WORLD JOURNAL OF PHARMACEUTICAL RESEARCH
SJIF Impact Factor 8.084
Volume 9, Issue 7, 2405-2412. Research Article ISSN 2277– 7105

MOLECULAR DOCKING STUDIES OF EPIFRIEDELANOL FROM GUAZUMA ULMIFOLIA AS A NATURAL ANTIHYPERLIPIDEMIC AGENTS

Danni Ramdhani* and Resmi Mustarichie

Department of Pharmaceutical Analysis and Medicinal Chemistry, Faculty of Pharmacy, Universitas Padjadjaran, Sumedang, Indonesia.

Article Received on
20 May 2020
Revised on 10 June 2020
Accepted on 30 June 2020
DOI: 10.20959/wjpr20207-17996

***Corresponding Author**
Danni Ramdhani
Department of Pharmaceutical Analysis and Medicinal Chemistry, Faculty of Pharmacy, Universitas Padjadjaran, Sumedang, Indonesia.

ABSTRACT

Objective: Natural compounds derived from plants have been known to have activity on our body's metabolism, one of which is the *Guazuma ulmifolia* (Jati Belanda plant). This plant is known empirically by the people of Indonesia has an activity to reduce obesity and LDL cholesterol value in the body. The character of natural compounds that have low toxicity, and high specificity has become the focus of research. This study aims to investigate the mechanism of antihyperlipidemic activity of Epifriedelanol compounds from *Guazuma ulmifolia* with three molecular targets; Niemann Pick C1 Like1 protein (NPC1L1), Lanosterol 14α-Demethylase (LDM), and Squalene Synthase (SqS) known to be implicated in the physiology of hyperlipidemia. The interactions of Epifriedelanol were compared with the interactions of their respective co-crystallized native ligands at the active sites of these receptors. **Materials and Methods:** Molecular docking studies began by downloading the receptor file as a target on the Protein Data Bank (PDB). The ligand structure was obtained from pubchem and zinc.docking. The receptor and ligand setup was done with discovery studio software, pyrx, MgTool, followed by docking and visualization processes using AutoDock Vina and Discovery Studio Visualizer. **Results:** Molecular docking obtained the value of binding affinity of Epifriedelanol against Niemann Pick C1 Like1 protein (NPC1L1) -2.5 kcal/mol receptors, Lanosterol 14α-Demethylase (LDM) receptor -11.2 kcal/mol, and with the Squalene Synthase (SqS) reseceptor -10.3 kcal/mol. **Conclusions**: Molecular docking studies show that epifriedelanol from the *Guazuma ulmifolia* plant has a strong

affinity and potential as an antihyperlipidemic through the mechanism of inhibiting LDM and SqS receptors.

KEYWORDS: Molecular Docking, Antihyperlipidemic, *Guazumae ulmifolia*, epifriedelanol, NPCL1, LDM, SqS.

INTRODUCTION

Hyperlipidemia is a medical condition wherein the parameters of plasma lipids are increased including triglycerides, cholesterol, cholesterol esters and phospholipids and/or plasma lipoproteins, including lipo proteins with very low density lipoprotein and low density lipoproteins, and decreased high-density lipoprotein levels. Hyperlipidemia is closely related to an increase in the value of oxidative stress which results in increased oxygen free radical production. This can cause changes in low density lipoproteins and can trigger the development of atherosclerosis and cardiovascular disease.[1,2]

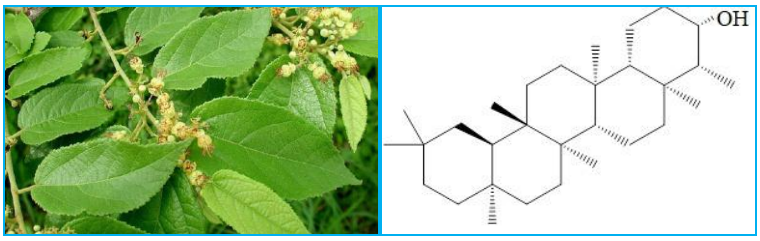

Figure 1: *Guazuma ulmifolia* and Structure of Epifriedelanol.

Molecular docking is a computational chemistry approach to helps search for molecules based on their free binding energy and proposes structural hypotheses about the analysis of ligand receptor interactions that plays an important role in identifying molecular targets (receptors) for different ligands.[3] In this study 6 selected molecular targets related to hyperlipidemia, were Niemann Pick C1 like1 protein (NPC1L1), lanosterol 14α-demethylase (LDM), and squalene synthase (SqS). Data on the crystal structure of each X-ray complex targets with each crystallized Original ligands are available from RSCB-Protein Data Bank.

The mechanism of cholesterol transport through the proximal small intestinal membrane is the main process of absorption of cholesterol into the body. This process involves transporting cholesterol across BBM by Niemann-Pick C1 Like-1 (NPC1L1). The role of NPC1L1 in intestinal sterol transfer through direct mechanism as a consequence of efforts to identify the molecular target of ezetimibe. NPC1L1 (PDB ID: 3QNT) is molecular target for cholesterol-lowering drugs ezetimibe.[4,5,6]

The cytochrome P450 enzyme plays an important role in the catalysis process of the biosynthesis of cholesterol removal processes from the l4α-methyl lanosterol group. Lanosterol 14*α*-demethylase (LDM) (PDB ID: 3LD6) inhibitors not only function as enzymatic mechanistic probes, but also as potential therapeutic agents for the treatment of hypercholesterolemia.[7,8]

Squalene synthase (SqS) has the function to catalyze the biosynthesis of squalene, as the main cholesterol precursor, through the reductive dimerization mechanism of two farnesyl diphosphate (FPP) molecules. Sqs are a key enzyme in cholesterol biosynthesis, therefore SqS is attractive as a target for therapeutic interventions for hyperlipidemia.[9,10]

MATERIALS AND METHODS

Software and Tools

Protein Data Bank (PDB), PubChem, ChemDraw Ultra 12.0, AutoDock Vina 1.1.2, MGL tools, Discovery Studio Visualizer, Pyrx.

Ligand Preparation

Epifriedelanol and various ligands (positive control) were used as ligands for docking studies were listed in Table 1.

Table 1: Ligands used in the study.

No	Ligand	Molecular Formula	References
1	Ezetimibe	$C_{24}H_{21}F_2NO_3$	[6]
2	Ketoconazole	$C_{26}H_{28}Cl_2N_4O_4$	[8]
3	INO	$C_{30}H_{31}ClN_2O_7$	[10]

Protein Preparation

Data on protein structure can be downloaded at Potein Data Bank (http://www.rcsb.org). The protein used is Niemann-Pick C1 Like-1 (NPC1L1; PDB ID: 3QNT), ATP citrate lyase (ACL; PDB ID: 3MWD), C-reactive protein (CRP; ID ID: 1B09), Lanosterol 14α-demethylase (LDM; PDB ID: 3LD6), Squalene synthase (SqS; PDB ID: 1EZF), Farnesoid X receptor (FXR; PDB ID: 1OSH). The file for each receptor in the PDB file format will first be converted to PDBQT file format using the MgL tools.

Docking Studies Using AutoDock Vina.

The energy-minimized structure of epifriedelanol and ligands (positive control) which will bind to the target protein using AutoDock Vina 1.1.2.[26] The receptor file that has been

modified and contains the charge of atoms and ligands must be changed first from the pdb format to PDBQT file format using the Open Babel program. For docking, all receptors are closed in the box, with a box spacing of 1 Å, keeping the receptors rigid and ligands as flexible molecules. The interaction energy between the ligand and receptor is calculated for all binding sites and is expressed as affinity (kcal/mol).

Protein–Ligand Interactions.s

Discovery studio visualizer is used to study interactions between protein-ligands. The docking output file is visualized in 2D interaction. Receptor ligand interactions contain informative data from intermolecular interactions, including types of hydrogen bonds, hydrophobics, and types of amino acids that interact on the active site.

RESULTS AND DISCUSSION

Ligand Preparation

The ligand structure was obtained from PubChem and zinc.docking and converted to pdb format using Open Babel. The physicochemical properties of ligands can be seen in table 2. All ligand structures are then minimized and stored in PDBQT format by the Pyrx program (Figure 2).

Table 2: Physiochemical parameters of ligand.

No	Ligand	Molecular Weight (Da)	Hydrogen Bond Donor	Hydrogen Bond Acceptor	Log P	Minimize Energy
1	Ezetimibe	409.43	2	3	4.888	977.00
2	Ketoconazole	531.44	0	7	4.206	547.92
3	INO	567.04	0	8	4.795	482.24

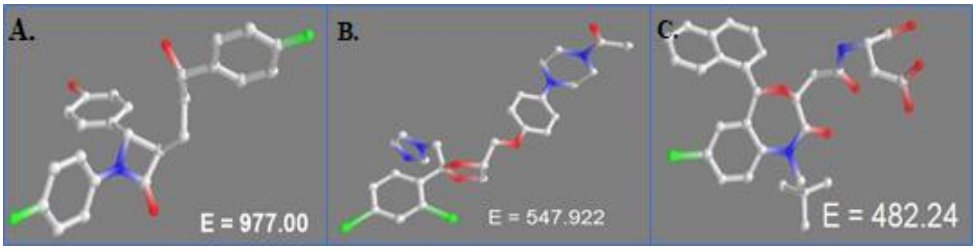

Figure 2: 3D ligand structure and energy minimized results (A) Ezetamide, (B) Ketoconazole, (C) INO.

Protein Preparation

Protein as a molecular target for antihyperlipidemia is downloaded from the Protein Data Bank and (PDB) is converted to PDBQT format using the AutoDock tool (Figure 3).

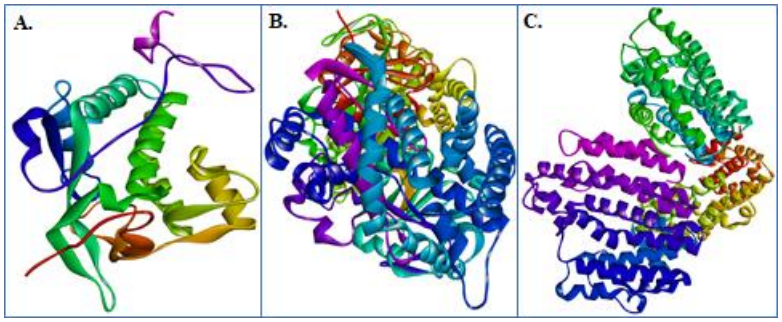

Figure 3: 3D structure of receptor, (A) Niemann-Pick C1 Like-1 (NPCL1), (B) Lanosterol 14α-Demethylase, (C) Squalene Synthase (SqS).

Docking Studies Using AutoDock Vina

Docking of Epifriedelanol into PDB structure of Niemann-Pick C1 Like-1 (PDB ID: 3QNT)

The results of molecular docking studies show that there are slight differences in the molecular bond interactions of the friedelanol and ezetimibe compounds in the same binding pocket in the NPCL1 receptor, with interactions on the amino acids Glu 238 and Gly 239 (figure 2.a and 2.b). Docking analysis by evaluating the score of binding affinity shows that Epi-friedelanol (-2.5 kcal / mol) has a lower affinity binding number compared to ezetimebe ligand (-4.9 kcal / mol). All docking observations showed that epi-fridelanol did not show antihyperlipidemic activity through a mechanism similar to ezetimibe, with an NPCL1 inhibitory mechanism.

Docking of Epifriedelanol into PDB structure of Lanosterol 14α-Demethylase (PDB ID: 3LD6).

Analysis of the interaction between the ketoconazole ligand and Epifredelanol in the same binding pocket at the Lanosterol 14α-Demethylase receptor was performed. Both ligands have the same binding interactions with amino acid residues Phe 234, Ile 379, Tyr 131 and Leu 134. Ketoconazole has hydrogen bonds with receptors on the amino acid Pro 376, and Met 487. Whereas Epifriedelanol has a pi-alkyl interaction with the amino acid Tyr 131 and Phe 234. Docing results were also evaluated through the binding affinity score where epifriedelanol had a binding affinity value (-11.2 kcal / mol) slightly greater than ketoconazole (-10.0 kcal / mol). This shows that the epifriedelanol ligand has antihyperlipidemic activity through the inhibitory mechanism of the Lanosterol 14α-Demethylase receptor.

Docking of Epifriedelanol into PDB structure of Squalene Synthase (PDB ID: 1EZF).

The results of interactions between the INO ligand and epifriedelanol on the Squalene Synthase receptor have relatively many similarities in amino acids such as the amino acids Tyr 73, Leu 76, Val 179, Pro 292, Leu 211, Ala 296 Thr 299, Arg 218, Asn 215 and Asp 80. INO ligands have hydrogen bonds with SqS receptors on Asn 215, Arg 218, and Thr 214. Whereas epifriedelanol ligands have pi-alkyl interactions on Met 295 and pi-sigma at Tyr 73. The results of binding affinity values indicate the proximity of docking scores between ligands INO (-10.2 kcal / mol) and epifriedelanol (-10.3 kcal / mol). It can be concluded that epifriedelanol ligands have the potential to have antihyperlipidemic activity through the mechanism of inhibition of Squalene Synthase receptors.

Table 4: Comparative binding affinity of different ligands with receptors.

No.	Receptor	Ligand	Binding Affinity (kcal/mol)
1	Niemann-Pick C1 Like-1 (NPCL1)	Ezetimibe Epi-friedelinol	-4.9 -2.5
2	Lanosterol14α-Demethylase (LDM)	Ketoconazole Epi-friedelinol	-10.0 -11.2
3	Squalene Synthase (SqS)	INO Epi-friedelinol	-10.2 -10.3

The interaction between the ligand and the receptor target is shown in Figure 4.

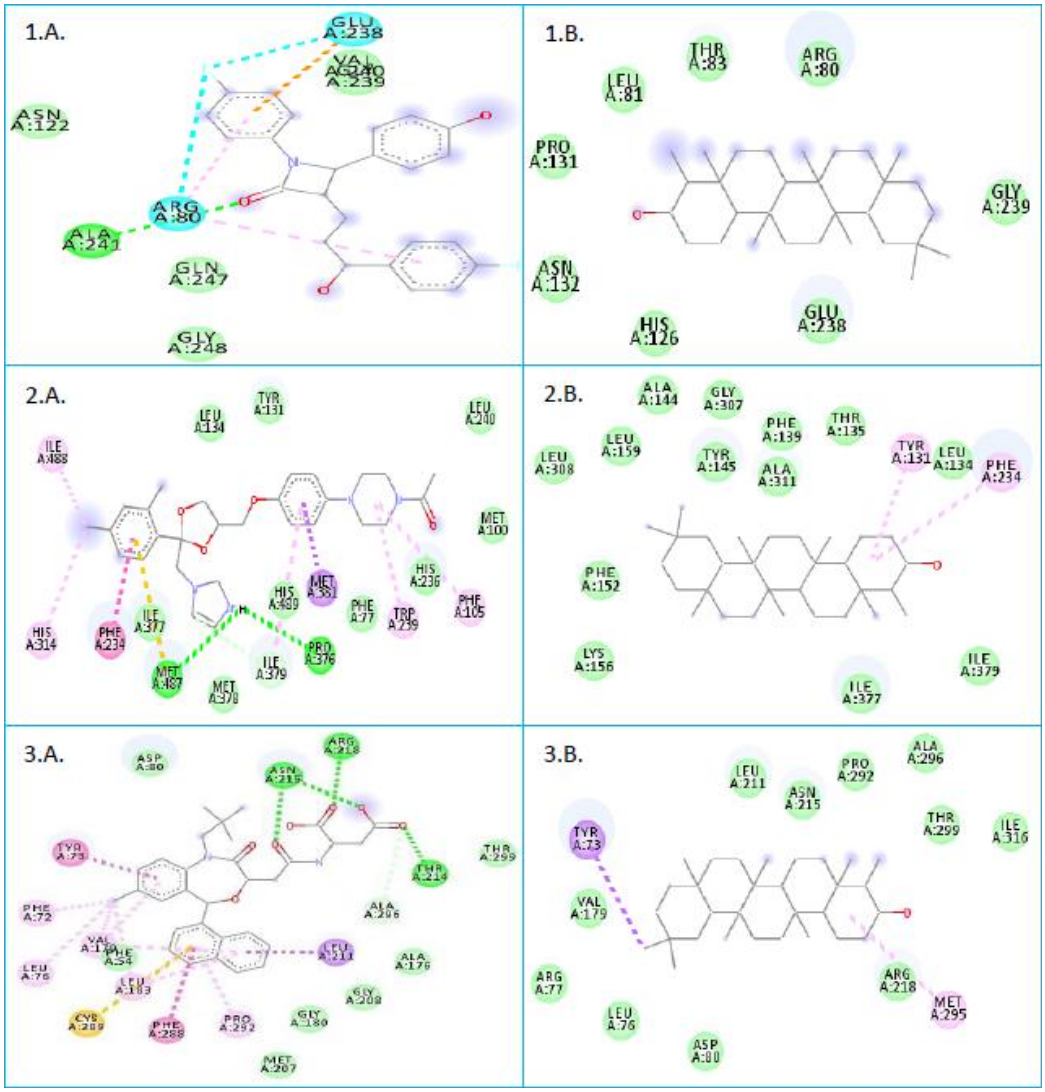

Figure 4: Interaction of ligands and target receptors. (1.A) Ezetemibe bound to NPCL1, (1.B) Epifriedelanol bound to NPCL1, (2.A) Ketoconazol bound to LDM, (2.B) Epifriedelanol bound to LDM, (3.A) INO bound to SqS, (3.B) Epifriedelanol bound to SqS.

CONCLUSIONS

Molecular docking studies of epifriedelanol compounds from the Guazuma ulmifolia plant have a strong affinity and potential as antihyperlipidemic agents through the mechanism of inhibiting LDM and SqS receptors.

REFERENCES

1. Ginghina, C., Bejan, I., Ceck, C. D. Modern risk stratification in coronary heart disease.J. Med. Life, 2011; 4(4): 377-86.

2. Jorgensen, T., Capewell, S., Prescott, E., Allender, S., Sans, S., Zdrojewski, T. Population-level changes to promote cardiovascular health. Eur. J. Prev. Cardiol., 2013; 20(3): 409-21.
3. Diller DJ, Merz KM Jr. High throughput docking for library design and library prioritization. Proteins: Structure, Function, and Bioinformatics, 2001; 43(2): 113-124.
4. Kwon HJ, Palnitkar M, Deisenhofer J The structure of the NPC1L1 N-terminal domain in a closed conformation. PLoS ONE, 2011; 6: e18722.
5. Ge L, Wang J, Qi W, Miao HH, Cao J, Qu YX, Li BL, Song BL The cholesterol absorption inhibitor ezetimibe acts by blocking the sterol-induced internalizationof NPC1L1. Cell Metab, 2008; 7: 508–519.
6. Arya N, Kharjul MD, Shishoo CJ, Thakare VN, Jain KS. Eur J Med Chem. Some molecular targets for antihyperlipidemic drug research, 2014.
7. Strushkevich N, Usanov SA, Park HW Structural basis of human CYP51 inhibition by antifungal azoles. J Mol Biol., 2010; 397: 1067–1078.
8. Gibbons GF The role of cytochrome P450 in the regulation of cholesterol biosynthesis. Lipids, 2002; 37: 1163–1170.
9. Pandit J, Danley DE, Schulte GK, Mazzalupo S, Pauly TA, Hayward CM, Hamanaka ES, Thompson JF, Harwood HJ Jr Crystal structure of human squalene synthase. A key enzyme in cholesterol biosynthesis. J Biol Chem, 2000; 275: 30610–30617.
10. Nikitakis A, Kourounakis AP QSAR of substituted morpholines with antioxidant and squalene synthase inhibitory activity. Med Chem Res., 2011; 20: 566–575.