies demonstrate its significance. The MDS in conjunction with docking investigations revealed the resistance to the first-line TB medication, isoniazid [29]. QSAR, docking studies, and QM/MM studies on *M. tuberculosis* inhibitors were good examples of M. tuberculosis molecular modeling investigations [30]. Several studies on the therapeutic target of *M. tuberculosis*, thymidine monophosphate kinase (TMPkinase), have recently been done [31]. TMPkinase inhibition was investigated using a combination of receptor independent 4D-QSAR formalism and 3D pharmacophore. Additionally, virtual screening (VS) based on molecular fingerprints was used to highlight the powerful anti-tubercular compounds against TMPkinase [32]. Similarly, VS has effectively recovered new IspF inhibitors using hierarchical filtering and docking experiments [33]. Pantothenate synthetase inhibitors were designed in-silico and tested for inhibitory effectiveness against the non-replicating persistent type of M. tuberculosis [34]. *Bonora et al.* investigated inhibitors targeting both HIV and tuberculosis proteases computationally [35]. Furthermore, the proteins essential for existence of TB are being considered potential druggable targets [36]. Aside from the above stated metabolic pathways, research has focused on the targets engaged in the manufacture of several constituents of the mycobacterium cell wall [37]. Furthermore, a considerable amount of study has been conducted to investigate the pathogenicity of various forms of *M. tuberculosis*, chiefly those helped in maintaining the latent anaerobic condition under host induced hypoxia [38-39]. As a result, it is evident that identification and validation are critical processes in *in-silico* drug design [40]. Despite improvements in anti-TB drug development, further therapeutic interventions with characteristics such as a unique mechanism of action, fast bactericidal efficacy, enhanced pharmacokinetic and pharmacodynamic qualities, minimal potential for drug-drug interactions, and an outstanding safety profile are still needed. Aside from these actions, there are also pragmatic considerations linked with them, such as compound stability, cost-effective

manufacture, a restricted action range, excellent tolerability, and a low incidence of resistance development [41]. Several previous attempts to find newer anti-TB medications have been done in this endeavour. Non rational approaches, like traditional and virtual screening methods, are included, and so are rational strategies, such as bioinformatics analysis. Table 3. lists some of the most recent successful narratives obtained using these methods. Among rational techniques, pharmacophore tactics have emerged as one of most imperative instruments in discovery of drugs. These methods have been used effectively and widely for the high throughput screening, scaffold hopping, lead identification and optimization. Concurrent developments in protein purification techniques, the availability of three-Dimensional structures in the protein data banks, have resulted in an improved knowledge of many proteins structural properties and its co-crystallized ligand. Virtual screening is another major progress in drug development that is a more direct and logical drug discovery technique than traditional experimental high-throughput screening, with the added benefit of being less expensive and more effective [42]. MDS have advanced to level that they can be used to successfully investigate the macromolecular structure and its function relationships. The effective blend of molecular docking and simulation research can result in the identification of new drugs.

Furthermore, these computational tools are extremely important for reducing the total budget and time period associated with drug development phase [43,44,45]. Despite the fact that various programs are widely accessible [46], the techniques necessary for the creation of pharmacophore models are still in the research stage. These approaches are divided as: direct methods and indirect approaches [47,48,49]. The direct technique uses either a protein or receptor ligand complex or only protein for model creation, while the indirect method uses a group of compounds active against a specific therapeutic target to derive the pharmacophoric properties required for biological activity. In the literature, the limitations of indirect approaches are well-recognized [50-55]. In general, these approaches create models using either ligand alone or protein-ligand multicomplexes. However, if database of multicomplexes is known pharmacophore modeling would be the natural approach analyze all of the protein-ligand complexes' relevant interactions at the same time. As a result, in this thesis, protein-ligand multi-complex-based generation of models being primarily used for *M. tuberculosis* structural proteins. The purpose was to see if pharmacophore models were similar across protein classes and if inhibitors had similar effects. Furthermore, on the basis of validation and knowledge, an attempt was made to uncover distinctive patterns that may be actively used for the creation of small molecules against the main therapeutic targets, ATP synthase and QcrB. The compounds that were ranked using computational approaches were validated experimentally.

Table 3. List of compounds showing anti-TB activity

Computational Approach	**Target Area**	**Ligand names**	**Ref.**
Homology modeling/Docking	ATP synthase	12i, 12l	[56]
Homology modeling/ Docking/VS	*Mycobacterium* multidrug-resistant protein	L1–L8	[57]
Homology modeling/Docking/ Pharmacophore modeling	Serine/Threonine protein kinases	T95, B31	[58]
Homology modeling/ Pharmacophore/VS/MD	DNA gyrase B	C1–C10	[59]
Pharmacophore modeling/ 3D QSAR/Docking	Enoyl-ACP-reductase	Cinnamic acid and cyclopropyl derivatives	[60]

Pharmacophore modeling/VS	L-alanine dehydrogenase	Compounds L1–L4	[61]
Docking	Enoyl-ACP- reductase	I1, I3, I4 y I5	[62]
VS/Pharmacophore modeling/ Docking/ Fingerprints	Thymidine Monophosphate Kinase	Compounds C1–C3	[63]
HTS/Docking	Protein kinase B	Compounds 11, 12, 15	[64]
QSAR	Cell wall synthesis	Compounds A, B	[65]
QSAR/HTS	SQ609	Cell wall synthesis	[66]
HTS/Whole-cell activity	CD39, CD117	Enoyl-ACP-reductase	[67]
HTS/Whole-cell activity	Compounds 1, 2	Pantothenate synthetase	[68]
HTS/Whole-cell activity	Oxadiazole-amide and 2-aminobenzothiazole core scaffold derivatives	Shikimate kinase	[69]

4. Conclusion

The application of advanced drug discovery approaches has revolutionized the fight against tuberculosis (TB), offering promising avenues for the development of new therapeutics. High-throughput screening, structure-based drug design, and computational modeling have significantly accelerated the identification of potential drug candidates. The integration of omics technologies—genomics, proteomics, and metabolomics—provides a comprehensive understanding of Mycobacterium tuberculosis, paving the way for targeted interventions. Drug repurposing and combination therapy strategies enhance treatment efficacy and offer solutions to overcome multidrug-resistant TB strains. Despite the progress, ongoing research and collaboration are essential to translate these innovative approaches into effective clinical therapies. This multifaceted and integrative strategy is crucial for addressing the global TB burden and achieving long-term control and eradication of the disease.

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5. References

1. Borsari C, Ferrari S, Venturelli A, Costi MP. Target-based approaches for the discovery of new antimycobacterial drugs. *Drug Discov. Today*, **201**7; *22*(3); 576-584.
2. Baldi A. Computational Approaches for Drug Design and Discovery, anOverview. *Sys. Rev. Pharm.***2010**; *1*(1): 99-105.
3. Burden FR. Quantitative Structure Activity Relationship (QSAR) using Guassian Processes. *J. Chem. Inform. Sci.***2001**; *41*(3): 830-835.
4. Perkins R, Fang H, Tong W, Welsh WJ. Quantitative Structure-Activity Relationship (QSAR), Perspective in Drug Discovery and Toxicology. *Envir. Toxi. Res.***2003**; *22*(8): 1666-1679.
5. Lill MA. Multi-Dimensional QSAR In Drug Discovery. *Drug. Dis. Today.***2007**; *12*(23-24): 1013-1017.

6. Verlinde Christophe LMJ, Hol GJ. Structure-Based Drug Design: Progress, Results and Challenges. *Curr. Bio.***1994**; *2*(7): 577-587.
7. Lengauer T, Lemmen C, Rarey M, Zimmermann M. Novel Technologies for Virtual Screening, *Drug. Discov. Tod.* **2004**; *9*(1): 27-34.
8. Meng XY, Zhang HX, Mezei M, Cui M. Molecular docking: a powerful approach for structure-based drug discovery. *Curr. Comput. Aided Drug Des.***2011;** *7*(2): 146-157.
9. Kroemer RT. Structure-Based Drug Design: Docking and Scoring. *Cur. Pro. Pep. Sci.* **2007**; *8*(4):312-328.
10. Pencheva T, Soumana O, Pajeva I, Miteva MA, Post-docking virtual screening of diverse binding pockets: Comparative study using DOCK, AMMOS, X-Score and FRED scoring functions. *Eur. J. Med. Chem.***2010**; *45*(6): 2622-28.
11. Welch W, Ruppert J, Jain AN. Hammerhead: Fast, Fully Automated Docking of Flexible Ligands to Protein Binding Sites. *Chem. Bio.***1996**; *3*(6): 449-462.
12. Ewing TJ, Makino S, Skillman AG, Kuntz ID. DOCK 4.0: Search strategies for automated molecular docking of flexible molecule databases. *J. Comput. Aided Mol.Des.***2001**; *15*: 411-428.
13. Rarey M, Kramer B, Lengauer T, Klebe G. A Fast Flexible Docking Method Using an Incremental Construction Algorithm. *J. Mol. Biol*. **1996**; *261*(3): 470-489.
14. Liu M, Wang S. MCDOCK: A Monte Carlo Simulation Approach to the Molecular Docking Problem. *J. Comput. Aided Mol. Des.***1999**; *13*: 435-451.
15. Huang SY, Zou X. Advances and Challenges in Protein-Ligand Docking. *Int. J. Mol. Sci.* **2010**; *11*(8): 3016-3034.
16. Goodsell DS, Morris GM, Olson AJ. Automated Docking of Flexible Ligands: Applications of AutoDock. *J. Mol. Recog.***1996**; *9*(1): 1-5.
17. Claussen H, Buning C, Rarey M, Lengauer T. FlexE: Efficient Molecular Docking Considering Protein Structure Variations. *J. Mol. Biol.* **2001**; *308*(2): 377-395.
18. McGann MR, Almond HR, Nicholl A, Grant A, Brown FK. Gaussian Docking Functions. *Biopoly.* **2003**; *68*(1): 76-90.
19. Halgren TA, Murphy RB, Friesner RA, Beard HS, et al. Glide: A New Approach for Rapid, Accurate Docking and Scoring. 2. Enrichment Factors in Database Screening. *J. Med. Chem.* **2004**;*47*(7):1750-1759.
20. Jones G, Willett P, Glen RC, Molecular Recognition of Receptor Sites using a Genetic Algorithm with a Description of Desolvation. *J. Mol. Biol.* **1995**; *245*(1): 43-53.
21. Trosset JY, Scheraga HA. PRODOCK: Software Package for Protein Modeling and Docking. *J. Comput. Chem.***1999**; *20*(4): 412-427.
22. Kastenholz MA, Pastor M, Cruciani G, Haaksma EE, Fox T. GRID/CPCA: A New Computational Tool to Design Selective Ligands. *J. Med. Chem.* **2000**; *43*(16): 3033-3044.
23. DeWitte RS, Shakhnovich EI. SMoG: de Novo Design Method Based on Simple, Fast, and Accurate Free Energy Estimates. Methodology and Supporting Evidence. *J. Am. Chem. Soc.* **1996;** *118*(47):11733-11744.
24. Schubert RC, Stultz MC. The Multi-Copy Simultaneous Search Methodology: A Fundamental Tool for Structure-Based Drug Design. *J. Comput. Aid. Mol. Des.***2009**; *23*(8):475-489.
25. McCarthy JD, Alvarez CJ. Automated Generation of MCSS-Derived Pharmacophoric DOCK Site Points for Searching Multiconformation Databases. *Proteins,* **2003**; *51*(2): 189-202.
26. Pearlman DA, Murcko AM. CONCERTS: Dynamic Connection of Fragments as an Approach to *de Novo* Ligand Design. *J. Med. Chem.* **1996;** *39*(8)*:* 1651-1663.

27. Hou T, Xu X. Recent Development and Application of Virtual Screening in Drug Discovery: An Overview. *Curr. Pham. Des.* **2004;** *10*(9):1011-1033.
28. Anderson AC. The Process of Structure-Based Drug Design. *Chem. Bio.***2003**; *10*(9): 787-797.
29. Cui T, Zhang L, Wang X, He ZG. Uncovering new signaling proteins and potential drug targets through the interactome analysis of Mycobacterium tuberculosis. *BMC genomics*. **2009**; *10*: 118.
30. Wahab HA, Choong YS, Ibrahim P, Sadikun A, Scior T. Elucidating isoniazid resistance using molecular modelling. . *J. Chem. Inf. Model.* **2009**; *49*(1): 97-107.
31. Neres J, Labello NP, Somu RV, Boshoff HI, et.al. Inhibition of siderophore biosynthesis in Mycobacterium tuberculosis with nucleoside bisubstrate analogues: structure− activity relationships of the nucleobase domain of 5′-O-[N-(salicyl) sulfamoyl] adenosine. *J. Med. Chem*. **2008**; *51*(17): 5349-5370.
32. Andrade CH, Pasqualoto KF, Ferreira EI, Hopfinger AJ. Rational design and 3D-pharmacophore mapping of 5′-thiourea-substituted α-thymidine analogues as mycobacterial TMPK inhibitors. *J. Chem. Inf. Model*. **2009**; *49*(4):1070-1078.
33. Kumar A, Chaturvedi V, Bhatnagar S, Sinha S, Siddiqi MI. Knowledge based identification of potent antitubercular compounds using structure based virtual screening and structure interaction fingerprints. *J. Chem. Inf. Model.***2009**; *49*(1): 35-42.
34. Ramsden NL, Buetow L, Dawson A, Kemp LA, et.al. A structure-based approach to ligand discovery for 2 C-methyl-D- erythritol-2, 4-cyclodiphosphate synthase: A target for antimicrobial therapy. *J. Med. Chem*. **2009**; *52*(8): 2531-2542.
35. Velaparthi S, Brunsteiner M, Uddin R, Wan B, et.al. 5- tert-Butyl-N-pyrazol-4-yl-4, 5, 6, 7-tetrahydrobenzo [d] isoxazole-3-carboxamide derivatives as novel potent inhibitors of Mycobacterium tuberculosis pantothenate synthetase: initiating a quest for new antitubercular drugs. *J. Med. Chem.* **2008**; *51*(7): 1999-2002.
36. Bonora S, Di Perri G. Interactions between antiretroviral agents and those used to treat tuberculosis. *Curr. Opin. HIV AIDS.* **2008**; *3*(3): 306-312.
37. Janin YL. Antituberculosis drugs: ten years of research, *Bioorg. Med. Chem.* **2007**; *15*(7): 2479-2513.
38. Levy J. The effects of antibiotic use on gastrointestinal function. *Am. J.Gastroenterol.* **2000**; *95* (1 Suppl): S8-S10.
39. Mdluli K, Spigelman M. Novel targets for tuberculosis drug discovery, *Curr. Opin. Pharmacol.* **2006**; *6*(5): 459-467.
40. Voskuil MI, Schnappinger D, Visconti KC, Harrell MI, et.al. Inhibition of respiration by nitric oxide induces a Mycobacterium tuberculosis dormancy program. *J. Exp. Med.* **2003**; *198*(5): 705-713.
41. Kumar A, Toledo JC, Patel RP, Lancaster Jr, Steyn AJ. Mycobacterium tuberculosis DosS is a redox sensor and DosT is a hypoxia sensor. *Proc. Natl. Acad. Sci. U S A***2007**; *104*(28): 11568-11573.
42. Hoagland DT, Liu J, Lee RB, Lee RE. New agents for the treatment of drug- resistant Mycobacterium tuberculosis. *Adv. Drug Deliv. Rev.* **2016**; *102*: 55- 72.
43. Hospital A, Goñi JR, Orozco M, Gelpí JL. Molecular dynamics simulations: advances and applications. *Adv. Appl. Bioinform. Chem.***2015;** 8: 37-47.
44. Wermuth CG. Pharmacophores: historical perspective and viewpoint from a medicinal chemist. *Methods and Principles in Med. Chem*. **2006**; *32*: 3.
45. Ekins S, Mestres J, Testa B. In silico pharmacology for drug discovery: applications to targets and beyond. *Br. J. Pharmacol.***2007**; *152* (1): 21-37.
46. Poptodorov K, Luu T, Hoffmann RD. Pharmacophore model generation software tools. *Methods and Principles in Med. Chem.***2006**; *32*:17.

47. Dror O, Shulman-Peleg A, Nussinov R, Wolfson HJ. Predicting molecular interactions in silico: A guide to pharmacophore identification and its applications to drug design. *Curr. Med. Chem.* **2004**; *11*(1): 71-90.
48. Khedkar SA, Malde AK, Coutinho EC, Srivastava S. Pharmacophore modeling in drug discovery and development: An overview. *Med. Chem*. **2007**; *3*(2):187- 197.
49. Wolber G, Seidel T, Bendix F, Langer T. Molecule-pharmacophore superpositioning and pattern matching in computational drug design. *Drug Discov. Today.***2008**; *13*(1-2): 23-29.
50. Kurogi Y, Guner OF. Pharmacophore modeling and three-dimensional database searching for drug design using catalyst. *Curr. Med. Chem.***2001**; *8*(9): 1035- 1055.
51. Guner OF. History and evolution of the pharmacophore concept in computer-aided drug design. *Curr. Top. Med. Chem.***2002**; *2*(12): 1321-1332.
52. Xiao Z, Varma S, Xiao YD, Tropsha A. Modeling of p38 mitogen-activated protein kinase inhibitors using the Catalyst™ HypoGen and k-nearest neighbor QSAR methods. *J. Mol. Graph. Model.* **2004**; *23*: 129-138.
53. Kirchmair J, Laggner C, Wolber G, Langer T. Comparative analysis of protein- bound ligand conformations with respect to catalyst's conformational space subsampling algorithms. *J. Chem. Inf. Model.***2005**; *45*(2): 422-430.
54. Kirchmair J, Wolber G, Laggner C, Langer T. Comparative performance assessment of the conformational model generators omega and catalyst: a large-scale survey on the retrieval of protein-bound ligand conformations. *J. Chem. Inf. Model.***2006**; *46*(4): 1848-1861.
55. Kirchmair J, Ristic S, Eder K, Markt P, et.al. Fast and efficient in silico 3D screening: toward maximum computational efficiency of pharmacophore-based and shape-based approaches. *J. Chem. Inf. Model.***2007**; *47*(6): 2182-2196.
56. Kristam R, Gillet VJ, Lewis RA, Thorner D. Comparison of conformational analysis techniques to generate pharmacophore hypotheses using catalyst. *J. Chem. Inf. Model.***2005**; *45* (2): 461-476.
57. Jain PP, Degani MS, Raju A, Ray M, Rajan MGR. Rational drug design based synthesis of novel arylquinolines as anti-tuberculosis agents. *Bioorg. Med. Chem. Lett.***2013**; *23*(22): 6097-6105.
58. Malkhed V, Mustyala KK, Potlapally SR, Vuruputuri U. Identification of novel leads applying in silico studies for Mycobacterium multidrug resistant (MMR) protein. *J. Biomol. Struct. Dyn*. **2014**; *32*(12): 1889-1906.
59. Kandasamy S, Hassan S, Gopalaswamy R, Narayanan S. Homology modelling, docking, pharmacophore and site directed mutagenesis analysis to identify the critical amino acid residue of PknI from Mycobacterium tuberculosis. *J. Mol. Graph. Model.* **2014**; 52: 11-19.
60. Maharaj Y, Soliman MES. Identification of Novel Gyrase B Inhibitors as Potential Anti-TB drugs: Homology Modelling, Hybrid Virtual Screening and Molecular Dynamics Simulations. *Chem. Biol. Drug Des.***2013**; *82*(2): 205-215.
61. Mondal S, Upamanyu N, Sen D. Hybrid computational simulation and modeling assisted structural analysis of anti-tubercular molecules. *Proc. Technol.* **2013**; *10*: 53-61.
62. Saxena S, Devi PB, Soni V, Yogeeswari P, Sriram D. Identification of novel inhibitors against Mycobacterium tuberculosisl-alanine dehydrogenase (MTB- AlaDH) through structure-based virtual screening. *J. Mol. Graph. Model.* **2014**; *47*: 37-43.
63. Izumizono Y, Arevalo S, Koseki Y, Kuroki M, Aoki S. Identification of novel potential antibiotics for tuberculosis by in silico structure-based drug screening. *Eur. J. Med. Chem.* **2011**; *46*(5): 1849-1856.

64. Kumar A, Chaturvedi V, Bhatnagar S, Sinha S, Siddiqi MI. Knowledge based identification of potent antitubercular compounds using structure based virtual screening and structure interaction fingerprints. *J. Chem. Inf. Model.***2008**; *49*(1): 35-42.
65. Naqvi A, Malasoni R, Srivastava A, Pandey RR, Dwivedi AK. Design, synthesis and molecular docking of substituted 3-hydrazinyl-3-oxo-propanamides as anti- tubercular agents. *Bioorg. Med. Chem. Lett.***2014**; *24*: 5181-5184.
66. Ventura C, Latino DARS, Martins F. Comparison of multiple linear regressions and neural networks based QSAR models for the design of new antitubercular compounds, *Eur. J. Med. Chem.* **2013**; *70*: 831-845.
67. Bogatcheva E, Hanrahan C, Nikonenko B, De-los-Santos G, et. al. Identification of SQ609 as a lead compound from a library of dipiperidines. *Bioorg. Med. Chem. Lett.***2011**; *21*(18): 5353-5357.
68. Vilchèze C, Baughn AD, Tufariello J, Leung LW, et. al. Novel inhibitors of InhA efficiently kill Mycobacterium tuberculosis under aerobic and anaerobic conditions. *Antimicrob. Agents Chemother.* **2011**; *55*(8): 3889-3898.
69. Kumar A, Casey A, Odingo J, Kesicki EA, et.al. A high-throughput screen against pantothenate synthetase (PanC) identifies 3-biphenyl-4-cyanopyrrole-2-carboxylic acids as a new class of inhibitor with activity against Mycobacterium tuberculosis.*PloS one*. **2013**; *8*(11):e72786.