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Research Article

Phytochemical profiling, molecular docking , and ADMET prediction of crude extract of *Atriplex nitens* Schkuhr for the screening of antioxidant and urease inhibitory

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ABSTRACT

Atriplex nitens Schkuhr (ANS), which grows naturally in arid and semi-arid regions of the world, is highly resistant to drought and salty environments and is used as food and animal feed. This study first performed phytochemical analysis and antioxidant and urease inhibition activities on the obtained methanol crude extract of ANS. The catechin and isoquercitrin were detected as the main compounds according to LC-MS/MS results. Oleic acid methyl ester (31.71%), palmitic acid methyl ester (25.87%), linoleic acid methyl ester (19.61%), and nonacosane (16.81%) were detected in GC-MS/MS analysis of extract. Posphomolybdenum reducing, DPPH˙ scavenging, and urease inhibition activities were found effective at 67.27±23.83, 7.85±0.44 and 6.58±0.48 µg/mL, respectively, of ANS extract. In this investigation, the biological activity and chemical composition of the ANS extract were initially examined. Molecular docking and ADMET prediction were performed on this plant's two most abundant components. It was found that the interaction with urease of isoquercitrin (MolDock score-121.42, binding affinity -8.60, and binding constant 0.62 µM) with urease determined a higher than. These two components have a negligible potential for toxicity. The Boiled Egg plot indicates a significant GIa for catechin. However, isoquercitrin does not exhibit BBB or GLa permeability. It was determined that the main component isoquercitrine may be effective against gastric diseases, and it was supported that it was not observed in the BBB and GLa systems.

Keywords: *Atriplex nitens* Schkuhr, Antioxidant activity, LC-MS/MS, GC-MS/MS, ADMET, Molecular Docking

1. INTRODUCTION

Antioxidants benefit the body's health by removing reactive oxygen and nitrogen species and preventing oxidative stress conditions that can lead to various diseases.[1] In particular, synthetic antioxidants (BHA, BHT, TBHQ) are used as food additives.[2] However, the tendency towards natural antioxidants is increasing due to their adverse effects on the body.[3] Natural antioxidants

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can be found in all tissues of plants. These compounds, which act as antioxidants in all tissues, are secondary metabolites known as carotenoids, phenols, flavonoids, and vitamins.[4]

Enzyme activity is supported or inhibited by several substances. By attaching itself to the molecule's active site, an inhibitor can stop the substrate from binding when it inhibits an enzyme. An inhibitor molecule and the substrate compete when binding to the active site. Thus, competitive inhibition is used to block the enzyme. The inhibitor attaches itself to the enzyme at a position other than the allosteric site in the noncompetitive inhibition mechanism. By stopping the substrate from attaching to the enzyme's active site, it inhibits the enzyme.[5]

Molecular docking studies are a method used to determine how a protein or biomolecule interacts with another molecule.[6] Drug design studies generally require high costs, meticulous work, and time. While compounds with high drug potential are identified in molecular docking studies, ligands with low binding capacity are eliminated and prevented from being used in other experiments, thus saving cost and time. This method is widely used because it provides great convenience in drug design.[7]

ADMET assesses the drug's safety, efficacy, absorption, elimination, metabolic characteristics, and toxicity by considering its adsorption, distribution, metabolism, elimination, and toxicity. ADMET was introduced because substances defined as drugs failed in clinical trials. Today, ADMET prediction is a computational method applied to select molecules with adsorption, distribution, metabolism, elimination, and toxicity values.[8,9] Physicochemical parameters and drug-likeness scores are related to water solubility, intestinal permeability, oral bioavailability, and pharmaceutical properties.[10] Cytochrome P450 (CYP), derived from pharmacokinetics-related proteins, is involved in the oxidative metabolism of compounds, in which oxidized molecules become polar and thus can be excreted, especially by the kidneys, over a period of time.[10]

Atriplex is a member of the naturally growing Chenopodiaceae family, which has more than 400 species and is highly resistant to drought and salty environments.[11,12] *Atriplex nitens* Schkuhr (ANS) is an annual herbaceous plant that spreads naturally in the provinces of Erzurum, Kars, Kayseri, Tokat, and Ankara in Turkey. In contrast, it spreads in the Southwest and Central Asia.[13,14] Although the ANS is widely known as a weed, it is used as a human and animal food source.[15,16] Salt-tolerant plants have traditionally been used for medicinal, nutritional, and even artisanal purposes. There is increasing interest in these species due to their high bioactive content. Salt-tolerant plants, with detailed research, will lead to their use as health products in various industries, functional foods, nutraceuticals, or active ingredients.[17] Studies have reported that extracts obtained from Atriplex species exhibit biological activities such as antioxidant, anticholinesterase, antibacterial, antifungal, and antiparasitic.[18-22] Atriplex species are rich in triterpenes, sterols and phytoecdysteroids, triterpene saponins, and phenolic compounds. Therefore, high activity can be seen due to these compounds.[18,23-25]

This study aimed to investigate the antioxidant and enzyme inhibition activities of the ANS plant, for which there is no comprehensive study, to determine its major components using phytochemical analysis methods and to observe the interactions of these major components with the urease enzyme. In addition, the effects of these significant molecules on the human body were evaluated by making ADMET predictions. With the results of these studies, it will be evaluated whether the plant can be used in traditional medicine or in the pharmaceutical industry

2. EXPERIMENTAL

2.1. Plant Material

The study was conducted in 2022 in the dry trial field of Iğdır University Agricultural Application and Research Center Directorate. *Atriplex nitens* Schkuhr (ANS) seeds were sown by hand in the lines made with a marker at a depth of 4-5 cm, with a row spacing of 45 cm and a distance of 10 cm between rows, following the rains at the end of March (24.03.2022), when the soil was tempered. No fertilizer or irrigation was applied during the growing period. Seed harvests were carried out manually at the end of September (25.09.2022), when the fruits on the plant turned yellow and ripened. After harvest, the seeds were brought to the laboratory and dried in an oven set at 40 °C until their weight was constant. Afterward, the seed samples were ground to 1 mm in diameter and made ready for analysis. Species identification was made by Muş Alparslan University, Faculty of Arts and Sciences, Department of Molecular Biology and Genetics, Lecturer, Prof. Dr. Murat Aydın Şanda.

2.2. Extraction of ANS

The ANS's seed part, harvested on September 29, was ground into powder and left for extraction in methanol solvent in a dark environment. The solvent sample mixture was filtered, then the solvent was removed using rotary evaporation, and the crude extract was obtained. The polarity index of methanol is 5.1. Also, among all alcohols, the boiling point of methanol is 65 degrees Celsius. Bioactive compounds obtained from plants include tannins, alkaloids, glycosides, lignans, terpinoids, etc. It belongs to various chemical groups, such as. Methanol is mainly used for the extraction of various polar compounds and has the highest extraction efficiency. Moreover, the non-polar compound is soluble

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even in methanol. Therefore, methanol is widely used in the extraction of bioactive compounds.

2.3. Total Phenol and Flavonoid Content

Gallic acid was utilized as a standard in the Folin-Ciocalteu phenol reagent method, which was used to quantify the total phenol content. Using the aluminum chloride method, standard quercetin was used to calculate the total flavonoid content.[26]

2.4. LC-MS/MS Analysis

We performed LC-MS/MS analysis to determine the phenolic contents and amounts of ANS crude extract, as our previously published article described. This analysis used LC-MS/MS with mass spectrometry coupled to HPLC (1260 Infinity II LC System).[27]

2.5. GC-MS/MS Analysis

ANS crude extract content analysis GC-MS/MS; It was performed using Agilent 7000 A GC/MS Triple Quad with 7890 GC, 7693 Autosampler, and 7697A Headspace Sampler. As a procedure, the initial temperature was determined at 50 °C and maintained for two minutes, then increased at 3 °C/min to 140 °C, 4 °C/min at 220 °C for 10 min, and constant at 4 °C/min to 270 °C and finally continued at 270 °C for 30 min. The ion temperature of the MS detector is 280 °C. The filtered sample with a 0.22 µm disposable syringe filter was run with a 1 mL He gas flow by injecting a volume of 1 µL at a ratio of 1:10. Agilent HP-5 (5%-phenyl)-methylpolysiloxane) (30m x 0.25 mm x 0.25 µm) was carried out on the GC column. 30 mg of the sample was taken, 2 ml MeOH was dissolved, and 2 mL n-hexane was added. Then, 1 ml KOH solution (1M) was added and mixed with a vortex device at 2500 rpm for 30 seconds. It was taken from the upper phase (n-hexane phase), which contained fatty acid methyl esters, filtered with 0.22 micron PVDF, and injected.

2.6. Antioxidant and Enzyme Inhibitions Activity Assays

The antioxidant activities of crude extract were examined with posphomolybdenum reducing[28] and free radical scavenging[29] activities and compared with ascorbic acid and BHT. The results were recorded as $A_{0.5}$ and IC_{50} (µg/mL).

The enzyme inhibition activity of urease was determined with a spectrophotometric method[30] and compared with thiourea as a standard drug. The activity result was expressed as IC_{50} (µg/mL).

2.7. Molecular Interaction Applications

In molecular docking studies, the structure of the molecules was drawn with Chem-Draw Ultra 15, and the minimum energy calculation was made with Chem3D 15 and then saved in Mol2 format. The RCSB PDB provided the urease [PDB ID: 4GY7] enzyme's 3D structure.[31] The molecules interacting with the enzyme's active sites were identified using the AutoDock Vina and Molegro Virtual Docker (MVD) programs. The MolDock score that produced the highest result was utilized to determine the molecule's location. Furthermore, the 2D and 3D structures of the interactions between enzymes and molecules were discovered using the Discovery Studio program. [27,32-35]

2.8. ADMET

SwissADME, an online server, was used to calculate bioavailability scores and pharmacokinetic features (https://www.swissadme.ch/).
Bioactivity scores were computed using the online Chemoinformatics features of the software Molinspiration (https://www. molinspiration.com).
Additionally, the OSIRIS Property Explorer (https://www.organic-chemistry.org/prog/peo/) was used to determine the toxicity risk characteristic of the selected compounds.[10]

2.9. Statistical Analysis

The activity results with standard deviations were provided for the triple-measured *in vitro* antioxidant and enzyme inhibitory activities. The analytical averages achieved with the IBM SPSS 20.0 software indicated a normal distribution, so a one-way ANOVA was used to analyze all the data. The data were subjected to the Tukey HSDa,b multiple comparison test for this inquiry. Based on the $p<0.05$ value, the statistical significance of the data was ascertained.

3.RESULTS and DISCUSSION

The obtained crude extract of chemical content, antioxidant, and enzyme inhibition activities were determined. Also, in LC-MS/MS analysis, molecular docking and ADMET studies were performed Since phenolic compounds detected in high amounts may affect the activity. It was observed that the total phenol (2.21±0.16 mg GAE/g extract) and total flavonoid (1.22±0.07 mg QE/g extract) amounts were low of ANS extract (Table 1)

Table 1. Bioactivity result of ANS

	Chemical content		Antioxidant activity*		Enzyme inhibition*
Sample/ Standard	**Total phenol** (mg GAE/g)	**Total flavonoid** (mg QE/g)	**Posphomolybdenum reducing** ($A_{0.5}$ µg/mL)	**Free radical scavenging** (IC_{50} µg/mL)	**Urease** (IC50 µg/mL)
ANS	2.21±0.16	1.22±0.07	67.27±23.83^a	7.85±0.44^a	6.58±0.48^a
Ascorbic acid	-	-	87.64±50.54^b	42.15±1.35^c	-
BHT	-	-	104.02±24.59^c	18.91±2.18^b	-
Thiourea	-	-	-	-	16.68±1.12^b

*p<0.05

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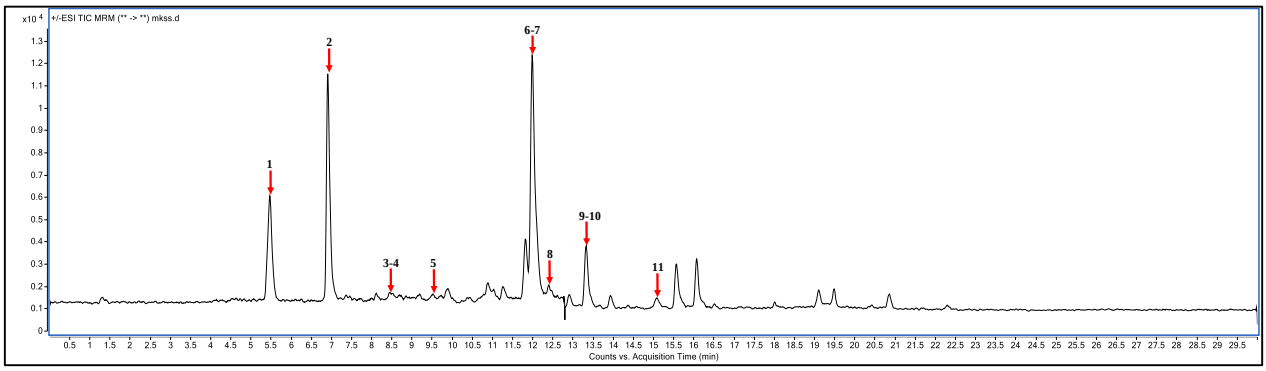

Figure 1. LC-MS/MS chromatogram of ANS [Protocatechuic acid (**1**), Catechin (**2**), Caffein (**3**), Vanillin (**4**), *o*-coumaric acid (**5**), Hesperidin (**6**), Isoquercitrin (**7**), Rutin (**8**), Kaempferol-3-glucoside (**9**), Fisetin (**10**), Naringenin (**11**)]

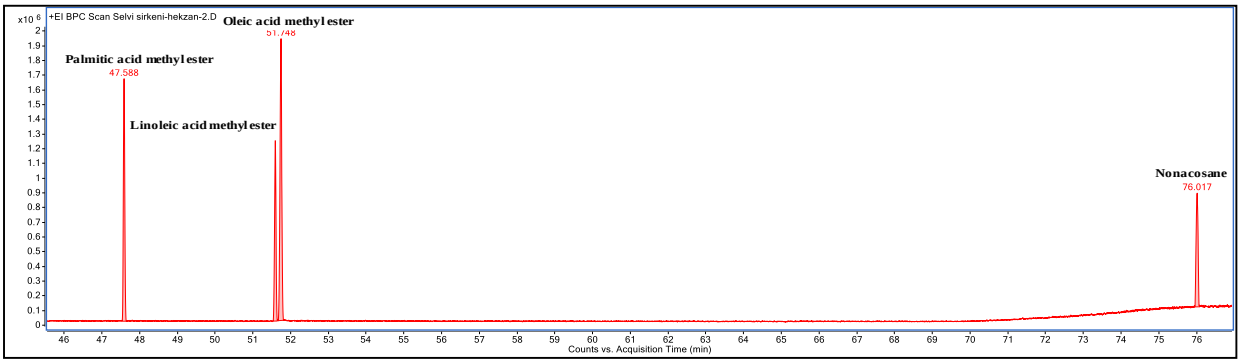

Figure 2. GC-MS/MS chromatogram of ANS

The antioxidant activities were tested as posphomolybdenum-reducing and free radical scavenging activities. According to the analysis results, ANS (67.27±23.83 µg/mL) showed higher activity than the standards as ascorbic acid (87.64±50.54 µg/mL) and BHT (104.02±24.59 µg/mL) in posphomolybdenum-reducing activity. The ANS (7.85±0.44 µg/mL) showed higher activity than the standards as ascorbic acid (42.15±1.35 µg/mL) and BHT (18.91±2.18 µg/mL) in free radical scavenging activity. According to these results, ANS extract exhibited high antioxidant activity (Table 1).

Table 2. LC-MS/MS result of ANS (µg/g extract)

No	R.T. (min.)	Compound	Concentration
1	5.490	Protocatechuic acid	18.83
2	6.912	Catechin	38237.54
3	8.459	Caffein	6.07
4	8.686	Vanillin	15.61
5	9.518	*o*-Coumaric acid	12.876
6	11.953	Hesperidin	22.44
7	11.985	Isoquercitrin	348.83
8	12.399	Rutin	109.50
9	13.333	Kaempferol-3-glucoside	46.55
10	13.318	Fisetin	8.39
11	15.075	Naringenin	86.41

It is known that the phenolic compounds present in the extracts affect the activity. Therefore, the plant extract we used in our study also contains high amounts of catechin and isoquercitrin components as phenolic components. It also increased activity in these components.

A urease inhibition test was performed for enzyme inhibition activity. Thiourea was used as standard. It showed higher activity than standard thiourea (16.68±1.12 µg/mL) in the urease inhibition test of ANS (6.58±0.48 µg/mL) (Table 1). The urease enzyme was used in molecular docking experiments for the compounds found in substantial quantities, as shown by the LC-MS/MS study results. According to LC-MS/MS phenolic content analysis, the highest amounts of catechin (38237.54 µg/g extract), isoquercitrin (348.83 µg/g extract), and rutin (109.50 µg/g extract) compounds were detected (Figure 1 and Table 2). In GC-MS/MS analysis, oleic acid, methyl ester; palmitic acid, methyl ester; linoleic acid, methyl ester, and nonacosane were determined at 31.71%, 25.87%, 19.61% and 16.81%, respectively (Figure 2 and Table 3).

Table 3. GC-MS/MS result of ANS

No	R.T.(min.)	Compound	%
1	47.59	Palmitic acid, methyl ester	25.87
2	51.60	Linoleic acid, methyl ester	19.61
3	51.75	Oleic acid, methyl ester	37.71
4	76.02	Nonacosane	16.81

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3.1. Molecular Interactions

Catechin interacting with the active site of the urease, five conventional HB interactions with ARG609, HIS593, ASP633, ALA636, ALA436; three carbon-HB interactions with ALA440, ALA436, ASP494; one amide-pi stacked interactions with ARG439; one alkyl interactions with ALA440; and six pi-alkyl interactions with HIS593, ALA440, ALA636, MET637, ARG439 were observed (Figure 3 and Table 5). The predicted values of the binding constant (2.77 µM), binding affinities (-7.70 kcal/mol), and MolDock score (-84.87) indicate the interactions between catechin and urease (Table 4).

Table 4. Interaction results of urease with catechin and isoquercitrin

Compound-Enzyme	Moldock Score	Binding Affinity (kcal/mol)	Binding Constant (µM)
Catechin-Urease	-84.87	-7.70	2.77
Isoquercitrin-Urease	-121.42	-8.60	0.62

A urease inhibition test was performed for enzyme inhibition activity. Thiourea was used as standard. It showed higher activity than standard thiourea (16.68±1.12 µg/mL) in the urease inhibition test of ANS (6.58±0.48 µg/mL) (Table 1). The urease enzyme was used in molecular docking experiments for the compounds found in substantial quantities, as shown by the LC-MS/MS study results. According to LC-MS/MS phenolic content analysis, the highest amounts of catechin (38237.54 µg/g extract), isoquercitrin (348.83 µg/g extract), and rutin (109.50 µg/g extract) compounds were detected (Figure 1 and Table 2). In GC-MS/MS analysis, oleic acid, methyl ester; palmitic acid, methyl ester; linoleic acid, methyl ester, and nonacosane were determined at 31.71%, 25.87%, 19.61% and 16.81%, respectively (Figure 2, Table 3).

Isoquercitrin interacting with the active site of the urease, seven conventional HB interactions with HIS593, ALA440, GLY550, GLN635, MET588, ARG439; one carbon-HB interactions with GLN635; two pi-pi stacked interactions with HIS593; and three pi-alkyl interactions with ALA440, ALA636, MET637 were observed (Figure 4-Table 6). Isoquercitrin's interactions with urease were estimated using the following metrics: binding constant (0.62 µM), binding affinities (-8.60 kcal/mol), and MolDock score (-121.42) (Table 6).

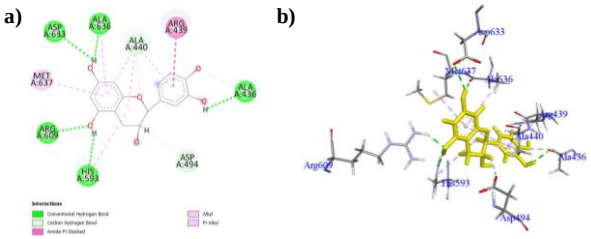

Figure 3. Images of the interactions of the catechin with urease a) 2D and b) general views

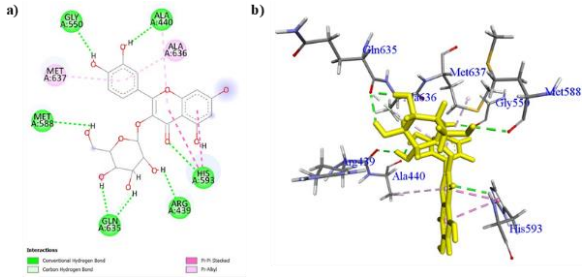

Figure 4. Images of the interactions of the isoquercitrin with urease a) 2D and b) general views

Table 5. Bond properties and distance of interaction of urease with catechin

No	Name	Distance	Category	Types	From	From Chemistry	To	To Chemistry
1	A:ARG609:HH22 - :[041:O2	2.12	HB	Conventional HB	A:ARG609:HH22	HD	:[041:O2	HA
2	:[041:H6 - A:HIS593:NE2	2.36	HB	Conventional HB	:[041:H6	HD	A:HIS593:NE2	HA
3	:[041:H8 - A:ASP633:OD2	2.74	HB	Conventional HB	:[041:H8	HD	A:ASP633:OD2	HA
4	:[041:H8 - A:ALA636:O	1.73	HB	Conventional HB	:[041:H8	HD	A:ALA636:O	HA
5	:[041:H11 - A:ALA436:O	2.14	HB	Conventional HB	:[041:H11	HD	A:ALA436:O	HA
6	A:ALA436:HA - :[041:O6	2.68	HB	Carbon HB	A:ALA436:HA	HD	:[041:O6	HA
7	A:ALA440:HA - :[041:O4	2.19	HB	Carbon HB	A:ALA440:HA	HD	:[041:O4	HA
8	:[041:H2 - A:ASP494:OD2	2.69	HB	Carbon HB	:[041:H2	HD	A:ASP494:OD2	HA
9	A:ARG439:C,O;ALA440:N - :[041	3.82	Hydrophobic	Amide-Pi Stacked	A:ARG439:C,O; ALA440:N	Amide	:[041	Pi-Orbitals
10	A:ALA440 - :[041	4.41	Hydrophobic	Alkyl	A:ALA440	Alkyl	:[041	Alkyl
11	A:HIS593 - :[041	5.20	Hydrophobic	Pi-Alkyl	A:HIS593	Pi-Orbitals	:[041	Alkyl
12	:[041 - A:ALA440	5.45	Hydrophobic	Pi-Alkyl	:[041	Pi-Orbitals	A:ALA440	Alkyl
13	:[041 - A:ALA636	5.26	Hydrophobic	Pi-Alkyl	:[041	Pi-Orbitals	A:ALA636	Alkyl
14	:[041 - A:MET637	4.65	Hydrophobic	Pi-Alkyl	:[041	Pi-Orbitals	A:MET637	Alkyl
15	:[041 - A:ARG439	4.45	Hydrophobic	Pi-Alkyl	:[041	Pi-Orbitals	A:ARG439	Alkyl
16	:[041 - A:ALA440	4.09	Hydrophobic	Pi-Alkyl	:[041	Pi-Orbitals	A:ALA440	Alkyl

* HB: Hydrogen Bond, HD: H-Donor, HA: H-Acceptor.

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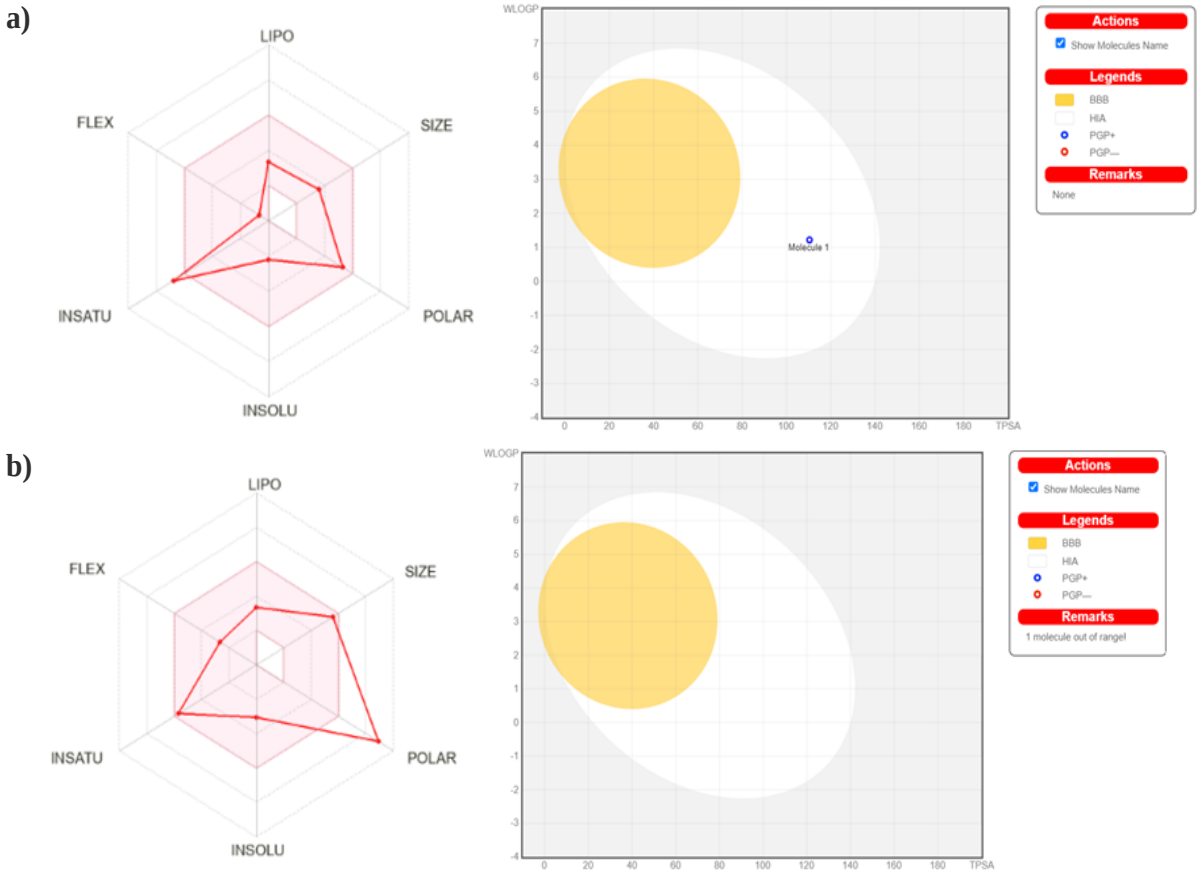
Table 6. Bond properties and distance of interaction of urease with isoquercitrin

No	Name	Distance	Category	Types	From	From Chemistry	To	To Chemistry
1	A:HIS593:HT1 - :[001:O4	2.50	HB	Conventional HB	A:HIS593:HT1	HD	:[001:O4	HA
2	:[001:H9 - A:ALA440:O	1.87	HB	Conventional HB	:[001:H9	HD	A:ALA440:O	HA
3	:[001:H10 - A:GLY550:O	1.88	HB	Conventional HB	:[001:H10	HD	A:GLY550:O	HA
4	:[001:H15 - A:GLN635:O	2.22	HB	Conventional HB	:[001:H15	HD	A:GLN635:O	HA
5	:[001:H18 - A:MET588:O	2.37	HB	Conventional HB	:[001:H18	HD	A:MET588:O	HA
6	:[001:H19 - A:ARG439:O	2.02	HB	Conventional HB	:[001:H19	HD	A:ARG439:O	HA
7	:[001:H20 - A:GLN635:O	1.85	HB	Conventional HB	:[001:H20	HD	A:GLN635:O	HA
8	:[001:H13 - A:GLN635:O	3.05	HB	Carbon HB	:[001:H13	HD	A:GLN635:O	HA
9	A:HIS593 - :[001	4.29	Hydrophobic	Pi-Pi Stacked	A:HIS593	Pi-Orbitals	:[001	Pi-Orbitals
10	A:HIS593 - :[001	4.08	Hydrophobic	Pi-Pi Stacked	A:HIS593	Pi-Orbitals	:[001	Pi-Orbitals
11	:[001 - A:ALA440	5.27	Hydrophobic	Pi-Alkyl	:[001	Pi-Orbitals	A:ALA440	Alkyl
12	:[001 - A:ALA636	5.36	Hydrophobic	Pi-Alkyl	:[001	Pi-Orbitals	A:ALA636	Alkyl
13	:[001 - A:MET637	4.37	Hydrophobic	Pi-Alkyl	:[001	Pi-Orbitals	A:MET637	Alkyl

* HB: Hydrogen Bond, HD: H-Donor, HA: H-Acceptor.

3.2. ADMET Results

Studying a drug's ADMET features entails examining its absorption, distribution, metabolism, excretion, and toxicity properties. As a result, an agent with drug-like properties is distributed throughout the body and absorbed in a time frame that allows for efficient metabolism.[36] On the other hand, after determining the physicochemical characteristics, an ADMET assessment may be completed using Lipinski's "five rules".[37,38]



[The ideal physicochemical region for oral bioavailability is the pink zone. In terms of polarity, 20 Å2<TPSA<130 Å2, SIZE: 150g/mol<MV <500 g/mol, LIPO (lipophilicity): −0.7<XLOGP3< +5.0, INSOLU (insolubility): −6<Log S (ESOL)<0, INSATU (insaturation): 0.25<Fraction Csp3<1, FLEX (flexibility): 0<Number of rotatable bonds<9. Molecules called points are thought to passively flow through the blood-brain barrier in boiled eggs. It is anticipated that molecules with points in Boiled-Egg's white will be absorbed passively by the digestive system. Molecules anticipated to be P-glycoprotein substrates are shown by blue dots. The red dots, on the other hand, represent compounds that are not anticipated to be P-glycoprotein substrates.]

Figure 5. Bioavailability radar chart and Boiled-egg graph of a) catechin, b) isoquercitrin

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SwissADME examination of the physicochemical characteristics revealed that components except for the isoquercitrin had scores that were within the norm and fit Lipinski's "five regimens" (Tables 7). The component's bioavailability radar charts were obtained online from SwissADME, as shown in Figure 3. Complete pinkness in the chosen parts denotes greater oral bioavailability and more pharmacological comparability. Accordingly, the isoquercitrin is more polar, whereas the catechin is more in saturation.[39,40] According to their pharmacokinetic characteristics, bioactive substances make for the best virtual filtration systems.[36] The GIa of catechin is considerable, according to the examination of the Boiled Egg plot (Figure 5). However, BBB and GLa permeability have not been found in isoquercitrin. P-gp is a membrane protein that extracts compounds from cells, so medications are not P-gp substrates. The tested catechin has met this condition, whereas isoquercitrin has not met this condition, according to the estimates offered in this regard (Table 7).

On the other hand, CYP (cytochrome P450), which is generated from proteins associated with pharmacokinetics, is engaged in the oxidative metabolism of medicines. Oxidized molecules can be removed because they become polar with time, especially by the kidneys. The examined components have not interacted with all CYP isoenzymes.[36,41] The catechin is also more drug-like and appropriate for use as medicine, according to a bioavailability score of 0.55. On the contrary, the low bioavailability scores of isoquercitrin (0.17) indicate their drug similarity is also low (Table 4). The GPCR ligand components were discovered to have high action. The bioactivity of catechin as an ion channel modulator has been considerable. The catechin and isoquercitrin were recognized as effective nuclear receptor ligands and potent all-purpose enzyme inhibitors. Catechin has a negligible inhibitory effect on protease inhibition (Table 7).

For potential medicinal applications, it is essential to determine a chemical substance's toxicity.[37,42-43] An *in silico* method can also be used to determine the risks associated with many different types of toxicity, such as mutagenicity, cancer, inflammation, and reproductive effectiveness. Catechin and isoquercitrin have a low potential for toxicity (Table 7).

Considering all these results, it is understood that the isoquercitrin compound can be used as a medicine according to Lipinski's rules. Because of this rule, the number of hydrogen donors and acceptors is high

Table 7. Predicted physicochemical and pharmacokinetic properties, lipinski parameters, bioactivity and bioavailability scores and toxicity risks of the catechin and isoquercitrin

Name	MW (100 ~ 600)	MV (500)	logS (-4 ~ 0.5)	logP(o/w) (0 ~ 3)	nHA (≤12)	nHD (≤7)	nRB (≤11)	TPSA (≤140)	%Abs (100%)	nVLip5R (≤1)
Catechin	290.27	261.13	-1.45	1.22	6	5	1	110.37	70.82	0.00
Isoquercitrin	464.38	416.64	-1.76	-0.54	12	8	5	210.50	36.38	2.00

Name	GI_a	BBB_p	$P\text{-}gp_s$	$CYP1A2_i$	$CYP2C19_i$	$CYP2C9_i$	$CYP2D6_i$	$CYP3A4_i$	Log K_p	Bioavailability Score
Catechin	High	No	Yes	No	No	No	No	No	-7.82	0.55
Isoquercitrin	Low	No	No	No	No	No	No	No	-8.88	0.17

Name	GPCR ligand	Ion channel modulator	Kinase inhibitor	Nuclear receptor ligand	Protease inhibitor	Enzyme inhibitor
Catechin	0.41	0.14	0.09	0.60	0.26	0.47
Isoquercitrin	0.07	-0.11	0.08	0.01	-0.07	0.47

Name	Mutagenicity	Tumorigenicity	Irritation	Reproductive
Catechin	LR	LR	LR	LR
Isoquercitrin	LR	LR	LR	LR

* MW: Molecular weight, MV: Molecular volume, logS: Log of the water solubility, logP(o/w): Log of the octanol/water partition coefficient, nHA: Number of HA, nHD: Number of HD, nRB: Number of rotatable bonds, TPSA: Topological polar surface, area %Abs = 109 − [0.345 × TPSA], nVLip5R: Number of violations according to the Lipinski "rule of five". GI_a: Gastrointestinal absorption, BBB_p: Blood-brain barrier permeant, $P\text{-}gp_s$: P-glycoprotein substrate, CYP: Cytochrome P450, Log K_p: skin permeation (cm/s). HR: Higher risk, MR: Medium risk, LR: Low risk.

4. CONCLUSION

ANS is a plant that is very resistant to drought and salty environments and is used in many areas. Biological activity and chemical content analysis of the ANS plant, which was harvested, ground, and extracted in methanol solvent, was performed. In chemical content analysis, the total phenol and flavonoid amounts were found to be low. According to the results of the applied antioxidant (posphomolybdenum reducing and free radical scavenging) and enzyme inhibition (urease) activities, it was seen that the ANS plant had high antioxidant and enzyme inhibition properties. According to the LC-MS/MS results, the experimental results were

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theoretically correct with molecular docking and ADMET applications for the catechin and isoquercitrin molecules detected in high amounts. Based on the ADMET results, these two components have minimal potential for toxicity. It is acknowledged that Lipinski's guidelines can be followed while using the isoquercitrin molecule as a medication since there are a lot of hydrogen donors and acceptors in accordance with this criterion. The MolDock score, binding affinity and constant of the molecular interaction of the isoquercitrin molecule with urease were determined to be higher than those of catechin. In later stages, active and drug precursor molecules can be discovered by isolating pure molecules from the ANS plant, which has not been studied sufficiently.

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Disclosure statement

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