3rd International Conference on Scientific and Academic Research

December 25-26, 2023 : Konya, Turkey

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# DFT, Molecular Docking, Molecular Dynamics Studies of (E,E)-3-Methyl-2,5-bis(4-methylbenzylidene)cyclopentano

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***Abstract –*** In this study, structural, spectroscopic and electronic properties of ( E , E )-3-methyl-2,5-bis(4-methylbenzylidene)cyclopentanone compound, which is reported to have antibacterial activity, were investigated using density functional theory, B3LYP level and 6-31G(d,p) basis set. Vibrational wavenumbers for the carbonyl group in the molecular structure were calculated by density functional theory and compared with the experimental results. Global descriptors determining the reactivity of the compound were calculated from HOMO-LUMO molecular orbital analysis. An insight into the bioactive properties of this compound was provided by drug-likeness and ADMET studies. On the other hand, the antibacterial properties of the compound were investigated by molecular docking studies of the proten-ligand interaction mechanism at the *Staphylococcus aureus* binding site. The results of the docking studies were validated by analysing the RMSD and RMSF graphs obtained from the molecular dynamics simulation study performed for 10 ns.

*Keywords – DFT, HOMO-LUMO, Molecular Docking, ADMET, Molecular Dynamic*

## I. INTRODUCTION

*Staphylococcus aureus*, a bacterial pathogen, can proliferate on the cell surface by interacting with specific receptors on the host cell surface through specific adhesion molecules localised on the cell walls [1, 2]. One of the most important adhesins of *Staphylococcus aureus* is fibronectin-binding protein-A (FnBPA) [3-5]. Inhibition of this protein is very promising for diseases caused by bacterial pathogenicity. Recently, the development of computer technologies has brought a different dimension to multidisciplinary studies and increased the interest in in silico studies [6]. By using in silico methods in new drug designs, the cost (time-expense-hazard) in experimental studies has been greatly reduced.

In this study, Mahdi et al. the structural and electronic properties of the compound (E,E)-3-methyl-2,5-bis(4-methylbenzylidene) cyclopentanone (MBMCP), which was synthesized by and predicted to have anti-bacterial properties [7], were examined theoretically. The use of MBMCP as a drug in living organisms was investigated by drug-likeness study while other pharmacological properties of MBMCP were evaluated by ADMET predictions. The fibronectin binding protein-A receptor (PDB:4b5z) protein selected as a target to investigate the antibacterial activity of MBMCP was retrieved from the data bank. Molecular docking study was performed to elucidate the protein-ligand (P-L) interaction mechanism between MBMCP and 4m5z. Molecular dynamics simulations (MDS) were performed to confirm the P-L interactions and to evaluate the stability over a slightly longer period (10 ns). Root mean square deviation (RMSD) and root mean square fluctuation (RMSF) analyses were used to evaluate stability.

## II. Materials and Method

In this study, the structural and electronic properties of MBMCP compound were investigated by using the DFT/B3LYP method and 6-31G(d) basis set with the help of Gaussian 09 program [8]. Gaussview software [9] was used for modeling of MBMCP compound and analysis of DFT calculation results. The drug-likeness properties of MBMCP were performed using the SWISSADME web tool according to Lipinsky's five criteria. ADMET predictions of MBMCP were obtained using the pkCSM web tool. Molecular docking study for P-L interactions between MBMCP and 4m5z was performed using UCSF Chimera software and AutoDock Vina tool [10] according to standard procedure [11]. BIOVIA Discovery Studio software was used to prepare the receptor before the docking study and to evaluate the results afterwards. Molecular dynamics simulations were performed using Gromacs v2023.3 at a period of 10 ns to verify P-L interactions and stability. The topology file for MDS was prepared using Swissparam web tool [12]. RMSD and RMSF analyses were used to assess stability.

The vibrational frequency modes of the MBMCP compound were calculated by DFT. The MBMCP molecule has 45 atoms and 129 vibrational modes. No virtual values were found in the calculated 129 vibrational modes, which means that the optimization process was successful. The theoretical results were scaled by 0.9614 to ensure agreement between the theoretically calculated values and the experimental values.

## III. Results

### *DFT Calculation*

The molecular geometry corresponding to the most stable structure of the MBMCP compound was calculated theoretically with the DFT/B3LYP/6-31G(d) basis set and the 3D view of the optimized structure is presented in Figure 1a. The optimized structure of MBMCP was used in other parts of the theoretical studies. The molecular orbital energies of compounds are directly related to their reactivity. The most important of these orbitals are the highest occupied orbital (HOMO) and the lowest empty orbital (LUMO). While HOMO orbitals exhibit electron donating properties, LUMO orbitals exhibit electron acceptor behavior. The HOMO orbital is localised everywhere in MBMCP except for the carbonyl group, while the LUMO orbital is localised over the whole molecule.

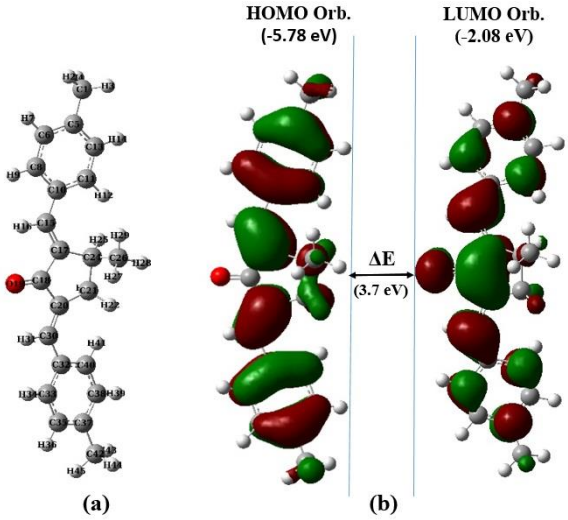


Fig.1 DFT calculation results of MBMCP, a) Optimized structure and b) 3D structure of HOMO-LUMO molecular orbitals of MBMCP

### *Drug-Likeness and ADMET Properties*

New drug designs are being made for many diseases that threaten the health of living organisms. In order to measure the effect of these designs on living organisms, different criteria have been put forward in the literature based on previous designs. One of these criteria is the criteria developed by Lipinski and called Lipinski's five rules. According to these criteria, the drug-likeness properties of MBMCP compound were evaluated and given in Table 1.

Table 1. Drug-likeness properties of MBMCP

| Lipinski's criteria | Accepted range | Value | result |
|---|---|---|---|
| Molecular Weight (Da) (MW) | ≤500 | 320.55 | ✓ |
| H-bond donors | ≤5 | 1 | ✓ |
| H-bond acceptors | ≤10 | 1 | ✓ |
| LogP | ≤5 | 6.05 | x |

In pharmacology, processes such as absorption, distribution, metabolism and toxicity are very important for new drug candidates. Newly designed drugs are evaluated through these processes before they enter the clinical trial phase. Nowadays, it is possible to model these processes mathematically by taking into account previous studies. These five processes are important in determining the bioavailability, safety and efficacy of a drug.

Table 2. ADMET analysis results obtained by using pkCSM tools.

| **Properties** | | **Properties** | |
|---|---|---|---|
| **Absorption** | | **Distribution** | |
| Water solubility (log mol/L) | -5.62 | VDss (human) (log L/kg) | 0.038 |
| Caco2 permeability (log Papp in $10^{-6}$ cm/s) | 1.22 | Fraction unbound (human) (Fu) | 0 |
| Human intestinal absorption (HIA+, %) | 92.57 | BBB permeability (log BB) | 0.743 |
| Skin Permeability (log Kp) | -2.70 | CNS permeability (log PS) | -0.785 |
| P-glycoprotein substrate | Yes | **Metabolism** | |
| P-glycoprotein I inhibitor | Yes | CYP2D6 substrate | No |
| P-glycoprotein II inhibitor | Yes | CYP3A4 substrate | Yes |
| **Toxicity** | | CYP1A2 inhibitior | No |
| AMES toxicity | No | CYP2C19 inhibitior | No |
| Max. tolerated dose (human) (log mg/kg/day) | -1.144 | CYP2C9 inhibitior | No |
| hERG I inhibitor | No | CYP2D6 inhibitior | No |
| hERG II inhibitor | No | CYP3A4 inhibitior | No |
| Oral Rat Acute Toxicity (LD50) (mol/kg) | 2.313 | **Excretion** | |
| Oral Rat Chronic Toxicity (LOAEL) (log mg/kg bw/day) | 2.072 | Total Clearance (log ml/min/kg) | 0.86 |
| Hepatotoxicity | No | Renal OCT2 substrate | No |
| Skin Sensitization | No | | |
| T. Pyriformis toxicity (log ug/L) | 1.041 | | |
| Minnow toxicity (log mM) | -0.903 | | |

ADMET predictions for MBMCP performed through the web-based online pkCSM tool are summarized in Table 2.

*Molecular Docking Study*

Molecular docking is used to predict the interactions between a target protein and a drug candidate ligand in new drug design through computer-based modeling. These studies evaluate the mechanisms of efficacy and selectivity of drugs by determining the potential binding site and how it interacts with which residues in this region. The molecular docking study between MBMCP and 4b5z receptor was performed with UCSF chimera and presented in Figure 2. 3D and 2D structures of the interactions of the P-L interaction were obtained with BIOVIA Discovery Studio visualization program and presented in Figure 2a and b.

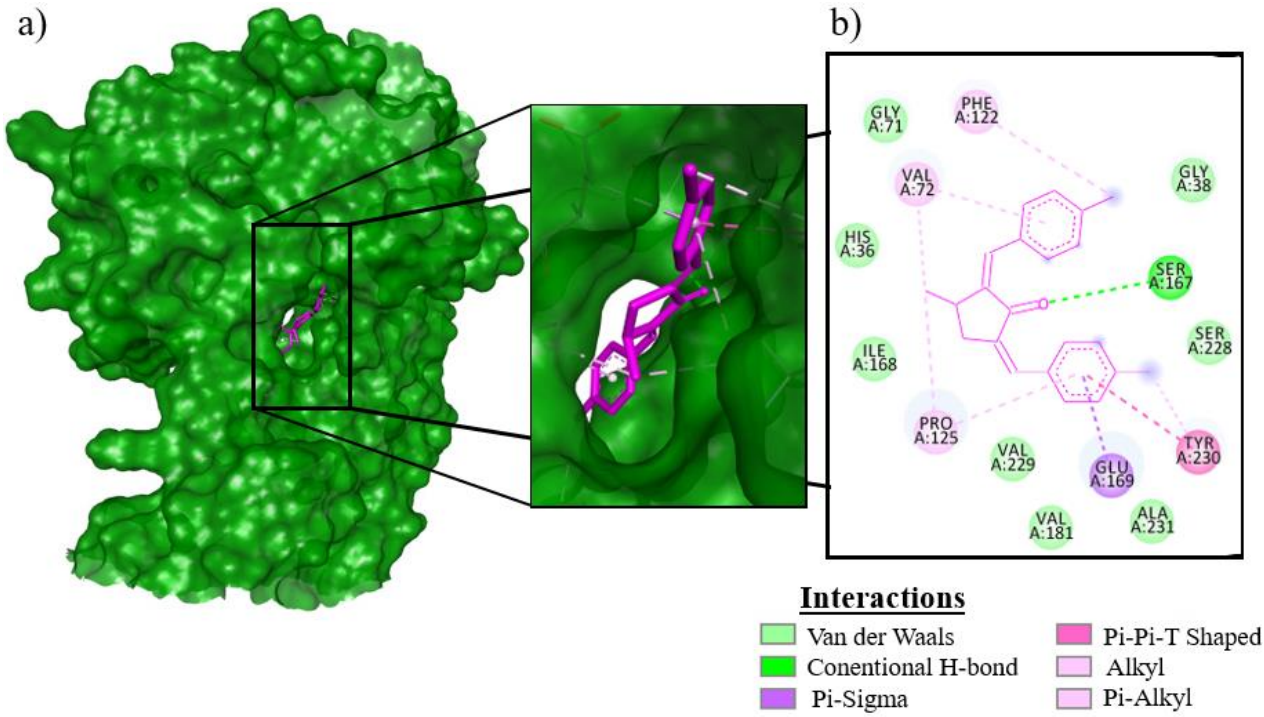


Fig. 2 P-L interactions after molecular docking study, a) 3D solid surface drawing of the interaction, (b) 2D interaction diagram of 4b5z with MBMCP.

Table 3. Summarative results of Post docking interactions between 4b5z with MBMCP

| **Protein** | **Ligands** | **ΔG (kcal/mol)** | **Hydrogen Bond interactions (Å)** | **Hydrophobic interaction (Å)** | **Van der Waals** |
|---|---|---|---|---|---|
| 4b5z | MBMCP | -8.5 | **Conventional H-Bond** Ser167(2.82) | **Pi-Pi-T shaped** Tyr230(4.62) **Pi-Sigma** Glu169(3.40) **Pi-Alkyl** Val72(4.79), Pro125(5.32), Phe122(4.87) Tyr230(4.74) | His36, Gly38, Gly71, Ile168, Val181, Ser228, Val229, Ala231, |

*Molecular Dynamic Simulation Study*

To investigate the stability of the conformation corresponding to the best binding obtained in the molecular docking study, MDS studies are performed over a slightly larger time period [13]. The basic data for the results obtained from MDS simulations are obtained by interpreting the RMSD and RMSF plots. The RMSD plot allows us to interpret how stable the ligand behaves with respect to the protein and the active site of the protein [14]. RMSF plots give information about the fluctuation of residues in the active site of the receptor. Low RMSF values mean high structural rigidity of the complexes [15]. For the analysis of the stability of MBMCP in the active cavity of the receptor, MDS study was performed with the help of Gromacs 2023.3 software and RMSD and RMSF plots are given in Figure 3a and b, respectively.

## IV. DISCUSSION

As a result of the theoretical optimization, the dihedral angles of C17-C18-C20-C21, C18-C20-C21-C24 and C20-C21-C24-C17 are found to be –

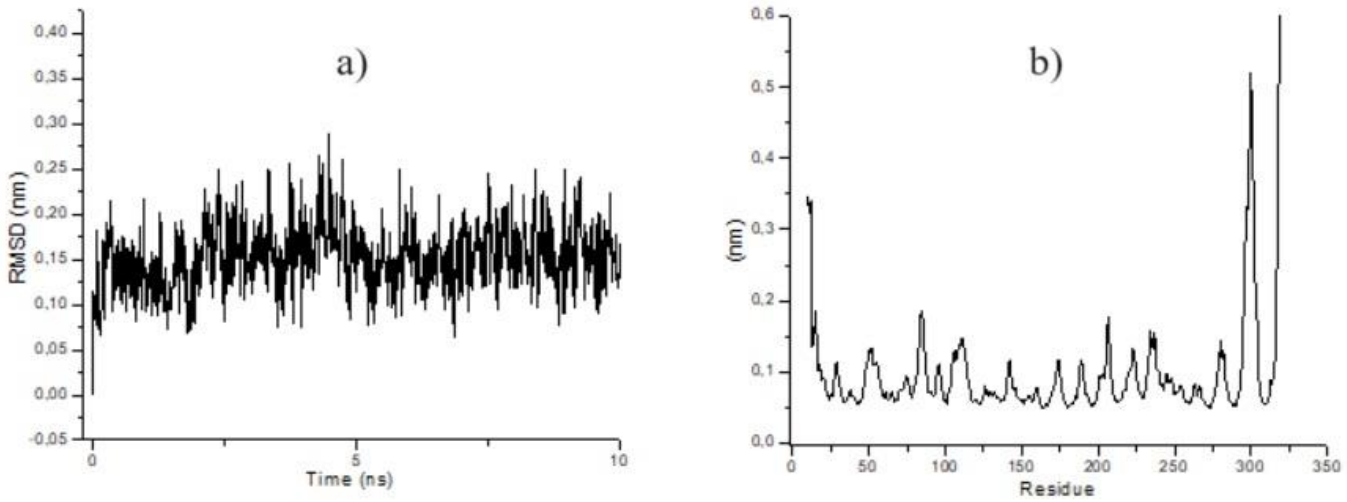


Fig.3 Graphs from the MDS study a) RMSD of protein and ligand, b) RMSF of the amino acid residues.

4.03°, 15° and -19°, respectively. These results showed that the geometric structure is non-planar, in agreement with the experimental results (). The dihedral angle between the ring plane of the C26 methyl group attached to the cyclopentanone ring (C20-C21-C24-C26) was calculated theoretically as 101° while it was 91° experimentally. Experimentally, 1 vibrational mode of C=O bond and 2 vibrational modes of C=C bond were observed at 1670, 1616 and 1596 cm -1, while the theoretical values of these vibrational modes were calculated as 1704, 1601 and 1595 $cm^{-1}$, respectively.

The energy difference between the HOMO-LUMO orbitals (ΔE) is very important as it is a descriptor of the chemical reactivity of the molecule. A high energy difference implies a high stability. In the theoretical study by DFT method, HOMO-LUMO molecular orbital energies and the energy difference between them were calculated as -5.78 eV, -2.08 eV and 3.7 eV, respectively. Based on these values, the ionization potential *(I)*, electron affinity *(A)*, chemical hardness *(η)*, chemical softness *(S)*, chemical potential *(μ)* and electronegativity *(χ)* were calculated as 8.04 eV, 2.31 eV, 2.86 eV, 0.18 $eV^{-1}$, -5.17 $eV^{-1}$ and 5.17 eV, respectively.

When the drug-likeness properties were investigated according to Lipinski criteria [16], MBMCP realized a violation. The intestinal (human) absorbance value is quite good at 92.57% as clearly seen in Table 2. In the distribution part of Table 2, the blood brain barrier (BBS) penetration and central nervous system (CNS) penetration values of MBMCP were estimated to be 0.743 and -0.785, respectively. The Ames toxicity result showed that the title compound is non-toxic.

Cytochrome P450 (CYP) enzymes are responsible for the metabolism of drugs in the body. Among these enzymes, the parameter for CYP3A4 is the most common and suggestive [17]. The term clearance refers to the rate at which the drug is excreted from the body. For the clearance value, above 15 mL/min/kg is considered high, between 5-15 mL/min/kg is considered medium and below 5 mL/min/kg is considered low [18]. According to Table 2, the excretion rate of MBMCP is low.

In the molecular docking study between P-L, the binding energy was calculated as -8.5 kcal/mol. When the molecular docking study analyzed the interactions between P-L, there was one conventional carbon hydrogen bond between the Ser167 residue and the carbonyl group of the ligand. In addition, Pi-Pi-T shaped with Tyr230 residue, Pi-Sigma with Glu169 residue, Val72, Pro125, Phe122 and Tyr230 residues interacted hydrophobically via Pi-Alkyl bonds. Most of the hydrophobic interactions occurred through aromatic rings in the laggin. It also interacted with residues His36, Gly38, Gly71, Ile168, Val181, Ser228, Val229 and Ala231 via Van der Waals bonds.

The RMSD plot in Figure 3a shows that MBMCP exhibits a stable behavior around the equilibrium point of 0.15 nm after 1 ns until the end of the simulation. The equilibrium between 0.1-0.3 nm is considered quite good for RMSD. The RMSF plot is very useful for the positional fluctuations of the residues in the active cavity of the protein. The RMSF plot plotted against the MDS trajectory reveals that the residues fluctuate around the

equilibrium point until almost the end of the simulation.

## V. CONCLUSION

In this study, the structural, spectroscopic and electronic properties of MBMCP compound, which is thought to have antibacterial activity, were theoretically investigated by DFT method. The HOMO-LUMO analysis provided information about the stability of the molecule and other global descriptors. The drug-likeness of MBMCP synthesised as a bioactive molecule was found to be a violation when evaluated according to Lipinski criteria. The AMES value in the ADMET analysis showed that there was no toxicity. Molecular docking study revealed that MBMCP travelled directly to the binding pocket of the 4b5z receptor and docked there with one hydrogen bond, 6 hydrophobic and 8 Van der Waals bond interactions. On the other hand, the stability of molecular docking was analysed by RMSD and RMSF graphs obtained from 10 ns MDS study. RMSD and RMSF showed that the ligand exhibited stable behaviour in the binding pocket. These results indicated that MBMCP may exhibit antibacterial properties.

## ACKNOWLEDGMENT

I would like to acknowledge the support provided by the project Grant No: 2013FBE013 at Pamukkale University.

## REFERENCES

[1] Telli AE, Doğruer Y: "Gıda Kaynaklı Bakteriyel Patojenlerde Patojeniteye Genel Bakış," Dicle Üniversitesi Veteriner Fakültesi Dergisi (2), 51-59 2013.

[2] Todar K: "Bacterial resistance to antibiotics (page 3)," Todar's online textbook of bacteriology, 4, 2011.

[3] Jönsson K, Signäs C, Müller Hp, Lindberg M: "Two different genes encode fibronectin binding proteins in Staphylococcus aureus: the complete nucleotide sequence and characterization of the second gene," European Journal of Biochemistry, 202(3), 1041-1048 1991.

[4] Massey RC, Kantzanou MN, Fowler T, Day NP, Schofield K, Wann ER, Berendt AR, Höök M, Peacock SJ: "Fibronectin-binding protein A of Staphylococcus aureus has multiple, substituting, binding regions that mediate adherence to fibronectin and invasion of endothelial cells," Cellular microbiology, 3(12), 839-851 2001.

[5] Stemberk V, Jones RP, Moroz O, Atkin KE, Edwards AM, Turkenburg JP, Leech AP, Massey RC, Potts JR: "Evidence for steric regulation of fibrinogen binding to Staphylococcus aureus fibronectin-binding protein A (FnBPA)," Journal of Biological Chemistry, 289(18), 12842-12851 2014.

[6] Ortega SS, Cara LCL, Salvador MK: "In silico pharmacology for a multidisciplinary drug discovery process," Drug metabolism drug interactions, 27(4), 199-207 2012.

[7] Mahdi F, Sid A, Bouchene R, Mosset P, Roisnel T: "(E, E)-3-Methyl-2, 5-bis (4-methylbenzylidene) cyclopentanone: synthesis, characterization, Hirshfeld surface analysis and antibacterial activity," Acta Crystallographica Section E: Crystallographic Communications, 75(4), 506-511 2019.

[8] *Frisch M, Trucks G, Schlegel H, Scuseria G, Robb M, Cheeseman J, Scalmani G, Barone V, Petersson G, Nakatsuji H: "Gaussian 16 Rev. C. 01, Gaussian 16 Rev C01," 2016.*

[9] Dennington R, Keith T, Millam JM: "GaussView Version 6.1, Roy Dennington, Todd A," Semichem Inc, Shawnee Mission, KS, 2016.

[10] Pettersen EF, Goddard TD, Huang CC, Couch GS, Greenblatt DM, Meng EC, Ferrin TE: "UCSF Chimera—a visualization system for exploratory research and analysis," Journal of computational chemistry, 25(13), 1605-1612 2004.

[11] Butt SS, Badshah Y, Shabbir M, Rafiq M, Biotechnology: "Molecular docking using chimera and autodock vina software for nonbioinformaticians," JMIR Bioinformatics, 1(1), e14232 2020.

[12] *Bugnon M, Goullieux M, Röhrig UF, Perez MA, Daina A, Michielin O, Zoete Vo: "SwissParam 2023: A Modern Web-Based Tool for Efficient Small Molecule Parametrization," Journal of Chemical Information, 63(21), 6469-6475 2023.*

[13] Vidal-Limon A, Aguilar-Toala JE, Liceaga AM: "Integration of molecular docking analysis and molecular dynamics simulations for studying food proteins and bioactive peptides," Journal of Agricultural Food Chemistry, 70(4), 934-943 2022.

[14] Zaki ME, Al-Hussain SA, Masand VH, Akasapu S, Bajaj SO, El-Sayed NN, Ghosh A, Lewaa I: "Identification of anti-SARS-CoV-2 compounds from food using QSAR-based virtual screening, molecular docking, and molecular dynamics simulation analysis," Pharmaceuticals, 14(4), 357 2021.

[15] Association AAP: National center for biotechnology information. pubchem compound summary for CID 12699, N-Nitroso-N-methylurea. retrieved 24. In.; 2020.

[16] Lipinski CA: "Lead-and drug-like compounds: the rule-of-five revolution," Drug discovery today: Technologies, 1(4), 337-341 2004.

[17] Zhou S-F, Xue CC, Yu X-Q, Li C, Wang G: "Clinically important drug interactions potentially involving mechanism-based inhibition of cytochrome P450 3A4 and the role of therapeutic drug monitoring," Therapeutic drug monitoring, 29(6), 687-710 2007.

[18] Loganathan T, Barathinivas A, Soorya C, Balamurugan S, Nagajothi T, Ramya S, Jayakumararaj R: "Physicochemical, Druggable, ADMET Pharmacoinformatics and Therapeutic Potentials of Azadirachtin-a Prenol Lipid (Triterpenoid) from Seed

Oil Extracts of Azadirachta indica A. Juss," *Journal of Drug Delivery Therapeutics*, 11(5), 33-46 2021.