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# Molecular Docking of South Sulawesi Propolis against Fructose 1,6-Bisphosphatase as a Type 2 Diabetes Mellitus Drug

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**Abstract.** Diabetes mellitus is one of the metabolic diseases, characterized by hyperglycemia, which is usually caused by endogenous glucose production through gluconeogenesis. Furthermore, fructose 1,6-bisphosphatase (FBPase), which is the last enzyme involved in gluconeogenesis, is used as inhibition target due to its relatively safe effect. In addition, It is known that propolis has shown antidiabetic activity through some sets of mechanisms due to its varied constituents. Therefore, this study aims to explore the antidiabetic activity of South Sulawesi propolis compounds against the allosteric site of FBPase (PDB ID: 3KC1) through molecular docking on Autodock Vina. The results show that 18 out of 30 propolis compounds outweigh AMP affinity. Furthermore, only two flavonoids showed 100% interaction similarity to the re-docked native ligand and AMP natural inhibition. These two compounds were Broussoflavonol F and Glyasperin A, which had docking score of -9 kcal/mol and -8.2 kcal/mol, respectively. This indicates that both compounds are capable of being used as FBPase inhibitors for the treatment of diabetes mellitus.

*Keywords:* Allosteric inhibition; Diabetes mellitus; Fructose 1,6-Bisphosphatase; Molecular docking; Propolis

## 1. Introduction

Diabetes mellitus is a world-wide metabolic disease that is characterized by hyperglycemia, which is usually caused by insulin secretion deficiency (Association, 2014; Abdillah and Suwarno, 2016). In severe hyperglycemia cases, the disease is worsened by the accompaniment of organ failures (Association, 2014; Seeberger and Rademacher, 2014). Among several classifications of the disease, type 2 diabetes mellitus (T2DM), for which insulin resistance is an additional symptom, is accounted for 90–95% of the total recorded cases. Most T2DM patients frequently go undiagnosed for many years, and the risk increases with age, obesity, and an unhealthy lifestyle (Moller, 2001; Association, 2014; Control, 2020). To date, several T2DM drugs have been developed and marketed, including thiazolidinediones and metformin groups. Unfortunately, the use of thiazolidinediones

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correlates with heart failure formation while metformin has the potential to produce lactic acidosis in its users (Singh et al., 2007; Lalau, 2010). Because hyperglycemia is a major characteristic of diabetes, recently administered therapies have worked to lower patients' blood sugar levels. Several drugs have been developed and marketed with different targets and mechanism of actions (Moller, 2001; Seeberger and Rademacher, 2014). One technique that shows a promising effect is to reducing the production of endogenous glucose in the gluconeogenesis pathway which is considered as the major contributor to high blood glucose levels (Seeberger and Rademacher, 2014).

Fructose 1,6-Bisphosphatase (FBPase) is known to be the penultimate enzyme in the gluconeogenesis pathway that catalyzes the hydrolysis of fructose 1,6-bisphosphate to fructose 6-phosphate by controlling the conversion of all substrates into glucose (Erion et al., 2005; Tsukada et al., 2009; Seeberger and Rademacher, 2014). Two reasons for choosing FBPase as an inhibition target are, that: (1) it does not directly involved in glycogenolysis, glycolysis, or the tricarboxylic acid cycle (Erion et al., 2005); and (2) the genetic deficiency of the compound in humans shows no severe anomaly in biochemical and clinical parameters (Matsuura et al., 2002; Seeberger and Rademacher, 2014). In regulating blood glucose levels, the inactive state of FBPase is naturally inhibited by AMP at the allosteric site, and by fructose 2,6-bisphosphate at the substrate part (Tsukada et al., 2009). This study focuses on the allosteric site, since its nature is not highly hydrophilic, unlike that of the substrate (Erion et al., 2005).

Propolis is a resinous material collected by honeybee from various plant, which has been preclinically proven for its variety of chemical constituent, exhibiting a wide range of biological activities, including antioxidant, antimicrobial, anti-inflammatory, and antidiabetic (Fuliang et al., 2005; Diva et al., 2019; Pratami et al., 2019). Propolis constituents include polyphenols, aromatic acids, terpenoids, steroids, and amino acids depending on its vegetation and geographical origin (Kumazawa et al., 2004; Miyata et al., 2020). Propolis has been shown to have antidiabetic properties in that it reduces the total cholesterol levels, decreases low and increases high-density lipoproteins, and regulates blood glucose levels (Fuliang et al., 2005). According to Miyata et al. (2020) there are several new compounds that have been obtained from South Sulawesi propolis through X-ray structure analysis (Miyata et al., 2020).

In modern drug discovery, virtual screening of constituents has become an important step in evaluating and reducing the number of compounds to be subjected to experimental testing (Seeliger and de Groot, 2010). There are two common methods of virtual screening in drug discovery: (1) molecular docking, which simulates small molecules to protein binding sites by assuming the receptor to be rigid and have a constant covalent length and angles, as well as a rotatable ligand bond (Trott and Olson, 2010); and (2) molecular dynamics, which evaluates every single atom during simulation. This technique, however, requires many processes and high-performance hardware (Suhartanto et al., 2018). In general, docking programs use a scoring function based on empirical free binding energies to measure conformation (Trott and Olson, 2010; Forli et al., 2016). Despite the fact that there is no scoring function that accurately measures binding affinity, due to its simplification and insufficient experimental data, fitness accuracy is reached by employing optimizers, such as those used in AutoDock (Trott and Olson, 2010; Seeberger and Rademacher, 2014).

This research aims to evaluate the antidiabetic activity of South Sulawesi propolis compounds from LC-MS/MS analysis and results published by Miyata et al. (2020) by inhibiting fructose 1,6-bisphosphatase at the allosteric site. Although there have been many molecular docking studies, the use of South Sulawesi propolis as a drug candidate for

diabetes mellitus has not been carried out. Therefore, this study is recommended as a reference for further in vitro research.

## 2. Methods

In selecting the three-dimensional crystal structure of Fructose 1,6-Bisphosphatase, several published structures were listed with complete crystallographic data, and those with $\Delta R \geq 0.05$ were eliminated. Fructose 1,6-Bisphosphatase (FBPase) complex with tricyclic inhibitor 19a (PDB ID: 3KC1) was selected as a receptor based on its resolution, completeness (from EDS), Real-Space Correlation Coefficient (RSCC), and Real-Space R-value (RSR) (Warren et al., 2012). The three dimensional crystal structure of FBPase with complex tricyclic inhibitor 19a (PDB ID: 3KC1) was obtained from Protein Data Bank (http://www.rcsb.org) in *.pdb format (Tsukada et al., 2010). The protein was loaded onto Visual Molecular Dynamics to remove water and separate bound ligands. Autodock Tools 1.5.6 was utilized to assign polar hydrogen, employ Gasteiger charges, and convert the protein into *.pdbqt format (Trott and Olson, 2010; Forli et al., 2016). Two groups of propolis compounds were obtained from Miyata et al. (2020) publication and LC-MS/MS analysis of ethanolic extract. Lipinski's Rule of Five (RO5) was utilized in ligand selection criteria by employing SwissADME (http://www.swissadme.ch/) to evaluate pharmacokinetic properties (Lipinski et al., 1997; Daina et al., 2017). The selected propolis compounds are shown in Table 1 with KM codes for those published by Miyata et al. (2020), and wr codes for those obtained from LC-MS/MS analysis. Inhibitor 19a is an inhibitor bound with FBPase in the 3KC1.pdb structure, whereas AMP is an approved drug for inhibiting FBPase according to drugbank.com (Wishart et al., 2006; Tsukada et al., 2010). Both inhibitor 19a and AMP were used as positive controls. Furthermore, both propolis compounds and controls were drawn on MarvinSketch by employing the MMFF94 force field as minimization energy and converted into 3D structures in *.pdb format. Autodock Tools 1.5.6 was used to assign polar hydrogen and to create *.pdbqt files (Trott and Olson, 2010; Forli et al., 2016).

Autodock Vina is an open-source molecular docking program that utilized the global particle swarm and Broyden-Fletcher-Goldfarb-Shanno (BFGS) optimization (Trott and Olson, 2010; Pasaribu et al., 2017). Molecular docking is a computational procedure that predicts the non-covalent binding of macromolecules (receptors) and small molecules (ligands) within a measured search space through optimization algorithm and scoring function calculation (Trott and Olson, 2010). First, redocking was performed to validate the simulation of the protein target. The Gridbox size was obtained from the largest ligand (Seeberger and Rademacher, 2014). The optimum search spaces (Gridbox) measured in $x$, $y$, and $z$-dimensions, were all 18 Å, with grid spacing adjusted to 1.0 Å. To measure validation, the PyMol 3D (Quad Buffer) was used to calculate the root-mean-square deviation of atomic positions (RMSD) (Harborne et al., 2015). The inhibitor 19a binding sites was found on the following coordinates: $x$ = 20.511, $y$ = 2.471, and $z$ = 48.728. The molecular docking was performed after an acceptable RMSD value from redocking was obtained. To interpret and compare molecular interactions between ligands and receptors, the selected molecules were analyzed using Ligplot+ for 2D visualization.

**Table 1** Accepted ligands based on Lipinski's Rule of Five criteria

| No | Formula | Code | Compound Name | Molecular Weight (g/mol) | MLogP | H Bond Donor | H Bond Acceptor |
|---|---|---|---|---|---|---|---|
| 1 | $C_{22}H_{22}O_7$ | KM01 | Sulabiroin A | 398.41 | 2.16 | 0 | 7 |
| 2 | $C_{23}H_{26}O_7$ | KM02 | Sulabiroin B | 414.45 | 1.96 | 0 | 7 |
| 3 | $C_{25}H_{38}O_7$ | KM03 | 2',3'-dihydro-3'-hydroxypapuanic acid | 450.57 | 2.07 | 3 | 7 |
| 4 | $C_{25}H_{36}O_6$ | KM04 | (–)-papuanic acid | 432.55 | 2.79 | 2 | 6 |
| 5 | $C_{23}H_{32}O_6$ | KM05 | (–)-isocalolongic acid | 404.50 | 2.37 | 2 | 6 |
| 6 | $C_{25}H_{36}O_6$ | KM06 | Isopapuanic acid | 432.55 | 2.79 | 2 | 6 |
| 7 | $C_{24}H_{32}O_6$ | KM07 | Isocalopolyanic acid | 416.51 | 2.58 | 2 | 6 |
| 8 | $C_{25}H_{26}O_7$ | KM08 | Glyasperin A | 422.47 | 2.09 | 4 | 6 |
| 9 | $C_{25}H_{26}O_7$ | KM09 | Broussoflavonol F | 422.47 | 2.09 | 4 | 6 |
| 10 | $C_{20}H_{20}O_5$ | KM10 | (2S)-5,7-dihydroxy-4'-methoxy-8-prenylflavanone | 354.40 | 2.04 | 2 | 5 |
| 11 | $C_{16}H_{12}O_7$ | KM11 | Isorhamnetin | 316.26 | -0.31 | 4 | 7 |
| 12 | $C_{15}H_{20}O_4$ | KM12 | (1'S)-2-trans,4-trans-abscisic acid | 264.32 | 1.44 | 2 | 4 |
| 13 | $C_{15}H_{20}O_4$ | KM13 | (1'S)-2-cis,4-trans-abscisic acid | 264.32 | 1.44 | 2 | 4 |
| 14 | $C_5H_{13}NO$ | wr01 | L-(+)-Valinol | 103.16 | 0.23 | 2 | 2 |
| 15 | $C_{17}H_{23}NO_3$ | wr02 | 1,2,2-Trimethyl-3-[(4-methylphenyl)carbamoyl] cyclopentanecarboxylic acid | 289.37 | 2.61 | 2 | 3 |
| 16 | $C_{17}H_{23}NO_2$ | wr03 | Linalyl anthranilate | 273.37 | 3.63 | 1 | 2 |
| 17 | $C_{20}H_{28}O_2$ | wr04 | Yucalexin B7 | 300.44 | 3.73 | 0 | 2 |
| 18 | $C_{25}H_{30}O_9$ | wr05 | Robustaol A | 474.50 | 0.57 | 5 | 9 |
| 19 | $C_{21}H_{18}N_6S$ | wr06 | 1,5-Dimethyl-4-[[(2-methyl-6-phenylthieno[2,3-d]pyrimidin-4-yl)hydrazinylidene]methyl]pyrrole-2-carbonitrile | 386.47 | 2.26 | 1 | 6 |
| 20 | $C_{25}H_{30}O_8$ | wr07 | Kadsurin | 458.50 | 2.33 | 0 | 8 |
| 21 | $C_{22}H_{31}NO_2$ | wr08 | 5-Hydroxymethyl tolterodine | 341.49 | 3.69 | 2 | 3 |
| 22 | $C_{25}H_{28}O_6$ | wr09 | Dulxanthone C | 424.49 | 2.4 | 2 | 6 |
| 23 | $C_{24}H_{34}O_4$ | wr10 | 9'-Carboxy-alpha-tocotrienol | 386.52 | 3.95 | 2 | 4 |
| 24 | $C_{15}H_{18}O_4$ | wr11 | Enokipodin D | 262.3 | 0.63 | 1 | 4 |
| 25 | $C_{21}H_{20}O_6$ | wr12 | Mollicellin H | 101.00 | 2.5 | 2 | 6 |
| 26 | $C_{25}H_{26}O_6$ | wr13 | Glyurallin B | 422.47 | 2.09 | 4 | 6 |
| 27 | $C_{21}H_{32}O_4$ | wr14 | ([8]-Paridyl acetate) | 348.48 | 3.78 | 0 | 4 |
| 28 | $C_{25}H_{26}O_6$ | wr15 | Macarangin | 422.47 | 2.09 | 4 | 6 |
| 29 | $C_{21}H_{38}O_3$ | wr16 | 3,4-Bis(octyloxy)benzaldehyde | 362.55 | 4.34 | 0 | 3 |
| 30 | $C_{25}H_{36}O_6$ | wr17 | Oleandrigenin | 432.55 | 3.05 | 2 | 6 |

## 3. Results and Discussion

### *3.1. Docking Score*

In this research, the inhibition was evaluated through molecular interaction formed between propolis and fructose 1,6-bisphosphatase at the allosteric site, in order to determine the potential antidiabetic activity of the compounds. Both propolis groups were derived from *Tetragonula biroi aff.* In South Sulawesi. First, 30 of the compounds were selected by Lipinski's Rule of Five (RO5). This method was adopted since its their physicochemical parameters were related to the compounds solubility in water and intestinal permeability (Lipinski, 2004). According to the structure selection, several published pdb files of FBPase were listed, and those with $\Delta R \geq 0.05$ were eliminated to avoid an overfit model. In order to select the FBPase structure, four facets were used as criteria, including resolution, completeness from electron density server, Real-Space Correlation Coefficient (RSCC), and Real-Space R-value (RSR) (Warren et al., 2012). The fructose 1,6-bisphosphatase PDB ID: 3KC1 was considered to be the best, since its ΔR, resolution, completeness, RSCC, and RSR were 0.034, 2.25 Å, 95.9%, 0.98, and 0.1 respectively.

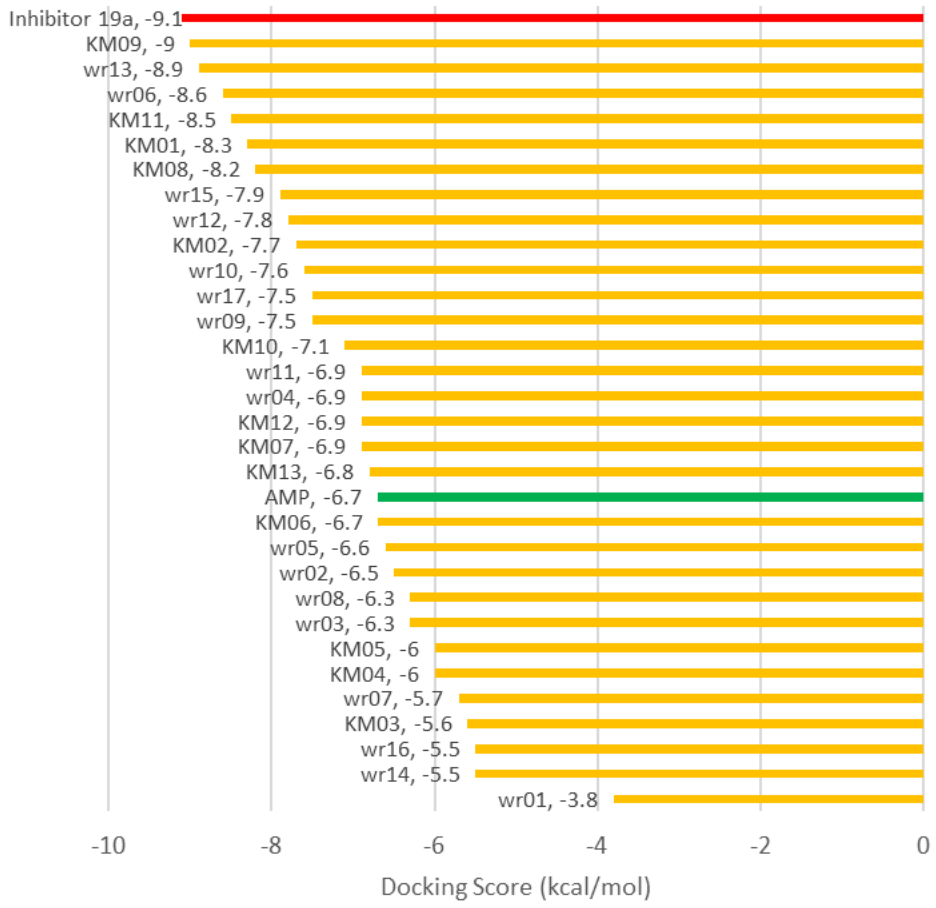


**Figure 1** Docking score of propolis compounds and the controls at allosteric site of human FBPase

In order to confirm the FBPase allosteric site, the re-docking of inhibitor 19a was conducted as a native ligand of the 3KC1.pdb structure by utilizing the rms_cur module in PyMol. The RMSD of 0.257 Å was obtained by re-docking, indicating that the confirmation was accurately acceptable. A good pose of ligand docking was achieved with an RMSD value of less than 2 Å (Marcou and Rognan, 2007). From the research by Tsukada et al. (2010), inhibitor 19a was created based on the cavity of the allosteric site at FBPase with the

addition of the amide group as an inhibition activity booster of the ligand. Furthermore, the amide group generated hydrogen bonding networks from several amino acids indicating that the half concentration of inhibitor 19a was improved to 1 nM (Tsukada et al., 2010). In this research, two positive ligand controls were used, namely inhibitor 19a and AMP, which were designed and natural, respectively.

The docking result of the compounds against the allosteric site at FBPase are presented in Figure. 1. According to this research, the direct correlation between Lipinski's parameter and the docking scores of the propolis was not determined. However, out of 30 of the compounds analyzed, 18 showed better affinity compared to AMP, although, they were lower than that of inhibitor 19a. As expected, one-third of the propolis compounds that outweighed the docking score of AMP were categorized as flavonoids, which have antidiabetic activity (Vinayagam and Xu, 2015; Ghorbani, 2017; Sarian et al., 2017). Furthermore, for an indepth exploration of this antidiabetic activity, the molecular interaction of propolis compounds at the FBPase allosteric site needs to be evaluated.

### *3.2. Molecular Interactions*

Previously, the molecular interaction of positive controls against the allosteric site of FBPase was analyzed. The results showed similar interaction at 12 residues from docking inhibitor 19a and AMP to FBPase, which included Glu20, Gly21, Ala24, Gly26, Thr27, Gly28, Glu29, Leu30, Lys112, Tyr113, Arg140, and Met177. In addition, the distinct interaction of Val-17 and Thr-31 was found in inhibitor 19a and was absent in AMP. According to Kaur et al. (2017), the natural inhibition of AMP towards the allosteric site provided a hydrogen bond formed between the phosphate group and Val17, Thr27, Gly28, Glu29, Leu30, Thr31, Lys112, Tyr113 and Arg140 whereas the purine group formed hydrophobic interactions with Val17, Glu20, Gly21, Thr31, and Met177 (Kaur et al., 2017). Based on this information, our findings were compared with the controls and the reference. Figure 2 presents the molecular interaction of inhibitor 19a and AMP at the allosteric site of FBPase.

From each group, five propolis compounds were selected that have a docking score lower than AMP to be analyzed in the molecular interaction. In the formation of hydrogen or hydrophobic bonds, all the selected propolis compounds were found to interact with Glu21, Ala24, Leu30, Arg140, and Met177. In addition, 90% of the selected propolis interacted with Gly26, 80% interacted with Thr31, and 60% interacted with Val17 and Lys112. Interestingly, all interactions formed with Thr27 were in the form of a hydrogen bond. Table 2 shows the molecular interaction formed, the similarity percentage of ligands, and the controls used.

By comparing the propolis compounds' interaction with the AMP in the allosteric site of FBPase, two compounds, namely Broussoflavonol F (KM09) and Glyasperin A (KM08), showed 100% similarity on interaction based on amino acid residues. Interestingly, propolis compounds other than these two mostly formed an interaction with Val160 and Asp178. Furthermore, KM09 and KM08 only formed an interaction with amino acid residues that were similar to those of the controls. Figure 2 presents the molecular interactions of Broussoflavonol F (KM09) and Glyasperin A (KM08) with the allosteric site. Regarding chemical classification, both KM08 and KM09 were categorized as flavonoids. It is known that flavonoid's antidiabetic activity acts differently based on its targets (Vinayagam and Xu, 2015; Sarian et al., 2017). One of the sites of action was fructose 1,6-bisphosphatase, which was reduced after reacting with the compounds (Ghorbani, 2017). Based on this finding, it is evident that the flavonoid structure may be used in an inhibitor FBPase design. Despite, there being many studies available about the antidiabetic activity of propolis, there is still limited information available about those two compounds.

**Table 2** List of molecular interaction between propolis compounds and the allosteric site of FBPase

| No | Ligands | Docking Score (kcal/mol) | Chemical Class | Hydrogen Bond | Hydrophobic Interaction | Interaction Similarity Based on Amino Acid Targets (%) | | |
|---|---|---|---|---|---|---|---|---|
| | | | | | | Inhibitor 19a | AMP | Reference of AMP |
| 1 | Reference of AMP (Kaur et al., 2017) | - | Purine nucleotides | $Val^{17}$, $Thr^{27}$, $Gly^{28}$, $Glu^{29}$, $Leu^{30}$, $Thr^{31}$, $Lys^{112}$, $Tyr^{113}$, $Arg^{140}$ | $Val^{17}$, $Glu^{20}$, $Gly^{21}$, $Thr^{31}$, $Met^{177}$ | - | - | 100 |
| 2 | AMP | -6.7 | Purine nucleotides | $Leu^{30}$, $Tyr^{113}$, $Arg^{140}$ | $Glu^{20}$, $Gly^{21}$, $Ala^{24}$, $Gly^{26}$, $Thr^{27}$, $Gly^{28}$, $Glu^{29}$, $Lys^{112}$, $Gly^{26}$ | 85.7 | 100 | 83.3 |
| 3 | wr10 | -7.6 | Prenol lipids | $Thr^{27}$, $Gly^{28}$, $Glu^{29}$, $Lys^{112}$ | $Val^{17}$, $Glu^{20}$, $Gly^{21}$, $Lys^{23}$, $Ala^{24}$, $Gly^{26}$, $Leu^{30}$, $Tyr^{113}$, $Arg^{140}$, $Met^{177}$, $Asp^{178}$ | 92.8 | 100 | 91.7 |
| 4 | KM02 | -7.7 | Aryltetralin lignans | - | $Val^{17}$, $Glu^{20}$, $Gly^{21}$, $Lys^{23}$, $Ala^{24}$, $Gly^{26}$, $Leu^{30}$, $Thr^{31}$, $Lys^{112}$, $Tyr^{113}$, $Arg^{140}$, $Met^{177}$, $Asp^{178}$, $Cys^{179}$ | 78.6 | 75 | 75 |
| 5 | wr12 | -7.8 | Depsides and depsidones | $Thr^{31}$, $Arg^{140}$, $Asp^{178}$ | $Gly^{21}$, $Ala^{24}$, $Gly^{26}$, $Gly^{28}$, $Leu^{30}$, $Tyr^{113}$, $Val^{160}$, $Met^{177}$, | 64.3 | 66.7 | 58.3 |
| 6 | wr15 | -7.9 | Flavonoids | $Thr^{27}$, $Gly^{28}$, $Glu^{29}$ | $Glu^{20}$, $Gly^{21}$, $Lys^{23}$, $Ala^{24}$, $Gly^{26}$, $Leu^{30}$, $Tyr^{113}$, $Arg^{140}$, $Met^{177}$, $Asp^{178}$, $Cys^{179}$ | 78.6 | 91.7 | 75 |
| **7** | **KM08** | **-8.2** | **Flavonoids** | **$Thr^{27}$, $Gly^{28}$, $Leu^{30}$, $Thr^{31}$, $Lys^{112}$** | **$Val^{17}$, $Glu^{20}$, $Gly^{21}$, $Ala^{24}$, $Arg^{25}$, $Gly^{26}$, $Glu^{29}$, $Tyr^{113}$, $Arg^{140}$, $Met^{177}$** | **100** | **100** | **100** |
| 8 | KM01 | -8.3 | Aryltetralin lignans | $Thr^{31}$ | $Val^{17}$, $Glu^{20}$, $Gly^{21}$, $Lys^{23}$, $Ala^{24}$, $Leu^{30}$, $Leu^{34}$, $Tyr^{113}$, $Arg^{140}$, $Val^{160}$, $Met^{177}$ | 64.3 | 58.3 | 66. 7 |
| 9 | KM11 | -8.5 | Flavonoids | $Thr^{31}$, $Asp^{178}$, $Cys^{179}$ | $Glu^{20}$, $Gly^{21}$, $Ala^{24}$, $Gly^{26}$, $Gly^{28}$, $Leu^{30}$, $Tyr^{113}$, $Arg^{140}$, $Val^{160}$, $Met^{177}$, | 71.4 | 75 | 66.7 |
| 10 | wr06 | -8.6 | Thienopyrimidines | $Arg^{140}$ | $Val^{17}$, $Glu^{20}$, $Gly^{21}$, $Ala^{24}$, $Gly^{26}$, $Leu^{30}$, $Thr^{31}$, $Lys^{112}$, $Tyr^{113}$, $Leu^{159}$, $Val^{160}$, $Met^{177}$, $Asp^{178}$ | 71.4 | 75 | 75 |
| 11 | wr13 | -8.9 | Isoflavonoids | $Thr^{27}$, $Gly^{28}$, $Glu^{29}$, $Arg^{140}$, $Val^{160}$, $Asp^{178}$, $Cys^{179}$ | $Glu^{20}$, $Gly^{21}$, $Lys^{23}$, $Ala^{24}$, $Gly^{26}$, $Leu^{30}$, $Thr^{31}$, $Lys^{112}$, $Tyr^{113}$, $Met^{177}$, | 92.8 | 100 | 91.7 |
| **12** | **KM09** | **-9.0** | **Flavonoids** | **$Thr^{27}$, $Gly^{28}$, $Glu^{29}$, $Leu^{30}$, $Thr^{31}$, $Lys^{112}$** | **$Val^{17}$, $Glu^{20}$, $Gly^{21}$, $Ala^{24}$, $Gly^{26}$, $Tyr^{113}$, $Arg^{140}$, $Met^{177}$** | **100** | **100** | **100** |
| 13 | Inhibitor 19a | -9.1 | Phenol ethers | $Thr^{27}$, $Gly^{28}$, $Leu^{30}$, $Lys^{112}$, $Tyr^{113}$ | $Val^{17}$, $Glu^{20}$, $Gly^{21}$, $Ala^{24}$, $Gly^{26}$, $Glu^{29}$, $Thr^{31}$, $Arg^{140}$, $Met^{177}$ | 100 | 100 | 100 |

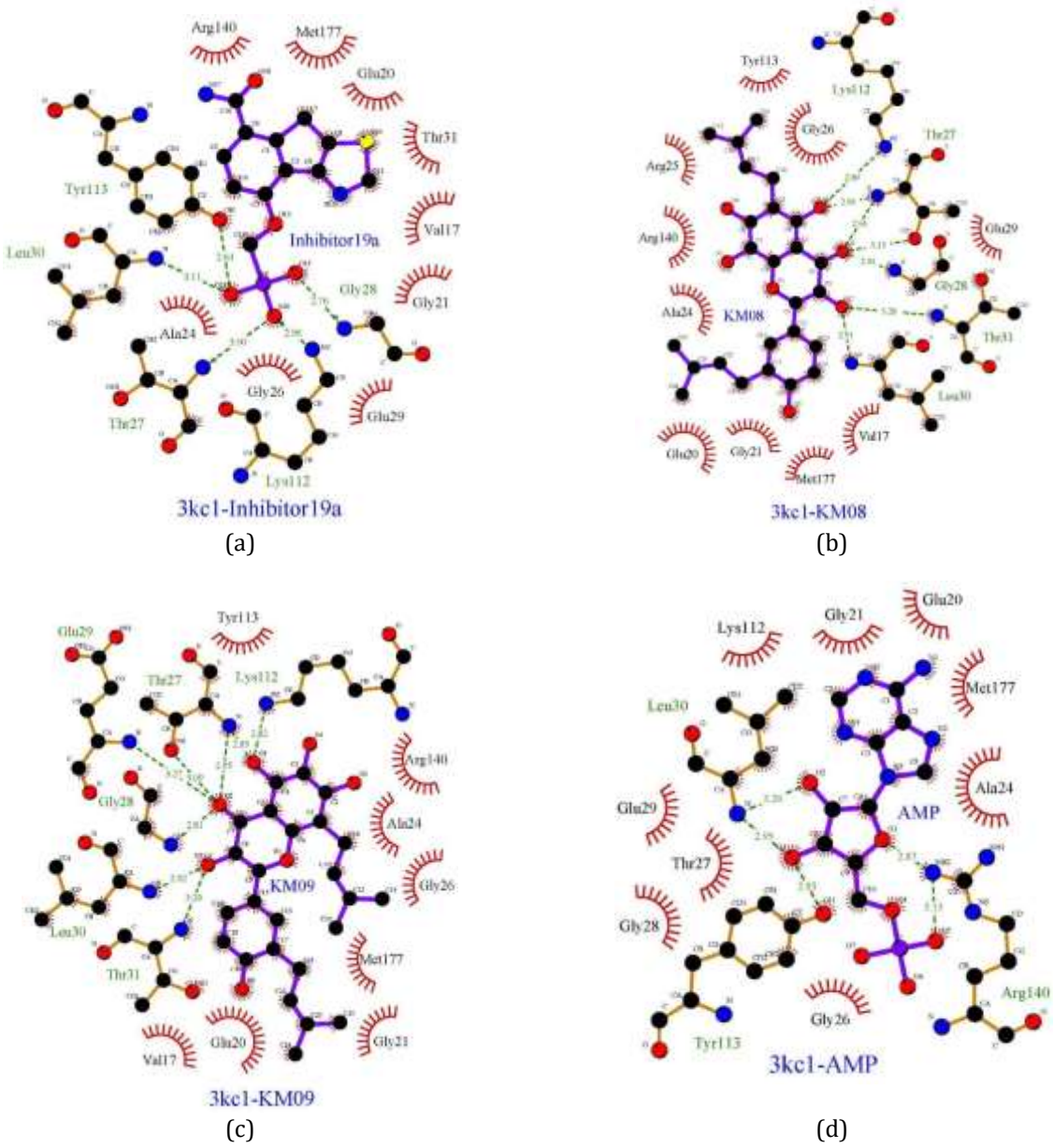


**Figure 2** Molecular interaction of: (a) inhibitor 19a; (b) KM08; (c) KM09; and (d) AMP with the allosteric site of human FBPase. The hydrogen bonds are shown in dashed green lines, while hydrophobic interactions are indicated by the half-moon red lines.

## 4. Conclusions

In this study, the in silico antidiabetic activity of South Sulawesi propolis was investigated. Among 30 selected propolis compounds, only 18 showed promising docking scores compared to AMP (-6.7 kcal/mol). Meanwhile, Broussoflavonol F and Glyasperin A showed docking scores of -9 kcal/mol and -8.2 kcal/mol, respectively, indicating 100% residue similarity in its interaction compared to the two re-docked positive controls and the AMP reference. Thus, both compounds have the potential to act against T2DM by inhibiting FBPase. Furthermore, the flavonoid structure is recommended for designing FBPase inhibitors. Finally, to ensure the validity of this finding, further research should be conducted by employing in vitro studies.

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