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Docking Studies, Synthesis, SAR and Anti-TB Activity of Glycinamido Analogues

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Mycolic acid is a crucial component of the *Mycobacterium tuberculosis* cell wall and mycolic acid methyltransferases (MAMTs) are essential for mycolic acids to mature. In the present study, an inhouse library of 330 ligands was designed taking glycinamido moiety as scaffold. Virtual screening was carried out with this library of compounds against MmaA1 as the target protein. About 55 hits were identified through docking, ADMET studies and these molecules were synthesized by the Schotten-Baumann reaction followed by a nucleophilic substitution reaction. All the compounds were subjected to *in vitro* anti-Tb screening by microplate alamar blue assay (MABA). The Mdb1, Mdb4 & Meb1 exhibited excellent activity against *M. tuberculosis* H37Rv bacilli strain with an MIC of 1.56 µg/mL. The SAR studies shows that the aryl ring attached directly to the nitrogen atom as present in 2(-N-substituted glycinamido) derivatives is essential for the compound to exhibit potent anti-TB activity.

Keywords: Glycinamido derivatives, Mycolic acids, Docking studies, *Mycobacterium tuberculosis*.

INTRODUCTION

The bacillus *Mycobacterium tuberculosis* is the primary cause of the deadly disease tuberculosis (TB). Tuberculosis patients cough out bacteria into the air, spreading the disease [1,2]. Most TB cases (pulmonary TB) involve the lungs, however, extrapulmonary TB can affect other parts of the body as well. According to the WHO TB Report 2022, 10.6 million people (95% UI: 9.9-11 million) are predicted to have contracted TB globally in 2021, an increase of 4.5% from 10.1 million (95% UI: 9.5-10.7 million) in 2020 [3], which indicates that the years of reduction have now reversed. At this time, drug-resistant tuberculosis (DR-TB) continues to threaten public health. Multidrug-resistant tuberculosis (MDR-TB) is characterized by resistance to rifampicin and isoniazid [4]. In order to treat both Rifampicin-resistant tuberculosis (RR-TB) and multidrug-resistant tuberculosis (MDR-TB), second-line medicine is required. In 2021, three nations accounted for 42% of the world's MDR/RR TB cases, with India accounting for 26% [5]. New drugs with novel mechanisms are therefore urgently required to treat TB, shorten the period of MDR-TB and RR-TB treatment and effectively aid in TB control [6,7].

The cell wall of *M. tuberculosis* is composed of a peptidoglycan (PG) layer that is covalently connected to arabinogalactan (AG), which acts as an attachment site for certain mycolic acids (MA). The cell wall core is commonly known as mycolyl-arabinogalactan-peptidoglycan (mAGP) complex [8]. The primary virulence factor is the mycolic acids, which provide *M. tuberculosis* an inherent resistance to most antibiotics [9]. The main regulators of *M. tuberculosis* cell wall formation are fatty acid synthases (FAS-I, FAS-II, KasA, KasB, MabA, InhA, HadABC), mycolic acid modifying enzymes (SAM-dependent methyltransferases), fatty acid activating and condensing enzymes (FadD32, Acc, Pks13), transporters (MmpL3) and transferases. FAS-I is involved in fatty acid production in eukaryotes, whereas FAS-II is only found in *M. tuberculosis* cells and is a target of anti-TB medicine [10]. Because these enzymes have no homologs in the mammalian system, enzymes involved in cell wall synthesis afford a plausible molecular target for fighting tuberculosis.

Methyl transferases, modify the meromycolyl chain through cyclopropanation and methylation. *M. tuberculosis* requires these alterations to maintain its pathogenicity, virulence and persistence [11]. Methyl transferase enzymes also help in the

oxygenation of the meromycoloyl chain, which is necessary for *M. tuberculosis* pathogenicity [12]. As a result, mycolic acid methyl transferases (MAMTs) may be promising targets for the discovery and development of novel, selective anti-TB drugs. Recently, a structure-based drug design study carried out in our research lab led to the identification of 3-(2-morpholinoacetamido)-*N*-(3,4-dihydro-4-oxoquinazolin-7-yl)-benzamide and its intermediates as inhibitors of the MmaA1 protein [13]. The intermediate compounds 3-(2-chloroacetamido) benzoic acid or 3-(N-substituted glycinamido) benzoic acid have shown an MIC of 25 µg/mL against *M. tuberculosis* H37Rv, in contrast to the parent molecule's activity of 100 µg/mL. Following more optimization, the derivatives of 3-(N-substituted glycinamido)benzoic acid were designed. Lead optimization study showed that compound 3-(2-(4-phenylpiperazin-1-yl)acetamido)benzoic acid has a docking score of -9.9 and exhibited a MIC of 1.6 µg/mL against *M. tuberculosis* H37Rv [14]. These intriguing findings led us to further advance our lead optimization study for the structural modification of glycinamido moiety considering it as a possible lead to identify potential inhibitors for tuberculosis.

EXPERIMENTAL

All chemicals and solvents were obtained from Merck and Aldrich Chemical Company (USA) and utilized as such. The SRS-EZMelt automated melting point instrument's open capillary tube was used to measure the melting points and are uncorrected. The IR spectra of the compounds were taken by using the KBr disc method on a Bruker ALPHA-T FT-IR spectrometer, the values are expressed in cm^{-1}. The ^{1}H & ^{13}C NMR spectra of the compounds were recorded using DMSO-d_6 on Bruker Avance 400 MHz and 100 MHz NMR spectrometers. The elemental analysis experiments were conducted using Carlo-Erba elemental analyzer.

Design of lead compounds and virtual screening: An attempt was made to prepare a library of compounds with different types of aryl units using various substituted anilines (A1-A4, A9-A11) and substituted phenols (A5-A8). The linker (B1-B3) between aryl and secondary amines was modified by increasing chain length. Hence, different secondary amines (C1-C10) were used including saturated heterocyclic moiety as depicted in Fig. 1. A total of 330 ligands were designed and subjected to virtual screening using Auto Dock.

Docking studies: In AutoDock, docking studies involves different stages, (i) preparation of coordinate files using AutoDockTools followed by (ii) precalculation of atomic affinities using AutoGrid and finally (iii) docking using AutoDock. Auto Dock requires grid maps for each atom type in the ligand which are calculated using AutoGrid. Other files required are pdbqt files containing proteins and ligands as well as docking parameter file. The first step involves the preparation of ligand and receptor coordinate files, which involves addition of polar hydrogen atoms, partial charges and atom types, in AutoDock specific coordinate file format, pdbqt.

Protein preparation: The MmaA1 protein was prepared for docking using AutoDock, the polar hydrogens and Kolman charges were added and the pdbqt file was generated.

Ligand preparation: A total of 330 ligands were modeled using ChemDraw Ultra 10.0 (Cambridge software). The 2D models generated were saved in pdb format and then converted to 3D format using Chem3D ultra10.0. The 3D models were subjected to energy minimization using molecular mechanics (MM2). The minimization was executed until the root mean square gradient value reached a value smaller than 0.001 kcal/mol. The ligands were converted from mol/mol2 to pdbqt and then submitted to docking using AutoDock 4.0 software [15].

Docking was performed on the inhouse library of designed compounds keeping ligands as flexible and receptors as semi-flexible using AutoDock. Pose analysis was done for the least energy conformation of the docked molecules in the output file containing 5 best-docked poses using PyMOL software [16] and Discovery studio [17].

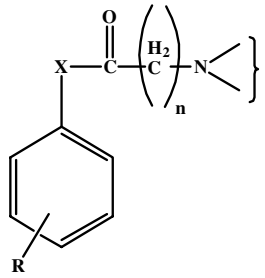
ADMET studies: Clinical failures in later phases of drug discovery are caused by substances with undesired characteristics and poor ADMET (absorption, distribution, metabolism, excretion and toxicity) profiles. The ADMET properties for the compounds were predicted by using Swiss ADME online server [18], pkCSM [19] and Molinspiration [20] softwares. pkCSM offers a platform for analyzing and improving pharmacokinetic and toxicological characteristics using a user-friendly and open-source online interface. A web-based program called Molinspiration (www.molinspiration.com) was used to predict the bioactivity score of synthesized compounds against common human receptors as GPCRs, ion channels, nuclear receptors, kinases and proteases.

Various physico-chemical characteristics, including molecular weight, TPSA (Topological Polar Surface Area), XLOGP3 (lipophilicity), drug-likeness and lead likeness, were predicted by SwissADME [21]. Drug and lead likeness predictions are based on TPSA, lipophilicity and the Lipinski rule of five. The other pharmacokinetic characteristics such as gastrointestinal (GI) absorption, blood-brain barrier (BBB) permeability and inhibition of certain cytochrome P450 (CYP) enzymes were also predicted by Swiss ADME. Due to pharmacokinetic related factors, CYP inhibition is one of the main causes of drug-drug interactions [22,23]. In Molinspiration, the four criteria for successful drug activity have been analyzed for all standard compounds and medications, activity on the G protein-coupled receptor (GPCR) ligand, modulation of ion channels, inhibition of kinases, proteases and other enzymes and activity on nuclear receptor ligands. The likelihood that a molecule will demonstrate significant biological activity depends on its bioactivity score. If the bioactivity score is more than 0.00, the compound is active, if the score is in between the values -0.50 to 0.00, the compounds are moderately active, if the score is less than -0.50, the compound is inactive [24].

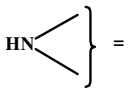
Synthesis: Based on literature search and SciFinder search investigations, the glycinamide analogs identified through the virtual screening study were all evaluated for novelty and synthetic viability.

General procedure for the synthesis of heterocyclic amine substituted 2-(*N*-substituted glycinamido) derivatives:

Synthesis of 2-(2-chloro acetamido) derivatives (step-1): Substituted amine (1 equiv.) was taken in a round bottom flask



X= N, O
R= A_1-A_{11}
n=1-3 represented by B



= heterocyclic amines C1-C10

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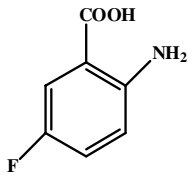

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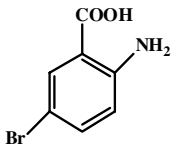

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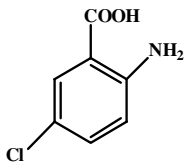

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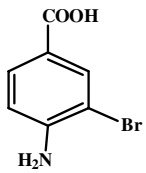

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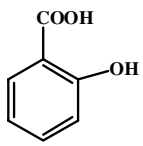

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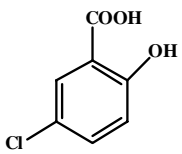

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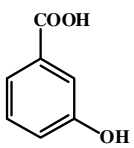

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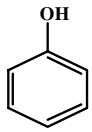

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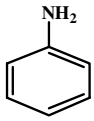

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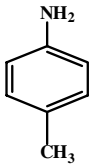

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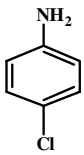
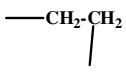
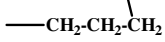


B1:

B2:

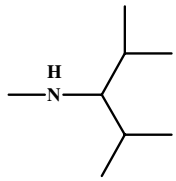


B_3



C_1

C_2



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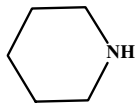

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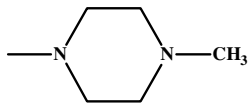
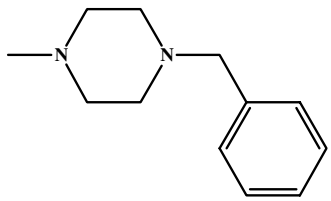
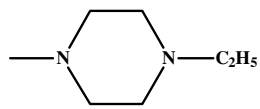


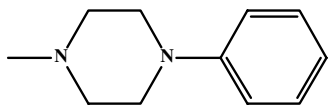
C_5



C_6



C_7



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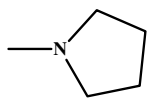

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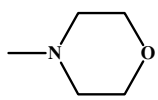


C_{10}

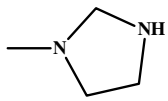


Fig. 1. Types of secondary amines for design of glycinamido analogues

and add 0.7 mL of *N*,*N*-diisopropylethylamine (DIPEA) and stirred at room temperature for 10-15 min. To this reaction mixture, chloroacetyl chloride (4 equiv.) was added dropwise by keeping round bottom flask under tap water as an exothermic reaction occurs [14,25]. Then heated for 10-15 min and monitored for the completion of the reaction by TLC. The reaction mixture was poured into crushed ice-cold water. An obtained white colour precipitate was recrystallized by equal volumes of glacial acetic acid and water.

Synthesis of heterocyclic amine substituted 2-(*N*-substituted glycinamido) derivatives (step-2): 2-(2-Chloroacetamido) derivative (1 equiv.) taken in a round bottom flask added to 5 mL ethyl acetate and kept for stirring at 80 ºC for 10 min. Then heterocyclic amine (2 equiv.) (**b1-b5**) was added and heated at 80 ºC for 2-3 h. The completion of the reaction was monitored by TLC. The solvent was evaporated under vacuum and the resulting residue was washed with hexane to obtain the target compound. The product was purified by recrystallization using hexane and ethyl acetate mixture (**Scheme-I**).

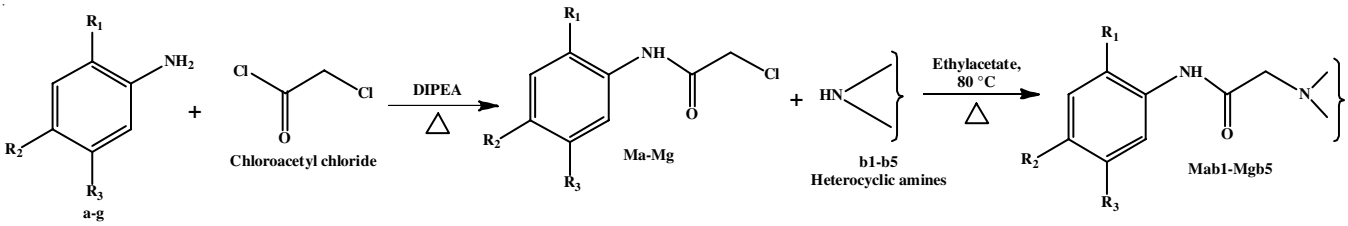
General procedure for the synthesis of heterocyclic amine substituted 2-(acetoxy) derivatives:

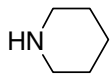
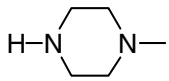
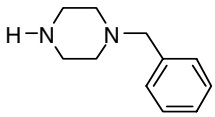
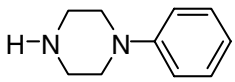
Synthesis of 2-chloroacetate derivatives (step-1): Substituted phenols (1 equiv.) taken in a round bottom flask were added to 0.7 mL of N,N-diisopropylethylamine (DIPEA) and stirred at room temperature for 10-15 min. To this reaction mixture, chloroacetyl chloride (4 equiv.) was added dropwise by keeping round bottom flask under tap water as an exothermic reaction occurs. Then heated for 10-15 min and monitored for the completion of the reaction by TLC [14,25]. The reaction mixture was poured into crushed ice-cold water. An obtained white colour precipitate was further recrystallized by equal volumes of glacial acetic acid and water.

Synthesis of heterocyclic amine substituted 2-(acetoxy) derivatives (step-2): 2-Chloroacetate derivative (1 equiv.) was mixed with 5 mL ethyl acetate in a round bottom flask with constant stirring at 80 ºC for 10 min. Then heterocyclic amine (2 equiv.) (**b1-b5**) was added and heated again at 80 ºC for 2-3 h. The completion of the reaction was monitored by TLC. The solvent was evaporated under a vacuum and the resulting residue was washed with hexane to obtain the target compound (**Scheme-II**). The product was purified by recrystallization using hexane and ethyl acetate mixture.

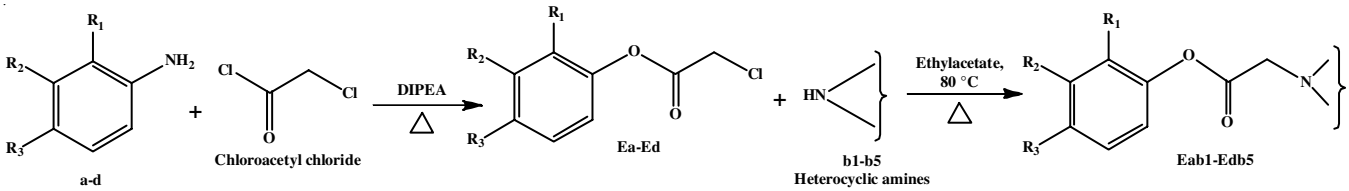
Anti-TB activity: The synthesized compounds (**Mab1-Mgb5** and **Eab1-Edb5**) were tested for *in vitro* anti-TB activity against *Mycobacterium tuberculosis* H37Rv by micro-plate alamar blue assay (MABA) [26]. This method employs a thermally stable reagent, is non-toxic and exhibits a good correlation with the proportion [27] and BACTEC [28] radiometric methods.

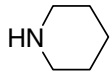
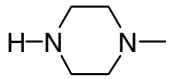
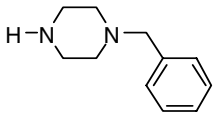
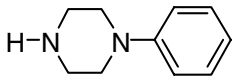
In present work, the compounds have been coded as follows: The inhouse library of 330 compounds were assigned codes as A1B1C1 to A11B3C10. The 55 hits identified from docking and *in silico* ADMET studies had been coded as ABC-1 to ABC-55. The 55 hit molecules were further divided into two groups based on their core structure as 2-(N-substituted glycinamido) derivatives and assigned compound codes as **Mab1-Mgb5** (35 compounds), while the second group are 2-(acetoxy) derivatives coded as **Eab1-Edb5** (20 compounds). The compounds have been assigned codes for ease of identification and no other specific criteria has been used for their classification.



Compound	R_1	R_2	R_3	Compound	Amines
Ma	COOH	F	H	b1	
Mb	COOH	Br	H	b2	
Mc	COOH	Cl	H	b3	
Md	Br	COOH	H	b4	
Me	H	H	H	b5	
Mf	H	CH_3	H		
Mg	H	Cl	H		

Scheme-I



Compound	R_1	R_2	R_3	Compound	Amines
Ea	COOH	H	H	b1	
Eb	COOH	H	Cl	b2	
Ec	H	COOH	H	b3	
Ed	H	H	H	b4	
				b5	

Scheme-II

RESULTS AND DISCUSSION

Chemistry: The synthesis of target compounds involves two steps. Step-1 involves the synthesis of intermediate, 2-(2-chloroacetamides) by using Schotten Baumann's reaction, which entails subsituted amines with chloroacetyl chloride. Then, step-2 undergoes a nucleophilic substitution reaction with a corresponding heterocyclic amine to obtain the target compound.

A total of 35 heterocyclic amine substituted 2-(N-substituted glycinamido) derivatives (**Mab1-Mgb5**) were synthesized. The products were obtained in good yield with a range of 60-99%. The synthesized compounds were characterized by using IR, ^{1}H NMR and ^{13}C NMR techniques. The physico-chemical constants were given in Table-1 and the spectral data contain-ing diagnostic information is presented in Table-2.

The IR spectrum of heterocyclic amine substituted 2-(N-substituted glycinamido) derivatives shows the key signals for

TABLE-1
PHYSICO-CHEMICAL PROPERTIES OF THE COMPOUNDS **Mab1** TO **Mgb5**

Compd.	Yield (%)	m.p. (°C)	Relative molecular mass	m.f.	Compd.	Yield (%)	m.p. (°C)	Relative molecular mass	m.f.
Mab1	99.50	122.2	280.29	$C_{14}H_{17}FN_2O_3$	**Mdb4**	66.50	166.3	418.28	$C_{19}H_{20}N_3O_3Br$
Mab2	69.80	Semi-solid	295.31	$C_{14}H_{18}FN_3O_3$	**Mdb5**	62.10	Semi-solid	418.28	$C_{13}H_{15}N_2O_3Br$
Mab3	76.82	Semi-solid	371.41	$C_{20}H_{22}FN_3O_3$	**Meb1**	54.23	Semi-solid	218.29	$C_{13}H_{18}N_2O$
Mab4	54.40	156.8	357.38	$C_{19}H_{20}FN_3O_3$	**Meb2**	84.22	215.9	233.31	$C_{13}H_{19}N_3O$
Mab5	99.35	143.8	266.27	$C_{13}H_{15}FN_2O$	**Meb3**	95.62	182.3	309.41	$C_{19}H_{23}N_3O$
Mbb1	83.42	122.2	341.20 mg	$C_{14}H_{17}N_2O_3Br$	**Meb4**	65.28	161.4	295.38	$C_{18}H_{21}N_3O$
Mbb2	82.20	Semi-solid	356.20 mg	$C_{14}H_{18}N_3O_3Br$	**Meb5**	74.23	Gummy resin	204.27	$C_{12}H_{16}N_2O$
Mbb3	78.31	Semi-solid	432.32 mg	$C_{20}H_{22}N_3O_3Br$	**Mfb1**	78.32	Gummy resin	232.32	$C_{14}H_{20}N_2O$
Mbb4	84.62	156.8	418.29 mg	$C_{19}H_{20}N_3O_3Br$	**Mfb2**	85.26	Gummy resin	247.34	$C_{14}H_{21}N_3O$
Mbb5	66.20	Semi-solid	327.18 mg	$C_{13}H_{15}N_2O_3Br$	**Mfb3**	92.35	Gummy resin	323.43	$C_{20}H_{25}N_3O$
Mcb1	93.40	192.5	296.75	$C_{14}H_{17}N_2O_3Cl$	**Mfb4**	76.24	Gummy resin	309.41	$C_{19}H_{23}N_3O$
Mcb2	81.58	Gummy resin	311.76	$C_{14}H_{18}N_3O_3Cl$	**Mfb5**	65.21	Gummy resin	218.29	$C_{13}H_{18}N_2O$
Mcb3	86.20	187.8	387.86	$C_{20}H_{22}N_3O_3Cl$	**Mgb1**	91.23	Semi-solid	252.74	$C_{13}H_{17}N_2OCl$
Mcb4	78.30	194.2	373.83	$C_{19}H_{20}N_3O_3Cl$	**Mgb2**	75.26	Semi-solid	267.11	$C_{13}H_{18}N_3OCl$
Mcb5	63.20	Gummy resin	282.08	$C_{13}H_{15}N_2O_3Cl$	**Mgb3**	84.25	Semi-solid	343.85	$C_{19}H_{22}N_3OCl$
Mdb1	88.20	Semi-solid	341.20	$C_{14}H_{17}N_2O_3Br$	**Mgb4**	72.31	Semi-solid	329.82	$C_{18}H_{20}N_3OCl$
Mdb2	79.34	Semi-solid	355.05	$C_{14}H_{18}N_3O_3Br$	**Mgb5**	59.12	Semi-solid	238.71	$C_{12}H_{15}N_2OCl$
Mdb3	84.22	Semi-solid	431.08	$C_{20}H_{22}N_3O_3Br$					

TABLE-2
SPECTRAL DATA OF THE COMPOUNDS **Mab1** TO **Mgb5**

Compd.	IR (KBr, ν_{max}, cm^{-1})	NMR data	Elemental analysis (%)		
				Req.	Found
Mab1	1190 (C-F *str.*), 1228 (C-O *str.*), 1451 (C=C *str.*), 1520 (C=O *str.*), 1689 (C=O *str.*), 2954 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.65 (d, 6H, *J* = 4.8 Hz) 2.98 (s, 4H), 4.95 (m, 2H), 7.68 (m, 4H), 10.35 (1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 22.15, 22.57, 44.0, 88.54, 95.71, 155.22, 204.33, 215.80	C H O	59.99 6.11 17.12	59.98 6.09 17.11
Mab2	1135 (C-F *str.*), 1224 (C-O *str.*), 1417 (C=C *str.*), 1519 (C=O *str.*), 1689 (C=O *str.*), 3388 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 2.51 (s, 3H) 2.74 (m, 8H) 3.16 (s, 2H), 7.4 (s, 1H), 7.69 (d, 1H *J* = 7.6 Hz), 8.69 (m, 1H), 11.87 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 49.85, 52.91, 60.89, 71.68, 117.63, 121.12, 165.61, 167.94, 169.02, 212.57	C H O	56.94 6.14 16.25	56.92 6.13 16.23
Mab3	1189 (C-F *str.*), 1248 (C-O *str.*), 1454 (C=C *str.*), 1515 (C=O *str.*), 1696 (C=O *str.*), 3417 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 2.50 (s, 4H) 2.79 (s, 4H) 3.07 (s, 2H), 3.24 (s, 2H) 7.32 (m, 3H), 7.41 (d, 2H *J* = 6.8 Hz) 7.53 (d, 1H *J* = 5.6 Hz) 8.63 (m, 2H), 12.18 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 43.28, 51.56, 61.89 127.75, 128.77, 129.50, 131.16, 137.42	C H O	64.68 5.97 12.92	64.67 5.96 12.91
Mab4	1143 (C-F *str.*), 1248 (C-O *str.*), 1504 (C=C *str.*), 1596 (C=O *str.*), 1676 (C=O *str.*), 3144 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 2.68 (s, 4H) 3.22 (m, 6H) 6.76 (s, 1H) 6.88 (m, 2H) 6.94 (m, 2H) 7.20 (m, 1H), 7.57 (m 1H) 7.93 (s, 1H) 8.73 (d, 1H *J* = 8.8 Hz), 9.2 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 48.32, 53.41, 75.64, 108.7, 116.5, 119.19, 129.58, 139.98, 150.51, 166.69	C H O	63.85 5.64 13.43	63.83 5.62 13.41
Mab5	1190 (C-F *str.*), 1241 (C-O *str.*), 1423 (C=C *str.*), 1596 (C=O *str.*), 1667 (C=O *str.*), 3475 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.83 (s, 4H) 3.08 (s, 4H) 4.41 (s, 2H) 7.50 (m, 1H) 7.692 (m, 1H) 9.1 (s, 1H), 12.0 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 24.20, 44.93, 61.84, 117.35, 122.31, 136.04, 147.87, 158.62, 165.54	C H O	58.64 5.68 18.03	58.63 5.67 18.02
Mbb1	1038 (C-Br *str.*), 1358 (C-O *str.*), 1447 (C=C *str.*), 1579 (C=O *str.*), 1697 (C=O *str.*), 2950 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.5 (s, 2H), 1.6 (s, 4H), 2.9 (s, 2H), 7.69 (m, 1H), 7.98 (s, 1H), 8.01 (s, 1H), 8.25 (s, 1H), 10.3 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 62.4, 92.3, 121.9, 124.5, 125.4, 133	C H O	49.28 5.02 14.07	49.27 5.01 14.06
Mbb2	1248 (C-Br *str.*), 1358 (C-O *str.*), 1504 (C=C *str.*), 1576 (C=O *str.*), 1667 (C=O *str.*), 3450 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 2.35 (s, 2H) 2.54 (s, 2H), 2.70 (d, 2H,) 2.72 (d, 2H, *J* = 8.8 Hz), 2.77 (d, 2H), 3.13 (s, 2H), 7.64 (d, 1H *J* = 6.8 Hz) 8.08 (s, 1H), 8.56 (s, 1H), 8.58 (s, 1H), 12.7 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 42.58, 45.09, 49.82, 51.17, 53.02, 61.09, 114.38, 119.18, 121.74, 133.96, 135.27, 139.56, 168.53, 169.40	C H O	47.20 5.09 13.47	47.19 5.07 13.46
Mbb3	1130 (C-Br *str.*), 1357 (C-O *str.*), 1573 (C=C *str.*), 1609 (C=O *str.*), 1668 (C=O *str.*), 2819 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 2.46 (s, 2H) 2.91 (d, 2H *J* = 10 Hz), 3.35 (s, 2H), 3.41 (s, 2H), 7.29 (s, 1H) 7.44 (t, 2H, *J* = 6.8 Hz), 7.81 (s, 1H), 8.08 (s, 1H) 8.5 (s, 1H), 8.54 (s, 1H), 13.49 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 50.42, 52.76, 53.41, 62.25, 62.55, 63.1, 113.92, 118.10, 121.37, 127.31, 127.33, 127.63, 128.61, 128.72, 134.02, 137.73, 138.7, 139.57, 169.55, 169.81	C H O	55.57 5.13 11.10	55.55 5.12 11.09
Mbb4	1145 (C-Br *str.*), 1253 (C-O *str.*), 1448 (C=C *str.*), 1500 (C=O *str.*), 1596 (C=O *str.*), 2917 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 2.5 (s, 2H), 2.6 (s, 2H) 3.3 (t, 10H, *J* = 5.6 Hz) 6.9 (s, 2H), 7.2 (m, 2H) 7.5 (dd, 1H, *J* = 2.4 Hz) 8.0 (s, 2H), 8.56 (s, 1H). 8.58 (s, 1H), 13.5 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 40.21, 40.41, 43.24, 46.11, 48.28, 113.98, 115.72, 116.5, 120.51, 129.42, 129.59, 139.19, 140.21, 149.04, 150.51	C H O	54.56 4.82 11.48	54.54 4.81 11.46
Mbb5	1025 (C-Br *str.*), 1582 (C=O *str.*), 1693 (C=O *str.*), 1305 (C-O *str.*), 3323 (COOH), 1501 (C=C *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.05 (m, 4H), 3.06 (s, 2H) 3.16 (s, 1H) 7.36 (m, 2H) 8.07 (s, 1H), 8.53 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 43.4, 49.06, 49.43, 61.92, 99.14, 127.73, 128.77, 128.9, 129.4, 137.5	C H O	47.72 4.62 14.67	47.71 4.60 14.66
Mcb1	1035 (C-Cl *str.*), 1300 (C-O *str.*), 1506 (C=C *str.*), 1584 (C=O *str.*), 1673 (C=O *str.*), 2990 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.428 (s, 2H) 1.556 (d, 2H, *J* = 4.8 Hz) 1.657 (d, 2H, *J* = 4.8 Hz), 3.008 (t, 4H *J* = 5.2 Hz), 3.59 (s, 1H), 7.53 (dd1H, *J* = 2.4 Hz), 7.93 (d, 1H, *J* = 2.8 Hz), 8.61 (d, 1H *J* = 9.2 Hz); ^{13}C NMR (100 MHz, DMSO-d_6): δ 22.60, 24.73, 44.41, 54.41, 121.37, 126.49, 130.90, 132.31, 139.33, 168.33	C H O	56.66 5.77 16.17	56.65 5.75 16.15
Mcb2	1089 (C-Cl *str.*), 1296 (C-O *str.*), 1495 (C=C *str.*), 1623 (C=O *str.*), 1685 (C=O *str.*), 3214 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 2.25 (s, 3H) 2.562 (t, 8H *J* = 4.8 Hz), 2.782 (s, 2H) 3.618 (s, 1H), 7.45 (1H, *J* = 2.8 Hz), 7.95 (t, 1H, *J* = 2.8 Hz), 8.61 (d, 1H *J* = 8.8 Hz), 13.24 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 45.59, 51.48, 53.03, 61.21, 121.11, 126.23, 131.07, 139.10, 169.26	C H O	53.93 5.82 15.40	53.92 5.82 15.38
Mcb3	1084 (C-Cl *str.*), 1343 (C-O *str.*), 1582 (C=C *str.*), 1623 (C=O *str.*), 1694 (C=O *str.*), 2814 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 3.08 (s, 8H) 3.123 (s, 2H), 13.36 (s, 1H) 3.54 (s, 2H), 7.252 (s, 2H) 7.323 (s, 2H) 7.63 (d, 1H, *J* = 2.4 Hz), 7.95 (d, 1H, *J* = 2.4 Hz), 8.64 (d, 1H *J* = 8.8 Hz); ^{13}C NMR (100 MHz, DMSO-d_6): δ 51.8, 54.2, 61.8, 62.9, 116.5, 124.3, 125.0, 127.2, 128.4, 129.3, 130.2, 138.6, 140.3, 168.5, 169.3	C H O	61.93 5.72 12.38	61.91 5.70 12.36
Mcb4	1150 (C-Cl *str.*), 1232 (C-O *str.*), 1498 (C=C *str.*), 1575 (C=O *str.*), 1636 (C=O *str.*), 3387 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 2.48 (t, 4H, *J* = 5.6 Hz) , 3.32 (s, 2H) 3.42 (t, 4H, *J* = 5.6 Hz) 6.79 (s, 1H) 6.94 (d 2H, *J* = 7.2) 7.27 (d, 2H *J* = 2.4 Hz) 7.81 (m 1H) 7.93 (d, 1H, *J* = 2.4 Hz) 8.32 (m, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 48.1, 52.2, 61.3, 114.2, 116.5, 121.3, 125.4, 129.3, 130.8, 142.5, 149.2, 168.2, 169.3	C H O	61.04 5.39 12.84	61.03 5.38 12.83

Mcb5	1144 (C-Cl *str.*), 1251 (C-O *str.*), 1499 (C=C *str.*), 1596 (C=O *str.*), 1679 (C=O *str.*), 3412 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.83 (m, 4H J = 6.8 Hz) 2.823 (s, 2H), 3.11 (t, 4H J = 6.8 Hz), 7.375 (d, 1H, J = 2.4 Hz), 7.89 (d, 1H, J = 2.4 Hz), 8.63 (d, 1H J = 8.8 Hz); ^{13}C NMR (100 MHz, DMSO-d_6): δ 24.21, 44.97, 54.30, 120.94, 131.01, 139.26	C H O	55.23 5.35 16.98	55.22 5.34 16.97
Mdb1	1029 (C-Br *str.*), 1371 (C=C *str.*), 1593 (C=O *str.*), 1688 (C=O *str.*), 2943 (COOH), 3450 (-NH *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.54 (s, 6H) 1.63 (s, 2H), 1.96 (s, 2H), 3.11 (s, 2H) 6.73 (d, 2H J = 6.4 Hz) 7.57 (m 1H), 10.06 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 22.93, 23.75, 26.33, 44.09, 54.65, 62.67, 106.53, 114.35, 119.67, 129.58, 134.02, 136.77, 148.16, 169.40	C H O	49.28 5.02 14.07	49.26 5.01 14.05
Mdb2	1090 (C-Br *str.*), 1560 (C=C *str.*), 1627 (C=O *str.*), 1691 (C=O *str.*), 2822 (COOH), 3412 (-NH *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.888 (s, 3H) 2.197 (t, 4H) 3.176 (s, 4H), 6.77 (t, 2H, J = 8.4 Hz) 7.59 (d, 1H J = 1.6 Hz) 7.849 (t, 1H J = 1.6 Hz), 8.31 (d, 1H J = 8.4 Hz), 9.99 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 43.47, 46.08, 52.66, 53.10, 55.22, 61.70, 106.44, 114.45, 119.45, 130.30, 133.54, 149.35, 169.12	C H O	47.20 5.09 13.47	47.18 5.07 13.46
Mdb3	1143 (C-Br *str.*), 1369 (C=C *str.*), 1520 (C=O *str.*), 1626 (C=O *str.*), 2827 (COOH), 3443 (-NH *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.893 (s, 2H) 2.945 (d, 6H J = 4.4 Hz) 3.178 (s, 1H) 3.515 (t, 1H J = 4 Hz), 7.351 (m, 5H) 7.624 (t, 2H J = 1.6 Hz), 7.868 (d, 1H, J = 1.6 Hz), 8.09 (s, 1H), 8.30 (s, 1H), 9.01 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 51.12, 53.16, 61.75, 62.45, 106.44, 114.45, 129.35, 134.22, 137.12, 138.53, 149.38, 169.09	C H O	55.57 5.13 11.10	55.56 5.11 11.08
Mdb4	1090 (C-Br *str.*), 1371 (C=C *str.*), 1595 (C=O *str.*), 1629 (C=O *str.*), 3176 (COOH), 3444 (-NH *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.99 (s, 4H) 3.01 (t, 4H J = 5.2 Hz) 3.89 (t, 2H J = 5.2 Hz), 6.82 (m, 4H) 6.95 (m, 4H) 7.21 (m, 1H) 7.240 (d, 1H), 7.63 (dd, 1H, J = 1.6 Hz)), 7.871 (d, 1H J = 2 Hz); ^{13}C NMR (100 MHz, DMSO-d_6): δ 44.67, 48.09, 60.22, 106.37, 114.49, 116.10, 119.70, 129.44, 130.42, 134.32, 151.43, 167.14, 170.81	C H O	54.56 4.82 11.48	54.55 4.81 11.47
Mdb5	1023 (C-Br *str.*), 1373 (C=C *str.*), 1595 (C=O *str.*), 1627 (C=O *str.*), 2985 (COOH), 3449 (-NH *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.802 (m, 4H) 3.05 (s, 4H), 3.25 (s, 2H) 6.71 (d, 1H J = 8.4 Hz) 7.57 (d, 1H J = 7.6 Hz), 7.83 (s, IH) 8.05 (s 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 24.11, 44.71, 54.20, 59.20, 106.60, 114.30, 129.53, 130.0, 133.49, 133.96, 147.61	C H O	47.72 4.62 14.67	47.70 4.61 14.65
Meb1	1309 (C-N *str.*), 1549 (C=C *str.*), 1657 (C=O *str.*), 3386 (-NH *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.89 (s, 2H), 2.2 (s, 4H) 2.36 (s, 2H), 2.68 (s, 2H) 3.01 (s, 2H) 7.04 (s, 1H), 7.29 (t, 2H, J = 7.6 Hz), 7.62 (s, 2H), 9.8 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 21.85, 43.15, 44.78, 45.79, 54.13, 58.85, 61.51, 120.23, 123.97, 129.34, 138.94, 168.85, 172.98	C H O	71.53 8.31 7.33	71.51 8.29 7.31
Meb2	1221 (C-N *str.*), 1549 (C=C *str.*), 1741 (C=O *str.*), 3341 (-NH *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.9 (s, 3H), 2.33 (s, 8H) 3.03 (s, 2H), 7.07 (s, 1H) 7.30 (t, 2H, J = 8 Hz), 7.61 (t, 2H J = 7.6 Hz) 9.7 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 43.29, 45.11, 45.80, 51.70, 52.04, 54.3, 61.66, 119.95, 124.0, 129.41, 129.17, 138.90, 168.68	C H O	71.53 8.31 7.33	71.52 8.30 7.32
Meb3	1313 (C-N *str.*), 1548 (C=C *str.*), 1671 (C=O *str.*), 3348 (-NH *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 2.59 (s, 4H), 3.06 (s, 2H) 3.55 (s, 2H), 4.24 (s, 2H) 7.06 (t, 1H J = 7.2 Hz), 7.33 (m, 3H), 7.42 (s, 2H), 7.61 (s, 2H), 9.06 (s, 1H), 9.9 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 51.8, 53.061.1, 61.9, 121.6, 127.2, 128.4, 128.8, 128.9, 138.5, 138.6, 168.5	C H O	73.76 7.49 5.17	73.75 7.48 5.16
Meb4	1221 (C-N *str.*), 1587 (C=C *str.*), 1739 (C=O *str.*), 3385 (-NH *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 2.63 (s, 4H), 3.07 (s, 4H) 4.1 (s, 2H), 7.06 (t, 1H J = 7.2 Hz) 7.32 (m, 4H) 7.42 (s 2H) 7.6 (s, 2H), 9.2 (s, 1H), 10.0 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 49.27, 50.77, 61.76, 119.99, 124.02, 128.79, 129.13, 129.60, 131.59, 138.98	C H O	73.19 7.17 5.42	73.18 7.16 5.41
Meb5	1312 (C-N *str.*), 1553 (C=C *str.*), 1741 (C=O *str.*), 3447 (-NH *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.84 (m, 2H), 1.96 (m, 4H) 2.2 (s, 2H), 3.07 (t, 2H J = 7.2 Hz) 7.09 (m, 1H) 7.32 (m, 2H) 7.62 (m, 2H), 11.0 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 23.28, 24.20, 54.56, 62.94, 11986, 120.24, 129.35, 162.94	C H O	70.56 7.90 7.83	70.55 7.89 7.81
Mfb1	1225 (C-N *str.*), 1518 (C=C *str.*), 1741 (C=O *str.*), 3399 (-NH *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.53 (m, 2H), 1.66 (m, 6H) 2.25 (s, 3H), 2.98 (t, 4H J = 5.6 Hz) 7.11 (d, 1H J = 8 Hz) 7.49 (m, 1H) 8.74 (s, 2H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 21.3, 24.6, 25.2, 53.4, 61.4, 121.5, 129.2, 135.5, 136.8, 168.5	C H O	72.38 8.68 6.89	72.37 8.67 6.88
Mfb2	1375 (C-N *str.*), 1404 (C=C *str.*), 1668 (C=O *str.*), 3358 (-NH *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 2.28 (s, 3H), 2.38 (m, 9H) 3.32 (s, 2H), 7.11 (d, 2H J = 8 Hz) 7.52 (d, 2H J = 7.2 Hz) 8.74 (s, 2H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 21.9, 43.27, 45.89, 51.85, 61.1, 121.5, 29.2, 134.5, 136.8, 168.5	C H O	67.98 8.56 6.47	67.97 8.55 6.45
Mfb3	1302 (C-N *str.*), 1495 (C=C *str.*), 1565 (C=O *str.*), 3028 (-NH *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 2.21 (s, 3H), 2.36 (t, 4H, J = 5.6 Hz) 2.48 (t, 4H J = 5.6 Hz), 3.22 (s, 3H) 3.68 (s, 3H) 7.21 (m, 5H) 7.33 (t, 2H, J = 7.2 Hz) 7.56 (d, 2H J = 8 Hz); ^{13}C NMR (100 MHz, DMSO-d_6): δ 21.3, 51.8, 53.0, 61.1, 61.9, 121.5, 127.2, 128.4, 128.8, 129.2, 135.5, 136.8, 138.6, 168.5	C H O	74.27 7.79 4.95	74.25 7.78 4.93
Mfb4	1312 (C-N *str.*), 1553 (C=C *str.*), 1741 (C=O *str.*), 3447 (-NH *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 2.50 (s, 3H), 3.17 (m, 8H) 3.33 (s, 2H), 6.87 (t, 1H J = 8 Hz) 6.99 (d, 3H J = 8 Hz) 7.25 (dd, 5H J = 7.2 Hz); ^{13}C NMR (100 MHz, DMSO-d_6): δ 21.3, 43.26, 46.21, 61.1, 61.9, 116.45, 120.35, 129.57, 150.64	C H O	73.76 7.49 5.17	73.74 7.47 5.16

Mfb5	1252 (C-N *str.*), 1552 (C=C *str.*), 1689 (C=O *str.*), 3346 (-NH *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.69 (m, 4H) 2.26 (s, 3H), 2.51 (t, 4H, *J* = 7.2 Hz), 3.1 (s, 2H), 7.21 (d, 2H *J* = 8 Hz) 7.52 (dd, 2H *J* = 7.2 Hz); ^{13}C NMR (100 MHz, DMSO-d_6): δ 21.3, 22.9, 57.2, 61.1, 121.5, 129.2, 135.5, 136.8, 168.5	C H O	71.53 8.31 7.33	71.51 8.30 7.31
Mgb1	1028 (C-Cl *str.*), 1248 (C-N *str.*), 1457 (C=C *str.*), 1699 (C=O *str.*), 3388 (-NH *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.54 (s, 2H) 1.88 (s, 4H), 2.95 (t, 4H, *J* = 5.2 Hz) 4.3 (s, 2H), 7.31 (d, 2H *J* = 8.4 Hz), 7.63 (d, 2H, *J* = 8.4); ^{13}C NMR (100 MHz, DMSO-d_6): δ 23.05, 23.88, 25.71, 44.05, 54.36, 62.69, 121.53, 128.98, 133.3, 137.90, 169.33	C H O	58.31 6.78 5.98	58.29 6.76 5.97
Mgb2	1012 (C-Cl *str.*), 1242 (C-N *str.*), 1518 (C=C *str.*), 1686 (C=O *str.*), 3269 (-NH *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.89 (s, 3H) 2.20 (s, 4H), 3.50 (s 2H), 7.35 (dd, 2H *J* = 7.2 Hz) 7.68 (d, 2H *J* = 4.8 Hz); ^{13}C NMR (100 MHz, DMSO-d_6): δ 45.5, 51.81, 54.56, 61.85, 121.51, 127.43, 129.26, 138.01, 168.92, 172.82	C H O	61.78 6.78 6.33	61.76 6.76 6.32
Mgb3	1049 (C-Cl *str.*), 1244 (C-N *str.*), 1538 (C=C *str.*), 1682 (C=O *str.*), 3408 (-NH *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 2.87 (s, 4H) 3.04 (s, 4H), 3.1 (s, 2H) 3.98 (s, 2H) 7.31 (m, 5H) 7.491 (s, 2H), 7.714 (d, 2H, *J* = 8.8 Hz); ^{13}C NMR (100 MHz, DMSO-d_6): δ 49.34, 51.45, 60.41, 61.94, 116.50, 121.57, 127.50, 127.70, 128.76, 128.98, 129.43, 130.87, 137.57, 138.02, 168.34	C H O	66.37 6.45 4.65	66.35 6.44 4.63
Mgb4	1028 (C-Cl *str.*), 1248 (C-N *str.*), 1457 (C=C *str.*), 1699 (C=O *str.*), 3388 (-NH *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.88 (s, 2H) 2.87 (t, 2H, *J* = 4.8 Hz), 3.06 (t, 2H, *J* = 4.4 Hz), 6.78 (t, 2H *J* = 7.2 Hz) 6.92 (d, 3H *J* = 8 Hz), 7.20 (m, 4H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 49.1, 51.5, 61.1, 114.3, 120.4, 129.6, 129.9, 133.3, 136.6, 149.6, 168.5	C H O	65.55 6.11 4.85	65.53 6.09 4.84
Mgb5	1145 (C-Cl *str.*), 1250 (C-N *str.*), 1594 (C=C *str.*), 1691 (C=O *str.*), 3487 (-NH *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.82 (dd, 4H, *J* = 18 Hz) 2.8 (s, 4H), 3.07 (s 2H) 3.1 (s, 2H), 7.31 (d, 2H *J* = 4.2 Hz) 7.61 (dd, 2H *J* = 4.2 Hz); ^{13}C NMR (100 MHz, DMSO-d_6): δ 24.50, 58.39, 66.22, 121.55, 127.75, 128.57, 128.9, 129.14, 137.69, 167.73	C H O	60.38 6.33 6.70	60.38 6.32 6.69

1232 (C-N *str.*), 1498 (C=C *str.*), 1575 (C=O *str.*), 1636 (C=O *str.*), 2950 (COOH), 3387 (-NH). The ^{1}H NMR spectrum of compounds **Mab1-Mgb5** showed signals for a singlet around δ 3.0-3.36 ppm due to the -CH_2 peak of acyl and a singlet at δ 10.0 ppm due to -NH and aromatic protons at δ 6.4.-8.6 ppm. ^{13}C NMR spectrum of compounds **Mab1-Mgb5** showed characteristic signals δ 62.1, 116.5 and 168.36 ppm confirming the formation of the target compounds.

A total of 20 heterocyclic amine substituted 2-acetoxy derivatives (**Eab1-Edb5**) were synthesized. The products were obtained in good yield with a range of 60-90% (Table-3). The synthesized compounds were identified by using IR, ^{1}H and ^{13}C NMR spectral data. In IR spectra of heterocyclic amine substituted 2-chloroacetates show the key signals for 1055 (C-O *str.*), 1493 (C=C *str.*), 1595 (C=O *str.*), 1664 (C=O *str.*), 2974 (COOH) (Table-4). The ^{1}H NMR spectra of compounds **Eab1-Edb5** showed characteristic signals for singlet around δ 3.16-3.46 due to the -CH_2 peak of acyl and aromatic protons at δ 6.86-7.8 ppm. ^{13}C NMR spectra of the synthesized compounds **Eab1-Edb5** showed a characteristic signal δ 51.8, 115.7 and 151.12 ppm confirming the formation of target compounds (Table-4).

Docking studies: MmaA1 is an essential methyl transferase enzyme involved in the maturation of mycolic acids [29]. The active pocket amino acids involved in the recognition of S-Adenosyl methionine residues are ASP 19, TRP 30, TYR 32, GLY71, TRP 74, GLY 76, ALA 77 and PHE 135 which were predicted from CASTp [30], SiteMap [31] and protein-ligand docking using PatchDock [32] with S-adenosyl-N-decyl-aminoethyl (SADAE). SADAE is a highly effective bisubstrate inhibitor of the mycolic acid methyl transferases of *M. tuberculosis* [33]. The 330 compounds from the in-house library of compounds were subjected to virtual screening using AutoDock and 55 hits were obtained. The obtained docking scores ranged from -10.5 to -5.8. The docking outcomes were visually assessed and prioritized based on the dock score, the docking results of the top 10 compounds are given in Table-5.

The docking data suggest that most of the ligands bind to protein active site residues *via* TRP 74, TRP 30, TRP 32, GLY

TABLE-3
PHYSICO-CHEMICAL PROPERTIES OF THE COMPOUNDS **Eab1** TO **Edb5**

Compd.	Yield (%)	m.p. (°C)	Relative molecular mass	m.f.	Compd.	Yield (%)	m.p. (°C)	Relative molecular mass	m.f.
Eab1	81.66	Semi-solid	263.29	$C_{14}H_{17}NO_4$	**Ecb1**	61.23	Gummy resin	263.29	$C_{14}H_{17}NO_4$
Eab2	80	Semi-solid	278.30	$C_{14}H_{18}N_2O_4$	**Ecb2**	78.23	Gummy resin	278.30	$C_{14}H_{18}N_2O_4$
Eab3	78.78	Semi-solid	354.40	$C_{20}H_{22}N_2O_4$	**Ecb3**	89.60	Gummy resin	354.40	$C_{20}H_{22}N_2O_4$
Eab4	90.62	Semi-solid	340.37	$C_{19}H_{20}N_2O_4$	**Ecb4**	94.2	Gummy resin	340.37	$C_{19}H_{20}N_2O_4$
Eab5	59.13	Semi-solid	249.26	$C_{13}H_{15}NO_4$	**Ecb5**	75.12	Gummy resin	249.26	$C_{13}H_{15}NO_4$
Ebb1	72.51	Semi-solid	297.73	$C_{14}H_{16}NO_4Cl$	**Edb1**	69.23	Gummy resin	219.28	$C_{13}H_{17}NO_2$
Ebb2	78.12	Gummy resin	312.75	$C_{14}H_{17}N_2O_4Cl$	**Edb2**	68.24	Semi-solid	234.29	$C_{13}H_{18}N_2O_2$
Ebb3	82.3	Semi-solid	388.84	$C_{20}H_{21}N_2O_4Cl$	**Edb3**	85.47	Gummy resin	310.39	$C_{19}H_{22}N_2$
Ebb4	86.34	88.9	374.82	$C_{19}H_{19}N_2O_4Cl$	**Edb4**	74.23	151.9	296.36	$C_{18}H_{20}N_2O_2$
Ebb5	62.32	Semi-solid	283.71	$C_{13}H_{14}NO_4Cl$	**Edb5**	62.35	Gummy resin	205.25	$C_{12}H_{15}NO_2$

TABLE-4
SPECTRAL DATA OF THE COMPOUNDS **Eab1** TO **EDb5**

Compd.	IR (KBr, ν_{max}, cm^{-1})	NMR data	Elemental analysis (%)		
				Req.	Found
Eab1	1218 (C-O *str.*), 1458 (C=C *str.*), 1588 (C=O *str.*), 1653 (C=O *str.*), 2945 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.55 (s, 6H) 2.98 (t, 2H, *J* = 4.8 Hz) 3.165 (s, 2H), 5.0 (s, 2H), 8.7-6.79 (m, 4H) 5.0 (s, 2H) 10.38 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 24.3, 25.57, 56.22, 61.99, 113.69, 117.34, 119.94, 130.72, 135.27 136.34, 162.16, 168.36	C H O	63.87 6.51 24.31	63.86 6.49 24.30
Eab2	1251 (C-O *str.*), 1460 (C=C *str.*), 1588 (C=O *str.*), 1666 (C=O *str.*), 3425 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 2.07 (s, 2H) 2.51 (s, 2H), 2.75 (s, 4H) 3.16 (s, 2H), 6.8 (m, 2H), 7.34 (t, 2H, 7.8 (dd, 1H, *J* = 7.2 Hz) 10.34 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 42.85, 49.47, 52.47, 62.52 113.53, 116.99, 118.56, 120.0, 134.45, 136.32, 160.24, 168.20.	C H O	60.42 6.52 23	60.40 6.50 22.98
Eab3	1254 (C-O *str.*), 1458 (C=C *str.*), 1588 (C=O *str.*), 1627 (C=O *str.*), 2828 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 2.568 (s, 4H) 2.71 (s, 3H) 3.085 (s, 2H) 3.17 (s, 1H, CH_2) 7.72-6.68 (m, 9H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 49.46, 51.32, 52.92, 61.94, 116.46, 117.39, 119.44, 127.80, 128.90, 129.46, 130.29, 130.60, 132.78, 137.21, 137.48, 162.49, 172.80	C H O	67.78 6.26 18.06	67.77 6.25 18.04
Eab4	1055 (C-O *str.*), 1493 (C=C *str.*), 1595 (C=O *str.*), 1664 (C=O *str.*), 2974 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 3.22 (s, 8H), 3.36 (s, 2H), 7.73-6.7 (m, 9H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 43.09, 46.0, 116.51, 116.72, 117.72, 120.47, 129.47, 130.57, 150.52	C H O	67.05 5.92 18.80	67.02 5.90 18.79
Eab5	1298 (C-O *str.*), 1482 (C=C *str.*), 1610 (C=O *str.*), 1648 (C=O *str.*), 2977 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.80 (m, 4H), 3.279 (t, 2H, *J* = 6.8 Hz) 3.411 (t, 2H, *J* = 6.8 Hz), 4.253 (s, 2H) 6.9 (m, 4H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 24.08, 26.05, 45.32, 46.45, 113.41, 117.95, 120.01, 131.96, 136.12, 150.01, 161.54, 172.37	C H O	62.64 6.07 25.67	62.22 6.06 25.65
Ebb1	1021 (C-Cl *str.*), 1035 (C-O *str.*), 1478 (C=C *str.*), 1579 (C=O *str.*), 1633 (C=O *str.*), 3393 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.54 (m, 6H) 3.0 (t, 4H, *J* = 5.6 Hz), 4.1 (s, 1H) 6.7 (s, 1H) 7.203 (dd, 1H, *J* = 2.8HZ), 7.6 (d, 1H, *J* = 2.8 Hz); ^{13}C NMR (100 MHz, DMSO-d_6): δ 22.09, 23.88, 44.05, 54.36, 121.53, 128.98, 137.90, 169.33	C H O	56.48 5.42 21.49	56.47 5.40 21.48
Ebb2	1098 (C-Cl *str.*), 1285 (C-O *str.*), 1472 (C=C *str.*), 1579 (C=O *str.*), 1632 (C=O *str.*), 3413 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.63 (d, 3H *J* = 5.2 Hz), 1.89 (m, 4H), 2.2 (s, 2H), 2.45 (t, 2H) 2.63 (s, 2H), 6.1 (s, 1H), 6.59 (dd, 1H, *J* = 2.8 Hz), 7.0 (d, 1H, *J* = 2.8 Hz); ^{13}C NMR (100 MHz, DMSO-d_6): δ 45.56, 51.81, 52.49, 54.56, 61.85, 121.51, 127.43, 128.98, 129.26, 138.01, 168.92, 172.82	C H O	53.77 5.48 20.46	53.76 5.47 20.45
Ebb3	1021 (C-Cl *str.*), 1032 (C-O *str.*), 1450 (C=C *str.*), 1638 (C=O *str.*), 1744 (C=O *str.*), 3357 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.87 (s, 2H), 2.44 (s, 6H), 2.89 (s, 3H), 3.43 (s, 3H), 6.65 (s, 1H), 7.15 (d, 1H, *J* = 2.8 Hz), 7.27 (m, 5H), 7.6 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 43.21, 50.90, 60.41, 61.94, 116.50, 121.57, 127.50, 127.70, 128.76, 129.43, 130.87, 137.57, 138.02, 168.34	C H O	61.78 5.44 16.46	61.77 5.43 16.45
Ebb4	1150 (C-Cl *str.*), 1232 (C-O *str.*), 1498 (C=C *str.*), 1575 (C=O *str.*), 1636 (C=O *str.*), 3387 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.9 (s, 2H), 2.57 (s, 2H), 3.1 (s, 4H), 3.14 (s, 3H), 6.63 (s, 1H) 6.85 (m, 3H) 6.95 (d, 1H, *J* = 0.8 Hz), 7.16 (s, 1H), 7.2 (m, 1H) 7.61 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 49.5, 51.9, 55.4, 114.3, 149.6, 121.35, 122.3, 125.5, 129.6, 130.7, 131.2, 135.2, 148.9, 149.8, 168.0, 172.5	C H O	60.88 5.11 17.07	60.87 5.10 17.06
Ebb5	1144 (C-Cl *str.*), 1241 (C-O *str.*), 1475 (C=C *str.*), 1573 (C=O *str.*), 1630 (C=O *str.*), 3437 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.81 (s, 4H), 3.08 (s, 4H), 3.69 (s, 2H), 6.62 (s, 1H) 7.11 (d, 1H, *J* = 2.8 Hz), 7.57 (d, 1H 2.8 Hz); ^{13}C NMR (100 MHz, DMSO-d_6): 23.57, 24.50, 44.99, 54.18, 58.39, 121.55, 127.75, 128.57, 129.14, 137.69, 167.73, 174.29	C H O	55.04 4.97 22.56	55.03 4.96 22.55
Ecb1	1295 (C-O *str.*), 1460 (C=C *str.*), 1568 (C=O *str.*), 1593 (C=O *str.*), 3427 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.52 (s, 4H), 1.78 (s, 1H) 2.93 (s, 4H), 3.1 (s, 2H), 6.7 (d, 1H *J* = 3.2 Hz) 6.82 (d, 1H, *J* = 1.6 Hz), 7.21 (m, 2H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 24.5, 25.7, 44.29, 48 .43, 113.75, 116.65, 117.05, 117.33, 128.74, 130.01, 157.76, 169.38	C H O	63.87 6.51 24.31	63.86 6.49 24.30
Ecb2	1295 (C-O *str.*), 1403 (C=C *str.*), 1504 (C=O *str.*), 1623 (C=O *str.*), 2814 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.85 (s, 3H), 2.18 (s, 4H) 2.34 (s, 4H), 3.32 (s, 2H) 6.7 (d, 1H *J* = 7.2 Hz) 6.83 (d, 1H, *J* = 1.6 Hz), 7.31 (s, 2H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 43.86, 46.02, 53.28, 55.04, 114.05, 116.88, 117.58, 129.04, 130.01, 137.62, 157.84, 169.41	C H O	60.42 6.52 23	60.41 6.51 22.99
Ecb3	1296 (C-O *str.*), 1451 (C=C *str.*), 1622 (C=O *str.*), 1734 (C=O *str.*), 2826 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 2.35 (s, 4H), 2.55 (s, 2H) 3.05 (s, 2H), 3.48 (m, 4H) 6.74 (s 1H) 6.84 (s, 1H), 7.3 (m, 7H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 52.82, 53.4, 60.99, 62.45, 127.4, 127.67, 128.63, 129.79, 130.53, 137.55, 138.38, 158.20, 167.8, 169.38	C H O	67.78 6.26 24.31	67.77 6.25 24.29
Ecb4	1235 (C-O *str.*), 1499 (C=C *str.*), 1597 (C=O *str.*), 1627 (C=O *str.*), 2914 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.86 (s, 2H), 2.0 (s, 2H) 2.51 (t, 2H), 2.97 (s, 2H), 3.14 (s, 2H) 6.7 (s, H) 6.96 (m, 5H), 7.17 (m, 2H), 7.34 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 44.67, 48.77, 49.16, 116.34, 119.84, 129.45, 137.44, 151.49, 157.36, 157.80, 169.53	C H O	67.05 5.92 18.80	67.03 5.90 18.78
Ecb5	1273 (C-O *str.*), 1449 (C=C *str.*), 1586 (C=O *str.*), 1609 (C=O *str.*), 2946 (COOH)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.81 (s, 4H), 3.07 (s, 4H), 3.38 (s, 2H) 6.82 (dd, 3H *J* = 8.4 Hz) 7.18 (t, 1H, =7.6 Hz); ^{13}C NMR (100 MHz, DMSO-d_6): δ 24.30, 24.55, 45.80, 45.95, 58.24, 114.24, 116.46, 117.83, 120.35, 128.77, 140.01, 157.26, 168.85	C H O	62.64 6.07 25.67	62.24 6.06 25.66

Edb1	1249 (C-O *str.*), 1476 (C=C *str.*), 1660 (C=O *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.67 (m, 6H), 2.95 (t, 4H J = 5.6 Hz) 3.55 (s, 2H), 6.77 (m, 2H) 6.96 (m, 5H), 7.16 (m, 2H), 9.0 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 23.14, 24.31, 53.05, 57.22, 59.15, 115.71, 119.19, 129.78, 157.83, 163.67	C H O	71.21 7.81 14.59	71.20 7.79 14.57
Edb2	1252 (C-O *str.*), 1472 (C=C *str.*), 1664 (C=O *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 2.23 (s, 3H), 2.42 (s, 8H) 3.06 (s, 2H), 5.97 (s, 1H) 8.01 (s, 2H), 8.08 (s, 1H), 8.18 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 45.5, 53.3, 53.8, 58.01, 59.20, 60.87, 162.56, 167.95, 171.17	C H O	66.64 7.74 13.66	66.63 7.73 13.65
Edb3	1237 (C-O *str.*), 1473 (C=C *str.*), 1631 (C=O *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 1.2 (s, 2H), 2.42 (s, 8H), 4.85 (s, 2H) 4.94 (s, 2H) 6.86 (s, 5H), 6.9 (s, 4H), 9.41 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 49.36, 50.74, 52.42, 77.11, 115.71, 128.9, 129.0, 129.49, 129.80, 130.38	C H O	73.52 7.14 10.31	73.51 7.13 10.30
Edb4	1231 (C-O *str.*), 1498 (C=C *str.*), 1666 (C=O *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 3.2 (s, 2H) 3.54 (s, 2H), 3.68 (s, 1H), 4.4 (s, 2H) 6.86 (s, 2H), 7.01 (s, 1H), 7.29 (s, 6H), 9.29 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 43.04, 44.63, 48.82, 4 8.56, 52.31, 115.70, 116.32, 116.51, 120.45, 129.52, 129.62, 151.12	C H O	72.95 6.80 10.80	72.93 6.79 10.78
Edb5	1235 (C-O *str.*), 1470 (C=C *str.*), 1640 (C=O *str.*)	^{1}H NMR (400 MHz, DMSO-d_6): δ 2.1 (s, 4H), 2.42 (s, 4H) 3.46 (s, 2H), 7.3 (s, 3H) 7.5 (m, 2H); ^{13}C NMR (100 MHz, DMSO-d_6): δ 23.3, 54.2, 56.6, 121.6, 125.6, 129.83, 151.2, 168.0	C H O	70.22 7.37 15.59	70.21 7.36 15.58

TABLE-5
LIGANDS SHOWING DOCK SCORE WITH INTERMOLECULAR INTERACTIONS AND BOND DISTANCES

IUPAC name and compound No.	Docking score	Intermolecular interactions	Bond distance (Å)
2-(2-(4-Benzylpiperazin-1-yl)acetamido)-5-fluorobenzoic acid (**Mab3**)	-10.6	**Hydrogen bonds**	
		TRP74:H-M1:O	3.32
		TRP74:H-M1:O	3.06
		GLY75:H-M1:O	3.01
		M1:H-GLY71:O	3.39
		M1:H-PHE135:O	3.37
		Pi-Pi interactions	
		VAL31:M1	3.67
		M1:LEU94	5.43
		Halogen interaction	
		LYS51:H-M1:F	2.29
2-(2-(4-Benzylpiperazin-1-yl) acetamido)-5-chloro benzoic acid (**Mcb3**)	-10.2	**Hydrogen Bonds**	
		M2:H-TRP30:O	3.75
		TRP74:H-M2:O	3.97
		pi-pi interactions	
		PHE22-M2	4.25
		TYR32-M2	5.78
		HIS98-M2	5.35
		ALA137-M2	3.86
		Halogen interaction	
		TRP74:H-M2:Cl	3.48
2-(2-(4-Benzylpiperazin-1-yl)acetamido)-5-bromobenzoic acid (**Mbb3**)	-9.9	**Hydrogen Bonds**	
		M3:H-TRP30:O	2.57
		M3:H-GLU136:O	2.09
		THR33:H-M3:O	3.80
		GLY73:H-M3:O	3.58
		ALA137:H-M3:O	3.32
		pi-pi interactions	
		VAL131-M3	3.99
		ASP139-M3	4.45
		Halogen interaction	
		ALA77:H-M3:Br	3.26
5-Fluoro-2-(2-(4-phenylpiperazin-1-yl)acetamido) benzoic acid (**Mab4**)	-9.2	**Hydrogen Bond**	
		GLY73:H-M4:O	3.65
		HIS98:H-M4:O	3.24
		M4:H-GLY71:O	3.21
		pi-pi Interactions	
		TYR32-M4	4.48
		M4-ALA137	4.98
		Halogen interaction	
		ASP139-M4:F	3.05

2-(4-(4-Benzylpiperazin-1-yl)butanamido)-5-bromobenzoic acid (**Mbb2**)	-9.2	**Hydrogen Bond**	
		SER134:H-M5:O	2.83
		M5:H-VAL70:O	2.18
		M5:H-PHE135:O	2.50
		GLU136:H-M5:O	3.58
		M5:H-GLY71:O	3.41
		pi-pi Interactions	
		VAL31-M5	3.89
		VAL70-M5	3.81
		M5-ALA77	4.82
		Halogen interaction	
		TRP122-M5:Br	4.11
		PHE149-M5:Br	4.39
2-(2-(4-Phenylpiperazin-1-yl)acetamido)-5-bromobenzoic acid (**Mbb4**)	-9.1	**Hydrogen Bonds**	
		HIS98:H-M6:O	3.53
		M6:H-SER134:O	3.68
		pi-pi Interactions	
		LYS143:H-M6	3.44
		ALA137:H-M6	4.48
		PHE149-M6	3.85
		TRP122-M6	4.85
		TYR146-M6	4.62
		Halogen interaction	
		ALA137-M6:Br	4.48
2-(2-(4-Benzylpiperazin-1-yl)acetamido)-5-chlorobenzoic acid (**Mcb3**)	-9.1	**Hydrogen Bonds**	
		TRP74:H-M7:O	3.47
		M7:H-GLY71:O	3.18
		M7:H-PHE135:O	3.48
		pi-pi Interactions	
		PHE135:H-M7	3.55
		ALA77:H-M7	3.86
		VAL31-M7	3.71
		M7-VAL52	4.11
5-Fluoro-2-(2-(4-methylpiperazin-1-yl)acetamido) benzoic acid (**Mab2**)	-8.7	**Hydrogen Bonds**	
		TRP74:H-M8:O	3.34
		M8:H-GLY71:O	3.51
		M8:H-PHE135:O	3.52
		pi-pi Interactions	
		VAL31-M8	3.69
		halogen interaction	
		Lys51:C-M8:F	3.27
5-Fluoro-2-(3-(imidazolidin-1-yl)propanamido) benzoic acid (**Mhh1**)	-8.7	**Hydrogen Bonds**	
		M9:H-TRP30:O	2.38
		M9:H-CYS72:O	2.44
		M9:H-THR93:O	2.77
		pi-pi Interactions	
		VAL31-M9	3.58
5-Bromo-2-(3-(piperidin-1-yl)propanamido) benzoic acid (**Mih3**)	-8.6	**Hydrogen Bonds**	
		TRP74:H-M10:O	3.32
		M10:H-GLY71:O	3.41
		M10:H-PHE135:O	3.52
		pi-pi Interactions	
		VAL31-M10	3.22
		halogen interaction	
		Lys51:C-M10:F	3.27
		Alkyl Interactions	
		ALA137-M10	3.86

71, PHE 135, LYS 51, ALA 137, ALA77, HIS 98. Three key interactions play a vital role in binding of the ligands with the protein. They are hydrogen bonding, pi-pi interactions and halogen bonding. The amino acids like TRP 74, TRP 30, TRP 32, GLY 71, PHE 135, LYS 51, ALA137 are majorly involved in the hydrogen bonding, TRP 122, TRP 32, TRP 137, VAL 31, ALA 77, ALA137, PHE 135 are involved in pi-pi interactions, whereas LYS 51, TRP 122, PHE 149, ASP139 are involved in the halogen bonding with receptor.

The ligand A1B1C5 is the highest scoring ligand with a binding affinity of -10.6 kcal and involved in hydrogen bond interactions with residues TRP 74, GLY 71, GLY 75, PHE 135, pi-pi interactions with VAL 31, LEU 94 and halogen bond with LYS 51 of MmaA1 protein which is shown in Fig. 2.

In ligand-receptor interaction, the carboxylic acid group in the benzoic acid moiety of 2-(N-substituted glycinamido benzoic acid) plays a pivotal role. Especially, when the C=O group is involved in H-bond interactions with the residues TRP

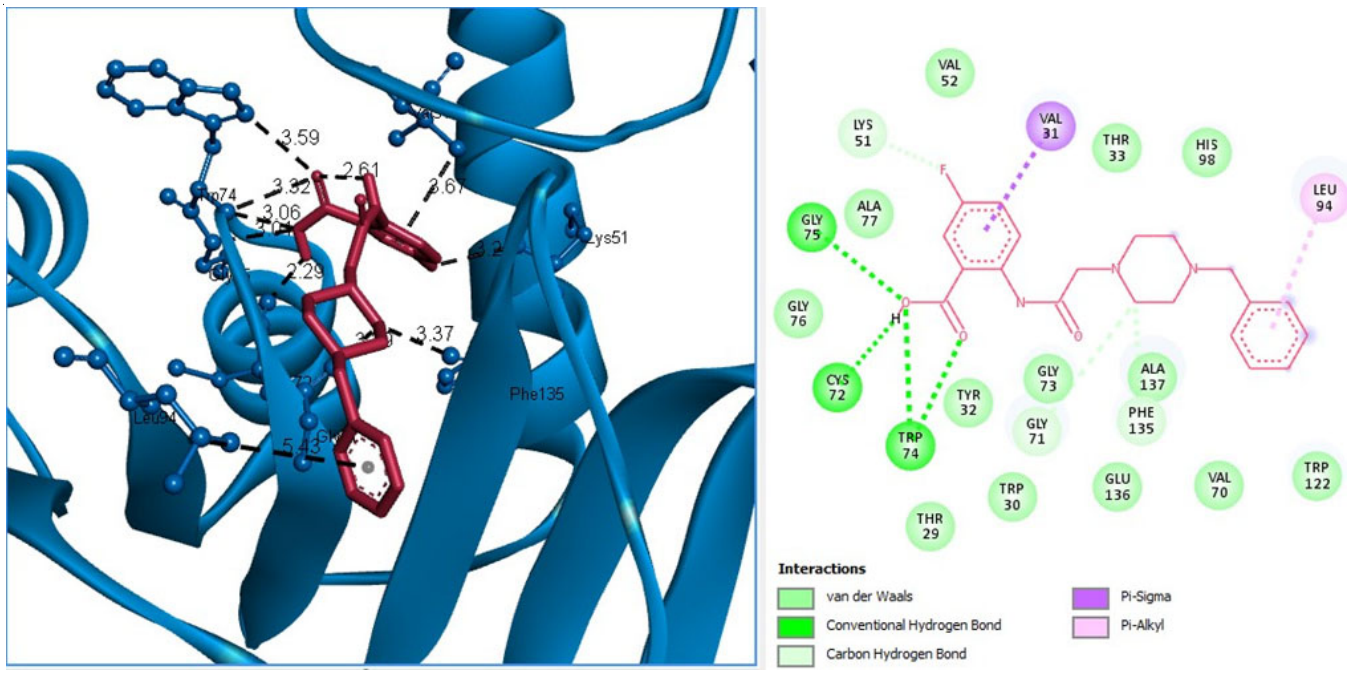


Fig. 2. 3D and 2D view of binding interactions of 2-(2-(4-benzylpiperazin-1-yl) acetamido)-5-fluorobenzoic acid (**Mab3**) with MmaA1 protein

74, TYR 32, GLU 136 and pi-pi interactions with VAL 31 of the MmaA1 protein. The halogen atoms on the benzoic acid also interacts with various amino acid residues of the protein like F bond with LYS 51, Br bond with VAL 31, ALA 137, TRP 122 and Cl bond with PHE 22, TYR 32, VAL 52.

In designed compounds, the compounds possessing aryl group (A1-A11), linker (B1) and secondary amine (C3, C4, C5, C7, C8) exhibited good drug-receptor interactions among the 330 compounds and exhibited good docking scores. The docking analysis indicated them as suitable pharmacophores for lead identification and further processing. Representative docking poses for top-scored compounds from 1-4 are given in Figs. 2-5 and top-scored compounds 5-10 are given in Figs. 6-11.

ADME studies: The lead likeness criteria are molecular weight < 500, TPSA 20 Å^2 < to < 130 Å^2, XLOGP < 5, number of hydrogen bond donors < 5, number of hydrogen bond acceptors < 10 based on the Lipinski's rule of five. These results showed that all the ligands have drug-like properties and ABC-

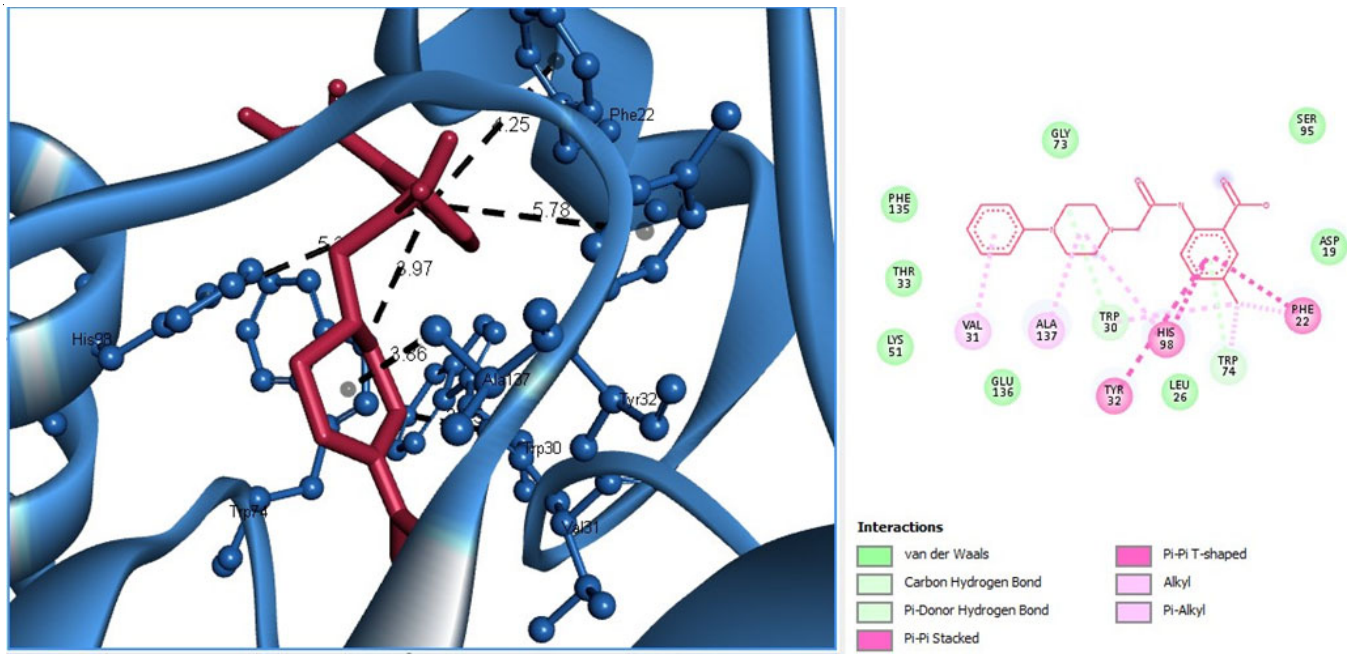


Fig. 3. 3D and 2D view of binding interactions of 2-(2-(4-phenylpiperazin-1-yl) acetamido)-5-chlorobenzoic acid (**Mcb3**) with MmaA1 protein

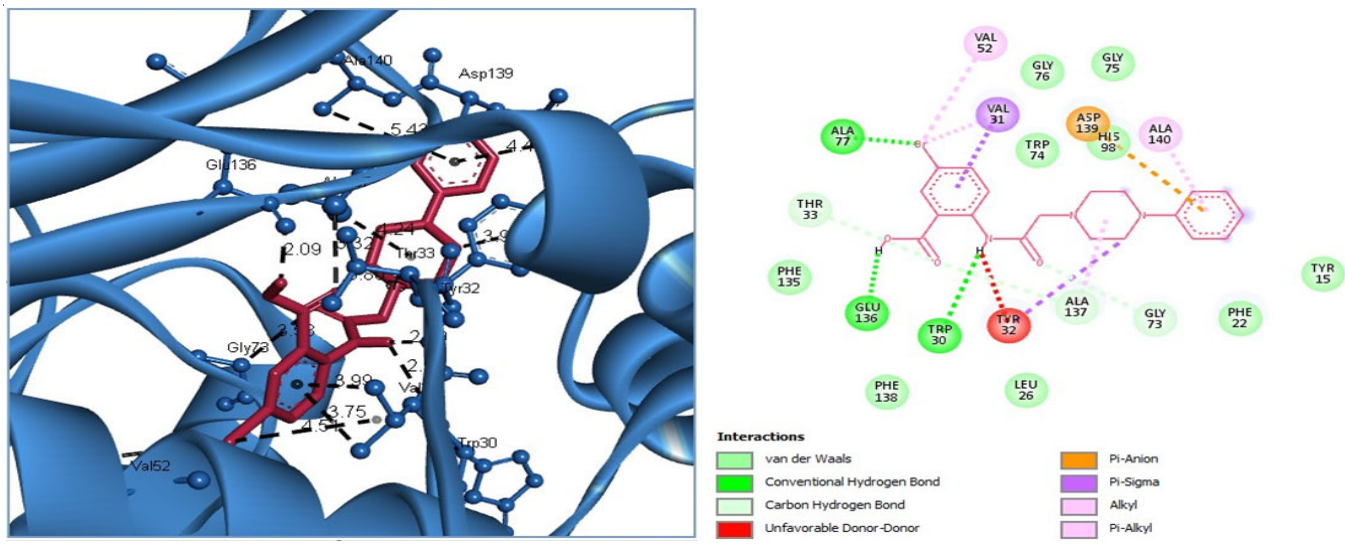


Fig. 4. 3D and 2D view of binding interactions of 2-(2-(4-phenylpiperazin-1-yl) acetamido)-5-bromobenzoic acid (**Mbb3**) with MmaA1 protein

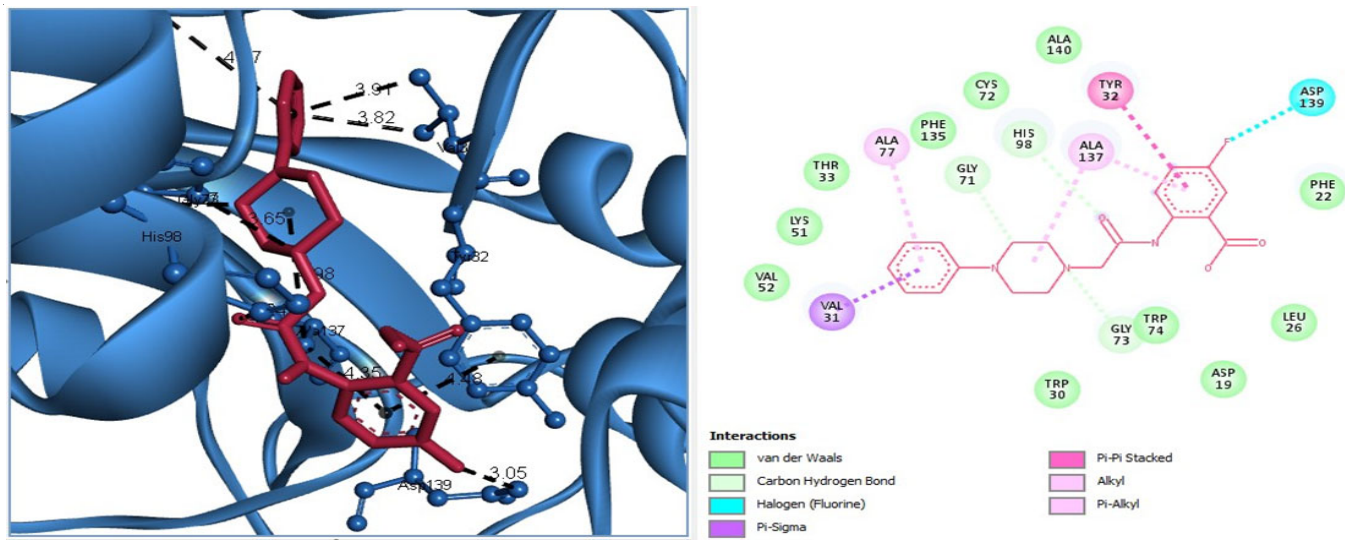


Fig. 5. 3D and 2D view of binding interactions of 5-fluoro-2-(2-(4-phenylpiperazin-1-yl) acetamido) benzoic acid (**Mab4**) with MmaA1 protein

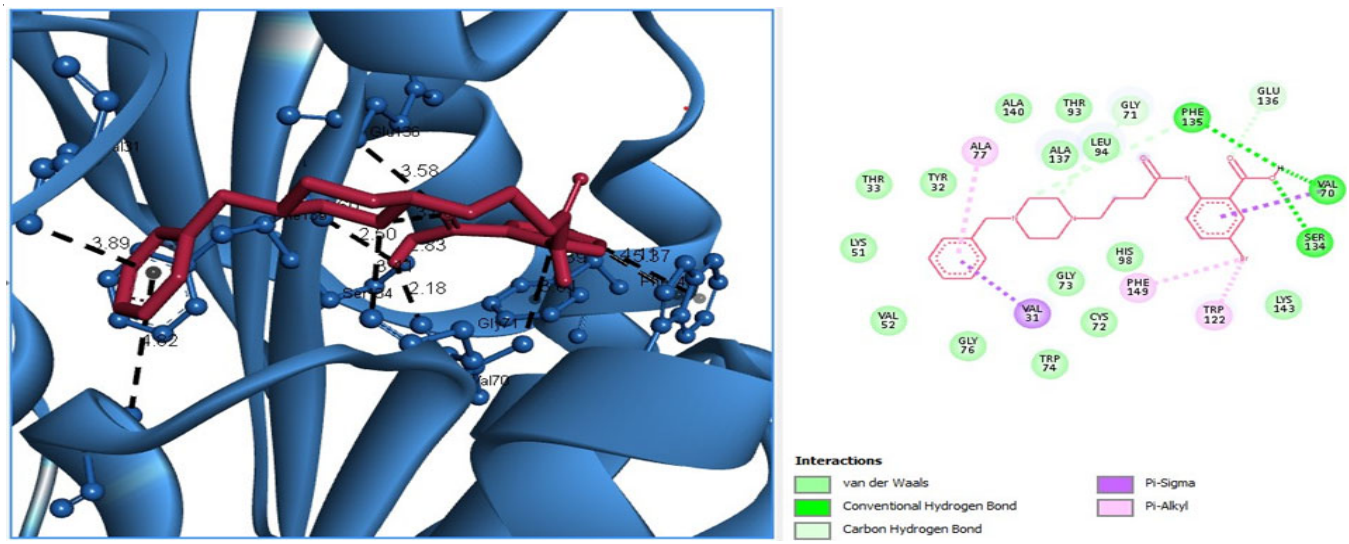


Fig. 6. 3D and 2D view of binding interactions of 2-(4-(4-benzylpiperazin-1-yl) butanamido)-5-bromobenzoic acid (**Mbb2**) with MmaA1 protein

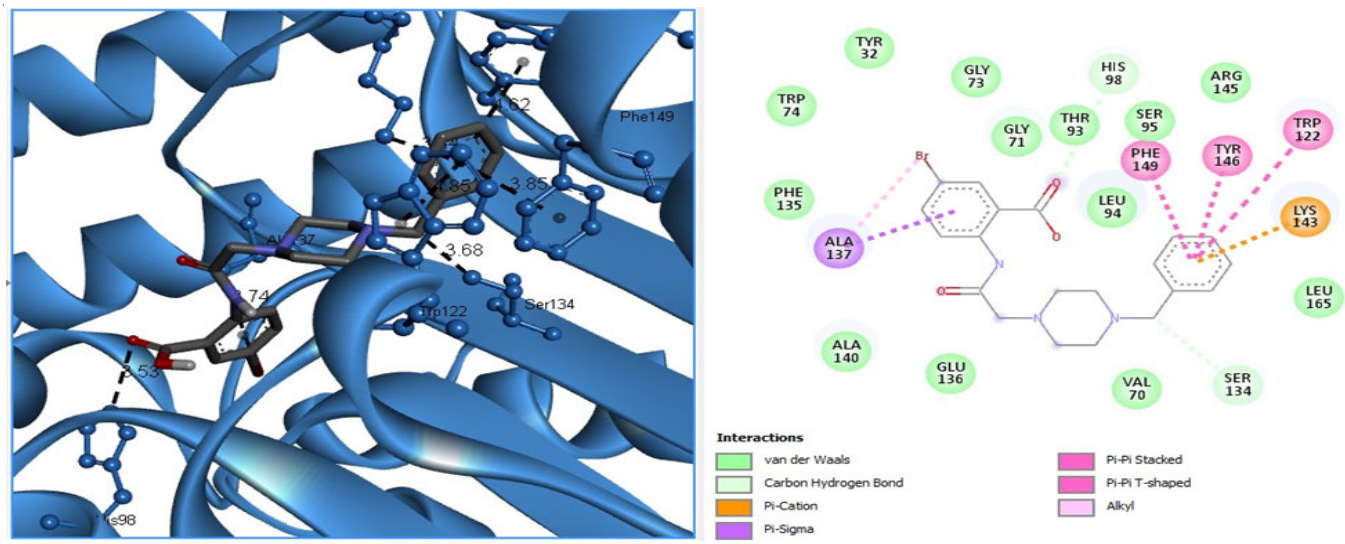


Fig. 7. 3D and 2D view of binding interactions of 2-(2-(4-benzylpiperazin-1-yl) acetamido)-5-bromobenzoic acid (**Mbb4**) with MmaA1 protein

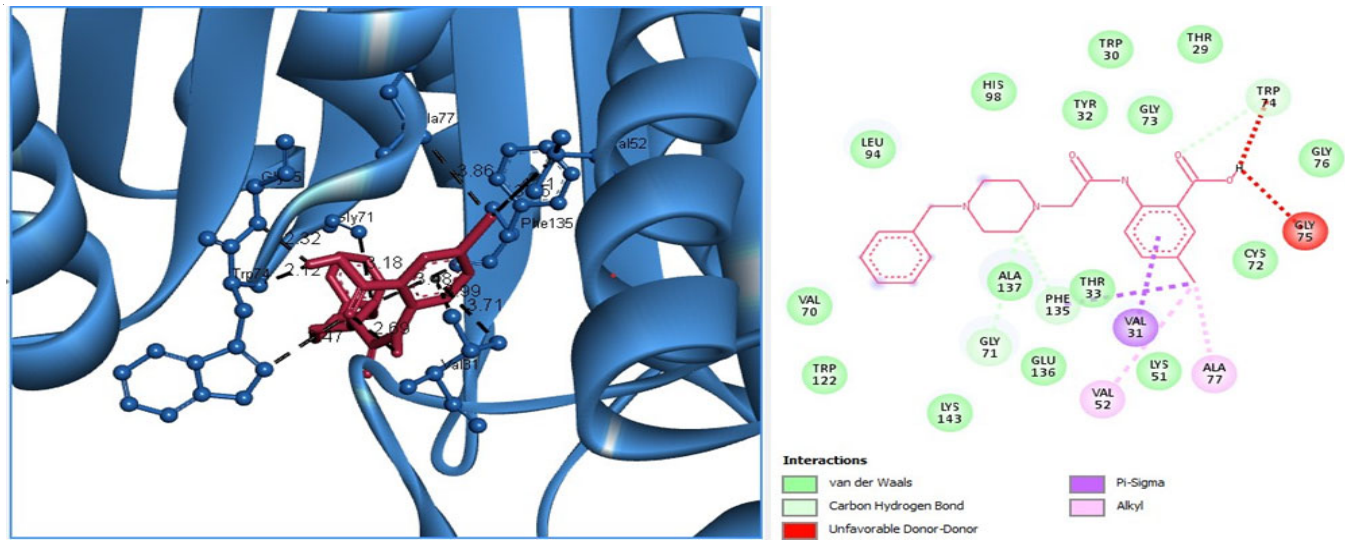


Fig. 8. 3D and 2D view of binding interactions of 2-(2-(4-benzylpiperazin-1-yl) acetamido)-5-chlorobenzoic acid (**Mcb3**) with MmaA1 protein

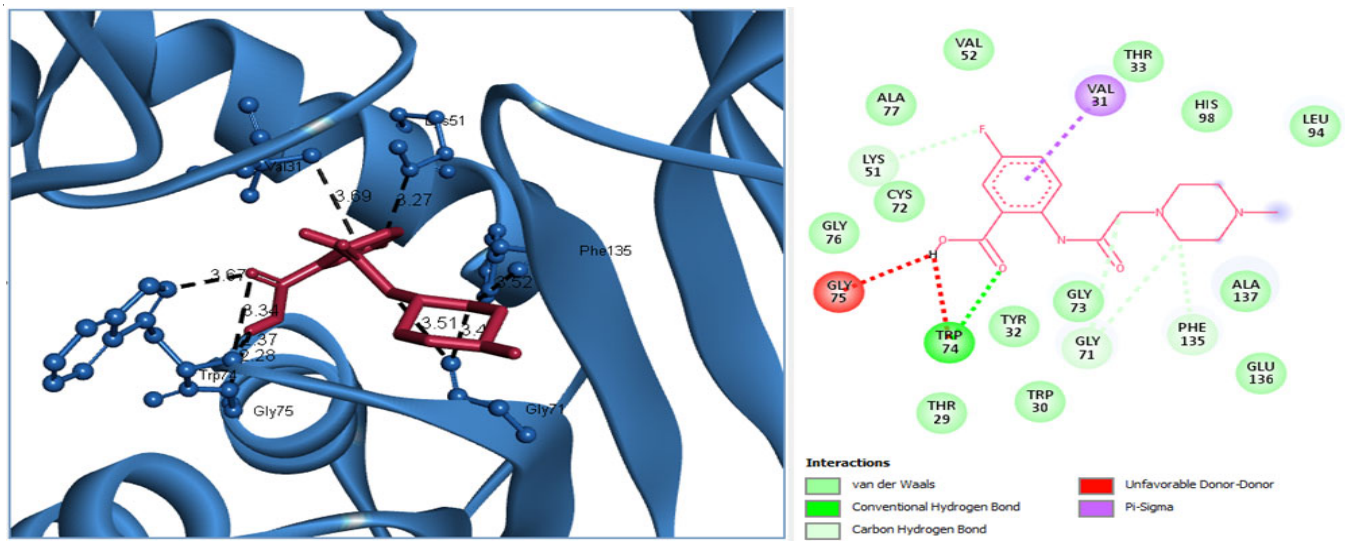


Fig. 9. 3D and 2D view of binding interactions of 5-fluoro-2-(2-(4-methylpiperazin-1-yl)Acetamido) benzoic acid (**Mab2**) with MmaA1 protein

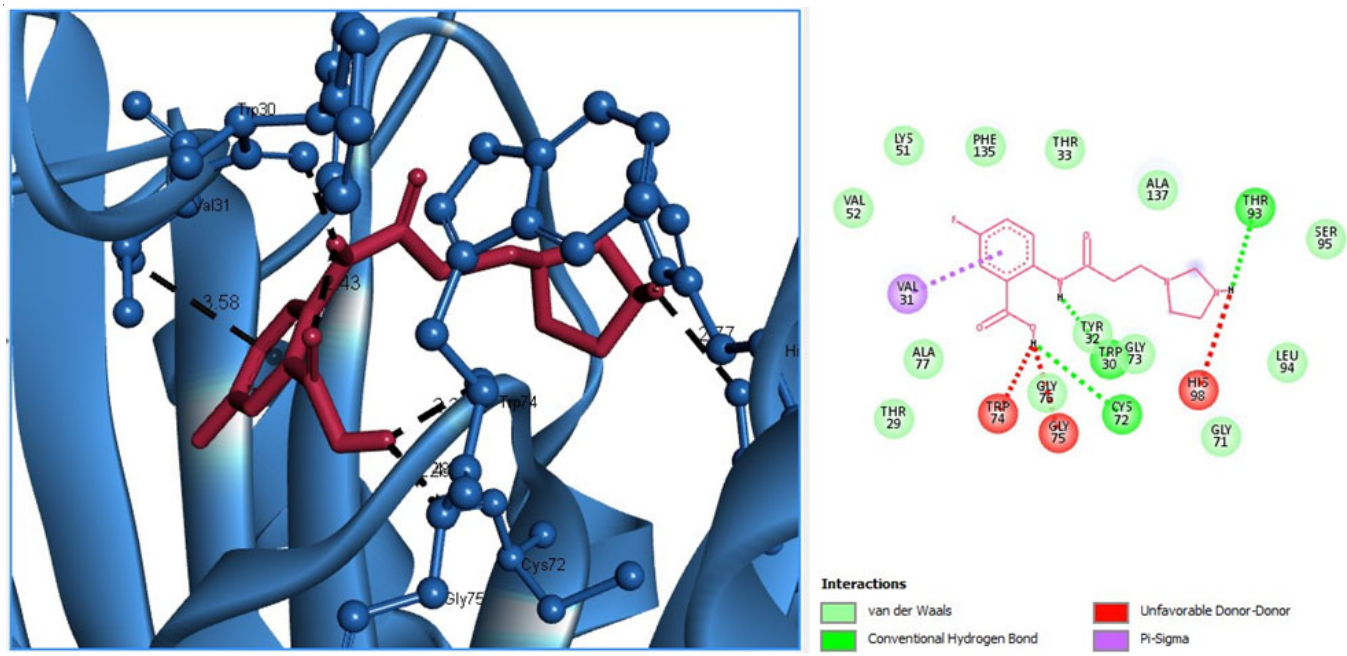


Fig. 10. 3D and 2D view of binding interactions of 5-fluoro-2-(3-(imidazolidin-1-yl)propanamido) benzoic acid (**Mhh1**) with MmaA1 protein

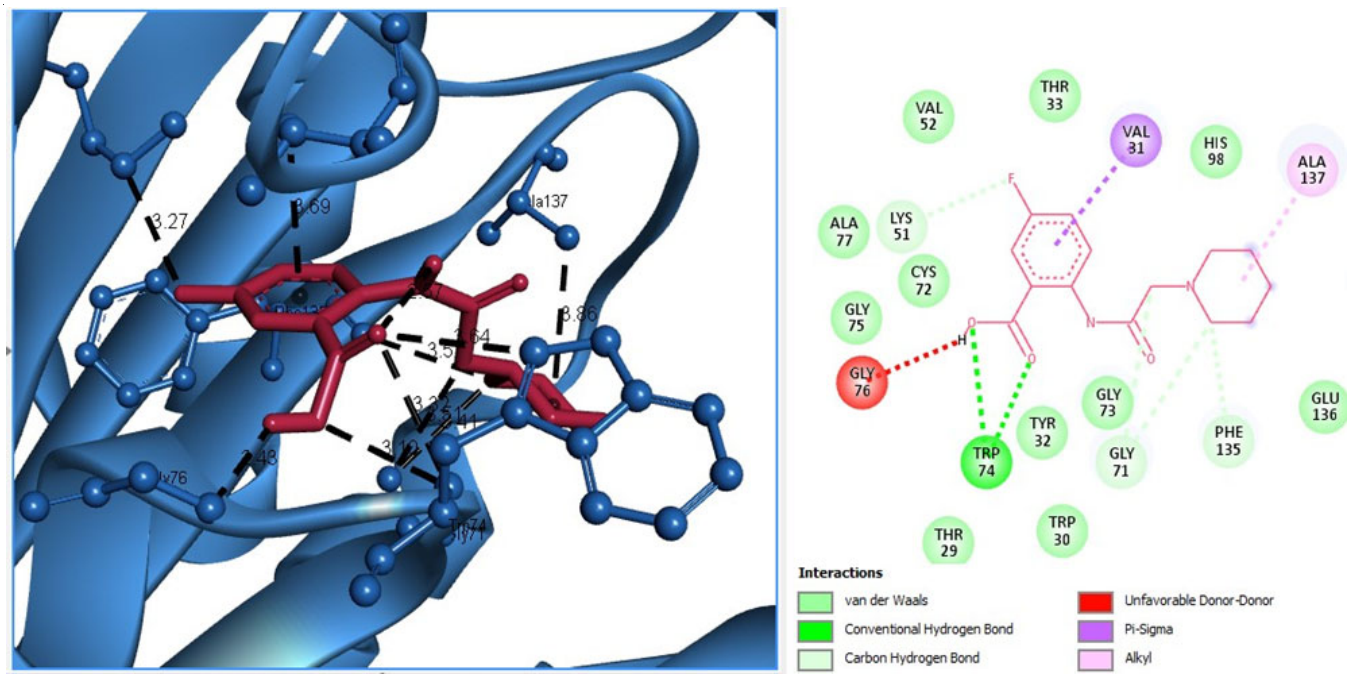


Fig. 11. 3D and 2D view of binding interactions of 5-bromo-2-(3-(piperidin-1-yl)propanamido) benzoic acid (**Mih3**) with MmaA1 protein

1, 2, 5, 6, 10, 11, 12, 15, 16, 20, 21, 22, 24, 27, 30, 31, 32, 34, 35, 38, 39, 43, 44, 49, 51, 52, 53, 54 have lead like properties according to SWISS ADMET study. Compounds ABC-3, 4, 8, 9, 13, 14, 18, 19, 28, 38, 39, 41, 43, 44, 46, 50, 51, 53, have no CYP inhibition. All the ligands have high GI absorption and the ligands ABC-2, 7, 12,17, 22, 23, 25, 27, 32, 33, 37, 42, 47, 48 have no (blood brain barrier) BBB permeation. The bioactivity score in Molinspiration data showed that the ligands exhibit biological action because of interaction with GPCR, nuclear receptor, ion channel modulator and other protease and enzyme inhibitors. The ligands ABC-3, 4, 13, 14, 22, 23, 24, 26, 27, 28, 29, 30, 33, 34, 38, 39, 43 have high probability for GPCR, ABC-26, 27, 30 exhibited ion modulation property, ABC-21-35 showed high probability for nuclear receptor, whereas ABC-23, 38 has protease inhibition property and ABC-21-23, 25-28, 30-33, 35 has enzyme inhibition property. The predicted ADMET results of various physico-chemical and pharmacokinetic parameters of the compounds ABC-1 to ABC-55 are given in Tables 6-11. The compounds identified from the virtual screening study have good ADME qualities that are within an acceptable range, indicating that they have drug-like properties.

TABLE-6
SWISS ADME PREDICTED RESULTS SHOWING VARIOUS PHARMACOKINETIC DESCRIPTORS OF THE MOLECULES **ABC-1** TO **ABC-55**

Compd.	H-bond donor	H-bond acceptor	TPSA (Å^2)	XLOGP3	Drug likeness	Lead likeness
ABC-1	2	5	69.64	-0.04	Yes	Yes
ABC-2	2	6	72.88	-1.12	Yes	Yes
ABC-3	2	5	72.88	0.66	Yes	No
ABC-4	2	5	72.88	0.66	Yes	No
ABC-5	2	5	69.64	-0.45	Yes	Yes
ABC-6	2	4	69.64	0.74	Yes	Yes
ABC-7	2	5	72.88	-0.53	Yes	No
ABC-8	2	5	72.88	0.96	Yes	No
ABC-9	2	4	72.88	1.25	Yes	No
ABC-10	2	4	69.64	0.14	Yes	Yes
ABC-11	2	4	69.64	0.82	Yes	Yes
ABC-12	2	5	72.88	-0.60	Yes	Yes
ABC-13	2	5	72.88	0.90	Yes	No
ABC-14	2	4	72.88	1.19	Yes	No
ABC-15	2	4	69.64	0.46	Yes	Yes
ABC-16	2	4	69.64	0.19	Yes	Yes
ABC-17	2	5	72.88	-1.08	Yes	No
ABC-18	2	5	72.88	0.41	Yes	No
ABC-19	2	4	72.88	0.70	Yes	No
ABC-20	2	4	69.64	-0.17	Yes	Yes
ABC-21	1	5	66.84	-0.17	Yes	Yes
ABC-22	1	6	70.08	-1.20	Yes	Yes
ABC-23	1	6	70.08	0.29	Yes	No
ABC-24	1	5	70.08	0.58	Yes	Yes
ABC-25	1	5	66.84	-0.52	Yes	No
ABC-26	0	2	20.31	0.64	Yes	No
ABC-27	1	6	70.08	-0.57	Yes	Yes
ABC-28	1	6	70.08	0.92	Yes	No
ABC-29	1	5	70.08	1.21	Yes	No
ABC-30	1	5	66.84	0.10	Yes	Yes
ABC-31	1	5	66.84	0.12	Yes	Yes
ABC-32	1	6	70.08	-0.91	Yes	Yes
ABC-33	1	6	70.08	0.29	Yes	No
ABC-34	1	5	70.08	0.58	Yes	Yes
ABC-35	1	5	70.08	0.58	Yes	Yes
ABC-36	0	3	29.54	2.59	Yes	No
ABC-37	0	4	32.78	1.55	Yes	No
ABC-38	0	4	32.78	3.05	Yes	Yes
ABC-39	0	3	32.78	3.33	Yes	Yes
ABC-40	0	3	29.54	2.23	Yes	No
ABC-41	1	2	32.34	2.71	Yes	No
ABC-42	1	3	35.58	1.68	Yes	No
ABC-43	1	3	35.58	2.47	Yes	Yes
ABC-44	1	2	35.58	2.76	Yes	Yes
ABC-45	1	2	32.34	2.35	Yes	No
ABC-46	1	2	32.34	2.96	Yes	No
ABC-47	1	3	35.58	1.93	Yes	No
ABC-48	1	3	35.58	1.93	Yes	No
ABC-49	1	2	35.58	3.12	Yes	Yes
ABC-50	1	2	32.34	2.60	Yes	No
ABC-51	1	2	32.34	3.34	Yes	Yes
ABC-52	1	3	35.58	2.31	Yes	Yes
ABC-53	1	3	35.58	3.10	Yes	Yes
ABC-54	1	2	35.58	3.39	Yes	Yes
ABC-55	1	2	32.34	2.98	Yes	No

TABLE-7
SWISS ADME PREDICTED RESULTS SHOWING VARIOUS PHARMACOKINETIC DESCRIPTORS OF THE MOLECULES **ABC-1** to **ABC-55**

Compd.	GI absorption	BBB permeant	Inhibitor				
			CYP1A2	CYP2C19	CYP2C9	CYP2D6	CYP3A4
ABC-1	High	Y	N	N	N	N	N
ABC-2	High	N	N	N	N	N	N
ABC-3	High	Y	N	N	N	Y	N
ABC-4	High	Y	N	N	N	Y	N
ABC-5	High	Y	N	N	N	N	N
ABC-6	High	Y	N	N	N	N	N
ABC-7	High	N	N	N	N	N	N
ABC-8	High	Y	N	N	N	Y	N
ABC-9	High	Y	N	N	N	Y	N
ABC-10	High	Y	N	N	N	N	N
ABC-11	High	Y	N	N	N	N	N
ABC-12	High	N	N	N	N	N	N
ABC-13	High	Y	N	N	N	Y	N
ABC-14	High	Y	N	N	N	Y	N
ABC-15	High	Y	N	N	N	N	N
ABC-16	High	Y	N	N	N	N	N
ABC-17	High	N	N	N	N	N	N
ABC-18	High	Y	N	N	N	Y	N
ABC-19	High	Y	N	N	N	Y	N
ABC-20	High	Y	N	N	N	N	N
ABC-21	High	Y	N	N	N	N	N
ABC-22	High	N	N	N	N	N	N
ABC-23	High	N	N	N	N	N	N
ABC-24	High	Y	N	N	N	N	N
ABC-25	High	N	N	N	N	N	N
ABC-26	High	Y	N	N	N	N	N
ABC-27	High	N	N	N	N	N	N
ABC-28	High	Y	N	Y	N	Y	N
ABC-29	High	Y	N	N	N	N	N
ABC-30	High	Y	N	N	N	N	N
ABC-31	High	Y	N	N	N	N	N
ABC-32	High	N	N	N	N	N	N
ABC-33	High	N	N	N	N	N	N
ABC-34	High	Y	N	N	N	N	N
ABC-35	High	Y	N	N	N	N	N
ABC-36	High	Y	N	N	N	N	N
ABC-37	High	N	N	N	N	N	N
ABC-38	High	Y	N	Y	N	Y	N
ABC-39	High	Y	N	Y	N	Y	N
ABC-40	High	Y	N	N	N	N	N
ABC-41	High	Y	N	N	N	Y	N
ABC-42	High	N	N	N	N	N	N
ABC-43	High	Y	N	N	N	Y	N
ABC-44	High	Y	N	N	N	Y	N
ABC-45	High	Y	N	N	N	N	N
ABC-46	High	Y	N	N	N	Y	N
ABC-47	High	N	N	N	N	N	N
ABC-48	High	N	N	N	N	N	N
ABC-49	High	Y	Y	Y	N	Y	N
ABC-50	High	Y	N	N	N	Y	N
ABC-51	High	Y	Y	Y	N	Y	N
ABC-52	High	Y	N	N	N	N	N
ABC-53	High	Y	Y	Y	N	Y	Y
ABC-54	High	Y	Y	Y	N	Y	Y
ABC-55	High	Y	Y	N	N	Y	N

TABLE-8
pkCSM PREDICTED RESULTS SHOWING VARIOUS PHARMACOKINETIC DESCRIPTORS OF THE MOLECULES **ABC-1** TO **ABC-55**

Compd.	H-bond donor	H-bond acceptor	TPSA (Å^2)	LOGP	Drug likeness	Compd.	H-bond donor	H-bond acceptor	TPSA (Å^2)	LOGP	Drug likeness
ABC-1	2	3	115.93	1.90	Yes	**ABC-29**	1	5	156.09	2.76	Yes
ABC-2	2	4	121.69	0.70	Yes	**ABC-30**	1	4	115.27	2.03	Yes
ABC-3	2	4	156.75	2.28	Yes	**ABC-31**	1	4	111.33	1.77	Yes
ABC-4	2	4	150.39	2.28	Yes	**ABC-32**	1	5	117.09	0.53	Yes
ABC-5	2	3	109.57	1.55	Yes	**ABC-33**	1	5	152.15	2.10	Yes
ABC-6	2	3	125.64	2.57	Yes	**ABC-34**	1	5	145.78	2.11	Yes
ABC-7	2	4	131.39	1.33	Yes	**ABC-35**	1	4	104.97	1.38	Yes
ABC-8	2	4	166.45	2.17	Yes	**ABC-36**	0	3	96.01	2.07	Yes
ABC-9	2	4	160.09	2.90	Yes	**ABC-37**	0	4	101.77	0.83	Yes
ABC-10	2	3	119.27	2.18	Yes	**ABC-38**	0	4	136.83	2.40	Yes
ABC-11	2	3	122.69	2.46	Yes	**ABC-39**	0	4	130.46	2.41	Yes
ABC-12	2	4	127.83	1.22	Yes	**ABC-40**	0	3	89.65	1.68	Yes
ABC-13	2	4	162.89	2.79	Yes	**ABC-41**	1	2	96.45	2.11	Yes
ABC-14	2	4	156.52	2.79	Yes	**ABC-42**	1	3	102.21	0.87	Yes
ABC-15	2	3	115.71	2.07	Yes	**ABC-43**	1	3	137.26	2.44	Yes
ABC-16	2	4	125.64	2.57	Yes	**ABC-44**	1	3	130.93	2.44	Yes
ABC-17	2	4	131.39	1.33	Yes	**ABC-45**	1	2	90.08	1.72	Yes
ABC-18	2	4	166.45	2.90	Yes	**ABC-46**	1	2	102.81	2.41	Yes
ABC-19	2	4	160.09	2.90	Yes	**ABC-47**	1	3	108.57	1.18	Yes
ABC-20	2	3	119.27	2.18	Yes	**ABC-48**	1	3	143.63	2.75	Yes
ABC-21	1	4	111.36	1.77	Yes	**ABC-49**	1	3	137.26	2.75	Yes
ABC-22	1	5	117.09	0.53	Yes	**ABC-50**	1	2	96.45	2.02	Yes
ABC-23	1	5	152.15	2.10	Yes	**ABC-51**	1	2	106.75	2.76	Yes
ABC-24	1	5	145.78	2.11	Yes	**ABC-52**	1	3	112.51	1.52	Yes
ABC-25	1	4	104.97	1.38	Yes	**ABC-53**	1	3	147.52	3.09	Yes
ABC-26	1	4	121.63	2.42	Yes	**ABC-54**	1	3	141.20	3.10	Yes
ABC-27	1	5	127.39	1.191	Yes	**ABC-55**	1	2	100.39	2.37	Yes
ABC-28	1	5	162.45	2.76	Yes						

***In vitro* antitubercular activity:** All the synthesized compounds (**Mab1-Mgb5**) and (**Eab1-Edb5**) were screened for anti-TB activity by MABA assay against *M. tuberculosis* H37Rv and the results are given in Tables 12 & 13.

Compounds **Mab4**, **Mbb3**, **Mbb4**, **Mcb3**, **Mdb3**, **Mdb5**, **Edb1**, **Mfb1**, **Mfb3** and **Mgb2** showed good anti-TB activity with an MIC of 12.5 µg/mL. Compounds **Mdb1**, **Mdb4** and **Meb1** exhibited excellent anti-TB activity by MABA against *M. tuberculosis* H37Rv bacilli strain with an MIC of 1.56 µg/mL.

SAR of glycinamido analogues: From the *in silico* and *in vitro* results, it is very clear that the aryl ring attached directly to nitrogen atom as present in 2(-N-substituted glycinamido) derivatives (**Mab1-Mgb5**) is essential for the compounds to exhibit potent anti-TB activity than 2-chloro acetates (**Eab1-Edb5**). Substitution of hydrogen on the aromatic ring with halogens improves the activity. The order of potency of halogens is Br > F = Cl. The activity of compounds **Mdb1** and **Mdb2-Mdb5** compounds shows the importance of the position of halogen and electron-withdrawing group on the aromatic ring. It clearly explains that *ortho*-substitution of bromine improves anti-TB activity. The anti-TB activity of the compound varies with the type of heterocyclic amine. Change in the amine altered the activity in the order, piperidine > phenylpiperazine = benzyl piperazine > methyl piperazine > pyrrolidine. Compounds **Meb1**, **Edb1**, **Mdb1** (piperidine), **Mdb4**, **Mab4**, **Mcb4** (phenyl piperazine), **Mbb3** (benzyl piperazine), **Mgb2** (methyl piperazine) and **Mdb5** (pyrrolidine) showed the highest potency.

Conclusion

In present study, a unique class of glycinamido analogues were designed, synthesized, computationally characterized and screened for anti-Tb activity. A library of 330 ligands was created and subsequently submitted to Auto Dock using flexible ligands and semi-flexible receptors. By using *in silico* virtual screening and ADMET studies, 55 hit molecules were identified. The molecular docking study revealed that the MmaA1 protein residues TRP 74, TRP 30, TRP 32, GLY 71, PHE 135, LYS 51, ALA 137, ALA77, HIS 98, VAL 31, PHE149, ASP 139 have significant hydrogen bonding interactions, pi-pi interactions, halogen interactions with the ligands, yielding good docking scores ranging from -10.5 to -8.0. According to Lipinski's rule of 5 in ADME testing, all the compounds were compliant. The *in silico* ADMET data indicates that ligands ABC-21 to ABC-35 attained good bioactivity score according to Molinspiration. The 55 hit molecules (**Mab1-Mgb5** and **Eab1-Edb5**) were successfully synthesized in two steps. Among the synthesized compounds, 10 glycinamide analogs showed good anti-TB activity by MABA against *M. tuberculosis* H37Rv bacilli strain with an MIC of 12.5 µg/mL. Compounds **Mdb1**, **Mdb4**, **Meb1** and **Edb1** exhibited excellent anti-TB activity by MABA against *M. tuberculosis* H37Rv bacilli strain with a MIC of

TABLE-9
pkCSM PREDICTED RESULTS SHOWING VARIOUS PHARMACOKINETIC DESCRIPTORS OF THE MOLECULES **ABC-1** TO **ABC-55**

Compd.	GI absorption	BBB permeant	Inhibitor				
			CYP1A2	CYP2C19	CYP2C9	CYP2D6	CYP3A4
ABC-1	97.16	-1.08	N	N	N	N	N
ABC-2	69.12	-1.144	N	N	N	N	N
ABC-3	97.64	-1.144	N	N	N	N	N
ABC-4	97.14	-0.96	N	N	N	N	N
ABC-5	97.55	-1.057	N	N	N	N	N
ABC-6	96.19	-1.062	N	N	N	N	N
ABC-7	98.66	-1.126	N	N	N	N	N
ABC-8	98.68	-1.124	N	N	N	N	N
ABC-9	96.17	-0.945	N	N	N	N	N
ABC-10	96.58	-1.039	N	N	N	N	N
ABC-11	96.26	-1.041	N	N	N	N	N
ABC-12	69.02	-1.105	N	N	N	N	N
ABC-13	96.74	-1.103	N	N	N	N	N
ABC-14	96.24	-0.925	N	N	N	N	N
ABC-15	96.65	-1.019	N	N	N	N	N
ABC-16	95.69	-1.068	N	N	N	N	N
ABC-17	98.16	-1.132	N	N	N	N	N
ABC-18	96.17	-1.13	N	N	N	N	N
ABC-19	95.67	-0.951	N	N	N	N	N
ABC-20	96.08	-1.045	N	N	N	N	N
ABC-21	97.52	-0.12	N	N	N	N	N
ABC-22	70.74	-0.89	N	N	N	N	N
ABC-23	96.5	-0.89	N	N	N	N	N
ABC-24	97.5	-0.14	N	N	N	N	N
ABC-25	97.91	-0.17	N	N	N	N	N
ABC-26	95.5	-0.14	N	N	N	N	N
ABC-27	74.47	-1.075	N	N	N	N	N
ABC-28	94.48	-1.074	N	N	N	N	N
ABC-29	95.48	-0.313	N	N	N	N	N
ABC-30	95.89	-0.16	N	N	N	N	N
ABC-31	98.03	0.001	N	N	N	N	N
ABC-32	75.91	-0.881	N	N	N	N	N
ABC-33	97	-0.879	N	N	N	N	N
ABC-34	98.01	-0.199	N	N	N	N	N
ABC-35	98.42	-0.193	N	N	N	N	N
ABC-36	94.28	0.221	N	N	N	N	N
ABC-37	80.74	0.043	N	N	N	N	N
ABC-38	93.26	0.41	N	N	N	N	N
ABC-39	94.26	0.429	Y	N	N	Y	N
ABC-40	94.67	0.235	N	N	N	N	N
ABC-41	92.59	0.336	N	N	N	N	N
ABC-42	71.67	0.011	N	N	N	N	N
ABC-43	93.25	0.386	N	N	N	Y	N
ABC-44	92.75	0.415	Y	N	N	Y	N
ABC-45	92.98	0.323	N	N	N	N	N
ABC-46	84.34	-0.029	N	N	N	N	N
ABC-47	92.96	0.304	N	N	N	N	N
ABC-48	93.44	0.355	N	N	N	N	N
ABC-49	92.94	0.352	N	N	N	N	N
ABC-50	93.35	0.291	N	N	N	N	N
ABC-51	91.5	0.291	N	N	N	N	N
ABC-52	93.97	-0.054	N	N	N	N	N
ABC-53	91.98	0.342	N	N	N	N	N
ABC-54	91.48	0.339	Y	Y	N	Y	N
ABC-55	91.89	0.278	N	N	N	N	N

TABLE-10
MOLINSPIRATION PREDICTED RESULTS SHOWING VARIOUS PHYSICO-CHEMICAL DESCRIPTORS OF THE MOLECULES **ABC-1** TO **ABC-55**

Compd.	H-bond donor	H-bond acceptor	TPSA (Å^2)	LOGP	Molecular weight	Drug likeness
ABC-1	2	5	69.64	2.60	280.29	Yes
ABC-2	2	6	72.87	1.59	295.31	Yes
ABC-3	2	6	72.87	2.99	371.41	Yes
ABC-4	2	6	72.87	3.29	357.38	Yes
ABC-5	2	5	69.64	2.10	266.27	Yes
ABC-6	2	5	69.64	3.25	341.20	Yes
ABC-7	2	6	72.87	2.23	356.22	Yes
ABC-8	2	6	72.87	3.63	432.32	Yes
ABC-9	2	6	72.87	3.93	418.29	Yes
ABC-10	2	5	69.64	2.75	327.18	Yes
ABC-11	2	5	69.64	3.12	296.75	Yes
ABC-12	2	6	72.87	2.10	311.77	Yes
ABC-13	2	6	72.87	3.50	387.87	Yes
ABC-14	2	6	72.87	3.80	373.84	Yes
ABC-15	2	5	69.64	2.61	282.73	Yes
ABC-16	2	5	69.64	3.07	341.20	Yes
ABC-17	2	6	72.87	2.05	356.22	Yes
ABC-18	2	6	72.87	3.45	432.32	Yes
ABC-19	2	6	72.87	3.75	418.29	Yes
ABC-20	2	5	69.64	2.56	327.18	Yes
ABC-21	1	5	66.84	2.16	263.29	Yes
ABC-22	1	6	70.08	1.15	278.31	Yes
ABC-23	1	6	70.08	2.54	354.41	Yes
ABC-24	1	6	70.08	2.84	340.38	Yes
ABC-25	1	5	66.84	1.66	249.27	Yes
ABC-26	1	5	66.84	2.82	297.74	Yes
ABC-27	1	6	70.08	1.80	312.75	Yes
ABC-28	1	6	70.08	3.20	388.85	Yes
ABC-29	1	6	70.08	3.50	374.82	Yes
ABC-30	1	5	66.84	2.31	283.71	Yes
ABC-31	1	5	66.84	2.10	263.29	Yes
ABC-32	1	6	70.08	1.09	278.31	Yes
ABC-33	1	6	70.08	2.49	354.41	Yes
ABC-34	1	6	70.08	2.79	340.38	Yes
ABC-35	1	5	66.84	1.60	249.27	Yes
ABC-36	0	3	29.54	2.22	219.28	Yes
ABC-37	0	4	32.78	1.20	234.30	Yes
ABC-38	0	4	32.78	2.60	310.40	Yes
ABC-39	0	4	32.78	2.90	296.37	Yes
ABC-40	0	3	29.54	1.71	205.26	Yes
ABC-41	1	3	32.34	2.42	218.30	Yes
ABC-42	1	4	35.37	1.41	233.31	Yes
ABC-43	1	4	35.37	2.80	309.41	Yes
ABC-44	1	4	35.57	3.10	295.39	Yes
ABC-45	1	3	32.34	1.92	204.27	Yes
ABC-46	1	3	32.34	2.87	232.33	Yes
ABC-47	1	4	35.57	1.85	247.34	Yes
ABC-48	1	4	35.57	3.25	247.32	Yes
ABC-49	1	4	35.57	3.55	309.41	Yes
ABC-50	1	3	32.34	2.37	218.30	Yes
ABC-51	1	3	32.34	3.14	252.75	Yes
ABC-52	1	4	35.57	2.08	267.76	Yes
ABC-53	1	4	35.57	3.48	343.86	Yes
ABC-54	1	4	35.57	3.78	329.83	Yes
ABC-55	1	3	32.34	2.59	238.72	Yes

TABLE-11
MOLINSPIRATION PREDICTED RESULTS SHOWING BIOACTIVITY SCORE OF THE COMPOUNDS **ABC-1** TO **ABC-55**

Compd.	GPCR ligand	Ion channel modulator	Kinase inhibitor	Nuclear receptor ligand	Protease inhibitor	Enzyme inhibitor
ABC-1	-0.05	-0.16	-0.23	-0.11	-0.17	-0.09
ABC-2	-0.01	-0.13	-0.10	-0.13	-0.17	-0.11
ABC-3	0.02	-0.15	-0.08	-0.08	-0.05	-0.09
ABC-4	0.02	-0.15	-0.09	-0.07	-0.09	-0.14
ABC-5	-0.09	-0.16	-0.24	-0.11	-0.16	-0.09
ABC-6	-0.21	-0.26	-0.37	-0.32	-0.34	-0.18
ABC-7	-0.16	-0.23	-0.23	-0.33	-0.33	-0.19
ABC-8	-0.09	-0.22	-0.19	-0.24	-0.17	-0.16
ABC-9	-0.10	-0.22	-0.20	-0.23	-0.22	-0.21
ABC-10	-0.26	-0.26	-0.38	-0.33	-0.34	-0.20
ABC-11	-0.06	-0.14	-0.31	-0.18	-0.22	-0.12
ABC-12	-0.03	-0.12	-0.17	-0.20	-0.21	-0.13
ABC-13	0.01	-0.14	-0.15	-0.14	-0.08	-0.11
ABC-14	0.01	-0.14	-0.15	-0.13	-0.13	-0.16
ABC-15	-0.10	-0.15	-0.32	-0.19	-0.21	-0.13
ABC-16	-0.19	-0.32	-0.40	-0.43	-0.30	-0.16
ABC-17	-0.15	-0.28	-0.26	-0.44	-0.29	-0.17
ABC-18	-0.08	-0.26	-0.21	-0.33	-0.14	-0.14
ABC-19	-0.09	-0.27	-0.23	-0.32	-0.19	-0.19
ABC-20	-0.24	-0.33	-0.42	-0.46	-0.30	-0.17
ABC-21	-0.01	-0.01	-0.38	0.03	-0.09	0.09
ABC-22	0.03	0.01	-0.23	0	-0.08	0.07
ABC-23	0.08	-0.04	-0.15	0.06	0.06	0.05
ABC-24	0.08	-0.05	-0.16	0.07	-0.01	-0.01
ABC-25	-0.05	-0.01	-0.41	0.02	-0.07	0.07
ABC-26	0.03	0	-0.35	0.04	-0.10	0.06
ABC-27	0.07	0.02	-0.21	0.01	-0.09	0.03
ABC-28	0.08	-0.03	-0.18	0.03	0.01	0.02
ABC-29	0.07	-0.04	-0.18	0.03	-0.05	-0.04
ABC-30	0	0.01	-0.37	0.05	-0.08	0.05
ABC-31	-0.06	-0.05	-0.42	0.07	-0.12	0.07
ABC-32	-0.02	-0.03	-0.27	0.04	-0.11	0.05
ABC-33	0.04	-0.07	-0.18	0.09	0.04	0.04
ABC-34	0.04	-0.08	-0.19	0.10	-0.03	-0.03
ABC-35	-0.11	-0.05	-0.45	0.07	-0.10	0.05
ABC-36	-0.28	-0.06	-0.60	-0.38	-0.36	-0.11
ABC-37	-0.20	-0.02	-0.40	-0.38	-0.33	-0.09
ABC-38	0.03	-0.06	-0.15	-0.07	0	-0.02
ABC-39	0	-0.07	-0.16	-0.10	-0.11	-0.09
ABC-40	-0.36	-0.09	-0.66	-0.42	-0.36	-0.15
ABC-41	-0.33	-0.24	-0.48	-0.70	-0.44	-0.29
ABC-42	-0.25	-0.18	-0.28	-0.68	-0.40	-0.26
ABC-43	0	-0.18	-0.06	-0.30	-0.06	-0.14
ABC-44	-0.02	-0.18	-0.08	-0.31	-0.14	-0.20
ABC-45	-0.41	-0.28	-0.52	-0.76	-0.44	-0.34
ABC-46	-0.32	-0.32	-0.46	-0.63	-0.44	-0.32
ABC-47	-0.24	-0.26	-0.27	-0.61	-0.40	-0.30
ABC-48	-0.21	-0.21	-0.12	-0.61	-0.26	-0.23
ABC-49	-0.05	-0.26	-0.12	-0.31	-0.17	-0.26
ABC-50	-0.40	-0.35	-0.50	-0.68	-0.45	-0.38
ABC-51	-0.26	-0.22	-0.41	-0.62	-0.41	-0.28
ABC-52	-0.19	-0.17	-0.23	-0.60	-0.38	-0.26
ABC-53	0	-0.18	-0.08	-0.30	-0.08	-0.18
ABC-54	-0.01	-0.18	-0.08	-0.31	-0.16	-0.23
ABC-55	-0.33	-0.24	-0.45	-0.67	-0.42	-0.33

Note: The active compounds are highlighted in green color.

TABLE-12
ANTI-TB ACTIVITY OF THE COMPOUNDS **Mab1** TO **Mgb5**

Compd.	MIC against MtbH37Rv (MABA assay) (µg/mL)	Compd.	MIC against MtbH37Rv (MABA assay) (µg/mL)
Mab1	25.0	**Mdb4**	1.56
Mab2	25.0	**Mdb5**	12.5
Mab3	25.0	**Meb1**	1.56
Mab4	12.5	**Meb2**	25.0
Mab5	25.0	**Meb3**	25.0
Mbb1	25.0	**Meb4**	25.0
Mbb2	25.0	**Meb5**	25.0
Mbb3	12.5	**Mfb1**	12.5
Mbb4	12.5	**Mfb2**	25.0
Mbb5	25.0	**Mfb3**	12.5
Mcb1	25.0	**Mfb4**	25.0
Mcb2	25.0	**Mfb5**	25.0
Mcb3	12.5	**Mgb1**	25.0
Mcb4	25.0	**Mgb2**	12.5
Mcb5	25.0	**Mgb3**	25.0
Mdb1	1.56	**Mgb4**	25.0
Mdb2	25.0	**Mgb5**	25.0
Mdb3	12.5	Rifampicin	3.125

TABLE-13
ANTI-TB ACTIVITY OF THE COMPOUNDS **Eab1** TO **Edb5**

Compd.	MIC against M. tbH37Rv (MABA assay) (µg/mL)	Compd.	MIC against M. tbH37Rv (MABA assay) (µg/mL)
Eab1	25	**Ecb1**	25.0
Eab2	25	**Ecb2**	25.0
Eab3	25	**Ecb3**	25.0
Eab4	25	**Ecb4**	25.0
Eab5	25	**Ecb5**	25.0
Ebb1	25	**Edb1**	12.5
Ebb2	0	**Edb2**	25.0
Ebb3	25	**Edb3**	25.0
Ebb4	25	**Edb4**	25.0
Ebb5	25	**Edb5**	25.0
		Rifampicin	3.125

1.56 µg/mL when compared with standard Rifampicin. The obtained SAR clarifies the essential structural elements that improve the potency and drug-like characteristics of the glycinamido analogues.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this article.

REFERENCES

1. https://www.nhs.uk/conditions/tuberculosis-tb/#:~:text=Tuberculosis%20(TB)%20is%20caused%20by,a%20long%20period%20of%20time
2. T.B. Facts, https://www.cdc.gov/tb/worldtbday/history.html
3. World Health Organization (WHO), Global Tuberculosis Report (2022); https://www.who.int/publications/i/item/9789240061729
4. P. Miotto, Y. Zhang, D.M. Cirillo and W.C. Yam, *Respirology*, **23**, 1098 (2018); https://doi.org/10.1111/resp.13393
5. TB India Report (2022); http://www.tbcindia.gov.in (2022). Accessed on 25.03.2022.
6. S. Tiberi, M. Muñoz-Torrico, R. Duarte, M. Dalcolmo, L. D'Ambrosio and G.-B. Migliori, *Pulmonology*, **24**, 86 (2018); https://doi.org/10.1016/j.rppnen.2017.10.009
7. C. Perrin, K. Athersuch, G. Elder, M. Martin and A. Alsalhani, *BMJ Glob. Health*, **7**, e007490 (2022); https://doi.org/10.1136/bmjgh-2021-007490
8. P.J. Brennan, *Tuberculosis*, **83**, 91 (2003); https://doi.org/10.1016/s1472-9792(02)00089-6
9. O. Zimhony, J.S. Cox, J.T. Welch, C. Vilchèze and W.R. Jacobs Jr, *Nat. Med.*, **6**, 1043 (2000); https://doi.org/10.1038/79558
10. M.S. Glickman, J.S. Cox and W.R. Jacobs Jr., *Mol. Cell*, **5**, 717 (2000); https://doi.org/10.1016/S1097-2765(00)80250-6
11. M.A. Forrellad, L.I. Klepp, A. Gioffré, J. Sabio y García, H.R. Morbidoni, M.P. Santangelo, A.A. Cataldi and F. Bigi, *Virulence*, **4**, 3 (2013); https://doi.org/10.4161/viru.22329
12. K. Takayama, C. Wang and G.S. Besra, *Clin. Microbiol. Rev.*, **18**, 81 (2005); https://doi.org/10.1128/CMR.18.1.81-101.2005
13. H. Veeravarapu, V. Malkhed, K.K. Mustyala, R. Vadija, R. Malikanti, U. Vuruputuri and M.K.K. Muthyala, *Mol. Divers.*, **25**, 351 (2021); https://doi.org/10.1007/s11030-020-10107-0
14. H. Veeravarapu, M. Tirumalasetty, S.P. Kurati, U. Wunnava and M.K.K. Muthyala, *Bioorg. Med. Chem. Lett.*, **30**, 127603 (2020); https://doi.org/10.1016/j.bmcl.2020.127603
15. G.M. Morris, R. Huey, W. Lindstrom, M.F. Sanner, R.K. Belew, D.S. Goodsell and A.J. Olson, *J. Comput. Chem.*, **30**, 2785 (2009); https://doi.org/10.1002/jcc.21256
16. L. Schrodinger, The PyMOL Molecular Graphics System, Version 1.3r1 (2010).
17. BIOVIA, Dassault Systemes, Discovery Studio Visualizer, v21.1.0.20298; Dassault Systemes: Sandiego, CA, USA (2021).
18. A. Daina, O. Michielin and V. Zoete, *Sci. Rep.*, **7**, 42717 (2017); https://doi.org/10.1038/srep42717
19. D.E.V. Pires, T.L. Blundell and D.B. Ascher, *J. Med. Chem.*, **58**, 4066 (2015); https://doi.org/10.1021/acs.jmedchem.5b00104
20. Molinspiration Cheminformatics Free Web Services, Available online: https://www.molinspiration.com (accessed on 12 March 2023).
21. J.L. Dahlin, J. Inglese and M.A. Walters, *Nat. Rev. Drug Discov.*, **14**, 279 (2015); https://doi.org/10.1038/nrd4578
22. L. Di, *Expert Opin. Drug Metab. Toxicol.*, **10**, 379 (2014); https://doi.org/10.1517/17425255.2014.876006
23. C.A. Lipinski, F. Lombardo, B.W. Dominy and P.J. Feeney, *Adv. Drug Deliv. Rev.*, **23**, 3 (1997); https://doi.org/10.1016/S0169-409X(96)00423-1
24. A. Jarrahpour, J. Fathi, M. Mimouni, T.B. Hadda, J. Sheikh, Z. Chohan and A. Parvez, *Med. Chem. Res.*, **21**, 1984 (2012); https://doi.org/10.1007/s00044-011-9723-0
25. S. Tahlan, K. Ramasamy, S. Lim, S.A. Shah, V. Mani and B. Narasimhan, *Chem. Cent. J.*, **12**, 139 (2018); https://doi.org/10.1186/s13065-018-0513-3
26. S.G. Franzblau, R.S. Witzig, J.C. Mclaughlin, P. Torres, G. Madico, A. Hernandez, M.T. Degnan, M.B. Cook, V.K. Quenzer, R.M. Ferguson and R.H. Gilman, *J. Clin. Microbiol.*, **36**, 362 (1998); https://doi.org/10.1128/JCM.36.2.362-366.1998
27. C.B. Inderlied and M. Salfinger, Antimicrobial Agents and Susceptibility Tests: Mycobacteria, Manual of Clinical Microbiology, ASM Press, Washington, edn. 6, p. 1385 (1995).
28. M.A. Morgan, C.D. Horstmeier, D.R. DeYoung and G.D. Roberts, *J. Clin. Microbiol.*, **18**, 384 (1983); https://doi.org/10.1128/jcm.18.2.384-388.1983
29. S. Ali, A. Ehtram, N. Arora, P. Manjunath, D. Roy, N.Z. Ehtesham and S.E. Hasnain, *Front. Cell. Infect. Microbiol.*, **11**, 622487 (2021); https://doi.org/10.3389/fcimb.2021.622487
30. J. Dundas, Z. Ouyang, J. Tseng, A. Binkowski, Y. Turpaz and J. Liang, *Nucleic Acids Res.*, **34**, 116 (2006); https://doi.org/10.1093/nar/gkl282
31. T. Halgren, *Chem. Biol. Drug Des.*, **69**, 146 (2007); https://doi.org/10.1111/j.1747-0285.2007.00483.x
32. D. Schneidman-Duhovny, Y. Inbar, R. Nussinov and H.J. Wolfson, *Nucleic Acids Res.*, **33**, 363 (2005); https://doi.org/10.1093/nar/gki481
33. J. Vaubourgeix, J.F. Bardou, F. Boisssier, S. Julien, P. Constant, O. Ploux, M. Daffe, A. Quemard, L. Mourey, *J. Biol. Chem.*, **284**, 19321 (2009); https://doi.org/10.1074/jbc.M809599200