Molecular Docking Study from Lunacridine, Scopoletin and Skimmianine as Antidiabetes through α-Glucosidase Inhibitor

Adriani1,2*, Noorhamdani3, S Winarsih3 and T Ardyati4

1 Doctoral Program in Medical Science, Faculty of Medicine, Brawijaya University, Malang, Indonesia
2STKIP Pembangunan Indonesia Makassar, Indonesia
3Department of Microbiology Faculty of Medicine, Brawijaya University, Malang, Indonesia
4Department of Biology, Faculty of Mathematics and Science, Brawijaya University, Malang, Indonesia

*Corresponding author’s email: adrimarsya@gmail.com

Abstract. Diabetes is a metabolic syndrome disease characterized by hyperglycemia in patients. The use of medicinal plants for the treatment of diabetes can control blood levels because it contains anti-diabetic active substances. The medicinal plant as an anti-diabetic through inhibition of the α-glucosidase enzyme thereby reducing the absorption of glucose in the small intestine. Lunacridine, skimmianine and scopoletin are found in the Rutaceae family but there is no information about them as α-glucosidase inhibitors. The purpose of the study to determine the ability of lunacridine, skimmianine and scopoletin as inhibitors of α-glucosidase enzymes based on docking molecular studies. The research method is ligand and receptor preparation using Pymol and docking. The docking process uses Autodoc vina in Pyrx and using acarbose as controls. The docking results are visualized using Ligplot and Discovery studio software. The results showed that lunacridine, skimmianine, scopoletin interacted with α-glucosidase and various binding affinity value. The lunacridine binding affinity is close to the acarbose control and can cross cell membranes based on Lipinski rules. Lunacridine has an anti-diabetic ability through inhibition of α-glucosidase enzyme with the inhibitory value close to acarbose control

Keywords: Lunacridine, scopoletin, skimmianine, α-glucosidase inhibitor, docking

1. Introduction
Diabetes mellitus, the metabolic syndrome, is characterized by postprandial hyperglycemia which results in increased blood glucose levels. The cause of diabetes is insulin resistance or pancreatic beta-cell damage. Another trigger is the less active style and consumption of unhealthy foods can increase the prevalence of diabetes. Monitoring and controlling blood glucose levels is the most common method in the treatment of type 2 diabetes currently. One of the targets for the treatment of diabetes is the inhibition of α-glucosidase enzyme activity. α-glucosidase is an enzyme that plays a role in breaking down the 1.4 glycosidic bonds in carbohydrates into glucose so that facilitate absorption in
the small intestine [1, 2]. Inhibition of this enzyme can control blood glucose levels of diabetic patients shortly after eating.

α-glucosidase inhibitors are produced by microorganisms and medicinal plants, one of which is from Rutaceae family. Previous phytochemical research on this family reported various types of compounds, including lunacridine [3], skimmianine [4] steroid, saponin[5], scopoletin[6] and polyphenol [5,7]. Lunacridine, skimmianine, and scopoletin have an anticancer effect [8-10] but its potential as an α-glucosidase inhibitor is unknown.

Screening of active compound bioactivity from natural materials based on molecular docking is widely used. Molecular docking is a method of exploring new compounds that have the potential to be drug candidates based on bioinformatics modeling. In general, molecular docking is used to determine the interaction of ligands and receptors and the energy produced by these interactions. This method considered efficient because it provides fast results and does not differ greatly from laboratory results [11, 12]. The purpose of this study was to determine the activity of lunacridine, skimmianine, and scopoletin as α-glucosidase inhibitors through a molecular docking approach.

2. Methods

2.1. Receptor and ligands preparation

The 3D structure of the receptor is downloaded from the Protein Data Base (RCSB) GDP ID: 2QMJ [13] then prepared using PyMOL software. The preparation process involves removing water molecules and ligands attached and adding hydrogen atoms. The 3D structure of ligands (lunacridine, skimmianine and scopoletin) was downloaded from Pubchem, then the ligand structure was minimized using PyRx 0.8 software.

2.2. Molecular docking

The docking process begins with the preparation of ligands and receptors. the next step is setting the grid box on the active site of the receptor to get the best binding position between the ligand-receptor. Docking results are stored in PDB format while affinity binding values are saved in Microsoft Excel format. The ligand and receptor interactions formed were visualized using LigPlot +, PyMOL and Discovery studio.

2.3. Pharmacokinetics analysis

The pharmacokinetic analysis includes the test of the human intestine absorbs (HIA) and toxicity test. The ligand structure stored in the molfile format (*.mol) is inputted to the online PreADMET software (https://preadmet.bmdrc.kr/adme/), then submitted [14,15]. The ability of ligands to pass through cell membranes and interact with target proteins was analysis based on the Lipinski Rule of Five. The 3D ligand structure is inputted to the SCFBIO Server (Http://www.scfbio.ittd.res.in/software/drugdesign/lipinski.jsp) and then run to get the data [16].

3. Result and Discussion

3.1. Activity of α-glucosidase inhibitors from the active compound Rutaceae

The docking results showed that lunacridine, scopoletin, and skimmianine were able to inhibit the α-glucosidase enzyme activity. Lunacridine has the highest inhibitory activity compared to scopoletin and skimmianine (Table 1). This can be seen from the lunacridine binding affinity value of -6.1 and still under the acarbose control (-7.8). The binding affinity value shows the lowest energy used by receptors to interact with ligands. The more negative the affinity binding value is formed, the more stable the interaction ligand and receptor because the bonds formed are stronger [11,17].
Table 1. Binding affinity value, prediction of HIA and toxicity lunacridine, scopoletin and skimmianine from Rutaceae.

| Ligand      | Binding affinity value | HIA (%) | Toxicity |
|-------------|------------------------|---------|----------|
| Lunacridine | -6.1                   | 95.91   | Negative |
| Scopoletin  | -5.8                   | 93.92   | Negative |
| Skimmianine | -5.4                   | 97.89   | Negative |
| Acarbose    | -7.8                   | 0.00    | Negative |

The type and number of bonds formed between the ligand and the receptor affect the binding affinity value. Hydrogen bonds are factors that affect protein stability because it strengthens the interaction effect between proteins and ligands [18]. The more hydrogen bonds, the binding affinity value is higher. Lunacridine interacts with 2 amino acids namely Asp 203 and Asp 443 through hydrogen bonds. Skimmianine only interacts with 1 amino acid (Asp A542) and scopoletin not interact. Hydrogen bonds have more energy than hydrophobic bonds. The interaction will be stronger if the ligand interacts with key amino acid residues from α-glucosidase namely Asp A203, Asp A443, Tyr A299, Trp A406, Met A444, Phe A450, Lys A480, Asp A542, Phe A575, Gln A603 [19]. Acarbose as a controlled drug bound to Asp A542 key amino acid with a distance of 3.43Å and Asp A443.

Figure 1. Molecular interaction analysis of (a) lunacridine and (b) acarbose.
3.2. Pharmacokinetic analysis of lunacridine, skimmianine and scopoletin as drug candidate

The pharmacokinetic analysis of lunacridine, skimmianine and scopoletin using PreADMET software states that all three are well absorbed by the small intestine (Table 1). The value obtained above 70% indicates that active compounds can be absorbed high in the intestine [20]. The ability of a drug to pass through cell membranes and interact with target proteins is important in the treatment of a disease. The results of the analysis based on the Lipinski Rule of Five states that lunacridine, skimmianine, and scopoletin can pass through cell membranes. They have a molecular weight of ≤ 500 Da, the number of H-donors ≤ 5, the number of H-acceptors ≤ 10, and Log P ≤ 5 (Table 2). Acarbose as a controlled drug cannot pass through the cell membrane because it is not in any following with the provisions of the Lipinski rule of five. Ligand not in by following accordance with Lipinski rule parameter, it is most likely to cause problems when it enters the body [21].

Table 2. Lipinski rule of five (Ro5) of Lunacridine, Scopoletin and Skimmianine from Rutaceae.

| Ligand      | Molecule weight (Da) | H-donor | H-acceptor | Log P  |
|-------------|----------------------|---------|------------|--------|
| Lunacridine | 305                  | 1       | 5          | 2.436100 |
| Scopoletin  | 192                  | 1       | 4          | 1.333000 |
| Skimmianine | 259                  | 0       | 0          | 3.006799 |
| Acarbose    | 645                  | 14      | 19         | -8.564498 |

Molecular weight ≤ 500 Da, H-donor ≤ 5, H-acceptor ≤ 10, Log P ≤ 5 [22]

Ligands with a molecular weight of less than 500 Da are easier to cross cell membranes because they do not interfere with the diffusion process. The number of H-donors and H-acceptors is related to the hydrophobicity and absorption of water. Many H-donor bonds cause the solubility of ligands in water to increase so that absorbance in cell membranes is limited, thus inhibiting ligands from interacting with targets [23]. Log P is also related to ligand hydrophobicity in crossing the cell membrane. The higher the hydrophobicity of a drug the ability to exit across the cell membrane is reduced. Thus ligands will be toxic because they are longer in cells [24].

4. Conclusion

Based on the results of the study can be concluded that the compound lunacridine from the family Rutaceae is a promising candidate as an α-glucosidase inhibitor. The active compound from Rutaceae has potential as an anti-diabetic drug candidate.

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