

## DESIGN, IN SILICO SCREENING, AND MOLECULAR DOCKING OF A NOVEL BENZOTRIAZOLE DERIVATIVE AGAINST THE TYROSYL t-RNA SYNTHETASE OF STAPHYLOCOCCUS AUREUS

Patil Tejaswini D.<sup>1\*</sup> and Amrutkar Sunil V.<sup>2</sup>

<sup>1</sup>\*Department of Pharmaceutical Chemistry, Progressive Education Society's, Modern College of Pharmacy, Nigadi, Pune, Maharashtra- 411044.

<sup>2</sup>Department of Pharmaceutical Chemistry, Gokhale Education Society's Sir Dr. M. S. Gosavi College of Pharmaceutical Education and Research, Nashik, Maharashtra - 422005.

\*Corresponding Author: Patil Tejaswini D.

Department of Pharmaceutical Chemistry, Progressive Education Society's, Modern College of Pharmacy, Nigadi, Pune, Maharashtra - 411044.

Article Received on 25/04/2021

Article Revised on 14/05/2021

Article Accepted on 04/06/2021

### ABSTRACT

The emergence of resistance to currently available antimicrobial drugs necessitates the development of new molecules as a potential alternative. One of the most commonly used methods for designing active ligands to the fixed structure of a target protein is molecular docking. The aim of this study is to find the best docking confirmation of benzotriazole derivatives against the Aminoacyl t-RNA synthetases enzyme (PDB: 1JII) in order to predict their antimicrobial potential. In comparison to the reference ligand, BZT10 has the lowest binding energy. BZT4 and BZT8 also show considerable binding affinity towards Aminoacyl t-RNA synthetases (PDB: 1JII). In silico ADME research of benzotriazole was carried out using PreADMET online server. The study found that the pharmacokinetics were optimal, with good oral absorption and low toxicity.

**KEYWORDS:** Benzotriazole, Molecular docking, in-silico screening, antimicrobial activity, AutoDock, Aminoacyl-tRNA synthetases.

### INTRODUCTION

Computational chemistry has emerged as an important tool for the discovery and design of drug molecules in today's fast-developing era.<sup>[1,2]</sup>

CADD (Computer-aided drug design) is one of the computational tools used in personalised drug design, in which the drug molecule is docked on a receptor to check the possibility of binding of it.<sup>[3-5]</sup>

The CADD technique reduces the amount of time, money, and effort needed for drug development. Ligand-based drug design (LBDD) and structure-based drug design (SBDD) are two approaches to CADD[1,6-9]. With the availability of different receptor protein structures and known ligands with binding affinity, structure-based drug design (SBDD) is becoming a popular method.<sup>[10]</sup>

When the active site of a receptor is known, molecular docking can be used to predict which molecule would have the best binding pose and binding affinity.<sup>[11-13]</sup> Molecular docking is a technique for determining the structure of a molecule that will fit into a receptor pocket

with a high binding affinity.<sup>[14]</sup> For molecular docking, there are a number of open-source software options.<sup>[15,16]</sup>

Benzotriazole is a bicyclic compound. Benzotriazole is formed when the benzene ring is fused with a heterocyclic triazole.<sup>[17,18]</sup> It's one-of-a-kind because of its chemical properties, which include the ability to act as an electron donor and a precursor of radicals or carbanions. Condensation, addition reactions, and benzotriazolyl-alkylation are among the reactions it undergoes.<sup>[18,19]</sup> It is simple to add different groups and heterocycles to benzotriazole using this reaction, resulting in the development of a new pharmacophore. Due to its ease of synthesis and substitution into a pharmacologically active molecule, benzotriazole is a common heterocycle. It has antibacterial<sup>[20]</sup>, antiprotozoal, antiviral, antiulcer, anthelmintic, antiviral and antiproliferative characteristics.<sup>[21,22]</sup>

A lot of research has recently been done on this heterocycle to see what other biological properties it has, such as antioxidant, antiulcer, antitumor, anti-inflammatory, antimycobacterial, and antiviral agent.<sup>[16,23,24]</sup>

### Computational Method

#### Evaluation Physicochemical parameter

Online chemical property calculator Molinspiration (<http://www.molinspiration.com>) used to calculate physicochemical properties.

The PreADMET server (<http://preadmet.bmdrc.org/>) and SwissADME (<http://www.swissadme.ch>)<sup>[25]</sup> were used to predict pharmacologically relevant physicochemical properties.

#### Computational data software's:

AutoDock software 4.2, which is open source MGL tools (version 1.5.6)<sup>[26]</sup> designed by The Scripps Research Institute, is used for computer-assisted molecular simulation. The ligand molecule's structure is drawn using Chemdraw Ultra software V.12.0.2, a chemical molecule drawing tool.

The chemistry toolbox Open Babel (version 3.0.0)<sup>[27]</sup> is used to transform 2D structures into 3D structures that are dockable. The PyMol viewer tool and Discovery studio 3.5 visualizer were used to analyse docking results and visualise molecular interactions after docking (DS visualizer).

#### Target Preparation

A protein data bank (<https://www.rcsb.org>) was used to obtain the three-dimensional structure of Staphylococcus aureus Tyrosyl t-RNA synthetase (PDB: 1JJJ). PyMol [28] was used to investigate the target binding site. After deleting binding Ligand, the active site prediction and visualisation were done in the DS visualizer, and the protein molecule was saved.

In AutoDock, the protein molecule was further prepared for docking by removing the heteroatom and water molecule, as well as adding Kollman and Gasteiger charges and polar hydrogen.

### Ligand preparation

The study included 11 different benzotriazole derivatives. Chemdraw Ultra software V.12.0.2 was used to create 2D structures, which were saved in SDF format. Then, using Open Babel (version 3.0.0), it was converted to the docking-compatible file formats PDB and PDBQT.

### Molecular Docking

The population of possible conformations and orientations for the ligand at the binding site with the lowest binding energy was obtained using docking in AutoDock software 4.2. In Autodock, all ligand and protein structures were loaded.

The Lamarckian Genetic Algorithm (LGA) technique was used to determine protein fixed docking since all ligand bonds were set to be rotatable. A grid box with the dimensions of X: -11.687 Y: 17.275 Z: 91.740 Å was created to describe a docking pocket in a macromolecule, with a grid spacing of 0.375 Å, centred on X: 60 Y: 60 Z: 60 Å. After each docking, the best docking confirmation was chosen as the one with the lowest docked energy. The Biovia Discovery Studio 4.5 programme could be used to perform docking analysis in the form of 2D hydrogen-bond interactions.

### RESULTS

Physicochemical properties must be defined in order to predict the molecule's potential as a drug. The drug likeness is predicted by Lipinski's Rules.<sup>[29]</sup> Orally active drug compounds will have a molecular weight (MW) of 500 Da, an octanol-water partition coefficient (log P) of 5, a polar surface area (PSA) of 150, a number of hydrogen bond donors (HBDs) of 5, a number of hydrogen bond acceptors (HBAs) of 10, and a number of rotatable bonds (RBs) of 10.<sup>[30]</sup> Lipinski's rule, as well as hydrogen bond acceptor and donor, Log P, and TPSA, were shown in "Table 1".

**Table 1: Physicochemical Parameter and druglikeness of Benzotriazole Derivatives.**

Comp Code	Formula	MW	Rotatable bonds	H-bond acceptors	H-bond donors	TPSA	LOGP	Follow lipinski	Lipinski violations	Bioavailability Score
BZT1	C <sub>14</sub> H <sub>12</sub> N <sub>4</sub> O	252.27	4	3	1	59.81	2.16	YES	0	0.55
BZT3	C <sub>14</sub> H <sub>11</sub> ClN <sub>4</sub> O	286.72	4	3	1	59.81	2.61	YES	0	0.55
BZT2	C <sub>15</sub> H <sub>14</sub> N <sub>4</sub> O	266.3	4	3	1	59.81	2.48	YES	0	0.55
BZT4	C <sub>14</sub> H <sub>11</sub> N <sub>5</sub> O <sub>3</sub>	297.27	5	5	1	105.63	2.12	YES	0	0.55
BZT5	C <sub>16</sub> H <sub>15</sub> N <sub>5</sub> O <sub>2</sub>	309.32	6	4	2	88.91	1.38	YES	0	0.55
BZT6	C <sub>15</sub> H <sub>12</sub> N <sub>4</sub> O <sub>3</sub>	296.28	5	5	2	97.11	2.07	YES	0	0.56
BZT7	C <sub>14</sub> H <sub>11</sub> FN <sub>4</sub> O	270.26	4	4	1	59.81	2.32	YES	0	0.55
BZT8	C <sub>14</sub> H <sub>13</sub> N <sub>5</sub> O	267.29	4	3	2	85.83	1.24	YES	0	0.55
BZT9	C <sub>15</sub> H <sub>14</sub> N <sub>4</sub> O <sub>2</sub>	282.3	5	4	1	69.04	2.22	YES	0	0.55
BZT10	C <sub>14</sub> H <sub>11</sub> N <sub>5</sub> O <sub>3</sub>	297.27	5	5	1	105.63	2.07	YES	0	0.55
BZT11	C <sub>14</sub> H <sub>12</sub> N <sub>4</sub> O <sub>2</sub>	268.27	4	4	2	80.04	1.68	YES	0	0.55

Pharmacokinetic properties are evaluated in order to predict the behaviour of compounds that may one day be

used as pharmaceuticals. Drug pharmacokinetic properties such as absorption, distribution, metabolism,

and toxicity can be predicted using computational programmes such as PreADMET and SwissADME in “Table 2”.

**Table 2: Pharmacokinetic properties of Benzotriazole Derivatives.**

Comp code	BBB	Caco2++ Cell permeability	HIA	MDCK	PPB	Skin Permeability
BZT1	1.14254	20.5038	95.71427	22.4985	100	-3.48279
BZT2	0.741288	20.8373	95.75007	24.7306	100	-3.41304
BZT3	0.43395	21.672	95.98776	14.8835	99.55019	-3.52879
BZT4	0.0278888	20.9278	93.94095	5.64767	98.71858	-3.54964
BZT5	0.0153849	18.6163	94.09474	14.5328	80.33479	-3.92514
BZT6	0.230619	21.0645	95.95253	6.64531	98.53965	-3.8034
BZT7	0.327233	22.5743	95.71651	7.22047	91.96729	-3.75733
BZT8	0.207501	19.613	94.67105	21.174	79.88649	-3.81992
BZT9	0.542045	23.8975	96.11326	14.1237	90.16409	-3.69014
BZT10	0.161515	19.3246	93.94078	26.1458	97.45237	-3.52878
BZT11	0.190873	20.3284	93.5513	14.7102	88.81176	-3.9097

Oral drug absorption is predicted using the human intestinal absorption (HIA), Caco2+ cell model, and MDCK cell model.<sup>[31]</sup>

The blood-brain barrier (BBB) penetration rate indicates the expected concentration in the Central Nervous System (CNS). Plasma Protein Binding projects the drug's binding to plasma proteins. Skin permeability testing is important for transdermal drug delivery and determining skin toxicity.

The safety of a drug is also an important factor to consider when developing it.<sup>[32]</sup> The PreADMET server is used to assess ligand toxicity (“Table 3”). The Ames test predicted the mutagenicity of compounds. The carcinogenicity of a compound was tested in rats and mice to see if it had a propensity to cause cancer. Small fish, such as minnows (*Pimephales promelas*), medakas (*Oryzias latipes*), and *Daphnia*, are expected to develop acute toxicity.

**Table 3: Toxicity parameters of Benzotriazole Derivatives.**

Comp code	Algae at	Ames test	Carcino Mouse	Carcino Rat	Daphnia at	hERG inhibition	Medaka at	Minnow at
BZT1	0.0850593	mutagen	negative	negative	0.0975441	Medium risk	0.0163037	0.0236633
BZT2	0.0526432	mutagen	positive	negative	0.0732563	Medium risk	0.00978624	0.0196446
BZT3	0.0378092	mutagen	positive	negative	0.0508136	Medium risk	0.00519518	0.0114149
BZT4	0.0804334	mutagen	negative	positive	0.0673005	Medium risk	0.00883201	0.0253528
BZT5	0.0819518	mutagen	negative	negative	0.219471	Medium risk	0.0887555	0.129804
BZT6	0.0615102	mutagen	negative	negative	0.134503	Medium risk	0.0336539	0.041646
BZT7	0.0736706	mutagen	positive	positive	0.0994781	Medium risk	0.0174324	0.0189498
BZT8	0.101826	mutagen	positive	negative	0.209098	Medium risk	0.0767358	0.0980478
BZT9	0.067475	mutagen	negative	negative	0.113123	Medium risk	0.0230063	0.0399228
BZT10	0.0896459	mutagen	negative	positive	0.0758117	Medium risk	0.0110455	0.026048
BZT11	0.0716288	mutagen	negative	negative	0.134699	Medium risk	0.0322442	0.0407708

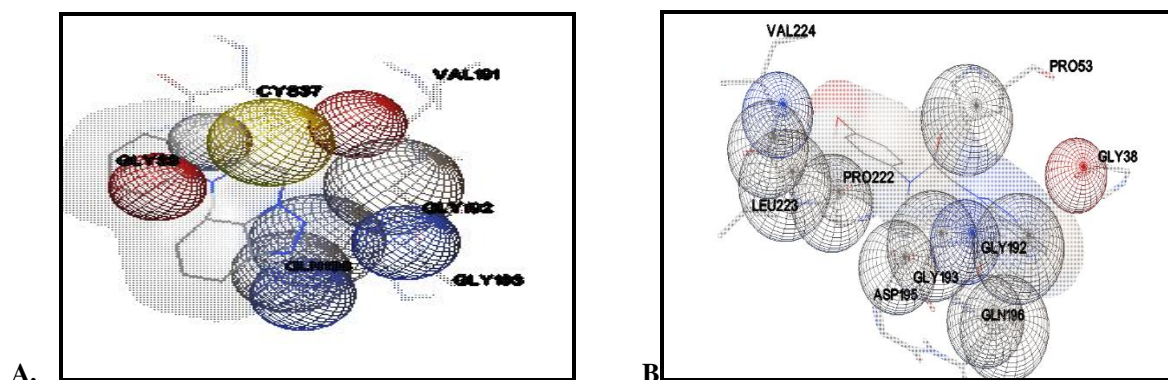
The docking scores obtained after docking in the AutoDock software are compared to find the lowest binding energy confirmation. The DS visualizer is used to determine the interaction of the best confirmation of ligand with the protein molecule. Table 4 shows amino acids that interact with ligands and the kind of interaction they have.

**Table 4: Docking score and Amino acid interaction of Benzotriazole Derivatives.**

Comp Code	Binding energy (kcal/mol)	Reference RMSD	Inhibition constant (nM)	Residue involved H-bond	Amino acid binding Residue
<b>BZT1</b>	-8.33	88.25	780.26	1 atom in H bond GLY193:HN1	CYS37, GLY38, VAL191, GLY192, GLY193, GLN 196
<b>BZT2</b>	-8.73	88.17	399.13	1 atom in H bond GLY193:HN1	CYS37, GLY38, THR75, ASN124, ASP177, VAL 191, GLY192, GLY193, ASP195, GLN196,
<b>BZT3</b>	-8.67	88.18	439.13	1 atom in H bond GLY193:HN1	CYS37, GLY38, THR75, ASN124, ASP177, GLN174, VAL 191, GLY192, GLY193, ASP195, GLN196,
<b>BZT4</b>	-9.26	86.81	162.71	3 atom in H Bond GLY38:HN1 ARG 88: HN1 LYS84:HZ1 0	GLY38, ASP40, ASP80, LYS84, ARG88, GLN196,
<b>BZT5</b>	-8.99	87.27	258.33	2 atomS in H bond ARG88: HN1	CYS37, GLY38, ASP40, TYR36, LYS84, ARG88, TYR170, GLN190, VAL 191, GLY192, GLN196
<b>BZT6</b>	-9.13	86.84	204.02	2 atoms in H bond ASP80: HN1 LYS84:HZ1 0	CYS37, GLY38, ASP40, ASP80, LYS84, ARG88, TYR170, GLN174
<b>BZT7</b>	-8.17	88.05	1.03 $\mu$ m	1 atom in H bond GLY193:HN1	CYS37, GLY38, ASN124, VAL 191, GLY192, GLY193, GLN196
<b>BZT8</b>	-9.23	87.65	171.16	3 atom in H bond GLY193:HN1 THR75: HN1 ASP177: HN1	CYS37, GLY38, TYR75, ASP177, GLN174, VAL 191, GLY192, GLY193, GLN196
<b>BZT9</b>	-8.38	87.95	729.54	1 atom in H bond GLY193:HN1	CYS37, GLY38, TYR75, ASP177, GLN174, VAL 191, GLY192, GLY193, GLN196
<b>BZT10</b>	-9.92	87.33	53.19	1 atom in H bond GLY38:HN1	CYS37, GLY38, TYR36, LEU70, THR75, LYS 84, ARG88, TYR170, GLY192, GLN196
<b>BZT11</b>	-7.74	91.03	2.11 $\mu$ m	2 atom in H bond GLY193:HN1 VAL224:HN1	GLY38, PRO53, GLY192, GLY193, ASP195, GLN196, PRO222, LEU223, VAL224
<b>Ref lig</b>	-10.36	88.28	876.54 pM	1 atom in H bond GLY193:HN1	CYS37, GLY38, TYR36, ASN124, THR75, TYR170, GLN174, GLN190, ILE200, GLN196, GLY192, GLY193

The 3D docking result of the Benzotriazole ligand with the active site of the 1jjj target protein, as well as the hydrophobic interaction pocket, is shown in Fig. 1. "Fig.2" depicts the 2D interaction of most active

derivatives of BZT10. The subsequent 2D interaction of BZT4 and BZT8 with substantial binding affinity is seen in Figures 3 and 4.



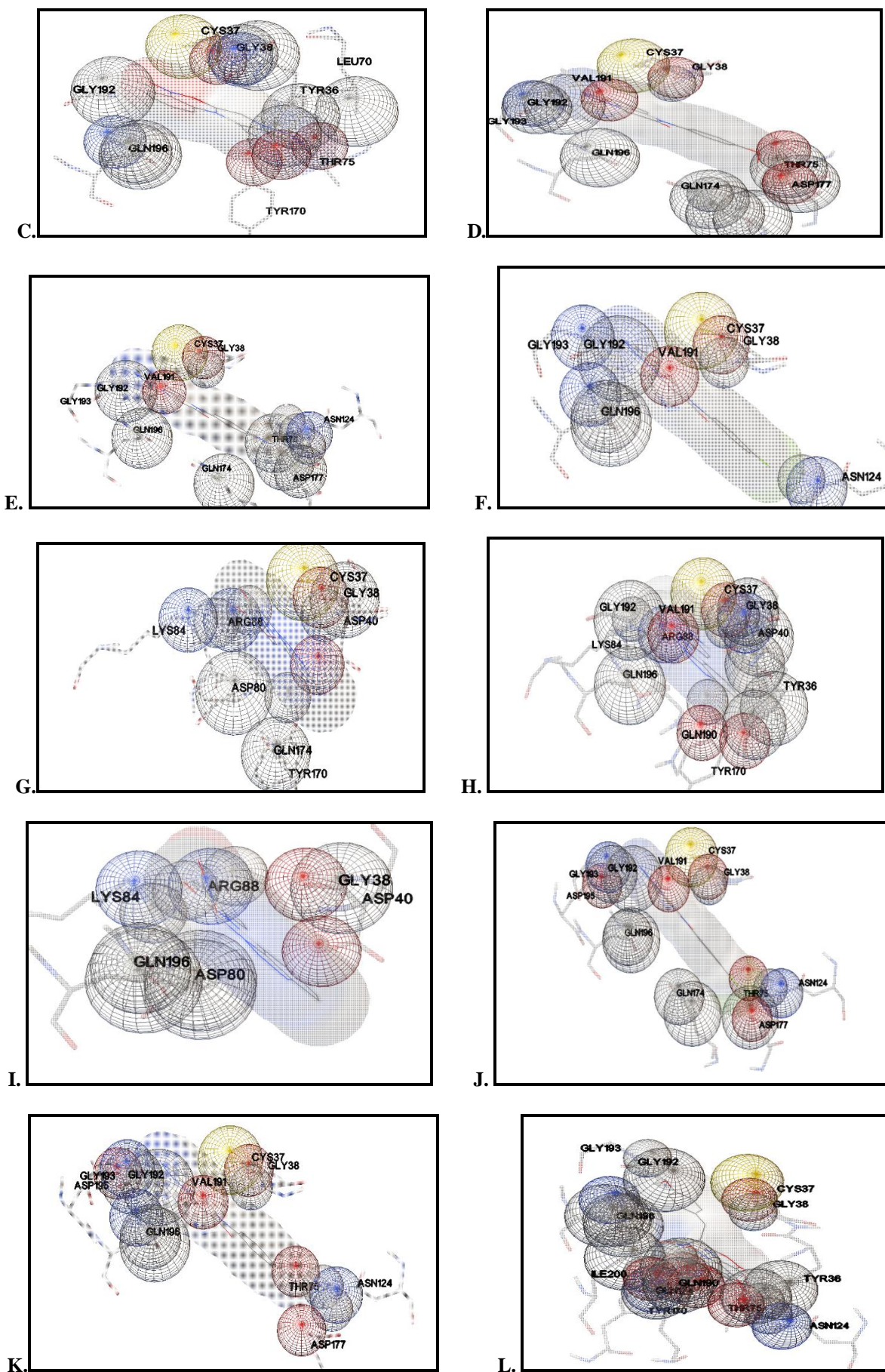


Figure 1: Interaction of Ligand BZT1 to BZT11 (A to K) and Ref Ligand with receptor 1JJJ.



## ACKNOWLEDGMENT

The author expresses gratitude to the management, principal, and faculty of Progressive Education Society's Modern College of Pharmacy and Gokhale Education Society's Sir Dr. M. S. Gosavi College of Pharmaceutical Education and Research for providing the study with the necessary facilities and support.

## REFERENCES

1. Ferreira LG, Dos Santos RN, Oliva G, Andricopulo AD. 2015. Molecular docking and structure-based drug design strategies. *Molecules*, 20(7): 13384–421.
2. Rezki N, Al-Blewi FF, Al-Sodies SA, Alnuzha AK, Messali M, Ali I, et al. 2020. Synthesis, Characterization, DNA Binding, Anticancer, and Molecular Docking Studies of Novel Imidazolium-Based Ionic Liquids with Fluorinated Phenylacetamide Tethers. *ACS Omega*, 5(10): 4807–15.
3. Shafiu S, Edache EI, Sani U, Abatyough M. 2017. Docking and Virtual Screening Studies of Tetraketone Derivatives as Tyrosine Kinase ( EGFR ) Inhibitors: A Rational Approach to Anti-Fungi Drug Design. *J Pharm Med Res*, 3(1): 78–80.
4. Pasupa K. 2019. The Review of Virtual Screening Techniques. *KMITL Inf Technol J.* (March), 1–21.
5. Meng, X. Y., Zhang, H. X., Mezei, M., & Cui M. 2011. Molecular docking: a powerful approach for structure-based drug discovery. *Current computer-aided drug design*. *Curr Comput Aided Drug Des*, 7(2): 146–57.
6. Pratyush Kumar, Asnani Alpana, Chaple Dinesh, Bais Abhinav. 2020. In silico screening and molecular docking of bioactive agents towards human coronavirus receptor. *J Pharm Res Ther*, 1(1): 41–5.
7. Chandel V, Raj S, Rathi B, Kumar D. 2020. In Silico Identification of Potent COVID-19 Main Protease Inhibitors from FDA Approved Antiviral Compounds and Active Phytochemicals through Molecular Docking: A Drug Repurposing Approach. Preprint, 7(March): 166–75.
8. Venkateshappa G, Shivaraja G, Kumar PR. 2020. Synthesis Characterization and Antimicrobial Activities Studies of N- { 2- ( 4-Chlorophenyl ) Acetyl } Amino Alcohols Derived From A-Amino Acids. *Int J Pharm Sci Drug Res*, 12(1): 11–6.
9. Kumar CJ, Vutla VR, Chapalamadugu SS, Reddy DP. 2017. Research Article Computer Aided Molecular Docking Studies on a Series of Benzothiazepines as Potential Anti Convalescent Agents. *Int J Pharm Sci Rev Res*, 47(04): 14–7.
10. Saleem H, Munir S, Mumtaz A, Ali R, Maqbool A, Malik D, et al. 2019. Identification of Potent Inhibitors against Potential Drug Target for Schizophrenia Trough Virtual Screening Approach. *Int J Sci Res Publ*, 9(6): 820–5.
11. Mishra V, Kashyap S, Hasija Y. 2015. Ligand based virtual screening for identifying potent inhibitors against viral neuraminidase: An in silico approach. *J Taibah Univ Sci*, 9(1): 20–6.
12. Sharma P, Mishra R, Vakil B. 2018. Virtual Screening and Docking Analysis of Novel lavonoidanalogues as Antipsoriaticagents. *Int J Curr Res Rev*, 10(3): 19–27.
13. Abdullahi M, Adeniji SE. 2020. In-silico Molecular Docking and ADME/Pharmacokinetic Prediction Studies of Some Novel Carboxamide Derivatives as Anti-tubercular Agents. *Chem Africa*, 3(4): 989–1000.
14. Somagond SM, Kamble RR, Kattimani PP, Joshi SD, Dixit SR. 2017. Design, synthesis, docking and in vitro antifungal study of 1,2,4-Triazole hybrids of 2-(aryloxy)quinolines. *Heterocycl Commun*, 23(4): 317–24.
15. Zeleke D, Eswaramoorthy R, Belay Z, Melaku Y. 2020. Synthesis and Antibacterial, Antioxidant, and Molecular Docking Analysis of Some Novel Quinoline Derivatives. *J Chem*, 2020; 1–16.
16. Baghel MS, Goswami K. 2015. Bcl-2 Targeted Structural Based Computer Aided Drug Design ( CAAD ) For Therapeutic Assessment of Ricin in Prostate Cancer. *Int J Pharm Sci Drug Res*, 7(2): 168–71.
17. Ren Y. 2014. Recent Development of Benzotriazole-based Medicinal Drugs. *Med Chem (Los Angeles)*, 4(9): 640–62.
18. Briguglio I, Piras S, Corona P, Gavini E, Nieddu M, Boatto G, et al. 2015. Benzotriazole: An overview on its versatile biological behavior. *Eur J Med Chem*, 97: 612–48.
19. Agha KA, Abo-Dya NE, Ibrahim TS, Abdel-Aal EH, Hegazy WA. 2016. Benzotriazole-mediated synthesis and antibacterial activity of novel n-acylcephalexins. *Sci Pharm*, 84(3): 484–96.
20. Jamkhandi CM, Disouza JI. 2012. Benzotriazole derivatives as antimicrobial agents. *Asian J Biochem Pharm Res*, 2(3): 123–30.
21. Suma B V., Natesh NN, Madhavan V. 2011. Benzotriazole in medicinal chemistry: An overview. *J Chem Pharm Res*. 3(6),375–81.
22. Parekh D V, Desai PS. 2012. Synthesis, characterization and antimicrobial activity studies of 5-(2-(5-benzoyl-1H-1,2,3-benzotriazole-1-yl)2-oxoethylamino)-2-hydroxy benzoic acid and their transition metal chelates. *Adv Appl Sci Res*, 3(4): 1992–6.
23. Singh VK, Rishishwar P, Bhardwaj P, Alok S. 2017. Benzotriazole: a Heterocyclic Molecule With Diversified Pharmacological Activities. *Int J Pharm Sci Res*, 8(2): 446–56.
24. Wan J, Peng-Cheng L V., Tian NN, Zhu HL. 2010. Facile synthesis of novel benzotriazole derivatives and their antibacterial activities. *J Chem Sci*, 122(4): 597–606.
25. Daina A, Michielin O, Zoete V. 2017. SwissADME: A free web tool to evaluate pharmacokinetics, drug-likeness and medicinal chemistry friendliness of

- small molecules. *Sci Rep*, 7(January), 1–13.
26. Allouche A. 2012. Software News and Updates Gabedit — A Graphical User Interface for Computational Chemistry Softwares. *J Comput Chem*, 32: 174–82.
  27. O’Boyle NM, Banck M, James CA, Morley C, Vandermeersch T, Hutchison GR. 2011. Open Babel. *J Cheminform*, 3(33): 1–14.
  28. DeLano WL. 2002. Pymol: An open-source molecular graphics tool. *News Protein Crystallogr*, 40(1): 82–92.
  29. Arora S, Lohiya G, Moharir K, Shah S, Yende S. 2020. Identification of Potential Flavonoid Inhibitors of the SARS-CoV-2 Main Protease 6YNQ: A Molecular Docking Study. *Digit Chinese Med*, 3(4): 239–48.
  30. Lipinski CA. 2004. Lead- and drug-like compounds: The rule-of-five revolution. *Drug Discov Today Technol*, 1(4): 337–41.
  31. Sahu M, Nerkar AG, Chikhale HU, Sawant SD. 2015. In silico screening, synthesis and pharmacological screening of quinazolinones containing oxazepinone ring as NMDA receptor antagonists for anticonvulsant activity: Part - I. *J Young Pharm*, 7(1): 21–7.
  32. Viana Nunes AM, das Chagas Pereira de Andrade F, Filgueiras LA, de Carvalho Maia OA, Cunha RLOR, Rodezno SVA, et al. 2020. preADMET analysis and clinical aspects of dogs treated with the Organotellurium compound RF07: A possible control for canine visceral leishmaniasis? *Environ Toxicol Pharmacol*. 80(August): 103470.