Chapter 1

Introduction

1.1 Background

Type 2 Diabetes Mellitus (T2DM) is a major global health concern, accounting for approximately 90–95% of all diabetes cases and affecting more than 530 million individuals worldwide. In Indonesia, the prevalence is estimated at around 11.7% of the adult population, corresponding to approximately 20 million individuals, with a higher incidence among those over 40 years of age (Bacha, 2024). T2DM is characterized by insulin resistance and impaired insulin secretion from pancreatic β-cells, resulting in persistent hyperglycemia that may lead to severe long-term complications (Lu et al., 2018).

One of the major therapeutic approaches in T2DM management is controlling postprandial hyperglycemia, which refers to the rapid increase in blood glucose following carbohydrate consumption (Berbudi et al., 2020). The digestion of dietary carbohydrates is facilitated by several enzymes, including intestinal α-glucosidases that hydrolyze terminal α-glycosidic bonds in oligosaccharides and disaccharides to produce absorbable monosaccharides (Gao et al., 2022). Human intestinal maltase-glucoamylase (MGAM) is one of the major α-glucosidase enzymes located in the brush border membrane of the small intestine and plays an important role in the final stage of dietary carbohydrate digestion. Consequently, inhibition of intestinal α-glucosidase activity can delay carbohydrate breakdown and reduce the rapid elevation of blood glucose following carbohydrate consumption, thereby contributing to improved postprandial glycemic control (Lebovitz, 2023).

Synthetic α-glucosidase inhibitors such as acarbose are widely used clinically to reduce postprandial glucose elevation. However, their use is frequently associated with gastrointestinal side effects, including bloating, flatulence, and diarrhea, which may affect patient adherence to long-term therapy (Liu et al., 2022). These limitations have encouraged continued research into naturally derived

compounds that may serve as potential alternatives or lead compounds for the development of new α-glucosidase inhibitors.

The human intestinal maltase-glucoamylase (MGAM) enzyme provides a relevant molecular target for investigating potential inhibitors of intestinal α-glucosidase activity. The experimentally determined structure of the N-terminal catalytic domain of human MGAM is available in the Protein Data Bank under PDB ID 2QMJ, providing a structurally characterized model for computational investigation of ligand binding. The catalytic activity of MGAM involves key catalytic residues, including Asp443, which functions as the catalytic nucleophile, and Asp542, which acts as the catalytic acid/base residue during α-glycosidic bond hydrolysis. Therefore, the structural information available for MGAM enables molecular docking studies to investigate the predicted binding behavior of potential inhibitors within the enzyme binding region and to examine their interactions with relevant active-site and catalytic residues.

Indonesia's traditional herbal medicine, known as JAMU, represents a rich source of bioactive compounds, particularly flavonoids and other polyphenols that have demonstrated antidiabetic potential (Salehi et al., 2023). Flavonoids possess hydroxyl-rich structures that may facilitate interactions with α-glucosidase enzymes and have also been associated with antioxidant properties that may help reduce oxidative stress associated with T2DM progression (Chen et al., 2022). Therefore, computational screening of JAMU-derived flavonoids against human intestinal MGAM may facilitate the identification of naturally derived compounds with potential inhibitory activity against intestinal α-glucosidase.

1.2 Objective

Below are the objectives the research aims to:

1. To retrieve and validate the three-dimensional structure of the N-terminal catalytic domain of human intestinal maltase-glucoamylase (MGAM; PDB ID: 2QMJ) for use in molecular docking simulations of flavonoid interactions.

2. To identify selected flavonoid compounds derived from Indonesian JAMU plants using the KNAPSACK Family Database and obtain their three-dimensional structures from PubChem in SDF format.
3. To prepare and visualize the three-dimensional structure of human intestinal MGAM using PyMOL and evaluate the stereochemical quality of the protein structure using PROCHECK through Ramachandran plot analysis.
4. To perform molecular docking simulations of the prepared flavonoids against the selected binding region of human intestinal MGAM (PDB ID: 2QMJ) using PyRx and evaluate their predicted binding affinities and binding poses.
5. To perform molecular re-docking of the co-crystallized acarbose ligand into the MGAM binding site to validate the selected molecular docking protocol by comparing the predicted and experimentally observed ligand binding poses.
6. To compare the docking results of the selected flavonoids with acarbose as the positive control based on their predicted binding affinities and protein–ligand interaction profiles within the MGAM binding region.
7. To analyze the predicted protein–ligand interactions using PyMOL and BIOVIA Discovery Studio Visualizer by identifying hydrogen bonds, hydrophobic interactions, van der Waals interactions, and contacts with relevant active-site and catalytic residues of human intestinal MGAM.
8. To evaluate the drug-likeness and pharmacokinetic characteristics of the selected flavonoids using Lipinski's Rule of Five and ADME screening based on available database information.
9. To conduct molecular dynamics simulations of selected flavonoid–MGAM complexes using CABS-flex 3.0 to assess protein structural flexibility and the stability of the predicted protein–ligand complexes.

1.3 Hypothesis

Null Hypothesis (H_0): Selected Indonesian JAMU-derived flavonoids **do not exhibit** predicted binding affinities comparable to or better than acarbose toward human intestinal maltase-glucoamylase (MGAM) and **do not form** structurally stable protein–ligand complexes during molecular dynamics simulations. The selected flavonoids also **do not demonstrate** favorable drug-likeness and ADME characteristics based on the applied screening criteria.

Alternative Hypothesis (H_1): Selected Indonesian JAMU-derived flavonoids **exhibit** predicted binding affinities comparable to or better than acarbose toward human intestinal maltase-glucoamylase (MGAM) and **form** structurally stable protein–ligand complexes during molecular dynamics simulations. The selected flavonoids also **demonstrate** favorable drug-likeness and ADME characteristics based on the applied screening criteria.