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Insilico studies of therapeutic agents in Phytocompounds obtained from *mondoro myristica* (African nutmeg) against *Mycobacterium tuberculosis*

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Abstract: **The highly adaptive cellular response of *Mycobacterium tuberculosis* to various antibiotics, combined with the high costs of clinical trials, hamper the development of novel antimicrobial agents with improved efficacy and safety. Subsequently, in silico drug screening methods are increasingly used in drug discovery and development and have proven useful for predicting the pharmacokinetics, toxicities, and targets of prospective new antimicrobial agents. In this investigation, we employed a reverse target fishing approach to identify potential hit targets and explore their interactions between *M. tuberculosis* and a promising new antituberculosis compound. Two of the four identified targets, Cyp130 and 1SFR, were strongly proposed as optimal drug targets for dormant M. tuberculosis, with Cyp130 exhibiting the highest binding affinity. The metabolic pathways associated with the selected target proteins were compared to previously published molecular mechanisms of Cyp130 in M. tuberculosis, confirming disruptions in the metabolism of proteins, cell wall components, and DNA. We also detailed the specific steps within these pathways that are inhibited and elaborated on the role of Cyp130 against dormant M. tuberculosis. This compound has previously shown promising in vitro safety and good oral bioavailability, both of which were supported by this in silico study. The pharmacokinetic properties and toxicity of the compound were predicted and evaluated using the online tools pkCSM and SwissADME, along with Discovery Studio software, further supporting its safety and bioavailability as an antimycobacterial agent.**

Keywords: **Mycobacterium tuberculosis, phytocompounds, *mondoro myristica*, ADMET.**

1. INTRODUCTION

Tuberculosis (TB), caused by *Mycobacterium tuberculosis* (MTB), remains a leading global health concern, responsible for millions of deaths annually despite extensive research and drug development efforts.[1,2] The persistence of the infection and the emergence of multidrug-resistant TB (MDR-TB) and extensively drug-resistant TB (XDR-TB) pose significant challenges to disease control.[3,4] The inefficacy of conventional drug discovery strategies, including target-based high-throughput screening, necessitates novel approaches such as in silico methods and molecular modeling to identify potential therapeutic candidates.[5] The literature survey highlights the shortcomings of traditional TB drug discovery methodologies and emphasizes the need for integrating computational approaches with experimental validation.[6] Advances in bioinformatics and systems biology have facilitated the identification of new drug targets, offering promising avenues for rational drug design.[7] However, despite these technological advancements, there is still an urgent need to develop effective,

fast-acting, and less toxic TB therapies. [5] Tuberculosis continues to be a major public health threat, with drug resistance exacerbating treatment challenges.[1] The prolonged treatment duration, coupled with patient non-compliance and adverse drug effects, underscores the necessity of discovering new anti-TB compounds. [2,8] Given the limitations of existing therapeutics, there is a critical need to explore alternative drug candidates, particularly those derived from natural sources, using modern computational techniques. [4] This study focuses on the molecular interactions of phytochemicals derived from *Mondoro myristica* with key enzyme targets in tuberculosis through molecular modeling. Computational tools and bioinformatics approaches were utilized to predict the efficacy of these phytochemicals as anti-TB agents, providing insights into their potential mechanisms of action. [5,9] The findings from this study aim to contribute to the ongoing search for novel, effective TB therapeutics with improved pharmacological profiles.[1]

2. MATERIALS AND METHODS

Preparation of phytochemicals

Thirty (30) phytochemicals from *Mondoro myristica* were compiled from previous reports through an extensive literature search performed on public databases which includes the PubMed, Scopus, Google Scholar, and Google databases.

Preparation of ligands

The 30 phytocompounds where retrieved in Structured Data Format (SDF) from *Mondoro myristica*, they were retrieved from the PubChem database (www.pubchem.ncbi.nlm.nih.gov). The phytocompounds where imported into the software: Open Babel in build in PyRx 0.8 and were further converted to the dockable PDBQT format.

Table 1. List of bioactive compounds derived from *Mondoro myristica*

S/N	COMPOUNDS
1	2,3-Anthracenedione
2	3,5-Dihydroxy-7-methoxy-2-(4-methoxyphenyl)-4H-chromen-4-one
3	3,10-dihydroxydecanoic Acid
4	6-Hydroxy-7-oxo-ferruginol
5	9-Octadecanamide
6	10-Hydroxy octadecadienoic acid
7	10-Methoxynormacusine
8	Alpha farnesene
9	alpha-Copaen-11-ol
10	Amycocyclopiazonic acid
11	Annonamine
12	Cornolactone B
13	Dihydrokaempferol 7-glucoside
14	Epicatechin 3-O-(3-O-methylgallate)
15	Estragole
16	Feruloyl putrescine
17	Granulatamide B
18	Ixoside
19	Lepidotol A
20	Linalool
21	Melicarpinone
22	Myristoyl chloride
23	Nerolidol
24	Oxalates
25	Oxiamycin
26	palmitic acid
27	Phytosterol
28	Sinensin
29	Squalene
30	Tryptamine

Preparation of protein

The 3D protein structures were retrieved from the Protein Data Bank (https://www.rcsb.org). After downloading retrieved structures, the native ligands were extracted, and water molecules removed using Discovery Studio 2022.

Table 2. Target proteins in tuberculosis

S/N	TARGET PROTEIN	PDB NUMBER	AMINO ACID RESIDUES	CO CYRSTALISED COMPOUND
1	Mycobacterium tuberculosis cytochrome (Cyp130)	2UVN	Leu 71	ECONAZOLE
			Pro 87	
			Pro 88	
			Phe 236	
			Thr 239	
			Met 240	
			Thr 247	
			Pro 289	
			Val 290	
			Phe 347	
			Cys 354	
			Leu 355	
			Gly 356	
			Ala 359	
			Ala 360	
			Val 393	
2	Mycobacterium tuberculosis mycolyl transferase gene (fbpB)	1SFR	Asp 40	BOVINE PROCARBOXY PEPTIDASE
			Leu 42	
			Arg 43	
			Ser 126	
			Leu 152	
			Leu 163	
			Leu 229	
			Phe 232	
			His 262	
			Trp 264	
			Trp 267	
3	Diaminopimelate decarboxylase (LysA)	2O0T	Cys 375	
			Glu 376	
			Ser 377	
			His 213	
			Arg 303	
			Try 405	
4	Adenosine Kinase (Adok)	2PKF	Val 49	
			Gln 172	
			Asn 195	
			Thr 223	
			Val 255	
			Asp 257	
			Phe 259	
			Ser 281	
			Leu 288	

Druglikeness screening

The selected compounds that were having higher binding affinity with target proteins in Alzheimer's disease were subjected to various drug-likeness filtering analysis. The drug-likeness analysis which includes Lipinski, Veber, Ghose, Egan and Muegge were performed on the SwissADME webserver.

The drug-likeness properties of the compound were screened using the Lipinski's rule (molecular mass (MM) less than 500 Da, no more than 5 hydrogen bond donors (HBD), no more than 10 hydrogen bond acceptors (HBA), and partition coefficient (log p).

ADMET (absorption, distribution, metabolism, excretion and toxicity) analysis

The compounds with the highest binding interaction with target proteins where subjected to ADMET (Absorption, Distribution, Metabolism, Excretion and Toxicity) analysis to study and analyze the **Absorption** – (How much of the phytocompound can be absorbed and how quickly), **Distribution**- (where is the phytocompound distributed within the body and what is the rate and extent of the distribution), **Metabolism**- (How fast can the phytocompound be metabolized, what is the mechanism of action and what metabolite is formed and is it active or toxic) **Elimination**- (How is the phytocompound excreted and how quickly) (**Toxicity**-Does this phytocompounds have a toxic effect to body systems or organs). [10]

3. RESULTS

Molecular docking results

The results of molecular docking against the selected enzymes in tuberculosis are shown in table 4.1 as represented by the docking scores. The docking scores of the compounds ranges from -4.2 to -8.7 for fbpB, -4.3 to -10.5 for Cyp130, -4.3 to -9.8 for Adok and -4.2 to -8.4 for LysA.

Table 3. Docking score of phytochemicals from *Mondoro mristica* against target proteins

S/N	COMPOUNDS	fbpB	Cyp130	Adok	LysA
1	2,3-Anthracenedione	-7.5	-8.5	-8.2	-6.8
2	3,5-Dihydroxy-7-methoxy-2-(4-methoxyphenyl)-4H-chromen-4-one	-7.8	-7.9	-7.9	-6.9
3	3,10-dihydroxydecanoic Acid	-5.8	-5.4	-5.7	-5.2
4	6-Hydroxy-7-oxo-ferruginol	-6.0	-8.7	-8.4	-8.1
5	9-Octadecanamide	-6.3	-5.8	-6.4	-4.5
6	10-Hydroxy octadecadienoic acid	-7.4	-8.7	-8.6	-7.2
7	10-Methoxynormacusine	-4.2	-4.3	-4.7	-4.7
8	Alpha farnesene	-6.3	-7.2	-6.8	-5.2
9	alpha-Copaen-11-ol	-6.6	-7.0	-6.9	-6.2
10	Amycocyclopiazonic acid	-8.7	-8.0	-7.4	-7.4
11	Annonamine	-6.6	-8.7	-8.1	-7.0
12	Cornolactone B	-7.0	-7.1	-7.5	-6.5
13	Dihydrokaempferol 7-glucoside	-8.3	-9.3	-9.5	-8.1
14	Epicatechin 3-O-(3-O-methylgallate)	-7.0	-9.5	-8.4	-8.0
15	Estragole	-5.4	-5.7	-6.0	-5.1
16	Feruloyl putrescine	-6.5	-7.4	-7.2	-5.9
17	Granulatamide B	-7.7	-7.8	-7.9	-6.1
18	Ixoside	-7.2	-8.6	-8.5	-7.7
19	Lepidotol A	-7.9	-9.5	-9.4	-7.5
20	Linalool	-5.2	-5.4	-6.2	-4.8
21	Melicarpinone	-7.1	-7.7	-7.5	-6.5
22	Myristoyl chloride	-5.3	-6.0	-5.7	-4.2
23	Nerolidol	-6.0	-6.5	-5.7	-4.2
24	Oxalates	-5.4	-4.4	-4.3	-4.5

Table 4. The Grid Box information of the Target Enzymes

Axis	FbpB	Cyp130	Adok	LysA
CENTER_X	50.92	54.39	55.95	2.24
CENTER_Y	52.48	53.16	-39.23	0.59
CENTER_Z	5.50	-28.08	29.80	42.47
DIMENSION_X	18.17	16.27	22.99	37.67
DIMENSION_Y	21.08	22.98	15.33	28.94
DIMENSION_Z	19.36	25.33	20.87	16.26

Table 5. Docking score of selected ligands with high binding affinity, co-crystallized compounds and reference drugs used in treatment of tuberculosis

S/N	COMPOUNDS	FbpB	Cyp130	Adok	LysA
1	2,3-Anthracenedione	-7.8	-6.8	-8.3	-6.8
2	3,5-Dihydroxy-7- methoxy-2-(4- methoxyphenyl)-4H-chromen-4-one	-7.8	-8.0	-7.9	-6.8
3	10-Hydroxy octadecadienoic acid	-6.1	-6.4	-6.6	-5.2
4	Amycocyclopiazonic acid	-8.8	-8.0	-7.4	-7.1
5	Annonamine	-8.3	-8.7	-8.3	-6.9
6	Dihydrokaempferol 7-glucoside	-8.6	-9.1	-9.5	-7.8
7	Epicatechin 3-0-(3- 0-methylgallate)	-8.9	-9.5	-8.7	-7.8
8	Granulatamide B	-7.9	-7.8	-8.0	-5.9
9	Ixoside	-7.6	-8.6	-8.5	-7.4
10	Lepidotol A	-9.1	-9.5	-9.4	-7.6
11	Melicarpinone	-7.1	-7.7	-7.6	-6.5
12	Oxiamycin	-8.7	-9.8	-9.4	-8.3
13	Phytosterols	-8.0	-10.5	-9.0	-6.9
14	Sinensin	-8.8	-9.4	-9.6	-8.2
15	Econazole	-8.0	-8.2	-7.5	-6.6
16	Ethambutol	-4.6	-5.6	-5.7	-4.3
17	Ethionamide	-5.2	-5.5	-6.3	-4.7
18	Isoniazid	-5.6	-5.6	-6.3	-4.9
19	Kanamycin	-7.2	-7.8	-7.2	-7.1
20	Pyrazinamide	-4.9	-5.5	-5.2	-4.6
21	Rifampicin	-4.8	-3.3	-7.6	-9.4

Interaction of 2UVN protein with top ligands

Table 6. Interaction of top ligands and (2UVN) protein

Compounds	Hydrogen bond Interaction		Hydrophobic interaction		Others	
	Numbers	Residues	Numbers	Residues	Num bers	Residues
3,5-Dihydroxy- 7-methoxy- 2-(4-methoxyphe nyl)-4H-chromen-4-one	3	TYR318(2.23) **THR247(3.75)** **CYS354(3.84)**	12	**ALA360 (3.94) (5.04)** LEU293 (5.36) MET251(3.79) LEU284 (5.08) **PRO289 (3.98) (5.49)**		

				PHE347(5.34) HIS352(5.30) **VAL290(5.20) (4.93)** **CYS354(4.86)**		
Amycocyclo Piazonic Acid			4	**PRO87(3.64) (4.65)** **VAL393 (5.10) (5.25)**		
Annonamine	2	GLY244(3.67) **CYS354(3.66)**	4	**PHE347(4.79)** **PRO289(4.87)** **CYS354(5.47)** **ALA360(4.72)**	1	**MET240(5. 87)**
Melicarpino Ne	4	ASN177(2.28) GLY243(2.38) SER388(2.36)	2	**VAL393(3.92)** **PRO87(5.01)**		

Table 7. Interaction of Top Ligands and 1SFR

Compounds	Hydrogen bond Interaction		Hydrophobic interaction		Others	
	numbers	Residues	Numbers	Residues	Numbers	residues
2,3-Anthracene Dione	1	**LEU152 (2.59)**	3	ALA167(4.38) (5.13) **LEU163(4.70)**		
3,5- Dihydroxy-7-methoxy-2-(4-methoxyphenyl)- 4H-chromen-4-One	3	**ARG43 (2.34)** **SER126 (2.10)** **HIS262 (2.78)**	10	LEU152(4.05) ILE164(4.41) LEU125(4.98) **TRY264(4.24)** **LEU42(5.03) (5.22)** **LEU229(5.40)** **(5.20)** ALA167(4.73)		
10-Hydroxy octadecadienoic acid			10	ALA167(4.62) (4.94) (4.01) LEU152(5.02) LEU164(4.66) LEU42(5.32) PHE78(5.34) LEU229(4.60) (4.45) HIS262(4.89)		
Amycocyclopiazonic Acid	1	Ser126(2.47)	5	Ala167(3.67)(4.68) Leu229(3.80)(4.81) Leu42(5.18)		

Annonamine	1	Leu163(3.58)	5	Leu42(3.54) Ala167(4.01)(4.84) Leu229(5.42)(5.10)		

Table 8. Interaction of Top Ligands and 2O0T

Compound s	Hydrogen bond Interaction		Hydrophobic interaction		Others	
	number s	Residues	Numbe rs	Residues	Numbers	residues
2,3- Anthracene dione						
3,5- Dihydroxy- 7-methoxy- 2-(4- methoxyph enyl)- 4H- chromen-4- one	4	ARG161(2.62) **HIS213(2.89)** **ARG303(2.25)** GLU197(3.65)	8	**TYR405(3.82)** **(4.56)** HIS114(5.06) ALA70(4.39) (4.85) ALA71(4.45) CYS93(4.58) TYR348(4.55)		
10- Hydroxy octadecadie noic acid	1	**ARG303(2.26)**	3	HIS114(3.54) PHE183(5.24) **HIS213(4.44)**		
Amycocycl opiazonic acid	2	ARG161(2.34) GLU97(2.24)	4	HIS114(3.57) ALA70(5.03) CYS43(4.58)(4.92)	1	AlA73(4.65)
Annonamin e	3	ARG344(2.29) **ARG303(3.72)** SER216(3.08)	3	TYR348(4.76) (3.99) **TYR405(4.95)**	1	HIS169(4.95)
Dihydrokae mpferol 7- glucoside	3	ARG344(2.22) GLU300(2.17) SER216(2.77)	1	TYR348(4.26)	1	**HIS213(3.92)**
Epicatechin 3-0-(3-0- methylgalla te)	5	ARG161(2.53) GLU300(2.87)(1.89)(3.41) **HIS213(3.17)**	6	ALA70(3.38)(4.10) PHE183(5.52) **HIS213(5.26)(** **5.05)** **TYR405(4.84)**		
Granulatam ide B	4	ALA73(2.23) ASP91(2.59) GLU97(2.67) GLU300(2.61)	6	ALA70(3.78)(3.63) **HIS213(3.73)** HIS114(4.62) CYS93(5.04) PHE183(4.81)		
Ixoside	5	GLY258(2.79) **ARG303(1.78)(2.30)** ARG344(2.16) **TYR405(2.00)**				

Lepidotol A			7	HIS114(3.82)(3.98) CYS93(5.41) **HIS213(4.75)(4.90)** PHE183(4.83) ALA70(4.69)	1	ALA73(4.69)
Melicarpin one	3	ARG161(2.67) **HIS213(3.44)** GLU97(3.16)	5	ALA70(4.22)(4.21) CYS93(4.61)(5.34) ALA71(5.01)		
Oxiamycin	5	**HIS213(2.80)(3 .50)** GLY257(3.00) GLU300(2.24)(2.87)	2	HIS114(3.75) LEU409(5.38)		
Phytosterol s	1	SER216(2.58)	4	HIS114(3.80) ALA70(5.13) CYS93(4.43) **HIS213(4.63)**		
Sinensin	7	**HIS213(2.53)** **ARG303(2.83)** SER216(2.43) GLY257(2.46) GLU300(2.59)(4.99) GLU179(2.18)			2	**HIS213(4.05)** GLU167(4.90)
Ethambutol	2	GLU300(1.93) ASP91(2.73)	1	**HIS213(3.89)**		
Ethionamid e			2	ALA70(3.73) **HIS213(4.51)**		
Isoniazid	3	GLU300(2.28)(2.35)(3.42)	2	HIS114(4.71) ALA70(5.12)	1	GLU300(3.83)
Kanamycin	8	**HIS213(2.96)(2**				
		.91)(3.88) **GLU257(2.72)** **ARG344(2.08)** **GLU300(2.51)(2.97)** **GLY257(3.28)**				
Pyrazinami de	4	**ARG161(2.51)** **GLU300(1.99)(2.29)(3.47)**	1	ALA70(4.39)	1	HIS213(4.07)
Rifampicin	5	**ARG161(2.62)** **SER216(2.18)** **ARG303(2.31)** **TYR348(3.57)** **TYR405(3.50)**			2	ARG161(4.39) HIS213(3.34)

Table 9: Interaction of Top Ligands and 2PKF

Compounds	Hydrogen bond Interaction		Hydrophobic interaction		Others	
	numbers	Residues	Number s	Residues	Number s	residues
2,3-Anthracene dione			1	**VAL255(4.77)**		
3,5-Dihydroxy- 7-methoxy-2-(4-methoxyph enyl)- 4H-chromen-4- one	3	**ASN195(2.11)** THR253(3.79) **SER281(3.67)**	6	**VAL255(3.70) (5.27)** VAL229(4.76) VAL243(5.06) **PHE259(4.58)** VAL284(4.62)		
10-Hydroxy octadecadie noic acid	3	VAL244(2.14) GLY256(2.88) **SER281(1.91)**	10	VAL243(4.40) (5.27) PRO252(5.15) **VAL255(4.56) (4.13)** ALA284(3.61) (5.25)(4.74)		

				LEU288(4.83) **PHE259(4.80)**		
Amycocycl opiazonic acid			1	**VAL255(4.88)**		
Annonamin e			4	**VAL255(4.70)** LEU288(5.44) VAL243(5.20) ALA284(4.20)		
Dihydrokae mpferol 7-glucoside	6	**THR223(2.34)** **(2.29)** GLY256(2.15) GLU198(2.38) PRO252(2.26) SER171(3.22)	6	VAL243(3.88) ALA284(3.82) **VAL255(5.12)** **(5.37)** VAL285(5.14) **LEU288(5.38)**		
Epicatechin 3-0-(3-0-methylgalla te)	5	GLY225(2.53) (2.36) GLU246(2.91) GLY228(2.32) ASP196(2.33)	12	**LEU288(3.97)** GLY225(4.05) PRO226(4.05)(5.37) ALA284(3.97) (5.14)		
				VAL243(4.67) **VAL255(5.21)** **(5.40)(5.30)** VAL285(4.42) VAL243(5.42)		
Granulatam ide B	1	**SER281(2.10)**	7	VAL243(3.56) (3.54) ALA284(3.61) (4.19) PRO226(5.10) **VAL255(5.08)** **LEU288(4.43)**		
Ixoside	4	GLY256(2.65) VAL244(2.14) (3.56) GLY225(2.48)				
Lepidotol A	2	**THR223(2.26)** GLY256(3.09)	12	**VAL255(3.59)** **(5.08)** PRO226(4.27) VAL243(4.74) (5.38) ALA284(4.93) (4.45) VAL285(5.22) **LEU288(5.15)** **(3.98)(4.80)** VAL229(5.18)		
Melicarpin one	2	GLY225(3.60) VAL244(3.73)	5	**VAL255(3.65)** **(4.88)** VAL243(4.97) **LEU288(4.86)** ALA284(4.65)		
Oxiamycin	2	GLY256(2.53) **ASP257(2.17)**	2	**VAL255(3.55) (4.89)**		
Phytosterol s			3	**VAL255(4.36)** ALA284(5.29) **LEU288(5.19)**		
Sinensin	5	THR225(2.43) GLY254(2.53) ARG260(2.32) PRO252(2.60) VAL244(2.26)	6	VAL243(3.77) ALA284(3.92) **VAL255(5.42)** **(5.42)** VAL285(5.24) **LEU288(5.42)**		
Ethambutol	2	SER281(2.12) GLY225(1.90)				
Ethionamid e	3	SER281(2.82)	3	VAL243(3.86)		

		GLY225(2.21) GLY228(2.21)		VAL285(4.25) LEU288(4.73)		
Isoniazid	4	GLY228(3.02) VAL244(2.74) (2.92) SER281(2.65)	4	PHE259(4.60) VAL229(5.10) VAL243(5.35) ALA284(4.58)		
Kanamycin	1	ARG260(2.94)				
Pyrazinami de	2	SER281(2.63) GLY228(2.81)	2	VAL243(3.44) ALA284(3.64)		
Rifampicin	6	GLY172(3.09) ASN195(2.80) GLY254(2.89) (2.88) GLY256(2.48) ASP257(2.02)				

THE (2D) AND (3D) SCHEMATIC INTERACTION OF LIGANDS AND TARGET PROTEIN

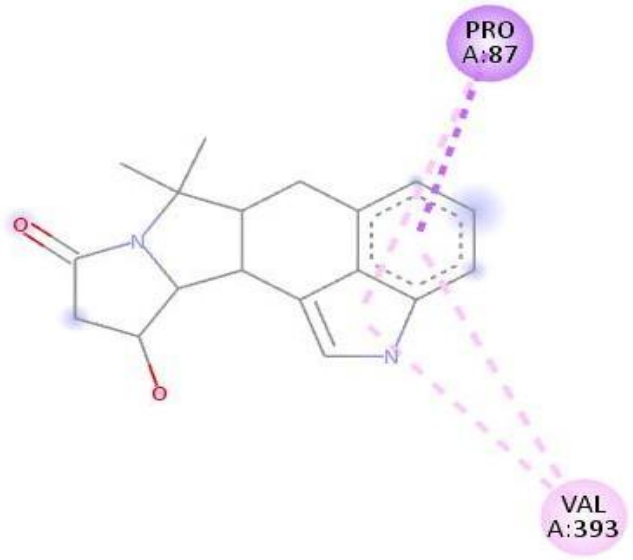

Figure 1. Amycocyclopiazonic Acid Complex

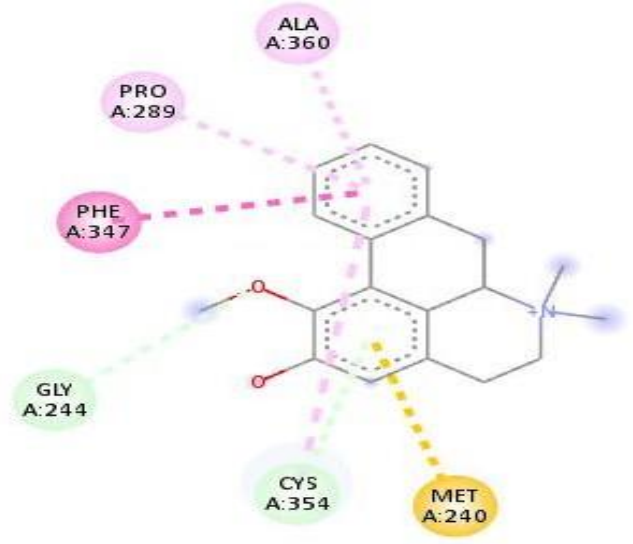

Figure 2. Annonamine Complex

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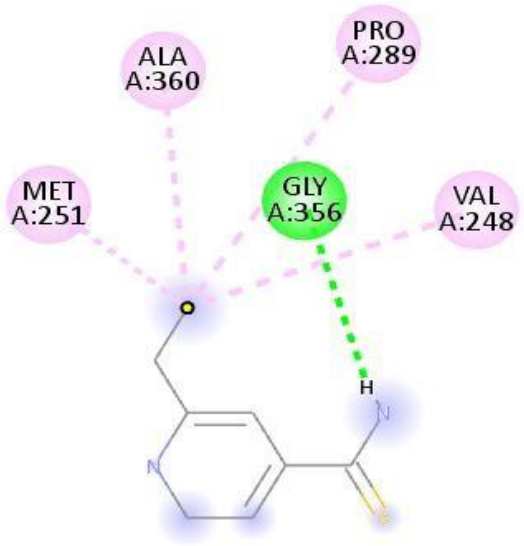

Figure 3. Ethionamide Complex

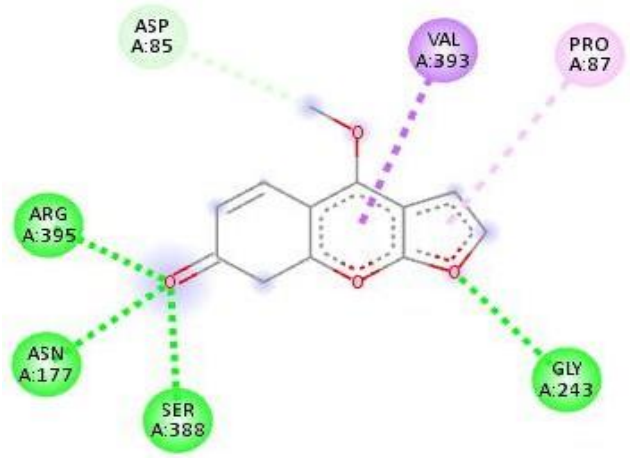

Figure 4. Melicarpinone Complex

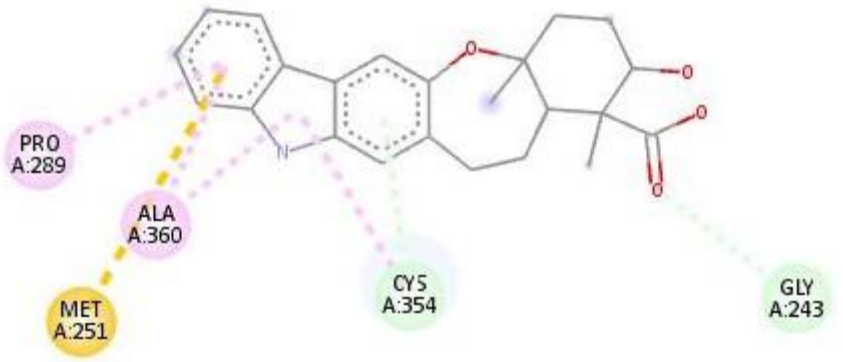

Figure 5. Oxiamycin Complex

DRUG-LIKENESS ANALYSIS

Drug-likeness test was carried out. Lipinski rule of five, which involves: ≤ 500 Da molecular weight (MM), ≤ 5 hydrogen bond donors (HBD), ≤ 10 hydrogen bond acceptors (HBA), partition coefficient (logP) score was applied to screen the top four phytocompounds derived with high binding interaction as shown in Table 4.5 All 4 phytocompounds passed the drug-

likeness filtering. This test was carried out to ensure that the phytocompounds possess drug-like properties; physiochemical properties. generally, a drug should not be too heavy that is why the molecular weight of drugs should be ≤500 molecular weight. A drug should have ≤5 hydrogen bond donors so it won't attack the biomolecules in the body and ≤10 hydrogen bond acceptors so it can be metabolized by the body.

Compounds with fewer (and preferably no) violations of these rules are considered drug-like and are more likely to be orally available. It was observed from the results that 4 of the top compounds (quercetin, epicatechin, oleandrin and quercetin-3-methyl ether) obeyed these rules as shown in (Table VII). The results therefore proves that the compounds possessed drug-like features, to be considered for future drug developments.

Drug likeness results of top compounds that exhibited high binding interaction with BChE target protein

Table 10. Drug likeness screening result of top compounds

Compounds	Lipinski	Ghose	Veber	Egan	Muegge	Passed
3,5-Dihydroxy-7-methoxy-2-(4-methoxyphenyl)- 4H-chromen-4-one	Yes	Yes	Yes	Yes	Yes	Passed
Amycocyclopiazonic acid	Yes	Yes	Yes	Yes	Yes	Passed
Annonamine	Yes	Yes	Yes	Yes	Yes	Passed
Melicarpinone	No	Yes	Yes	Yes	Yes	Passed
Oxiamycin	Yes	Yes	Yes	Yes	Yea	Passed
Ethambutol	Yes	Yes	Yes	Yes	Yes	Passed
Ethionamide	Yes	Yes	Yes	Yes	No	Passed

ADMET analysis of top compounds

Table 11. ADMET Analysis Result of Ethionamide

PROPERTY PREDICTED VALUE	MODEL NAME	VALUE
ABSORPTION (mol/L)	Human intestinal absorption	0.003
	P-glycoprotein substrate	0.008
	P-glycoprotein Inhibitors	0.002
	Lipophilicity	1.58
	Solubility	-2.60
	Caco2 permeability (log Papp)	-4.372
	Bioavailability Aqueous Solubility(logmol/L)	0.001(20%) 0.001(30%) 2.50e-03mol/L
DISTRIBUTION (L/kg)	VDss(Human)	1.951
	Fraction Unbound(human)	68.17%
	BBB permeability	0.903
	Plasma protein binding	28.86%
METABOLISM	CYP 1A2 inhibitor	0.767
	CYP2C19 inhibitor	0.101
	CYP2C9 inhibitor	0.037
	CYP2D6 inhibitor	0.022
	CYP3A4 inhibitor	0.06
EXCRETION(mL/min/kg)	CL	7.022
	t1/2	0.461
TOXICITY	AMES Toxicity	0.029
	Max. Tolerated dose (human)	0.034

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	hERG Blockers	
	Human hepatoxicity	0.026
	Carcinogenicity	0.699
	Respiratory Toxicity	0.087
	Skin Sensitization	0.11
		0.77

Table 12. ADMET analysis result of Amycocyclopiazonic Acid

PROPERTY PREDICTED VALUE	MODEL NAME	VALUE
ABSORPTION (mol/L)	Human intestinal Absorption	0.006 0.414
	P-glycoprotein substrate	0.042
	P-glycoprotein inhibitors	1.92
	Lipophilicity	-2.89
	Solubility	-4.802
	Caco2 permeability (log Papp)	0.009(20%) 0.102(30%)
	Bioavailability	1.30e-03mol/L
	Aqueous Solubility(logmol/L)	
DISTRIBUTION (L/kg)	VDss(Human)	0.96
	Fraction Unbound(human)	24.42%
	BBB permeability	0.974
	Plasma protein binding	76.22%
METABOLISM	CYP 1A2 inhibitor	0.069
	CYP2C19 inhibitor	0.79
	CYP2C9 inhibitor	0.657
	CYP2D6 inhibitor	0.053
	CYP3A4 inhibitor	0.741

Table 13. ADMET analysis result of Melicarpinone

PROPERTY PREDICTED VALUE	MODEL NAME	VALUE
ABSORPTION (mol/L)	Human intestinal absorption	0.001
	P-glycoprotein substrate	0.003
	P-glycoprotein inhibitors	0.861
	Lipophilicity	2.25
	Solubility	-2.47
	Caco2 permeability (log Papp)	-4.547
	Bioavailability	0.006(20%) 0.345(30%)
	Aqueous Solubility(logmol/L)	3.43e-03mol/L
DISTRIBUTION (L/kg)	VDss(Human)	0.894
	Fraction Unbound(human)	14.28%
	BBB permeability	0.019
	Plasma protein binding	84.63%
METABOLISM	CYP 1A2 inhibitor	0.984
	CYP2C19 inhibitor	0.738
	CYP2C9 inhibitor	0.345
	CYP2D6 inhibitor	0.306
	CYP3A4 inhibitor	0.435
EXCRETION (mL/min/kg)	CL	9.52
	t1/2	0.51
TOXICITY	AMES Toxicity	0.017

	Max. Tolerated dose(human)	0.164
	hERG Blockers	0.032
	Human hepatoxicity	0.731
	Carcinogenicity	0.959
	Respiratory Toxicity	0.964
	Skin Sensitization	0.487

Table 14. ADMET analysis result of Annonamine

PROPERTY PREDICTED VALUE	MODEL NAME	VALUE
ABSORPTION (mol/L)	Human intestinal absorption	0.945
	P-glycoprotein substrate	0.994
	P-glycoprotein inhibitors	0.001
	Lipophilicity	0.67
	Solubility	-3.98
	Caco2 permeability (log P_{app})	-5.495
	Bioavailability	0.999(20%)
	Aqueous Solubility(logmol/L)	0.977(30%)
		1.05e-04mol/L
DISTRIBUTION (L/kg)	VDss(Human)	3.042
	Fraction Unbound(human)	54.58%
	BBB permeability	0.257
	Plasma protein binding	39.69%
METABOLISM	CYP 1A2 inhibitor	0.425
	CYP2C19 inhibitor	0.021
	CYP2C9 inhibitor	0.001
	CYP2D6 inhibitor	0.227
	CYP3A4 inhibitor	0.04
EXCRETION (mL/min/kg)	CL	3.466
	t1/2	0.877
TOXICITY	AMES Toxicity	0.395
	Max. Tolerated dose(human)	0.949
	hERG Blockers	0.325
	Human hepatoxicity	0.031
	Carcinogenicity	0.032
	Respiratory Toxicity	0.385
	Skin Sensitization	0.118

Table 15. ADMET analysis result of Oxiamycin

PROPERTY PREDICTED VALUE	MODEL NAME	VALUE
ABSORPTION (mol/L)	Human intestinal absorption	0.009
	P-glycoprotein substrate	0.011
	P-glycoprotein inhibitors	0.004
	Lipophilicity	2.28
	Solubility	-5.35
	Caco2 permeability (log Papp)	-4.96
	Bioavailability	0.01 (20%)
	Aqueous Solubility(logmol/L)	0.59 (30%)
		4.47e-06mol/L
DISTRIBUTION (L/kg)	VDss (Human)	0.427
	Fraction Unbound(human)	1.982%
	BBB permeability	0.217
	Plasma protein binding	96.62%
METABOLISM	CYP 1A2 inhibitor	0.359
	CYP2C19 inhibitor	0.028
	CYP2C9 inhibitor	0.05
	CYP2D6 inhibitor	0.035
	CYP3A4 inhibitor	0.047
EXCRETION (mL/min/kg)	CL	4.146
	t1/2	0.268
TOXICITY	AMES Toxicity	0.025
	Max. Tolerated dose(human)	0.933
	hERG Blockers	0.026
	Human hepatoxicity	0.696
	Carcinogenicity	0.409
	Respiratory Toxicity	0.955
	Skin Sensitization	0.379

4. DISCUSSION

In molecular modelling, docking is used to predict the preferred orientation of a ligand when it binds to a target, forming a stable complex.[11] For virtual screening and molecular docking, we utilized AutoDock Vina—a program designed to accelerate and improve the accuracy of binding mode predictions. Table 2 presents the Cyp130 protein receptor and the ligands used in this study, while Table 3 shows the docking results between Cyp130 and the ligands. The binding affinities of Annonamine, Amycocyclopiazonic acid, Oxiamycin, Melicarpinone, and Ethinamide with Cyp130 are provided in Table 5. Tables 6 to 8 detail the chemical interactions between these ligands and key amino acid residues of Cyp130 (Leu71, Pro87, Pro88, Phe236, Thr239, Met240, Thr247, Pro289, Val290, Phe347, Cys354, Leu355, Gly356, Ala359, Ala360, and Val393) as well as those of the 1SFR receptor (Asp40, Leu42, Arg43, Ser126, Leu152, Leu163, Leu229, Phe232, His262, Trp264, and Trp267). Furthermore, Tables 9 and 10 summarize the bond numbers, types, and lengths between the ligands and the amino acid residues, including hydrogen bonds, hydrophobic interactions, and others.

Medicinal plants are a rich source of phyto-compounds with numerous therapeutic benefits. Many plant-derived compounds and extracts have demonstrated antimycobacterial activity, and oral administration remains the most convenient route for patients. Since a drug must be absorbed across the small intestine's epithelium to be effective, high intestinal absorption is crucial. The results in Tables 11–15 indicate that all the compounds exhibit high absorption based on empirical criteria. For instance, the absorption values for Annonamine, Amycocyclopiazonic acid, Oxiamycin, Melicarpinone, and Ethinamide were 0.945, 0.006, 0.009, 0.001, and 0.003, respectively, demonstrating their potential for intestinal uptake.

The blood–brain barrier (BBB) is commonly used to assess the extent of drug penetration into the central nervous system. Because experimental determination of BBB permeability is time-consuming and costly, in silico methods are often employed before chemical synthesis. The BBB value is defined as the ratio of the brain concentration to the blood concentration of a compound at a steady state. For peripherally acting drugs, limited BBB penetration is desirable to avoid adverse CNS effects. According to the results in Tables 11-15, the BBB values for Annonamine, Amycocyclopiazonic acid, Oxiamycin, Melicarpinone, and Ethinamide are 0.257, 0.974, 0.217, 0.019, and 0.903, respectively. Values below the standard empirical range of 0–0.3 are considered excellent (green), 0.3–0.7 medium (yellow), and 0.7–1.0 poor (red), indicating that these compounds are excellent candidates due to their low BBB permeability.

Enzymatic metabolism—the chemical biotransformation of drugs—is critical for drug conversion and clearance. The cytochrome P450 (CYP) enzyme system, including families such as 1A2, 2C19, 2C9, 2D6, and 3A4, plays a major role in this process. Our results (Tables 11–15) show that Annonamine, Amycocyclopiazonic acid, Oxiamycin, Melicarpinone, and Ethinamide act as inhibitors of these enzymes.

Oral delivery is the most common route of administration, relying on absorption through the digestive tract. P-glycoproteins, embedded in the cell membrane, facilitate the intracellular transport of many drugs; however, their inhibition can affect normal drug distribution. In vitro permeability studies using the Caco-2 cell line have demonstrated that these compounds are readily absorbed in the intestine. [10,12] All compounds—except Annonamine, which exhibited higher inhibition potentially affecting its systemic circulation—were reported to be non-inhibitory to P-glycoprotein, permeable through the BBB, and effectively absorbed in human intestinal tissue, as confirmed by studies using Caco-2 cell monolayers. [13-17]

Acute oral toxicity, expressed as the median lethal dose (LD_{50}), is the dose required to kill 50% of test animals within 24 hours. Chemicals are classified into four toxicity categories: Category I (highly toxic and irritant), Category II (moderately toxic and irritant), Category III (slightly toxic and irritant), and Category IV (non-toxic and non-irritant).[10,18] Based on our analysis, Annonamine and Ethionamide—with values of 0.395 and 0.029, respectively—fall under Category III (slightly toxic and irritant); Oxiamycin, with a value of 0.025, falls under Category IV (non-toxic and non-irritant); while Amycocyclopiazonic acid and Melicarpinone, with values of 0.024 and 0.017 respectively, are classified as Category I (highly toxic and irritant). Notably, none of the selected ligands exhibited AMES toxicity or carcinogenicity.[19] Mutagenicity, typically assessed by AMES toxicity tests, is a critical endpoint in evaluating chemical safety. [20] The observed effects on multiple enzymes suggest that these compounds may be rapidly degraded and excreted from the body [21,22] Considering all the parameters evaluated in this study, the compounds investigated appear to be promising enhancers of Cyp130 activity in the signalling pathway, thereby contributing to the treatment of *Mycobacterium tuberculosis*.

5. CONCLUSION

This study adopted an in-silico approach to analyze the binding profiles of newly designed compounds as potential anti-TB candidates. Using a template scaffold, ligand compounds were designed that exhibited improved binding affinities compared to both the scaffold template (6.8 kcal/mol) and the standard drug ethionamide (5.5 kcal/mol). All docking results for these ligands with the target protein ranged from –4.3 to –9.5 kcal/mol. Furthermore, drug-likeness and pharmacokinetic predictions revealed that the selected ligands adhered to Lipinski's rules, exhibited similar bioavailability, and demonstrated high gastrointestinal absorption, while toxicity assessments predicted them to be non-toxic in terms of carcinogenicity and cytotoxicity.

REFERENCES

[1] World Health Organization. Global tuberculosis control: WHO report 2012. Geneva: World Health Organization; 2012.

[2] Ahmad S, Mokaddas E. Current status and future trends in the diagnosis and treatment of drug-susceptible and multidrug-resistant tuberculosis. J Infect Public Health. 2014;7(2):75-91.

[3] Ayon NJ. High-Throughput Screening of Natural Product and Synthetic Molecule Libraries for Antibacterial Drug Discovery. Metabolites. 2023;13(5):625. doi:10.3390/metabo13050625.

[4] Sacchettini JC, Rubin EJ, Freundlich JS. Drugs versus bugs: in pursuit of the persistent predator Mycobacterium tuberculosis. Nat Rev Microbiol. 2008;6(1):41-52.

[5] Koul A, Arnoult E, Lounis N, Guillemont J, Andries K. The challenge of new drug discovery for tuberculosis. Nature. 2011;469:483-490. doi:10.1038/nature0965.

[6] Brötz-Oesterhelt H, Sass P. Postgenomic strategies in antibacterial drug discovery. Future Microbiol. 2010;5(10):1553-1579.

[7] Aderem A. Systems biology: its practice and challenges. Cell. 2005;121(4):511-513.

[8] Ali A, Hasan Z, McNerney R, Mallard K, Hill-Cawthorne G, Coll F, Nair M, Pain A, Clark TG, Hasan R. Whole genome sequencing based characterization of extensively drug-resistant Mycobacterium tuberculosis isolates from Pakistan. PLoS One. 2015;10(2):e011777.

[9] Ayanwale AO, Adabara NU, Adeniyi AK, Oyewole SO, Ogonna DW. Mechanism of drug resistance in Mycobacterium tuberculosis. 2020.

[10] Li AP. Screening for human ADME/Tox drug properties in drug discovery. Drug Discov Today. 2001;6(7):357-366.

[11] Lengauer T, Rarey M. Computational methods for biomolecular docking. Curr Opin Struct Biol. 1996;6(3):402-406.

[12] Fukunaga R. Epidemiology of tuberculosis and progress toward meeting global targets—worldwide, 2019. MMWR Morb Mortal Wkly Rep. 2021;70.

[13] Kumar A, Toledo JC, Patel RP, Lancaster JR 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.

[14] Halezeroğlu S, Okur E. Thoracic surgery for haemoptysis in the context of tuberculosis: what is the best management approach? J Thorac Dis. 2014;6(3):182.

[15] Danilchanka O, Sun J, Pavlenok M, Maueröder C, Speer A, Siroy A, Marrero J, Trujillo C, Mayhew DL, Doornbos KS, Muñoz LE, Herrmann ME, Ehrt S, Berens C, Niederweis M. An outer membrane channel protein of Mycobacterium tuberculosis with exotoxin activity. Proc Natl Acad Sci U S A. 2014;111(18):6750-6755. doi:10.1073/pnas.1400136111.

[16] Chung BKS, Dick T, Lee DY. In silico analyses for the discovery of tuberculosis drug targets. J Antimicrob Chemother. 2013;68(12):2701-2709.

[17] Chen ZL, Wei LL, Shi LY, Li M, Jiang TT, Chen J, Lui CM, Yang S, Tu HH, Hu YT, Gan L, Mao LG, Wang C, Li JC. Screening and identification of lncRNAs as potential biomarkers for pulmonary tuberculosis. Sci Rep. 2017;7(1):16751.

[18] Macalino SJY, Billones JB, Organo VG, Carrillo MCO. In silico strategies in tuberculosis drug discovery. Molecules. 2020;25(3):665.

[19] Pai M, Behr M. Latent Mycobacterium tuberculosis infection and interferon-gamma release assays. Microbiol Spectr. 2016;4(5):10-1128.

[20] World Health Organization. WHO consolidated guidelines on drug-resistant tuberculosis treatment. WHO/CDS/TB/2019.7. Geneva: World Health Organization; 2019.

[21] Vaidya S, Maniar J, Muley S, Koppikar G, Kulkarni M, Chowdhary A. Relationship between rpoB mutations and minimum inhibitory concentrations of rifampicin in multidrug-resistant strains of Mycobacterium tuberculosis. LIFE Int J Health Life Sci. 2015;1(1).

[22] Stehr M, Elamin AA, Singh M. Pyrazinamide: the importance of uncovering the mechanisms of action in mycobacteria. Expert Rev Anti Infect Ther. 2015;13(5):593-603. PubChem [Internet]. Bethesda (MD): National Center for Biotechnology Information. Available from: https://pubchem.ncbi.nlm.nih.gov.