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RESEARCH ARTICLE

In silico analysis of quercetin, gallic acid, oleanolic acid, and ursolic acid on diabetes mellitus

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ABSTRACT
Objective: Diabetes is a disease that occurs due to pancreatic β cells failing to produce enough insulin or the inability to use the produced insulin effectively in the body. According to the data of the IDF diabetes atlas, it has been reported that there are 537 million diabetic patients aged 20-79 worldwide in 2021 and this number is expected to reach 643 million in 2030 and 783 million in 2045. To control diabetes at such a severe level, scientists are trying to find various treatment methods. One of them is natural treatments. In this study, the affinity values of quercetin, gallic acid, oleanolic acid and ursolic acid, which have antidiabetic properties, to diabetes-related renin, cathepsin-d, T-PA, leptin, MASP-2, FABP4 proteins were investigated. **Methods:** For molecular docking analysis, unwanted residues and chains were deleted from the proteins with UCSF Chimera 1.15 software and prepared for docking by adding polar hydrogen atoms. Next, quercetin, gallic acid, oleanolic acid and ursolic acid used as ligands were brought to minimum energy conformation. Protein and ligands prepared for molecular docking were analyzed with Autodock Tools 1.5.6 software. Molecular docking results were viewed with BIOVIA Discovery Studio and protein plus software. Moreover, pkCSM software was used for ADME analysis. **Conclusion:** As a result, quercetin was found to be more effective than the other three flavonoids.

Keywords: flavonoids, diabetes mellitus, ADME, molecular docking

ÖZET
Şeker hastalığı üzerinde quercetin, gallik asit, oleanolik asit ve ursolik asitin in siliko analizi
Amaç: Diyabet, pankreatik β hücrelerinin yeterli seviyede insülin üretememesin ya da üretilen insülinin vücut tarafından etkili şekilde kullanılamaması sonucu ortaya çıkan bir hastalıktır. IDF diyabet atlasının verilerine göre 2021'de dünya çapında 20-79 yaş aralığında 537 milyon diyabetli hasta olduğu ve bu sayının 2030'da 643 milyon, 2045'te ise 783 milyon olmasının beklendiği bildirilmiştir. Bu derece ciddi düzeyde olan diyabet hastalığını kontrol altına alabilmek için bilim insanları çeşitli tedavi yöntemleri bulmaya çalışmaktadır. Bunlardan biri de doğal tedavilerdir. Bu çalışmada, antidiyabetik özelliklere sahip quercetin, gallik asit, oleanolik asit ve ursolik asidin diyabetle ilişkili renin, katepsin-d, T-PA, leptin, MASP-2, FABP4 proteinlerine afinite değerleri araştırıldı. **Yöntem:** Moleküler docking analizi için öncelikle UCSF Chimera 1.15 yazılımı ile proteinlerden istenmeyen kalıntılar ve zincirler silinmiş ve polar hidrojen atomları eklenerek docking için hazırlanmıştır. Daha sonra ligand olarak kullanılan kuersetin, gallik asit, oleanolik asit ve ursolik asit minimum enerji konformasyonuna getirilmiştir. Moleküler yerleştirme için hazırlanan protein ve ligand, Autodock Tools 1.5.6 yazılımı ile analiz edildi. Moleküler yerleştirme sonuçları, BIOVIA Discovery Studio ve protein plus yazılımı ile görüntülendi. Ayrıca ADME analizi için pkCSM yazılımı kullanılmıştır. **Sonuç:** Sonuç olarak, quercetin diğer üç flavonoidden daha etkili bulunmuştur.

Anahtar kelimeler: flavonoidler, şeker hastalığı, ADME, moleküler yerleştirme

INTRODUCTION

Diabetes is a metabolic disease that results from damage to insulin secretion, insulin action, or both, and hyperglycemia occurs. Diabetes is a severe and progressive disease, and when this disease cannot be controlled, acute complications such as ketoacidosis coma, nonketotic hyperosmolar coma, lactic acidosis coma, and hypoglycemia coma, as well as retinopathy, neuropathy, nephropathy, coronary artery disease, cerebrovascular disease, peripheral artery disease, diabetic foot, hyperlipidemia, necrobiosis lipodika diabeticorum and fungal infections, causing chronic complications and adversely affecting morbidity and mortality, emerges as a significant health problem [1]. Diabetes can occur in many ways. In the type of diabetes called type 1 diabetes, insulin deficiency is observed

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because of the autoimmune destruction of pancreatic beta cells. For the kind of diabetes called type 2 diabetes, the insulin response is inadequate as a result of insulin resistance, and insulin secretion is insufficient due to beta-cell deficiency. Gestational diabetes occurs due to pregnancy-related insulin resistance [1].

The renin-angiotensin system (RAS) is a system that triggers diabetic nephropathy. Diabetic nephropathy is a disease that results in kidney failure. Recently, this problem has been tried to be solved by using molecules that block RAS [2]. In the study of Öztürk et al. using renin-angiotensin blockers to prevent diabetic nephropathy, it was found that renal survival increased [3]. High levels of leptin in obesity and type 2 diabetes are thought to hurt cardiovascular health. For treatment, leptin levels are tried to be reduced with various antidiabetic drugs [4]. In a study by Li et al., it was observed that leptin-treated aortic smooth muscle cells increased MMP-2 level, cell proliferation, and reactive oxygen species. However, when cells were treated with the drug metformin, the adverse effects caused by leptin were inhibited [5]. The cathepsin-D level is high in diabetes. Cathepsin-D causes β-cell death and dysfunction in lysosomal/autophagic cell death in response to glucolipotoxicity in type 2 diabetes [3]. High serum fatty acid-binding protein 4 (FABP4) concentration is associated with obesity, type 2 diabetes, hypertension, cardiac dysfunction, renal dysfunction, dyslipidemia, atherosclerosis, and cardiovascular events. It has been shown in experimental models that the use of FABP4 inhibitors can be a therapeutic strategy against type 2 diabetes and atherosclerosis [6]. Mannose-binding protein-associated serine protease 2 (MASP-2), which is at high levels in diabetic patients, is one of the lectin pathway molecules. Lectin pathway molecules are involved in the complement system of the immune system, and high levels of proteins involved in this system are associated with vascular complications of diabetes such as cardiovascular diseases and diabetic nephropathy [7]. Type 2 diabetes is associated with endothelial dysfunction and impaired vasodilation due to defects in endothelial-derived nitric oxide and may cause diabetes by the role of T-PA protein on vascular walls in the pathogenesis of type 2 diabetes [8].

Phytotherapy is an important field of study in the fight against diabetes and it is thought to have an important role in the prevention of diabetes-related diseases [9]. Quercetin, a member of the flavonoid family, has antioxidant, anti-inflammatory, anti-allergic, anti-ulcer, anti-cancer as well as anti-diabetic properties [10]. Quercetin has been reported to have antidiabetic potential in several preclinical studies. Therefore, the inclusion of quercetin as a natural treatment product to prevent diabetes and its complications has gained considerable attention [11]. Rifaai et al. [12] reported that the protective effect of quercetin against beta-cell damage was exerted through its antioxidant, anti-inflammatory, and anti-apoptotic activities. In addition, quercetin can cause differentiation and regeneration of islet cells by stimulating pancreatic duct stem cells [12]. Remarkably, controlling diabetes is an important step in reversing damaged hepatic and pancreatic β-cells, stabilizing blood sugar levels, and normalizing enzymatic activity. Previous studies have reported that quercetin induces an improvement in pancreatic β cells and islets of Langerhans, which have a distinct appearance [12]. They also reported that there was a persistent increase in β-cell mass, heavy staining granules and significant endocrine and exocrine, and immunohistochemical staining of beta cells showed moderate insulin antigen positivity [13]. When we look at the relationship of quercetin to the proteins to be investigated within the scope of our study, it has been shown that it blocks the renin-angiotensin pathway in the study of Parichatikan et al [14]. The study of Tan et al. with quercetin showed that the level of leptin decreased [15]. Ursolic acid is one of the main components of traditional medicinal herbs. It has anti-cancer [16], antioxidative [17], anti-inflammatory [18], anti-allergic [19], immunomodulatory effects [20]. Zhang et al. reported that ursolic acid inhibited protein tyrosine phosphatase 1B, which negatively affects the insulin signalling pathway, and stimulated glucose uptake by stimulating insulin receptor phosphorylation [21]. In a study by Ma et al., targeting the renin-angiotensin pathway with ursolic acid, it was reported that the level of angiotensin type 1-related protein decreased, and the damage of diabetic nephropathy was alleviated in mice treated with ursolic acid [22]. Obesity is known to be a disease that triggers type 2 diabetes. Jia et al. reported that ursolic acid decreased the circulating leptin level and decreased the total white adipose tissue in the body, and as a result, they revealed that ursolic acid had both hypoglycemic and hypolipidemic effects [23]. He et al. exposed 3T3-L1 cells to ursolic acid and demonstrated that FABP4 levels decreased and ursolic acid could be a therapeutic agent in the prevention of obesity [24]. Oleanolic acid has promising pharmacological activity due to its hepatoprotective, anti-inflammatory, antioxidant, anti-hyperlipidemic, anti-ulcer, anti-microbial, hypoglycemic, and anti-cancer properties [25]. It is known that diabetes is associated with hyperglycemia. Hyperglycemia may trigger diabetic nephropathy by causing endoplasmic reticulum (ER) stress and oxidative stress. In vitro study with oleanolic acid, Lee et al. reported that oleanolic acid may be a potential therapeutic for diabetic nephropathy by increasing blood insulin secretion, increasing superoxide dismutase level, and reducing ER stress [26]. In the in vivo experiment of Ahn et al. reported that renin and angiotensin involved in the renin-angiotensin pathway were suppressed in mice fed oleanolic acid [27]. Gallic acid has multiple properties such as antibacterial [28], protective against cardiovascular diseases [29], anti-inflammatory [30], anti-tumor [31] and anti-diabetic [32]. It is one of the main flavonoids used in the pharmaceutical and biomedical sectors. In the study of Huang et al. reported that gallic acid reduced hyperglycemia in rats fed a high fructose diet [32]. In the study of Garud and Kulkarni, it was reported that gallic acid inhibited the

renin-angiotensin system and improved diabetic nephropathy in type 1 diabetes [33]. In the study of Hsu and Yen, male Wistar rats were fed a high-fat diet, and obesity was induced. Subsequently, it was determined that insulin and leptin levels were decreased, and adipose tissue and oxidative stress decreased in rats fed with gallic acid for 10 weeks [34].

Within the scope of this study, the affinity value of quercetin, gallic acid, oleanolic acid, and ursolic acid to diabetes-related renin, T-PA, MASP-2, FABP4, leptin, and cathepsin-D proteins is determined by molecular docking analysis and absorption, distribution, metabolism and excretion (ADME) analysis of these flavonoids is performed, and the body is administered. effects will be analyzed.

MATERIALS and METHODS

Molecular Docking and ADME analysis have an important place in discovering molecules with drug potential. As a result of molecular docking, it provides the lowest affinity score between the molecular interactions and the ligand and receptor complex structure. On the other hand, ADME analysis tests various parameters and determines the drug candidate's absorption, distribution, metabolism, excretion, and toxicity properties. In our study, UCSF Chimera 1.15, BIOVIA Discovery Studio, Autodock Tools 1.5.6, and Protein Plus software is used for molecular docking. For molecular docking analysis, firstly, unwanted residues and chains were deleted from the proteins used in Table 1 with UCSF Chimera 1.15 software and prepared for docking by adding polar hydrogen atoms. Next, quercetin, gallic acid, oleanolic acid and ursolic acid used as ligands were brought to minimum energy conformation. Protein and ligand prepared for molecular docking were analyzed with Autodock Tools 1.5.6 software. Molecular docking results were viewed with BIOVIA Discovery Studio and protein plus software. Moreover, pkCSM software was used for ADME analysis.

RESULTS

Molecular docking is a computational procedure that predicts the non-covalent binding of a macromolecule (receptor) and a small molecule (ligand). As a result of molecular docking, the receptor and ligand's binding compatibility and affinity are predicted. In this way, it is of great importance in the drug development process by predicting the protein binding properties of small molecules [35]. For example, Table 2 shows the molecular docking results of quercetin ligand with diabetes-associated proteins in order from highest binding affinity to lowest binding affinity. According to these results, while quercetin binds to the MASP2 protein with the highest affinity, it binds to the Leptin protein with the lowest affinity.

In Figure 1, the binding status of the quercetin molecule to the proteins associated with diabetes is shown by the molecular docking results in Table 2.

Quercetin with amino acids ILE56, HIS98, GLU77, ARG111, ILE109, GLN100, THR79, ASP81, ALA38, PHE62, VAL30, VAL28, MET25, ARG83, TYR24, ALA80, PHE21, PRO43, TYRR33, SER58, MET45 in FABP-4 protein were found to interact (Figure 2).

Quercetin interacts with the amino acids ILE22, VAL107, LEU110, ILE19, SER111, GLN114, ASN20, ASP24, and LEU23 in the Leptin protein (Figure 2).

Quercetin has been found to interact with amino acids HIS118, SER268, PHE164, SER289, TRP290, VAL288, GLY291, SER263, SER292, LEU210, CYS295, ASN294, MET293, ARG265, GLY266, CYS264, THR101 in the MASP2 protein (Figure 2).

Quercetin interacted with the amino acids THR18, TYR230, SER229, ALA228, MET302, PHE124, THR85, GLY227, VAL36, TYR83, SER41, VAL127, ASP38, PRO118, GLN19, LEU121, ALA122 in the Renin protein (Figure 2).

Quercetin interacted with amino acids HIS44, TYR90, GLY193, SER195, ASP194, CYS191, ILE213, TRP215, GLY216, VAL227, GLY226, ALA190, ASP189, GLY220, CYS219, GLY218, LEU217 in the T-PA protein (Figure 2).

Quercetin interacts with the amino acids GLY78, GLN13, SER79, TYR77, ASP32, SER35, and GLY34 in the A subunit of the Cathepsin-D protein; at the same time, it was determined that Cathepsin-D interacts with the amino acids ASP222, GLY224, SER226, THR225, PHE117, THR116, ALA120, PHE122 in the B subunit (Figure 2).

The affinity results of gallic acid for proteins associated with diabetes are given in Table 3. According to these results, gallic acid binds to T-PA protein with higher affinity than other proteins, with an affinity value of -6.3 kcal/mol.

In Figure 3, the binding status of the gallic acid molecule to the proteins associated with diabetes is shown by the molecular docking results in Table 3.

Gallic acid with amino acids ARG131, PHE21, ALA41, ALA38, SER60, PHE62, LYS63, SER58, THR65, ALA80, and PRO43 in FABP-4 protein were found to interact (Figure 4).

Gallic acid interacts with the amino acids ILE26, PRO27, GLY28, GLY115, PHE25, ASP24, GLN118, TRP122, and ASP119 is the Leptin protein (Figure 4).

Gallic acid has been found to interact with amino acids ASP262, CYS295, GLY302, SER263, GLY291, CYS264, SER292, ARG265, SER268, HIS118, SER289, VAL288, TRP290, VAL303 in the MASP2 protein (Figure 4).

Gallic acid interacted with the amino acids SER84, THR85, TYR83, PHE119, PHE124, VAL36, GLY227, ASP38, SER41, GLY40, and ASP225 is the Renin protein (Figure 4).

Gallic acid interacted with amino acids VAL227, GLN192, SER214, HIS44, SER195, ILE213, TRP215, GLY218, CYS191, CYS219, GLY220, ASP189, GLY216, GLY226, ALA190 in the T-PA protein (Figure 4).

Gallic acid interacts with the amino acids TYR77, ASP32, GLY34, SER35, GLY78, and SER79 in the A subunit of the Cathepsin-D protein; at the same time, it

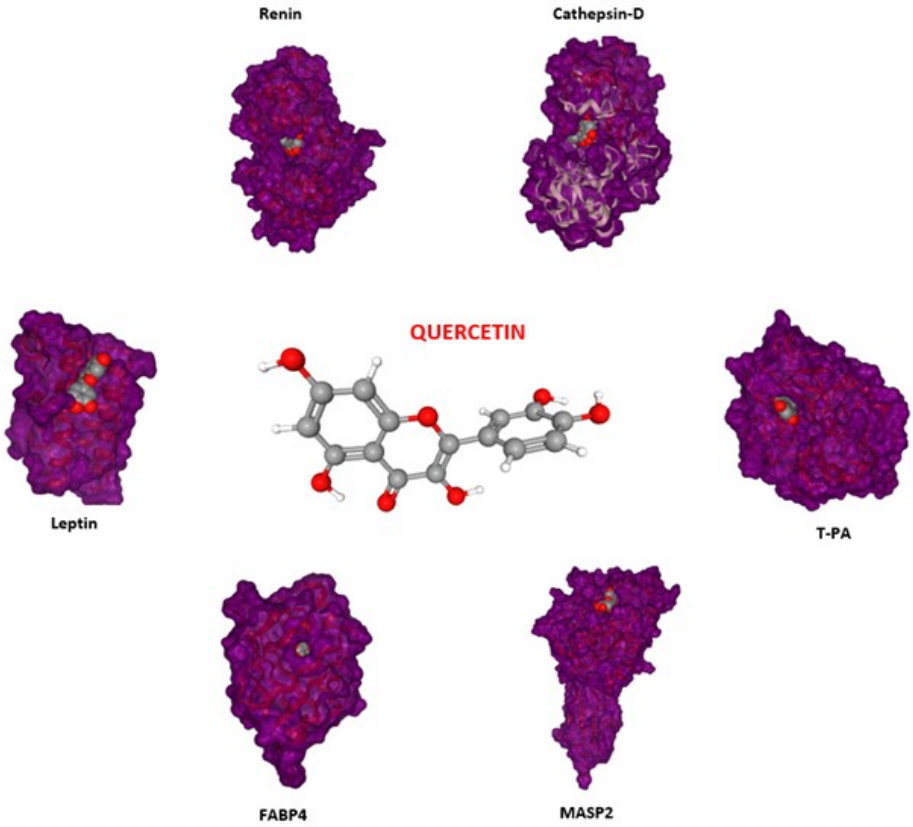

Figure 1. Molecular docking result of quercetin.

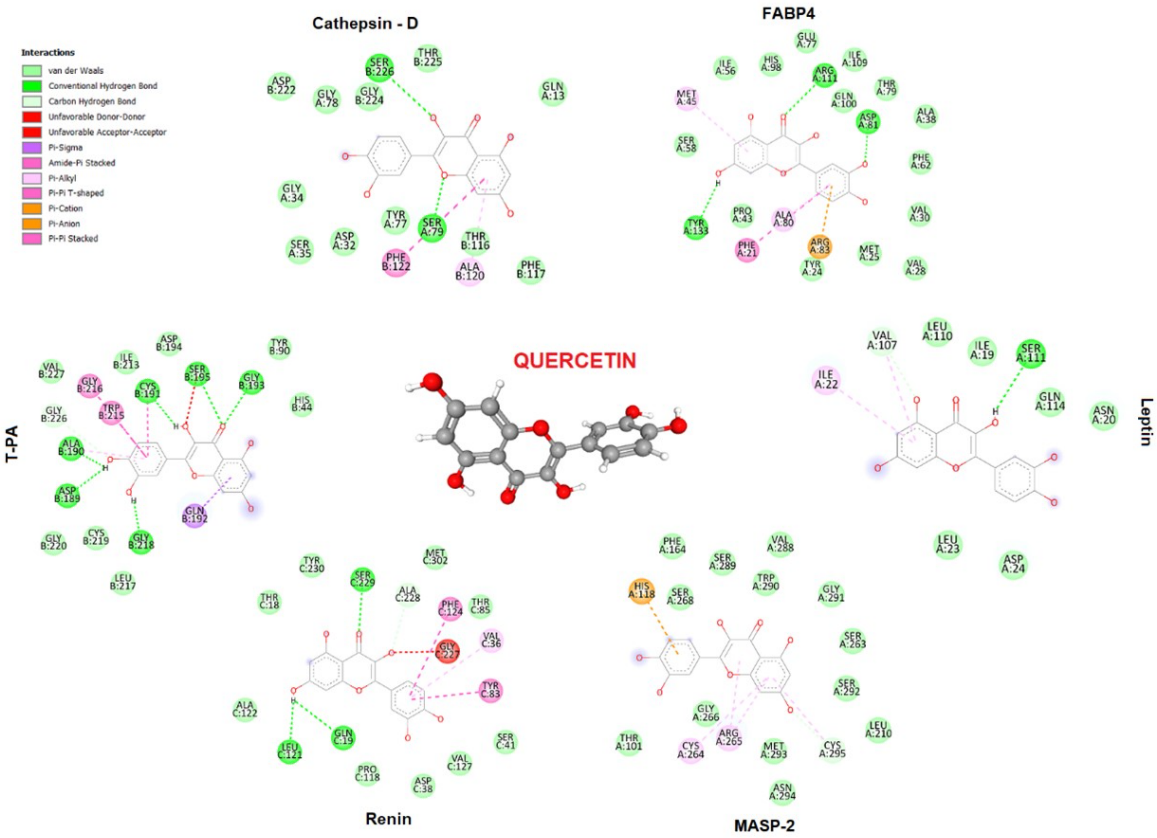

Figure 2. Protein interactions of quercetin.

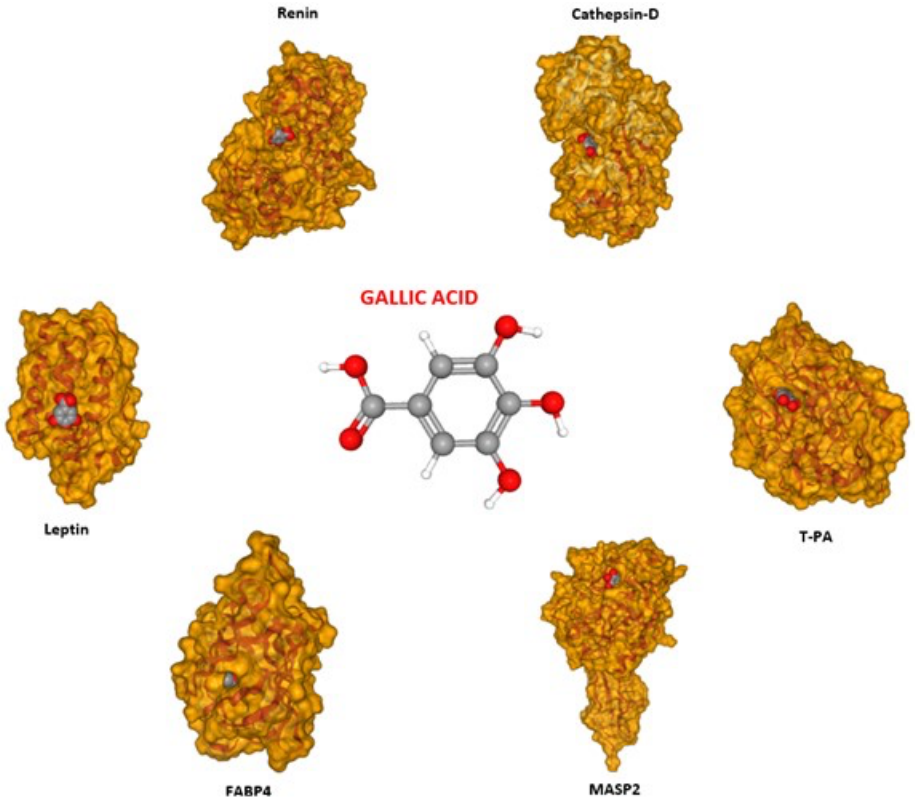

Figure 3. Molecular docking result of gallic acid.

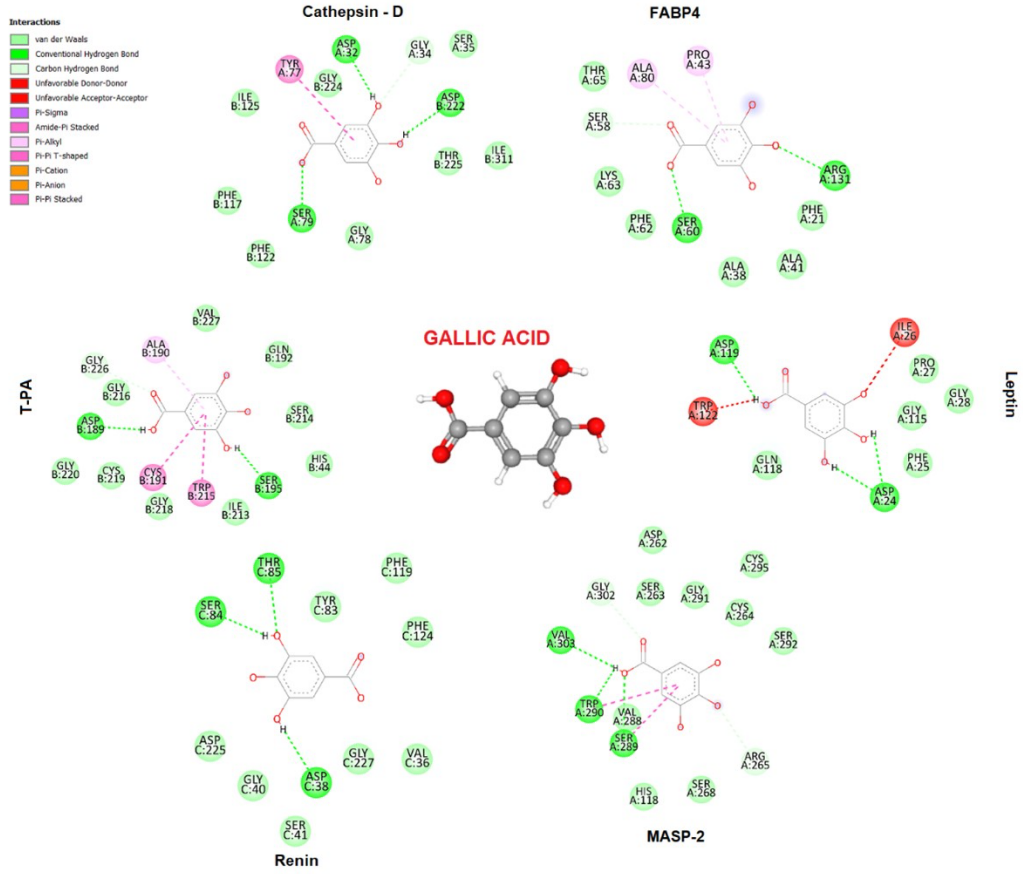

Figure 4. Protein interactions of gallic acid.

Table 1. Protein and ligand names.

	Protein and ligand names	Resolution
Proteins	Renin (PDB: 2V0Z)	2.20 Å
	Leptin (PDB: 1AX8)	2.40 Å
	Cathepsin-D (PDB: 4OD9)	1.90 Å
	FABP4 (PDB: 6LJW)	1.40 Å
	MASP2 (PDB: 1Q3X)	2.23 Å
	T-PA (PDB: 1RTF)	2.30 Å
Ligands	Quercetin (PubChem: 5280343)	-
	Gallic Acid (PubChem: 370)	-
	Oleanolic Acid (PubChem: 10494)	-
	Ursolic Acid (PubChem: 64945)	-

Table 2. Protein binding affinities and RMSD values of quercetin.

Proteins	Affinity (kcal/mol)	RMSD l.b.	RMSD u.b.
MASP2	-7.9	1.48	2.995
FABP4	-7.8	1.242	2.97
Renin	-7.7	1.557	6.619
T-PA	-7.6	1.268	2.956
Cathepsin-D	-7.3	1.521	7.177
Leptin	-6.5	2.354	7.123

Table 3. Protein binding affinities and RMSD values of gallic acid.

Proteins	Affinity (kcal/mol)	RMSD l.b.	RMSD u.b.
T-PA	-6.3	0.55	2.4
MASP2	-6.1	1.508	3.772
FABP4	-5.6	0.152	2.397
Renin	-5.6	1.278	2.818
Cathepsin-D	-5.5	0.235	2.564
Leptin	-4.8	1.547	2.745

Table 4. Protein binding affinities and RMSD values of oleanolic acid.

Proteins	Affinity (kcal/mol)	RMSD l.b.	RMSD u.b.
Renin	-7.2	1.871	3.99
Cathepsin-D	-7.1	1.888	3.075
MASP2	-6.9	1.651	2.126
Leptin	-6.8	1.731	8.327
T-PA	-6.7	1.83	3.188
FABP4	-6.3	2.014	8.466

Table 5. Protein binding affinities and RMSD values of ursolic acid.

Proteins	Affinity (kcal/mol)	RMSD l.b.	RMSD u.b.
Renin	-8.0	2.336	4.464
MASP2	-7.3	1.598	2.028
T-PA	-7.0	1.842	2.885
Cathepsin-D	-6.9	2.894	8.176
Leptin	-6.8	1.982	8.595
FABP4	-6.4	1.654	8.371

was determined that Cathepsin-D interacts with the amino acids GLY224, ASP222, THR225, ILE311, PHE122, PHE117, ILE125 in the B subunit (Figure 4).

The affinity results of oleanolic acid for proteins associated with diabetes are given in Table 4. According to these results, oleanolic acid binds to renin protein with a higher affinity than other proteins, with an affinity value of -7.2 kcal/mol.

In Figure 5, the binding status of the oleanolic acid molecule to the proteins associated with diabetes is shown by the molecular docking results in Table 4.

Oleanolic acid with amino acids GLU77, SER68, PHE69, GLU74, ILE70, ASP76, ILE67, PHE75, VAL78, and GLU66 in FABP-4 protein were found to interact (Figure 6).

Oleanolic acid interacts with the amino acids PHE25, ASP24, GLN118, GLY115, GLY28, PRO27, ILE32, PRO31, ASP119, and TRP122 in the Leptin protein (Figure 6).

Oleanolic acid has been found to interact with amino acids TRP112, ASN111, LYS176, ILE318, GLU70, CYS69, PRO67, LEU11, PRO12, and VAL68 in the MASP2 protein (Figure 6).

Oleanolic acid interacted with the amino acids TYR15, MET16, TYR230, SER229, ALA228, GLY227, MET302, THR85, PRO118, GLN19, PHE124, THR18, LEU121 in the Renin protein (Figure 6).

Oleanolic acid interacted with amino acids GLY193, GLN192, TRP215, GLY216, GLY218, ARG171, GLU48, PHE50, TYR90, CYS29, CYS45, TYR146, ARG26, LEU28, HIS44 in the T-PA protein (Figure 6).

Oleanolic acid interacts with the amino acids ASP11 in the A subunit of the Cathepsin-D protein; at the same time, it was determined that Cathepsin-D interacts with the amino acids LYS284, VAL259, GLU279, THR282, LEU283, PRO164, ASP163, ARG162, ASP280, ARG319 in the B subunit (Figure 6).

The affinity results of ursolic acid for proteins associated with diabetes are given in Table 5. According to these results, ursolic acid binds to renin protein with a higher affinity than other proteins, with an affinity value of -8.0 kcal/mol.

In Figure 7, the binding status of the ursolic acid molecule to the proteins associated with diabetes is shown by the molecular docking results in Table 5.

Ursolic acid with amino acids VAL78, GLU66, THR65, ILE67, GLU77, ASP76, SER68, PHE75, and PHE69 in FABP-4 protein were found to interact (Figure 8).

Ursolic acid interacts with the amino acids PHE25, GLN118, ASP24, GLY115, GLY28, PRO31, ASP119, PRO27, and TRP122 in the Leptin protein (Figure 8).

Ursolic acid has been found to interact with amino acids PHE196, ILE202, ASP201, PHE274, LEU275, TRP282, ARG,78, and THR79 in the MASP2 protein (Figure 8).

Ursolic acid interacted with the amino acids THR85, GLY86, TYR83, PHE119, PHE124, PRO118, VAL36, GLN19, LEU121, THR18, TYR15, TYR230, SER229, ALA228, GLY227 in the Renin protein (Figure 8).

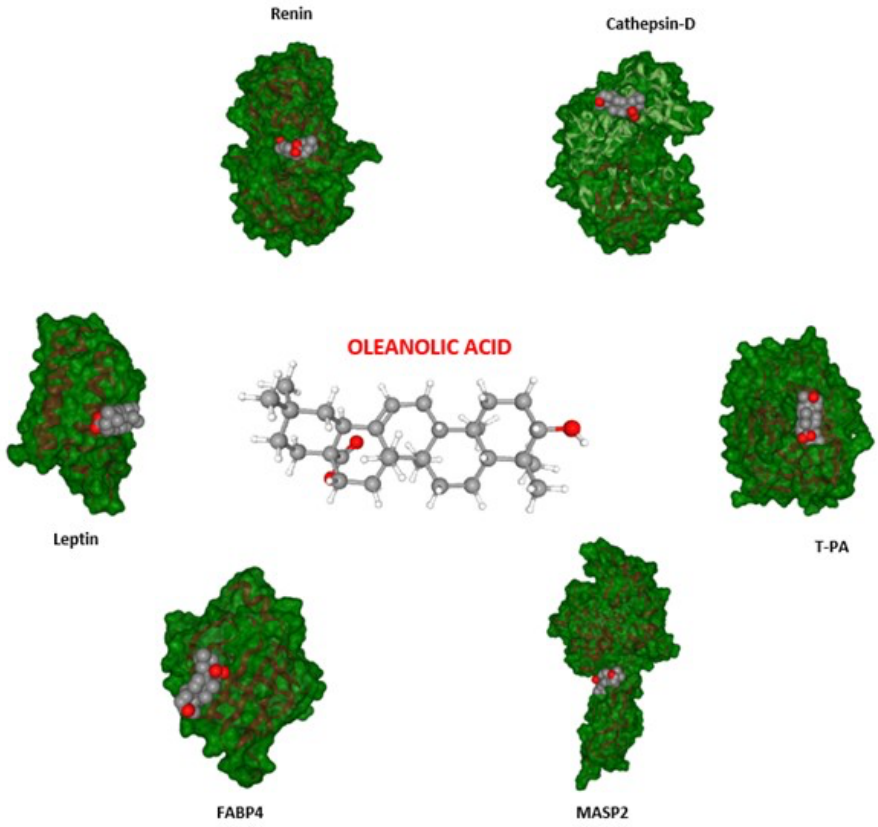

Figure 5. Molecular docking result of oleanolic acid.

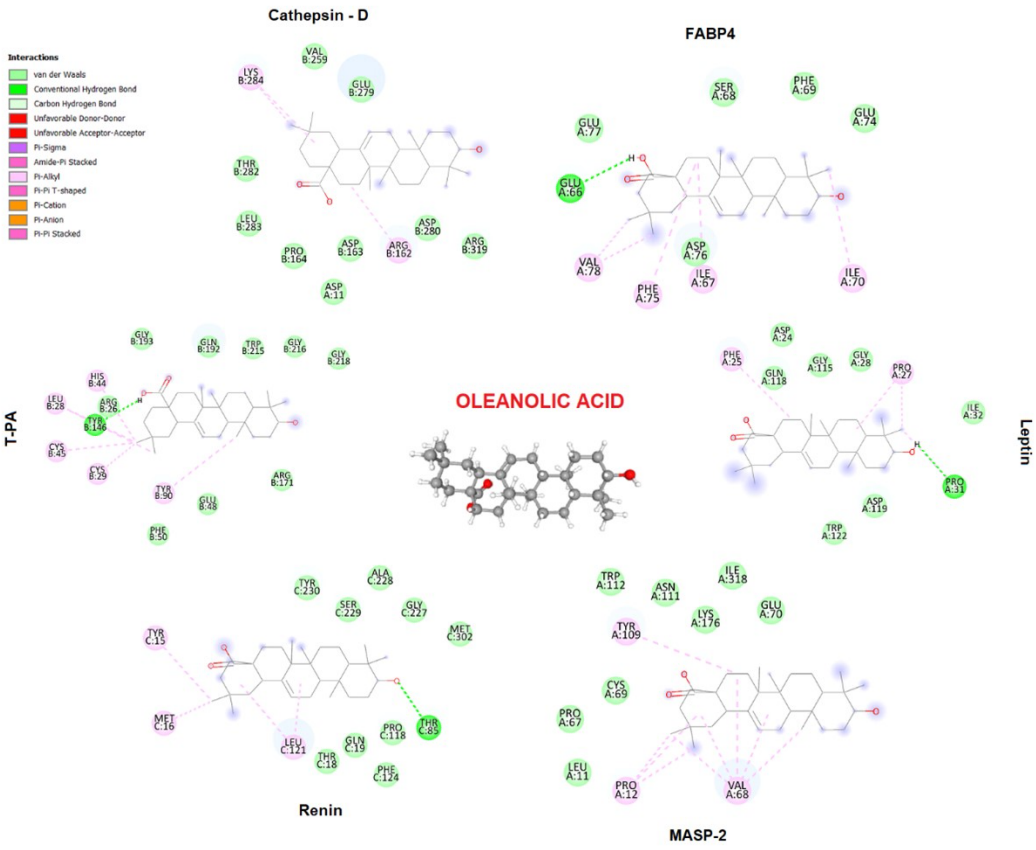

Figure 6. Protein interactions of oleanolic acid.

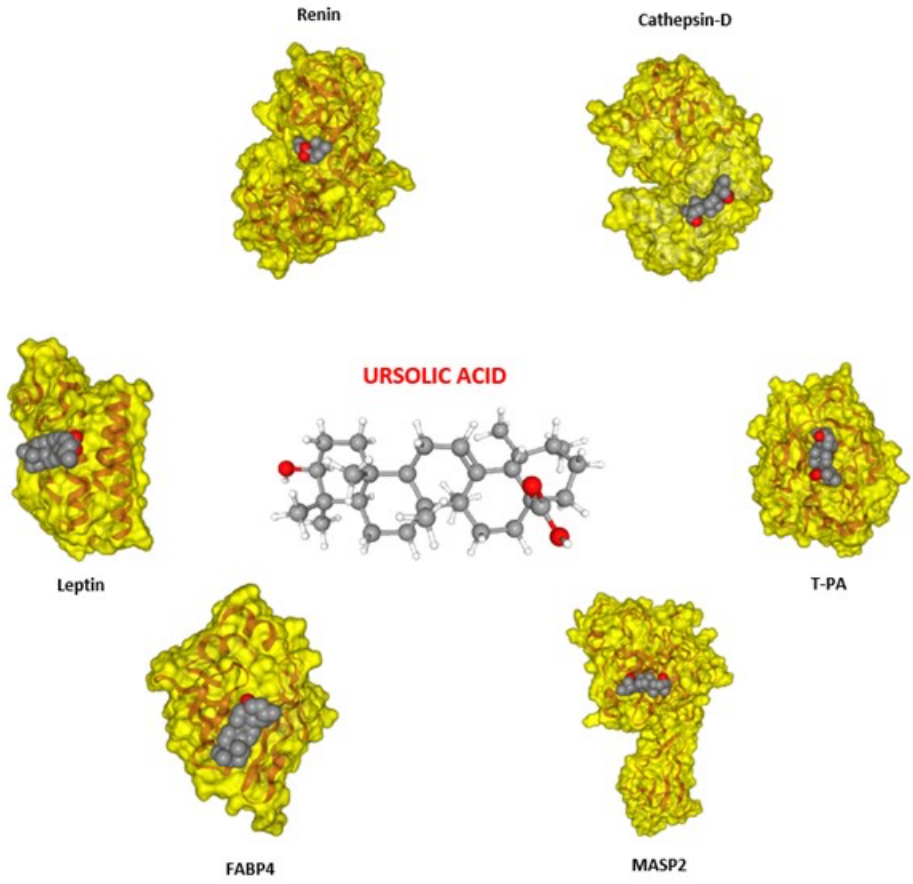

Figure 7. Molecular docking result of ursolic acid.

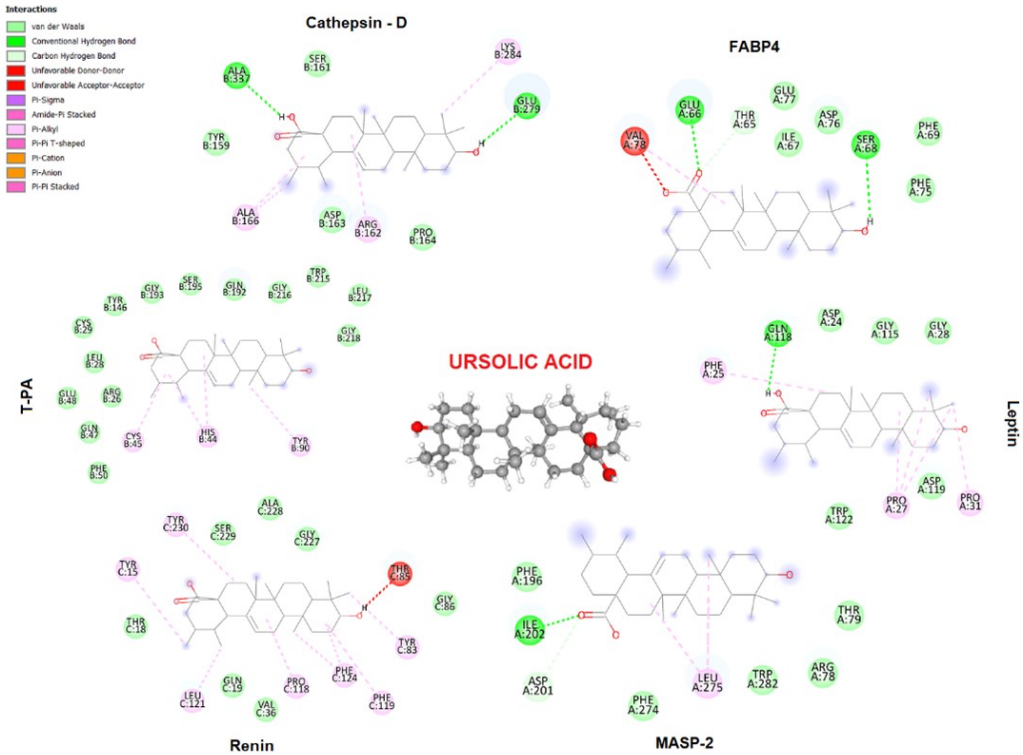

Figure 8. Protein interactions of ursolic acid acid.

Ursolic acid interacted with amino acids TYR90, HIS44, CYS45, PHE50, GLN47, ARG26, GLU48, LEU28, CYS29, TYR146, GLY193, SER195, GLN192, GLY216, TRP215, LEU217, GLY218 in the T-PA protein (Figure 8).

Ursolic acid interacts with the amino acids LYS284, GLU279, PRO164, ARG162, ASP163, ALA166, TYR159, ALA337, and SER161 in the B subunit of the Cathepsin-D protein (Figure 8).

As a result of ADME analysis, situations such as the solubility rate of the drug candidate molecule in water, the percent of absorption of the molecule from the intestine, whether it can pass through the blood-brain barrier, and how it interacts with liver enzymes such as Cytochrome-p450 can be analyzed. Therefore, ADME studies are important for us to understand whether molecules can be drug substances [36]. The results of the ADME analysis of quercetin, gallic acid, ursolic acid, and oleanolic acid are given in Table 6. When we look at the water solubility of these molecules, whether they are soluble in water or not is interpreted according to the following criteria: insoluble < -10 < poor < -6 < moderate < -4 < soluble < -2 < very < 0 < very good [37]. The solubility of quercetin in water was -2.925, the solubility of gallic acid in water was -2.56, the solubility of ursolic acid in water was -3.072, and the solubility of oleanolic acid in water was -3.074. As a result, these molecules dissolve in water. If the intestinal absorption criterion is below 30%, it is interpreted as poor absorption [37]. Intestinal absorption was 77.2% for quercetin, 43.7% for gallic acid, 100% for ursolic acid, and 99.9% for oleanolic acid. All flavonoids are above the 30% absorption criterion. Quercetin, gallic acid, ursolic acid and oleanolic acid have no blood brain barrier permeability. Cytochrome p450 enzymes (CYP enzymes) play a role in the elimination of drugs taken from the body, and inhibition of these enzymes delays the elimination of the drug from the body. Table 6 shows the effect of quercetin, gallic acid, ursolic acid, and oleanolic acid on the activities of CYP enzymes. Quercetin has been found to inhibit the CYP1A2 enzyme, which is one of the CYP enzymes. Gallic acid, ursolic acid, and oleanolic acid do not inhibit any CYP enzyme.

DISCUSSION

Diabetes is one of the important health problems today. According to IDF diabetes atlas data in 2021, 51 million in North America and the Caribbean, 32 million in South and Central America, 61 million in Europe, 73 million in the Middle East and North Africa, 24 million in Africa, and Southeast Asia. These patients range from 20–79 years of age [38]. The cost of this disease is also high, as there are so many cases. Therefore, scientists are also investigating alternative treatment methods to reduce the cost. One of these treatment methods is herbs that have been used since ancient times. Flavonoids found in plants have many biological properties. Examples of these biological properties are antioxidant, anti-tumour, anti-inflammatory, anti-diabetic, anti-ulcer, and anti-allergic properties. Quercetin, gallic acid, oleanolic acid, and ursolic acid flavonoids discussed in this study have many different biological properties as well as anti-diabetic properties [10, 32]. Due to these anti-diabetic properties, molecular docking analyzes of proteins involved in various complications caused by diabetes have been performed and it has been revealed which flavonoid can play a more active role in diabetes. When we look at the docking analysis of the MASP2 protein, it was determined that quercetin binds with an affinity of -7.9 kcal/mol, ursolic acid with -7.3 kcal/mol, oleanolic acid with -6.9 kcal/mol, and gallic acid with -6.1 kcal/mol (Table 2-5). According to these results, it is seen that the flavonoid that can best affect the MASP2 protein is quercetin. In the docking analysis of the FABP4 protein, it was determined that quercetin binds with an affinity of -7.8 kcal/mol, ursolic acid with -6.4 kcal/mol, oleanolic acid with -6.3 kcal/mol, and gallic acid with -5.6 kcal/mol (Table 2-5). From these results, it is seen that quercetin flavonoid is more effective on FABP4 protein. In the docking analysis of T-PA protein, it was determined that quercetin binds with -7.6 kcal/mol, ursolic acid -7.0 kcal/mol, oleanolic acid -6.7 kcal/mol, and gallic acid -6.3 kcal/mol (Table 2-5).

According to these results, it is seen that ursolic acid is the flavonoid that has the best effect on renin protein. As a result of docking to leptin protein, ursolic acid was bound with -6.8 kcal/mol, oleanolic acid -6.8 kcal/mol,

Table 6. The ADME parameters of quercetin, gallic acid, ursolic acid and oleanolic acid.

ADME Parameters	Quercetin	Gallic Acid	Ursolic Acid	Oleanolic Acid
Water solubility (log mol/L)	-2.925	-2.56	-3.072	-3.074
Intestinal absorption (human) (% absorbed)	77.20	43.73	100	99.93
Blood-brain barrier permeability	No	No	No	No
P-glycoprotein substrate	Yes	No	No	No
P-glycoprotein I inhibitor	No	No	No	No
P-glycoprotein II inhibitor	No	No	No	No
CYP1A2 inhibitor	Yes	No	No	No
CYP2C19 inhibitor	No	No	No	No
CYP2C9 inhibitor	No	No	No	No
CYP2D6 inhibitor	No	No	No	No
CYP3A4 inhibitor	No	No	No	No

quercetin -6.5 kcal/mol, and gallic acid -4.8 kcal/mol with affinity (Table 2-5). The flavonoids ursolic acid and oleanolic acid appear to have the best effect on leptin protein. In general, it is seen that quercetin has a greater effect on proteins associated with diabetes, while the effect of gallic acid is weaker than other flavonoids. In this case, quercetin could potentially be therapeutic that could be used to inhibit complications caused by diabetes. When we look at the ADME analysis of quercetin, gallic acid, ursolic acid, and oleanolic acid, they are all water-soluble. When intestinal absorption is considered, the highest absorption belongs to ursolic acid with 100%, while the lowest absorption belongs to gallic acid with 43.73%. According to the docking result, it is quercetin with an absorption rate of 77.20% (Table 6). By using quercetin in combination with new treatment methods, its absorption in the intestinal tract can be increased and more effective results can be obtained. At this point, in vitro studies can be designed. When we look at the effects on CYP 450 enzymes, other flavonoids, except quercetin, do not inhibit these enzymes. Quercetin appears to inhibit the CYP1A2 enzyme. Inhibition of the CYP1A2 enzyme has the property of inhibiting cancerization [39]. According to this result, the anti-tumour properties of quercetin were also revealed.

As a result of this study, it was found that quercetin flavonoid is more effective on diabetes. Clinical studies in the literature also suggest that quercetin flavonoid reduces the effects of diabetes [40]. However, in the future, the effect of quercetin on the genes in our study can be investigated in vivo.

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