Insilco screening and synthesis of tripeptides for tuberculosis

Dolly Chauhan, Priya Sinha, K. Nagarajan*, Parul Grover*, Praveen Dixit, Pankaj Bhatt,Pragati Gupta

KIET group of Institutions (KIET School of Pharmacy), Delhi NCR, Muradnagar, Ghaziabad, U.P., India 201206

Corresponding authors email: k.nagarajan@kiet.edu, parul.grover@kiet.edu

Abstract

One of the main diseases caused by Mycobacterium tuberculosis is tuberculosis (TB). It is necessary to create safer, smaller chain peptides in order to prevent TB from becoming widely and multi-drug resistant. Two fatty acid synthase systems (FAS), FAS-I and FAS-II, are involved in the production of mycolic acids, which are long-chain, acylated, and hydroxylated fatty acids (MA). Our investigation focuses on mycolic acid synthase, which is essential for Mycobacterium tuberculosis intracellular survival and lowers the apoptotic activity of macrophages. The primary lipid in the envelope, mycolic acid, makes up the exterior mycomembrane. Our goal is to investigate proline-based tripeptides as effective mycolic acid synthase inhibitors. As a crucial amino acid, proline has been used in various shorter chains of selected tripeptides for a thorough analysis of 3D interactions using the Swiss DOCK online tool (PDB code: 1l1e), Chimera, and Discovery studio visualizer. With a G value of (-8.92 kcal/mol), Pro-trp-tyr was determined to be the most promising lead as a powerful mycolic acid synthase inhibitor, followed by Pro-glu-gln and Pro-gly-his, which had G values of (-8.66 kcal/mol) and (-8.62 kcal/mol), respectively.

Key words: Tripeptides, Docking, Mycolic Acid Synthase.

I. Introduction

The intracellular pathogen Mycobacterium tuberculosis is what causes the infectious illness tuberculosis (Mtb). According to the WHO's 2018 report on tuberculosis, over 1.2 million people died from the disease and an estimated 10 million people contracted it. Around 50 antibiotics are now in the clinical pathway, according to WHO [1]. The pharmaceutical sector, however, is insufficient to address the issue of rising antibiotic resistance. This is mostly caused by elements including incomplete diagnosis, poor patient compliance, subpar medications, and mutation transfer [2]. The cell surface of mycobacterium possesses an odd waxy covering that is neither Gram-positive nor - negative and is primarily made of mycolic acid. This article describes the models developed for predicting antitubercular peptides using peptide sequence information [3]. Our creation of antitubercular peptides, which consider a number of sequence factors, including the amino acid composition, binary profile of terminal residues, and dipeptide composition, may provide an alternate method for combating antibiotic resistance [4]. The three major types of mycolic acids produced by M. tuberculosis

are alpha, methoxy, and keto. Alpha-mycolic acids, which have several cyclopropane rings, make up at least 70% of the mycolic acids in the body [5]. 10% to 15% of the mycolic acids in the organism are methyl-mycolic acids, which have several methoxy groups. The 10% to 15% of mycolic acids that are left over are keto-mycolic acids, which include several ketone groups. Mtb is inhaled as an aerosol and then travels to the lungs [6]. Mtb avoids interaction with potentially harmful lysosomal hydrolases and secures its survival by inhibiting the phagosome's acidification process and preventing its fusion with the lysosome after entering alveolar macrophages via receptor-mediated phagocytosis [7]. After Mtb has entered the lung interstitial tissue, dendritic cells or inflammatory monocytes move it to the pulmonary lymph nodes for T cell priming, which draws T and B cells to the lung parenchyma. This promotes the synthesis of several chemokines and cytokines as well as the attraction of more immune cells, such as lymphocytes, who will organise themselves into a globular structure with infected macrophages in the middle, resembling the shape of granuloma [8]. A situation where the mycobacteria can survive is created through the regulation of the immune response, even though it gives the host the ability to maintain the mycobacterial infection in a controlled microenvironment. Furthermore, established granulomas create a hypoxic environment near their center, which will induce mycobacteria to enter a dormant state—a state of low metabolic activity in which the bacteria do not multiply and display a phenotypic drug resistance. In many cases, the infection remains dormant [9]. However, a variety of genetic and environmental factors could cause this latent infection to become active TB weeks or decades down the road. Antitubercular peptide with high in vitro activity that is rich in proline and arginine was first reported in 2001. The antifungal peptide plectasin has been shown to be effective against Mycobacterium species by Erik et al. in 2018. Shamima et al. showed how amino acids can be used to make peptides that have activity in the detection, diagnosis, and treatment of TB. in 2019 [10]. Through a thorough docking investigation approach, our goal is to build a short chain peptide lead, preferably a tripeptide, as effective inhibitors of Mycobacterium tuberculosis Mycolic acid synthase.

II. Materials for experiments and methodology

The Configuration of the system in which docking study was performed along with other online tools of drug design was running on 1.80 GHZ Acer Laptop Intel®Core™ is processor, 8 GB RAM and 64-Bit window operating system.

Methodology

Pro-arg-asp, Pro-Ala-arg, pro-asp-cys, pro-cys-glu, pro-glu-gln, pro-gly-his, pro-his-ile, pro-ile-leu, pro-leu-met, pro-met-phe, pro-phe-ser, pro-ser-lys, pro-lys-trp, pro-trp-tyr, pro-tyr-val, pro-val-ala
were taken for the study and submitted to SWISS ADME online to examine the drug likeliness property of the Lipinski rule of five as well as in addition to testing the drug's metabolic profile against several cytochrome P450 inhibitors, all compounds' bioavailability were assessed against the substrate permeability glycoprotein (PGP) (CYP1 A2, CYP2C19, CYP2C9, CYP2D6, CYP3A4). As a general method of selecting the most effective potent molecule, the online Molinspiration cheminformatics tool was utilised to forecast the various physicochemical properties as well as the bioactivity score on enzyme inhibition for each of the seventeen smaller chain tripeptides. The same group of compounds were tested using the method 2 medication online PASS software to determine their activity against a particular

target and any potential harmful or negative consequences. Mycolic acid occurs within the fluid matrix in the form of free trehalose dimycolate (TDM) and trehalose monomycolate (TM), and PDB code was utilised to dock the selective dipeptides based on preliminary results with a target protein (1l1e). UCSF Chimera and the Biovia Discovery studio visualizer were used to further visualisation and analyse in-depth the bonding interaction in 3D and 2D.

III Results and Discussion

For all 16 selected smaller chain tripeptides, including Pro-arg-asp, Pro-Ala-arg, pro-asp-cys, pro-cys-glu, pro-glu-gln, pro-gly-his, pro-his-ile, pro-ile-leu, pro-leu-met, pro-met-phe, pro-phe-ser, pro-ser-lys, pro-lys-trp, pro-trp-tyr, pro-tyr-val, and pro-val-ala, we used the Swiss ADME tool to determine their physicochemical characteristics, including molecular weight, number of heavy atoms, number of aromatic heavy atoms, number of rotatable bonds, number of hydrogen bond donors, number of hydrogen bond acceptors, molar refractivity, total polar surface area, lipophilicity (consensus logP/o), water solubility (log S), and all ADME. Tables presenting these parameters and the findings for all 16 tripeptides are included.

The same group of compounds were also analyzed using cheminformatics tools from Molinspiration, and the outcomes are shown in a table. This tool revealed that the majority of the tripeptides were eliminated as general enzyme inhibitors based on relative bioactivity score, and that only six dipeptides (pro-trp-tyr, pro-met-phe, pro-gly-his, pro-glu-gln, pro-cys-glu, and pro-ala-arg) were chosen for further investigation. Additionally, the PASS online way 2 drug software tool was used for all 16 tripeptide molecules with their isomeric SMILES notation to assess the additional bioactivity of smaller chain tripeptides against a specific target for tuberculosis with the possibility of adverse and toxic effects prediction. A table presents the PASS activity value results. After comparing the PASS and Molinspiration data, only six dipeptides (pro-trp-tyr, pro-met-phe, pro-gly-his, pro-glu-gln, pro-cys-glu, and pro-ala-arg) were chosen for further docking exploration against the specific target mycolic acid synthase (PDB Code: 1l1e). Another table presents the findings of the docking investigation with the G value (kcal/mol).

Pro-trp-tyr, pro-met-phe, pro-gly-his, pro-glu-gln, pro-cys-glu, and pro-ala-arg all underwent thorough docking investigations. Pro-trp-tyr was discovered to be the most promising compound as a potent mycolic acid synthase inhibitor with a G value of -8.92 kcal/mol.

Conclusion

After SWISS ADME on certain parameters, 16 tripeptides are filtered from 6 tripeptides, and we checked their toxicity and perform docking on them.

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Conflict of interest

All authors declares that there is no competing interest.

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Tripeptides	Canonical SMILES	Formula	MW	Heavy atoms	Aromatic heavy atoms
Pro-arg-asp	OC(=O)C[C@@H](C(=O)O)NC(=O)[C@@H](NC(=O)[C@@H]1CCCN1)CCCN=C(N)N	C15H26N6O6	386.4	27	0
Pro-Ala-arg	OC(=O)[C@@H](NC(=O)[C@@H](NC(=O)[C@@H]1CCCN1)C)CCCN=C(N)N	C14H26N6O4	342.39	24	0
pro-asp-cys	SC[C@@H](C(=O)O)NC(=O)[C@@H](NC(=O)[C@@H]1CCCN1)CC(=O)O	C12H19N3O6S	333.36	22	0
pro-cys-glu	SC[C@@H](C(=O)N[C@H](C(=O)O)CCC(=O)O)NC(=O)[C@@H]1CCCN1	C13H21N3O6S	347.39	23	0
pro-glu-gln	OC(=O)CC[C@@H](C(=O)N[C@H](C(=O)O)CCC(=O)N)NC(=O)[C@@H]1CCCN1	C15H24N4O7	372.37	26	0
pro-gly-his	O=C(N[C@H](C(=O)O)Cc1[nH]cnc1)CNC(=O)[C@@H]1CCCN1	C13H19N5O4	309.32	22	5
pro-his-ile	CC[C@@H]([C@@H](C(=O)O)NC(=O)[C@H](Cc1cnc[nH]1)NC(=O)[C@@H]1CCCN1)C	C17H27N5O4	365.43	26	5
pro-ile-leu	CC[C@@H]([C@@H](C(=O)N[C@H](C(=O)O)CC(C)C)NC(=O)[C@@H]1CCCN1)C	C17H31N3O4	341.45	24	0
pro-leu-met	CSCC[C@@H](C(=O)O)NC(=O)[C@@H](NC(=O)[C@@H]1CCCN1)CC(C)C	C16H29N3O4S	359.48	24	0
pro-met-phe	CSCC[C@@H](C(=O)N[C@H](C(=O)O)Cc1ccccc1)NC(=O)[C@@H]1CCCN1	C19H27N3O4S	393.5	27	6
pro-phe-ser	OC[C@@H](C(=O)O)NC(=O)[C@H](Cc1ccccc1)NC(=O)[C@@H]1CCCN1	C17H23N3O5	349.38	25	6
pro-ser-lys	NCCCC[C@@H](C(=O)N[C@H](C(=O)O)Cc1c[nH]c2c1cccc2)NC(=O)[C@@H]1CCCN1	C22H31N5O4	429.51	31	9
pro-lys-trp	NCCCC[C@@H](C(=O)N[C@H](C(=O)O)Cc1c[nH]c2c1cccc2)NC(=O)[C@@H]1CCCN1	C22H31N5O4	429.51	31	9
pro-trp-tyr	Oc1ccc(cc1)C[C@@H](C(=O)O)NC(=O)[C@H](Cc1c[nH]c2c1cccc2)NC(=O)[C@@H]1CCCN1	C25H28N4O5	464.51	34	15
pro-tyr-val	O=C([C@H](Cc1ccc(cc1)O)NC(=O)[C@@H]1CCCN1)N[C@H](C(=O)O)C(C)C	C19H27N3O	377.4	27	6

		5	3		
pro-val-ala	CC([C@@H](C(=O)N[C@H](C(=O)O)C)NC(=O)[C@@H]1CCCN1)C	C13H23N3O4	285.34	20	0

Table 1: It represents the Canonical SMILES, Formula, Molecular weight, heavy atoms, Aromatic heavy atoms.

Table 2: It represents the Fraction Csp3, Rotatable bonds, H-bond acceptors, H-bond donors, MR, TPSA, iLOGP, XLOGP3, WLOGP, MLOGP, Silicos-IT Log P.

Tripeptides	Fraction Csp3	Rotatable bonds	H-bond acceptors	H-bond donors	MR	TPSA	iLOGP	XLOGP3	WLOGP	MLOGP	Silicos-IT Log P
Pro-arg-asp	0.67	13	8	7	97.47	209.23	-0.09	-6.35	-3.06	-2.01	-1.57
Pro-Ala-arg	0.71	11	6	6	90.89	171.93	0.66	-5.48	-2.51	-1.48	-0.99
pro-asp-cys	0.67	10	7	5	81.88	183.63	-0.15	-5.23	-2.18	-1.65	-0.85
pro-cys-glu	0.69	11	7	5	86.68	183.63	0.49	-4.87	-1.79	-1.38	-0.46
pro-glu-gln	0.67	13	8	6	91.28	187.92	0.1	-5.96	-2.46	-2.07	-1.07
pro-gly-his	0.54	9	6	5	79.19	136.21	0.43	-3.51	-1.99	-1.91	-0.11
pro-his-ile	0.65	11	6	5	98.42	136.21	0.87	-3.05	-0.58	-0.9	1.12
pro-ile-leu	0.82	11	5	4	96.21	107.53	2	-1.64	0.5	0.48	1.63
pro-leu-met	0.81	12	5	4	98.99	132.8	2.09	-2.32	0.21	0.24	1.52

						3					
pro-met-phe	0.53	12	5	4	109.06	132.83	2.18	-1.68	0.41	0.69	2.05
pro-phe-ser	0.47	10	6	5	93.02	127.76	0.88	-3.37	-1.35	-0.57	0.54
pro-ser-lys	0.5	13	6	6	120.84	149.34	1.09	-2	0.26	-0.1	2.1
pro-lys-trp	0.5	13	6	6	120.84	149.34	1.09	-2	0.26	-0.1	2.1
pro-trp-tyr	0.32	11	6	6	130.22	143.55	1.25	-0.75	1.08	0.58	2.74
pro-tyr-val	0.53	10	6	5	103.49	127.76	1.88	-1.71	0.02	0.17	1.31
pro-val-ala	0.77	8	5	4	76.98	107.53	1.33	-3.17	-0.91	-0.53	0.17

Table 3: It represents Consensus Log P, ESOL Log S, ESOL Solubility (mg/ml), ESOL Solubility (mol/I), ESOL Class, Ali Log S, Ali Solubility (mg/ml).

Tripeptides	Consensus Log P	ESOL Log S	ESOL Solubility (mg/ml)	ESOL Solubility (mol/l)	ESOL Class	Ali Log S	Ali Solubility (mg/ml)
Pro-arg-asp	-2.62	2.62	162000.0000	420.0000	Highly soluble	2.64	170000.0000
Pro-Ala-arg	-1.96	2.22	56200.0000	164.0000	Highly soluble	2.52	115000.0000
pro-asp-cys	-2.01	2.05	37200.0000	112.0000	Highly soluble	2.02	34900.0000
pro-cys-glu	-1.6	1.8	21900.0000	63.1000	Highly soluble	1.65	15400.0000
pro-glu-gln	-2.29	2.46	108000.0000	291.0000	Highly soluble	2.69	181000.0000
pro-gly-his	-1.42	0.88	2340.0000	7.5700	Highly soluble	1.23	5260.0000
pro-his-ile	-0.51	0.4	917.0000	2.5100	Highly soluble	0.75	2070.0000

pro-ile-leu	0.59	-0.2	217.0000	0.6340	Very soluble	-0.11	267.0000
pro-leu-met	0.35	0.18	550.0000	1.5300	Highly soluble	0.07	419.0000
pro-met-phe	0.73	-0.59	100.0000	0.2550	Very soluble	-0.6	99.5000
pro-phe-ser	-0.77	0.6	1390.0000	3.9700	Highly soluble	1.26	6400.0000
pro-ser-lys	0.27	-0.6	108.0000	0.2510	Very soluble	-0.61	105.0000
pro-lys-trp	0.27	-0.6	108.0000	0.2510	Very soluble	-0.61	105.0000
pro-trp-tyr	0.98	-1.85	6.5900	0.0142	Very soluble	-1.79	7.5800
pro-tyr-val	0.33	-0.61	93.2000	0.2470	Very soluble	-0.46	131.0000
pro-val-ala	-0.62	0.92	2350.0000	8.2400	Highly soluble	1.48	8620.0000

Table 4: It represents Ali Solubility (mol/I), Ali Class Silicos-IT LogSw, Silicos-IT Solubitity(mg/ml), Silicos-IT Solubility (mol/I), Silicos-IT class.

Tripeptides	Ali Solubility (mol/l)	Ali Class	Silicos-IT LogSw	Silicos-IT Solubility (mg/ml)	Silicos-IT Solubility (mol/l)	Silicos-IT class
Pro-arg-asp	441.0000	Highly soluble	-0.58	100.0000	0.2600	Soluble
Pro-Ala-arg	335.0000	Highly soluble	-1.23	20.4000	0.0596	Soluble
pro-asp-cys	105.0000	Highly soluble	-0.55	94.4000	0.2830	Soluble
pro-cys-glu	44.3000	Highly soluble	-0.94	39.5000	0.1140	Soluble
pro-glu-gln	487.0000	Highly soluble	-0.81	57.7000	0.1550	Soluble
pro-gly-his	17.0000	Highly soluble	-2.38	1.2800	0.0041	Soluble
pro-his-ile	5.6700	Highly soluble	-3.22	0.2200	0.0006	Soluble

pro-ile-leu	0.7810	Very soluble	-2.7	0.6750	0.0020	Soluble
pro-leu-met	1.1700	Highly soluble	-2.77	0.6080	0.0017	Soluble
pro-met-phe	0.2530	Very soluble	-4.44	0.0143	0.0000	Moderately soluble
pro-phe-ser	18.3000	Highly soluble	-3	0.3530	0.0010	Soluble
pro-ser-lys	0.2440	Very soluble	-5.25	0.0024	0.0000	Moderately soluble
pro-lys-trp	0.2440	Very soluble	-5.25	0.0024	0.0000	Moderately soluble
pro-trp-tyr	0.0163	Very soluble	-6.3	0.0002	0.0000	Poorly soluble
pro-tyr-val	0.3470	Very soluble	-3.39	0.1530	0.0004	Soluble
pro-val-ala	30.2000	Highly soluble	-1.49	9.2800	0.0325	Soluble

Table 5: It represents GI absorption, BBB permeant, Pgp substrate, CYP1A2 inhibitor, CYP2C19 inhibitor, CYP2C9 inhibitor, CYP2D6 inhibitor, CYP3A4 inhibitor.

Tripeptides	GI absorption	BBB permeant	Pgp substrate	CYP1A2 inhibitor	CYP2C19 inhibitor	CYP2C9 inhibitor	CYP2D6 inhibitor	CYP3A4 inhibitor
Pro-arg-asp	Low	No	No	No	No	No	No	No
Pro-Ala-arg	Low	No	No	No	No	No	No	No
pro-asp-cys	Low	No	No		No	No	No	No
pro-cys-glu	Low	No	No	No	No	No	No	No
pro-glu-gln	Low	No	No	No	No	No	No	No
pro-gly-his	Low	No	No	No	Yes	No	No	No
pro-his-ile	High	No	No	No	No	No	No	No
pro-ile-leu	High	No	Yes	No	No	No	No	No
pro-leu-met	High	No	Yes	No	No	No	No	No
pro-met-phe	High	No	Yes	No	No	No	No	No
pro-phe-ser	High	No	No	No	No	No	No	No
pro-ser-lys	Low	No	No	No	No	No	No	No

pro-lys-trp	Low	No	No	No	No	No	No	No
pro-trp-tyr	Low	No	No	No	No	No	No	No
pro-tyr-val	High	No	Yes	No	No	No	No	No
pro-val-ala	High	No	Yes	No	No	No	No	No

Table 6: It represents the physicochemical properties of tripeptides.

Tripeptides	log Kp (cm/s)	Lipinski violations	Ghose violations	Veber violations	Egan violations	Muegge violations	Bioavailability Score
Pro-arg-asp	-13.17	2	1	2	1	3	0.17
Pro-Ala-arg	-12.28	1	1	2	1	3	0.55
pro-asp-cys	-12.05	0	1	1	1	2	0.11
pro-cys-glu	-11.88	0	1	2	1	2	0.11
pro-glu-gln	-12.8	2	1	2	1	3	0.11
pro-gly-his	-10.68	0	1	0	1	1	0.55
pro-his-ile	-10.69	0	1	1	1	1	0.55
pro-ile-leu	-9.55	0	0	1	0	0	0.55
pro-leu-met	-10.14	0	0	1	1	1	0.55
pro-met-phe	-9.89	0	0	1	1	0	0.55
pro-phe-ser	-10.82	0	1	0	0	1	0.55
pro-ser-lys	-10.34	1	0	2	1	1	0.55
pro-lys-trp	-10.34	1	0	2	1	1	0.55
pro-trp-tyr	-9.67	1	1	2	1	1	0.55
pro-tyr-val	-9.82	0	0	0	0	0	0.55
pro-val-ala	-10.29	0	1	0	0	1	0.55

Table 7: Represents the toxicity data of tripeptides.

Tripeptides	PAINS alerts	Brenk alerts	Leadlikeness violations	Synthetic Accessibility
Pro-arg-asp	0	2	2	4.06
Pro-Ala-arg	0	2	1	3.73
pro-asp-cys	0	1	1	3.59
pro-cys-glu	0	1	1	3.6
pro-glu-gln	0	0	2	3.59
pro-gly-his	0	0	1	3.24
pro-his-ile	0	0	2	4.09
pro-ile-leu	0	0	1	3.69
pro-leu-met	0	0	2	3.89
pro-met-phe	0	0	2	3.84
pro-phe-ser	0	0	1	3.3
pro-ser-lys	0	0	2	3.97
pro-lys-trp	0	0	2	3.97
pro-trp-tyr	0	0	2	3.96
pro-tyr-val	0	0	2	3.45
pro-val-ala	0	0	1	3.1

Table 8: It represents the docking results of smaller change tripeptides.

According to docking results pro-trp-tyr, pro-met-phe, pro-gly-his, pro-glu-gln, pro-cys-glu, Pro-Ala-arg was seen the most promising Tripeptides.

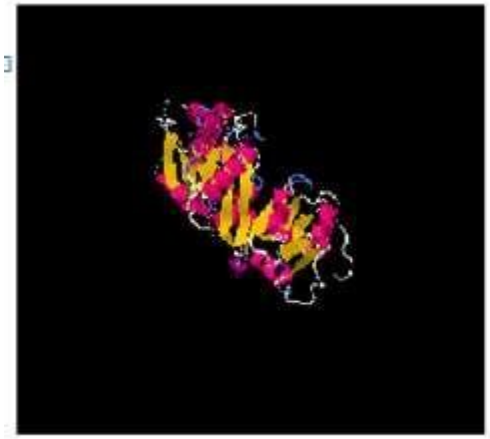
Tripeptides	Cluster	Element	Full Fitness (kcal/mol)	Estimated ΔG (kcal/mol)
pro-tyr-val	0	0	-2884.33	-7.4
pro-trp-tyr	0	0	-2875.49	-8.92
pro-lys-trp	0	0	-2886.49	-7.81
pro-ser-lys	0	0	-2885.86	-7.48
pro-phe-ser	0	0	-2866.05	-7.07
pro-met-phe	0	0	-2879.1	-8.14
pro-leu-met	0	0	-2892.57	-7.6
pro-ile-leu	0	0	-2886.03	-7.31
pro-his-ile	0	0	-2888.19	-7.75
pro-gly-his	0	0	-2896.64	-8.62
pro-glu-gln	0	0	-2933.17	-8.66
pro-cys-glu	0	0	-2907.12	-8.25
Pro-Ala-arg	0	0	-2992.81	-8.33
Pro-arg-asp	0	0	-3024.05	-7.43
pro-val-ala	0	0	-2886.75	-7.32

Images of selected Tripeptides

14

10

4



6

5

2

Note: 14=pro-trp-tyr, 10=pro-met-phe, 4=pro-cys-glu, 6=pro-gly-his, 5=pro-glu-gln, 2=Pro-Ala-arg.