IJPSR (2021), Volume 12, Issue 10 **(Research Article)**

INTERNATIONAL JOURNAL OF PHARMACEUTICAL SCIENCES AND RESEARCH

Received on 10 November 2020; received in revised form, 12 May 2021; accepted, 28 June 2021; published 01 October 2021

ANTIBACTERIAL ACTIVITY, PHARMACOKINETICS AND MOLECULAR DOCKING OF PHYTOCHEMICALS FROM *RICINUS COMMUNIS*` LEAF EXTRACT

Tsolanku Sidney Maliehe *, Sandile Nduduzo Mboyazi, Mduduzi Innocent Nqotheni, Sbongiseni Buthelezi and Jabulani Siyabonga Shandu

Department of Biochemistry and Microbiology, University of Zululand, Kwa Dlangezwa 3886, South Africa.

Keywords:

Ricinus communis, Antibacterial activity, Pharmacokinetics, Toxicity, Molecular docking

Correspondence to Author:
Tsolanku Sidney Maliehe

Department of Biochemistry and Microbiology, University of Zululand, Kwa Dlangezwa 3886, South Africa.

E-mail: sidttmaliehe@gmail.com

ABSTRACT: To evaluate the antibacterial activity, pharmacokinetics and molecular interaction of the compounds from *Ricinus communis*` leaf extract. Antibacterial activity was assessed by using the micro-dilution method. The chemical constituents of the methanolic leaf extract were assessed by using gas chromatography-mass spectrophotometry (GC-MS). The drug-like and toxicity properties of the phytocompounds were predicted by SwissADME and ADMET lab tools. AutoDock Vina was employed to investigate the binding strength of the ligand-receptor complexes. The extract displayed antibacterial activity with the lowest minimum inhibitory concentration of 1.56 mg/mL on *Bacillus spizizenii* (ATCC 6633*)*. The GC-MS showed volatile compounds, namely hexadecanoic acid, methyl ester (0.62%), tridecanoic acid (0.76%), pentafluoro propionic acid, nonyl ester (0.85%), 10-octadecanoic acid, methyl ester (2.93%) and cis-vaccenic acid (94.84%). The compounds did not inhibit isomers of cytochrome P450 (CPY) (CYP 2D6 and CYP 3A4). They were all predicted to have drug-like properties as they adhered to the Lipinski Ro5. The compounds did not show to induce any hepatotoxic effects. The compounds revealed the docking scores in the range of -4.4 to -5.7 kcal/mol. *R. communis* has the potential to be utilized as a source of therapeutic drug-like compounds.

INTRODUCTION: There is remarkable progress made towards reducing morbidity and mortality rates due to infectious diseases in most parts of the world. However, the emergence of new antimicrobial resistance mechanisms among pathogenic microorganisms does threaten human health [1].

QUICK RESPONSE CODE

DOI: 10.13040/IJPSR.0975-8232.12(10).5308-18

This article can be accessed online on www.ijpsr.com

DOI link: http://dx.doi.org/10.13040/IJPSR.0975-8232.12(10).5308-18

Major reasons for resistance to antimicrobials are an inappropriate use of antimicrobials, intake of improper dosages and excessive use of antimicrobials [2]. Microorganisms resist drug effects through enzymatic inactivation of the drug molecules, modification of target sites and extrusion by efflux pump [3]. Moreover, most of the allopathic drugs are costly and turn to induce adverse side effects [4]. Confronted with these challenges, it is imperative to search for new compounds with inhibitory activities characterized by high potency, improved mechanisms of action and less side effects. Medicinal plants are the predominantly used natural sources of pharmacologically active compounds in modern-

day drug discoveries [5]. The plant-based molecules are recognized for their broad spectrum of pharmacological activities that include antimicrobial and immune-suppressant effects, among others [6]. Their pharmacotherapeutic activities are due to their diverse bioactive compounds, including alkaloids, tannins, saponins, glycosides and flavonoids [7]. Nevertheless, many of the newly discovered compounds are banned at the clinical stages of drug discovery processes. This is mainly due to the poor pharmacokinetic properties and toxicity displayed by most of these compounds [8]. The pharmacokinetic properties of molecules describe their absorption, distribution, metabolism, and excretion characteristics. These parameters are summed up by the term ADME or better ADMET when toxicity profiles are included [9]. The evaluation of the ADMET parameters at the early stages of drug discovery and development can decrease the pharmacokinetic-related fiascos and financial losses. Although conventional methods are used to evaluate pharmacokinetics, they take a considerable length of time and are costly [10]. Recently, computational methods are considered the important primary step in the evaluation of suitable drug-like compounds and prevention of time wastage and expenses on drug candidates that would have undesirable effects [11]. Moreover, these computational tools are widely used in the analysis of the binding energetics, molecular interactions, and conformational changes [12]. Molecular docking is widely used to predict the conformation of ligands within the receptor proteins with a significant degree of certainty [13].

Ricinus communis is a medicinal plant from the Eurphobiceae family [14]. It is recognized for its therapeutic properties, including antimicrobial, anti-inflammatory, antioxidant, and wound healing properties [15]. The phytochemical screening has revealed the presence of different classes of compounds that include flavanols, alkaloids, glycosides, flavonoids, steroid and saponin in different parts [16]. Although many studies are documenting its pharmacological properties that include antimicrobial activity, there are limited studies on assessing pharmacokinetic properties and molecular interactions with its phytochemicals.

The study aimed to evaluate the antibacterial activity, ADMET properties and molecular interactions of phytochemicals from *Ricinus communis`* leaf extract with the target protein. The chemical constituents were identified using the standard biochemical techniques and gas chromatography-mass spectrophotometry (GC-MS). The *in-vitro* antibacterial activity was assessed by using the dilution method. Lastly, the ADMET properties of the identified phytochemicals and their molecular interaction with the target protein were investigated using SwissADME, ADMET lab and AutoDock Vina tools.

EXPERIMENTAL:

Study Area: The study was conducted in the Department of Biochemistry and Microbiology at the University of Zululand, KwaZulu-Natal, South Africa, from July 2019 to the end of February.

Chemicals and Media: The chemicals and media that were used in this study were of analytical grades. They were procured from Merck (Ltd) Pty and Sigma Aldrich Co. Ltd (Steinem, Germany).

Sample Preparation and Extraction: *Ricinus communis`* leaves were collected from Felix ton, Empangeni in KwaZulu-Natal, South Africa. The sample was transported to the University of Zululand, South Africa. The leaves were authenticated by Dr. Ntuliat, the University Herbarium. The plant was allocated the specimen voucher SNM01. Thereafter, the leaves were washed, dried at room temperature and milled to powder. Ten grams of the powdered material was extracted with methanol (100 mL) for 72 hours at the shaking incubator at 25°C, 110 rpm. The extract was filtered by Whatman no. 1 filter paper and evaporated under a stream of air at room temperature [17].

Antibacterial Activity: The minimum inhibitory concentration (MIC) of the extract was evaluated by serial microdilution using 96-well microplate [18]. The wells in row A were pipetted with 50 µL of Mueller-Hinton Broth (MHB) and had 50 µL of 50 mg/mL of the extract. Rows B through H was filled with 50 µL of MHB. A serial dilution was carried out with corresponding final concentration of 0.39 mg/mL. Thereafter, the wells were added with 50 µL of fresh bacterial suspension (10^6cfu/mL). The bacterial strains were *Proteus mirabilis* (ATCC

25933) and *Bacillus spizizenii* (ATCC 6633). Ciprofloxacin was used as positive control, while 10% dimethyl sulfoxide served as a negative control. The plate was sealed with parafilm and incubated at 37°C overnight. After incubation, the wells were added with 30 µL p-iodonitrotetrazolium violet (4 mg/mL). The MIC was taken as the lowest concentration that displayed clear wells, indicative of the absence of bacterial growth.

Phytochemical Analysis: The phytochemical screening of the crude extract was done for the detection of different classes of phytocompounds [20]. The evaluation of volatile phytochemicals was performed by using gas chromatography-mass spectrometry (THERMO Gas Chromatography TRACE ULTRA VER: 5.0, Germiston, SA). The helium gas flow rate was set to 1 mL per minute, with a spit ratio of 1:50. The temperature of the injection port was set to 250°C with the temperature of the detector put at 280°C. The column temperature was put at 40 °C for 1 min and then raised from 40 - 120°C. After that, 2 µL of the extract was injected for investigation. The mass spectra required in the scan mode were 70 eV. The identification of the phytochemicals was confirmed based on the retention indexes and mass spectra that were compared to those attained from the libraries. The evaluation was performed two times [15].

Physicochemical and Pharmacokinetic Properties: The SwissADME tool was used to evaluate the physicochemical and pharmacokinetic properties of the identified compounds. The simplified molecular-input line-entry system (SMILES) of each compound was obtained from Pub Chem and inserted into the SwissADME tool. Thereafter, the physicochemical properties such as the molecular weight (MW), number of hydrogen bond acceptors (N_{HBA}), hydrogen bond donors (N_{HBD}) and rotatable bonds (N_{RB}). The lipophilicity was evaluated using consensus logP (cLog*P*) estimation. The solubility (Log*S*) was investigated using three models, namely ESOL, Ali and SILICOS-IT. The pharmacokinetic properties such as human intestinal absorption (HIA), penetration of the blood-brain barrier (BBB), the interaction of compounds with P-glycoprotein (P-gp) and metabolism were predicted [20, 21].

Drug-Likeness Prediction: The drug-likeness of the compounds was investigated in accordance with the Lipinski rule of five (Ro5) using Swiss ADME tool [21]. According to Ro5, drug-like molecules ought to have MV ≤ 500 Da, log *P*-value≤ 5, N_{HBD} ≤ 5, and N_{HBA} must be ≤ 10. In addition, the N_{RB} ought to be ≤ 10 and compounds must not violate more than one of the Ro5 [22]. The bioavailability scores of the phytochemicals were computed based on their MV, cLog*P*, N_{HBD} and N_{HBA} [21, 22].

Bioactivity Score: Bioactivity scores of the compounds were predicted using the software Molin spiration score online. The bioactive scores were evaluated against G protein-coupled receptors (GPCR ligand), ion channel modulator, a kinase inhibitor, nuclear receptor ligand, protease inhibitor, and enzyme inhibitor [23, 24].

Toxicological study: The toxicological properties were calculated using the online server ADMET lab online tool. The simplified molecular-input line-entry system (SMILES) for each compound was procured from Pub Chem and inserted into ADMET lab. Thereafter, the human hepatotoxicity (HHT) and skin sensitivity (Skin Sen) of the compounds were evaluated [25].

Molecular Docking: Retrieval and Preparation of The Receptors: The three-dimensional structures of the three target receptor proteins were retrieved from the Protein Data Bank (PDB). The receptor protein was FtsZ (PDB ID 4DDQ), a protein that plays an important role in bacterial cell division [26].

The structures were loaded into the Bio *via* Discovery Studio 4.1 Visualizer. The water molecules, heteroatoms and ligands were deleted. Thereafter, polar hydrogen atoms were added to the protein [27].

Ligand Preparation: The structures of the identified compounds were procured from the National Centre for Biotechnology Information (NCBI) PubChem compound database. The ligands were energy minimized using Chimera 1.14. Thereafter, the PDB files of the structures were converted to PDBQT files using Auto Dock tools.

Molecular Docking and Visualisation of the Complexes: The docking of the target protein with

the ligands was done using AutoDock Vina [28]. Docking was performed to get a set of possible conformations and orientations for the ligand at the binding site. The ligand displaying the lowest binding affinity was chosen as the best conformation. The docked protein-ligand complexes were visualized using Bio *via* Discovery Studio 4.1 Visualizer. For analyzing the interaction in the complexes, surfaces were created close to the ligand and the hydrogen and other interactions were evaluated [27].

Statistical Analysis: The experiments were performed in triplicates and the data was reported as mean ± standard deviation. The analyses were done by one-way analysis of variance and regarded to be significantly different at $p < 0.05$.

RESULTS AND DISCUSSION:

Antibacterial Activity: The antibacterial activity of the extract is shown in **Table 1**. *B.* spizizenii (ATCC 6633) was more susceptible to the antibacterial effect of the extract, presenting a lower MIC of inhibition 1.56 mg/mL as compared to the MIC of *P. mirabilis* (ATCC 25933), which was 6.25 mg/mL.

It is generally accepted that Gram-negative bacteria are less susceptible to antimicrobials in comparison to the Gram-positive bacteria. The difference is associated with the properties of the outer membrane of the Gram-negative bacteria that have a surface with strong hydrophilicity that acts as a robust permeability barrier [29].

TABLE 1: MINIMUM INHIBITORY CONCENTRATION (mg/mL) OF THE EXTRACT

Bacteria	Extract	Ciprofloxacin
B. spizizenii(ATCC 6633)	1.56	1.56
P. mirabilis (ATCC 25933)	6.25	3.13

Phytochemical Classes from the Methanol Extract: The screening of different classes of the compounds in the leaf material was performed and the results are illustrated in **Table 2.** The presence of all classes (alkaloids, flavonoids, glycosides, saponins, steroids, tannins, and terpenoids) evaluated was observed**.** The screened phytochemicals are reported to possess pharmacologically important properties that include antibacterial and cytotoxic activities [30]. The antibacterial activity is owed to the presence of these phytochemicals.

TABLE 2: PRELIMINARY PHYTOCHEMICAL SCREENING OF THE LEAF MATERIAL

Phytochemicals	Results
Alkaloids	+ + +
Flavonoids	+ + +
Glycosides	+ +
Saponins	+
Steroids	+ +
Tannins	+ +
Terpenoids	+ +

Key: +: weakly positive; ++: moderately positive; +++: strongly positive

Volatile Compounds from the Extract: The methanolic extract under GC-MS chromatogram gave a yield of 5 volatile compounds **Table 3**. Hexadecenoic acid, methyl ester (0.62%), tridecanoic acid (0.76%), pentafluoro propionic acid, nonyl ester (0.85%), 10-octadecanoic acid, methyl ester (2.93%) and cis-vaccenic acid (94.84%) as the major component. Previous studies have reported all of these compounds to possess antimicrobial activity [31, 32]. Thus, the antimicrobial activity observed in this study was attributed to the presence of these compounds [35].

TABLE 3: THE CHEMICAL CONSTITUENTS OF THE EXTRACT

Compounds	Area (%)
Hexadecenoic acid, methyl ester	0.62
Tridecanoic acid	0.76
Pentafluoro propionic acid, nonyl ester	0.85
10-Octadecenoic acid methyl ester	2.93
cis-Vaccenic acid	94.84

Molecular Weight (MV) of the Compounds: The permeability and bioavailability of drug molecules are influenced by the molecular property called molecular weight (MV). The weightier the molecule (greater than 500 g/mol), the lower the permeability and bioavailability are and *vice versa* [18]. The molecular weights of the compounds are shown in **Table 4**. It was observed that all compounds have less than 500 g/mol, implying the potential for good bioavailability.

N_{hba} and N_{hbd} of the Compounds: Hydrogen bonds are recognized for their important role in membrane permeation and absorption of molecules in the gastro-intestines [36]. In addition, hydrogen bond interactions have an influence on the ligand-receptor binding affinity. Hydrogen bond strength in the molecules was estimated by calculating the N_{HBD} (sum of OHs and NHs) and N_{HBA} (sum of Os and Ns) and the results are presented in **Table 4**.

Molecules with $N_{HBD} \leq 5$ and $N_{HBA} \leq 10$ have high probabilities of membrane permeability and bioavailability [37]. This means that all compounds evaluated in this study have the potential to be bioavailable as they have $N_{HBD} \leq 5$ and $N_{HBA} \leq 10$.

TABLE 4: THE PHYSICOCHEMICAL PROPERTIES AND LIPOPHILICITY OF IDENTIFIED COMPOUNDS

Compounds	Properties				
	MV (g/mol)	N_{HBD}	N_{HBA}	N_{RB}	cLog*P*
Hexadecenoic acid, methyl ester	270.45	0	2	15	5.54
Tridecanoic acid	214.34	1	2	11	4.10
Pentafluoro propionic acid, nonyl ester	290.27	0	7	11	4.86
10-Octadecenoic acid methyl ester	296.49	0	2	16	5.93
cis-Vaccenic acid	282.46	1	2	15	5.68

N_{RB} of the Compounds: Rotatable bonds are defined as single bonds that are not found in aromatic rings but bound to nonterminal heavy atoms. The N_{RB} contributes to molecular rigidity or flexibility, which directly affects oral bio-availability and the strength of intermolecular interactions [38]. The N_{RB} were counted, and the results are shown in **Table 4**. The compounds had the N_{RB} in the range of 11 to16. According to [39], compounds with N_{RB} greater than 10 turns to display poor drug bioavailability. A high N_{RB} (greater than 10) enhance high flexibility, consequently leading to poor bioavailability.

Lipophilicity (cLog*P*) of the compounds: Lipophilicity (expressed as cLog*P*) is an important parameter that influences drug absorption in the body. High cLog*P*-values (5≤ Log*P*) is associated with poor absorption and *vice versa* [40]. The cLog*P*-values of the compounds are displayed in **Table 4.** Tridecanoic acid and pentafluoro propionic acid, nonyl ester were found to have a cLog *P*-value less than 5. This means these compounds have a high probability of being absorbed and bioavailable. Hexadecenoic acid, methyl ester, 10-octadecenoic acid methyl ester and cis-vaccenic acid have cLog*P*-value greater than 5, indicative of the likelihood of these compounds to have problematic permeability and bioavailability [41].

Aqueous Solubility of the Compounds: Aqueous solubility measures the compounds' solubility in water at the temperature of 25°C. Low solubility turns to be a limiting factor in drug discovery and development [11]. The solubility of the compounds was predicted and the results are shown in **Table 5**.

The results reveal the phytochemicals to be poorly soluble to soluble depending on the Log*'s* prediction model used. Thus, the compounds have the potential to be bioavailable in the body as they are all soluble to some degree [42].

TABLE 5: SOLUBILITY PREDICTIONS OF IDENTIFIED COMPOUNDS

Properties	Compounds				
	Hexadecenoic acid, methyl ester	Tridecanoic acid	Pentafluoro propionic acid, nonyl ester	10-Octadecenoic acid methyl ester	cis-Vaccenic acid
Log*S* (ESOL)	-5.02	-3.95	-4.51	-5.32	-5.41
Class	Moderately soluble	Soluble	Moderately soluble	Moderately soluble	Moderately soluble
Log*S* (Ali)	-7.77	-6.11	-6.03	-7.83	-8.26
Class	Poorly soluble	Poorly soluble	Poorly soluble	Poorly soluble	Poorly soluble
Log*S* SILICOS-IT	-6.01	-4.10	-4.68	-6.09	-5.39
Class	Poorly soluble	Moderately soluble	Moderately soluble	Poorly soluble	Moderately soluble

Gastrointestinal Absorption (GIA) of the Compounds: Gastrointestinal absorption (GIA) is a vital property for the drug molecules that are orally administered.

It depicts the absorption of molecules administered from the gastro-intestine into the bloodstream. All compounds except pentafluoro propionic acid, nonyl ester are predicted to be absorbed in the gastrointestinal tract **Table 6.** Since, most of the drug molecules are orally administered, they are required to be highly absorbed in the intestinal tissue [42].

Interestingly, all tested compounds except pentafluoro propionic acid, nonyl ester, can be easily absorbed in the intestines upon oral administration.

Blood-Brain Barrier (BBB) Permeability: The BBB is the endothelial cell layer found in the brain and is responsible for separating the brain from the blood [44]. The ability of the compounds to cross BBB was assessed and the results are shown in **Table 6**. Hexadecenoic acid, methyl ester and tridecanoic acid are presumed to permeate BBB, whereas pentafluoro propionic acid, nonyl ester, 10-octadecenoic acid methyl ester and cis-vaccenic acid are not able to cross BBB. BBB is important for drug molecules that target mainly the brain cells [45]. This implies that the three compounds that could cross the BBB can be effectively administered and used to treat brain-related disorders.

TABLE 6: PHARMACOKINETIC PARAMETER OF IDENTIFIED COMPOUNDS

Compounds	Properties							
	GIA	BBB Permeant	P-gp substrate	CYP 1A2 inhibitor	CYP 2C19 Inhibitor	CYP 2C9 Inhibitor	CYP 2D6 inhibitor	CYP 3A4 Inhibitor
Hexadecanoic acid, methyl ester	High	Yes	No	Yes	No	No	No	No
Tridecanoic acid	High	Yes	No	Yes	No	No	No	No
Pentafluoropropionic acid, nonyl ester	Low	No	No	No	Yes	No	No	No
10-Octadecenoic acid methyl ester	High	No	No	Yes	No	No	No	No
cis-Vaccenic acid	High	No	No	Yes	No	Yes	No	No

P-Glycoprotein (P-Gp) Substrates: P-gp in the cell membrane is responsible for drug transportation out of the cells [46]. The ability of the identified phytocompounds to act as substrates were predicted and the results were presented in **Table 6**. The five identified compounds were found not to be P-gp substrates. The efflux action of P-gp turns to render most cells resistant to the effect of drug molecules as it pumps them out of the cells and lowers their concentration, consequently their efficacy[47]. Since all compounds are P-gp substrates, it is highly possible that their pharmacologic effects can drastically be reduced the efflux action of P-gp.

Metabolism of the Compounds: There are a group of enzymes in the liver called cytochrome P450 (CPY) family that are responsible for drug metabolism of drugs and their excretion through bile or urine. Out of the 17 families of CYP enzymes, CYP 1, 2 and 3 are mainly responsible for metabolising drug molecules [48]. The effects of compounds on CYP 1A2, CYP 2C19, CYP 2C9, CYP 2D6 and CYP 3A4 are shown in Table 6. All compounds except pentafluoro propionic acid, nonyl ester were predicted inhibitors of CYP 1A2. However, they did not show the potential to inhibit CYP 2D6 and CYP 3A4. CYP 2C19 was only inhibited by pentafluoro propionic acid, nonyl ester cis-Vaccenic acid showed potential to inhibit CYP 2C9. The inhibition of these enzymes would affect the biodegradation of the compounds, consequently leading to adverse side effects[49].

Drug-Likeness of the Compounds: Drug-likeness is a concept that assesses the relationship between the structural, physicochemical and pharmaco-kinetic properties of the compounds [50]. The drug-likeness of the compounds was assessed using the Lipinski Ro5. The Lipinski Ro5 states that the drug-like compounds should have the MV, cLog*P*, nHBA, and n HBD of the compounds should be no more than 500 g/mol, 5, 10 and 5, respectively. Moreover, the compounds ought not to violate more than one of the Ro5 [22]. The drug-like properties of the identified compounds are displayed in **Table 7**. All compounds adhere to the Lipinski Ro5. This means that all compounds can be categorized as drug-like molecules.

TABLE 7: DRUG-LIKENESS AND BIOAVAILABILITY SCORES OF THE COMPOUNDS

Compounds	Lipinski's rule		
	Satisfactory	Number of violations	Bioavailability score
Hexadecanoic acid, methyl ester	Yes	1	0.55
Tridecanoic acid	Yes	0	0.56
Pentafluoropropionic acid, nonyl ester	Yes	0	0.55
10-Octadecenoic acid methyl ester	Yes	1	0.56
cis-Vaccenic acid	Yes	1	0.56

Bioavailability Scores of the Compounds: The bioavailability score defines the permeability and bioavailability characteristics of possible drug candidates [51]. All compounds showed a bio-availability score of 0.55 and 0.56 **Table 7**. Many factors that control the entry of drug molecules from the gastrointestinal tract to the blood system often reduce the amounts of the bioavailable drugs. However, the tested compounds have desirable bioavailability scores (≥ 0.50).

Bioactivity Scores of the Compounds: Bioactivity scores the compounds as the nuclear receptor ligand; kinase inhibitor and protease inhibitor were predicted by using Molinspiration. The predicted bioactivity scores are given in **Table 8**. The drug-like molecules are esteemed as active when the scores are greater than 0, moderately active when the scores are in the range of -5.0 - 0.0, and inactive when the scores are less than -5.0 [52].

All compounds showed to be moderately active as the kinase inhibitors. This implies that all compounds can be used to treat adverse conditions caused by hyperactive protein kinases. Pentafluoro propionic acid, nonyl ester and is-vaccenic acid were found to be active nuclear receptors and protease inhibitors, whereas the other ligands were moderately active. In accordance with the results, the compounds can reduce the hyper-active protease effects to some degree, which can lead to the managed biological homeostasis. Moreover, nuclear receptor molecules are used to regulate some metabolic disorders such as cancer [53]. Thus, the compounds have the potential to be utilized as therapeutic agents against such diseases**.**

TABLE 8: BIOACTIVITY SCORES OF THE IDENTIFIED COMPOUNDS

Compounds	Bioactivity scores		
	Kinase inhibitor	Nuclear receptor ligand	Protease inhibitor
Hexadecanoic acid, methyl ester	-0.34	-0.09	-0.13
Tridecanoic acid	-0.62	-0.15	-0.27
Pentafluoropropionic acid, nonyl ester	-0.42	0.16	0.18
10-Octadecenoic acid methyl ester	-0.25	-0.06	-0.02
cis-Vaccenic acid	-0.22	0.23	0.07

Toxicological Properties of the Compounds: Toxicity is a measure of adverse effects induced by compounds on living organisms and their ecosystem [11]. The human liver is the central site of drug metabolism and is thus exposed to the adverse effects that may be induced by various drug-like compounds [54]. The human hepatotoxicity (HHT) of the compounds was evaluated, and the results are demonstrated in **Table 9**. The compounds are all predicted to be non-hepatotoxic. This implies that the compounds have biosafety and the inability to cause liver dysfunction and failure**.**

TABLE 9: TOXICOLOGICAL PROPERTIES OF THE COMPOUNDS

Compounds	Toxicity	
	HHT	SkinSen
Hexadecanoic acid, methyl ester	Negative	Positive
Tridecanoic acid	Negative	Negative
Pentafluoropropionic acid, nonyl ester	Negative	Positive
10-Octadecenoic acid methyl ester	Negative	Positive
cis-Vaccenic acid	Negative	Positive

Skin reactions such as sensitization and irritation are widely known hazards of chemicals. Skin sensitization is a fatal reaction triggered by the repetitive exposure of the skin to the sensitizing compound [55]. The skin sensitivity to the identified phytochemicals was predicted and the results are shown in **Table 9.** Tridecanoic acid is the only compound found as non-skin sensitizer, whereas other compounds are skin sensitizers. This implies that only safety measures that include skin protectors ought to be considered upon the use of these compounds except when tridecanoic acid.

Binding Scores of the Compounds: Molecular docking uses scoring algorithms to predict the likelihood of ligands to bind to target proteins. The ligand-protein binding ability occurs spontaneously when the free energy charge is negative [56]. The binding scores of the ligand-receptor complexes are presented in **Table 10**. Pentafluoro propionic acid, nonyl ester-Fts Z interaction had the largest negative free energy values of -5.7kcal/mol followed by hexadecenoic acid, methyl ester-FtsZ (-4.77kcal/mol), cis-vaccenic acid-FtsZ (-4.77kcal/mol), tridecanoic acid-FtsZ (-4.57kcal/mol) and 10-octadecanoic acid, methyl ester-FtsZ (-4.47 kcal/mol) interactions.

The observed results suggested that all the tested ligands have the potential to form stable complexes with the target protein, consequently resulting in the inhibition of the bacteria. However, the drug used as a control cipro ofloxacin had the highest negative free energy (-7.5 kcal/mol), indicative of better binding ability in comparison to the test ligands.

TABLE 10: MOLECULAR DOCKING SCORES AND INTERMOLECULAR MOLECULAR INTERACTIONS BETWEEN THE LIGANDS AND THE RECEPTOR

Compounds	Binding scores (kcal/mol)	H-bonds interaction residues	Number of other interacting residues
Hexadecanoic acid, methyl ester	-4.7	GLY A:21, THR A:109, GLY A:108, GLY A:110	16
Tridecanoic acid	-4.5	GLY A:108, GLY A:110, GLY A:104, GLY A:21, THR A:109	11
Pentafluoropropionic acid, nonyl ester	-5.7	GLY A:21, GLY A:21, GLY A:108, GLY A:108, GLY A:108, GLY A:110, THR A:109, THR A:109	15
10-Octadecanoic acid, methyl ester	-4.4	ARG A:143, GLY A:108	11
cis-Vaccenic acid	-4.7	GLY A:22,VAL A:19, THR A:109, GLY A:110	14
Ciprofloxacin	-7.5	GLY A:104, GLY A:23, GLY A:110, GLY A:21	11

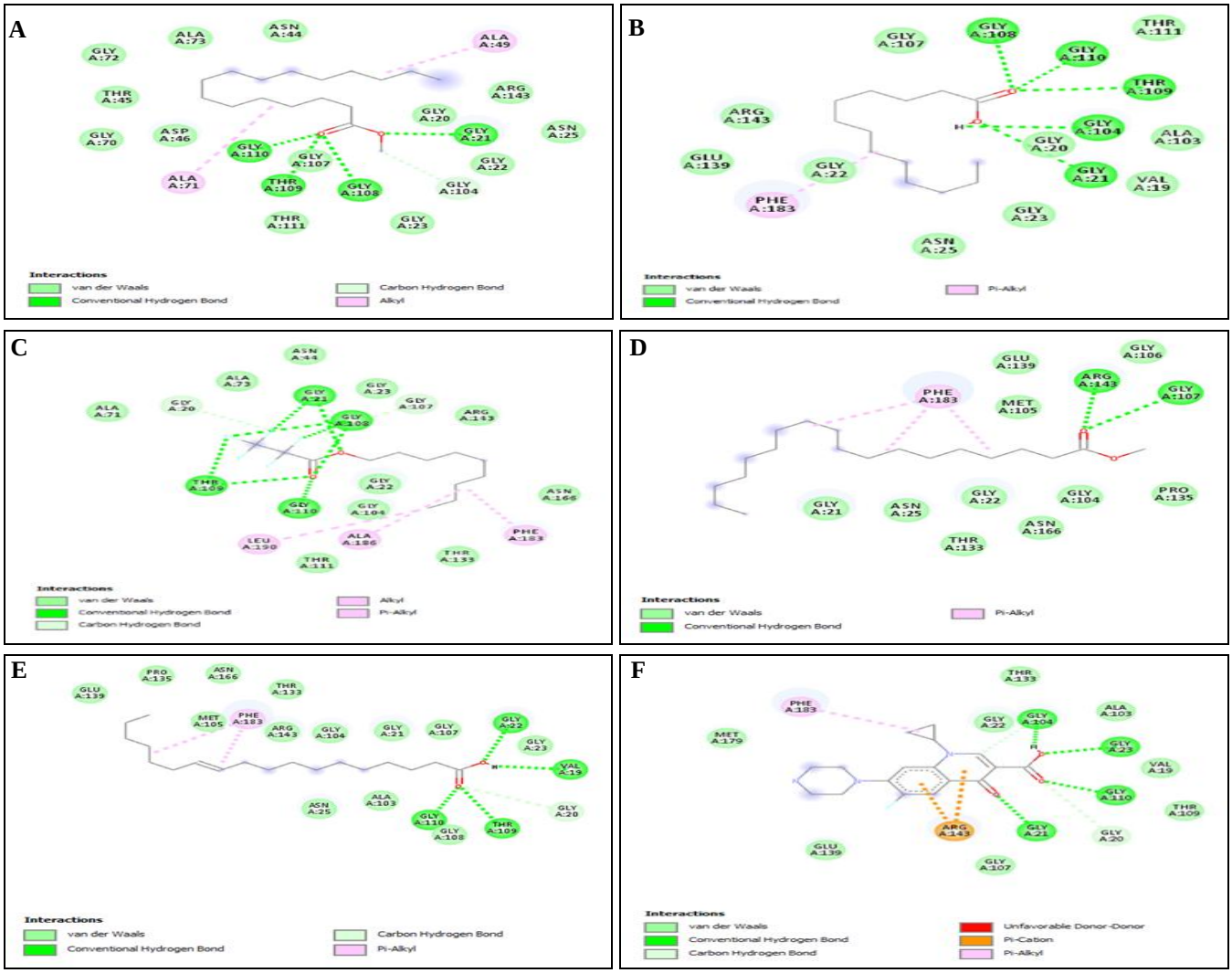

FIG 1: THE 2D STRUCTURES OF DOCKED COMPLEXES. (A) HEXADECANOIC ACID, METHYL ESTER-FTSZ, (B) TRIDECANOIC ACID-FTSZ, (C) PENTAFLUOROPROPIONIC ACID, NONYL ESTER-FTSZ, (D) 10-OCTADECANOIC ACID, METHYL ESTER-RECEPTOR, CIS-VACCENIC ACID-FTSZ, AND (F) CIPROFLOXACIN-FTSZ

Molecular Bonds of the Ligand-Receptor Complexes: The binding energy values of the ligand-receptor complexes are the results of the different types of molecular bonds formed. The various ligand-receptor interactions are displayed in **Table 10**, **Fig. 1** and **Fig. 2**. Hexadecenoic acid, methyl ester demonstrated four hydrogen bonds with different amino acid residues (GLY A:21, THR A:109, GLY A:108, and GLY A:110). The compound also formed van der Waals, carbon-hydrogen bonds and alkyl bonds. Tridecanoic acid interacted with FtsZ protein via five hydrogen bonds with GLY A: 108, GLY A: 110, GLY A: 104, GLY A:21 and THR A:109. It also showed pi-alkyl interaction with PHE A: 183 and van der Waals interactions with other amino acid residues. Pentafluoro propionic acid, nonyl ester formed eight hydrogen bonds, the residues GLY A: 21, GLY A:108, GLY A:110, and THR A:109. This ligand-receptor complex also formed van der Waals, carbon-hydrogen bond, alkyl, and pi-alkyl bonds. The10-Octadecanoic acid, methyl ester has two H- bonds with ARG A:143 and GLY A:1082, van der Waal interactions, and pi-alkyl bonds (PHE A: 183). the cis-Vaccenic acid-FtsZ complex is supported by four hydrogen bonds at residues GLY A:22, VAL A:19, THR A:109 and GLY A:110, van der Waals interactions, carbon-hydrogen bond, and pi-alkyl bonds. Ciprofloxacin formed four hydrogen bonds with GLY A: 104, GLY A:23, GLY A:110 and GLY A:21, van der Waal interactions, carbon-hydrogen bond, pi-alkyl bond (PHE A:183), pi-cation bonds (ARG A:143), and unfavorable bonds. The ligand-receptor complexes were steadied by hydrogen bonds that include O-H and C=O groups. The test ligands served as donors and acceptors. It was concluded that the ligands revealed antibacterial activity by interacting with the functional groups of the FtsZ protein [57].

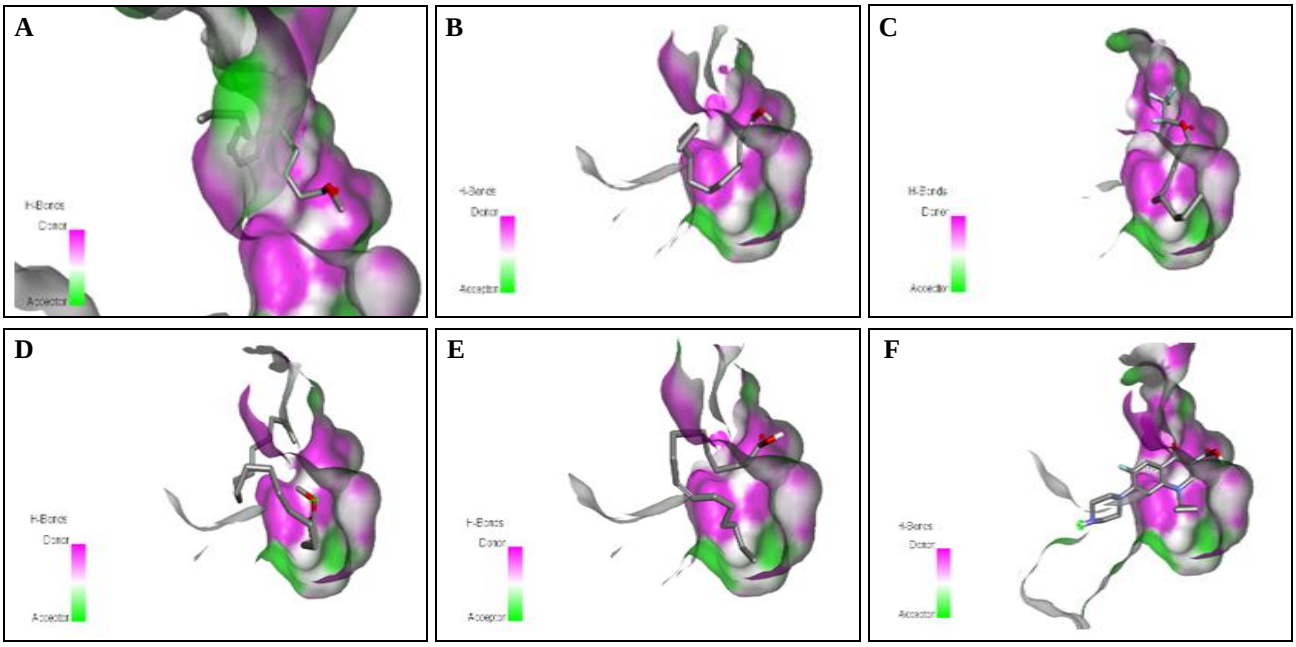

FIG 2: THE 3D STRUCTURES OF DOCKED LIGANDS INSIDE THE BINDING POCKET OF THE RECEPTOR. (A) HEXADECENOIC ACID, METHYL ESTER-FTSZ, (B) TRIDECANOIC ACID-FTSZ, (C) PENTAFLUORO PROPIONIC ACID, NONYL ESTER-FTSZ, (D) 10-OCTADECANOIC ACID, METHYL ESTER-FTSZ, CIS-VACCENIC ACID-FTSZ, AND (F) CIPROFLOXACIN-FTSZ

CONCLUSION: *B.* spizizenii (ATCC 6633) was more susceptible to the antibacterial effect of the extract with the lowest MIC value of 1.56 mg/ml. The volatile compounds obtained were hexadecenoic acid, methyl ester, tridecanoic acid, pentafluoro propionic acid, nonyl ester, 10-octadecanoic acid, methyl ester and cis-vaccenic acid, which are all reported to have antimicrobial activity. All compounds were predicted to be bioavailable and were also found to have drug-like properties. These compounds do not display any hepatotoxic effects. The ligands were predicted to demonstrate antibacterial activity by interacting with the functional groups of the FtsZ protein. For future studies, the *in vitro* and *in vivo* studies of the identified volatile phytocompounds are recommended.

ACKNOWLEDGEMENT: The authors acknowledge the Department of Biochemistry and Microbiology, University of Zululand and National Research Foundation of South Africa (Grant number: SFH 180531338367) facilities and funds to conduct this research.

CONFLICTS OF INTEREST: No conflict of interest associated with this work.

CONTRIBUTION OF AUTHORS: We declare that this work was done by the authors named in this article and the authors will bear all liabilities about claims relating to the content of this article.

REFERENCES:

1. De Oliveira DM, Forde BM, Kidd TJ, Harris PN, Schembri MA, Beatson SA, Paterson DL and Walker MJ: Antimicrobial resistance in ESKAPE pathogens. Clinical Microbiology Reviews 2020; 33(3): 00181-19.
2. Mama M, Mamo A, Usman H, Hussen B, Hussen A and Morka G: Inappropriate antibiotic use among inpatients attending Madda Walabu University Goba Referral Hospital, Southeast Ethiopia: implication for future use. Infection and Drug Resistance 2020; 13: 1403.
3. Farhat N, Ali A, Bonomo RA and Khan AU: Efflux pumps as interventions to control infection caused by drug-resistance bacteria. Drug Discovery Today 2020 Oct 1.
4. Parasuraman S: Herbal drug discovery: challenges and perspectives. Current Pharmacogenomics and Personalized Medicine Formerly Current Pharmac 2018; 16(1): 63-8.
5. Srivastava SK and Singh NK: General overview of medicinal and aromatic plants: A. Journal of Medicinal Plants 2020; 8(5): 91-3.
6. Mboyazi SN, Nqotheni MI, Maliehe TS and Shandu JS: *In vitro* antibacterial and *in silico* toxicity properties of phytocompounds from *Ricinus communis* Leaf Extract. Pharmacognosy Journal 2020; 12(5).
7. Yu Sheng Toh E, Lim CL, Pick Kiong Ling A, Chye SM and Koh RY: Overview of the Pharmacological Activities of Aframomum melegueta. Pertanika Journal of Tropical Agricultural Science 2019 Feb 1; 42(1.
8. He C and Wan H: Drug metabolism and metabolite safety assessment in drug discovery and development. Expert Opinion on Drug Metab & Toxi 2018; 14(10): 1071-85.
9. Mignani S, Rodrigues J, Roy R, Shi X, Ceña V, El Kazzouli S and Majoral JP: Exploration of biomedical dendrimer space based on *in-vivo* physicochemical parameters: Key factor analysis (Part 2). Drug Discovery Today 2019; 24(5): 1184-92.
10. Williams M, Mullane K and Curtis MJ: Addressing reproducibility: peer review, impact factors, checklists, guidelines, and reproducibility initiatives. In Research in the Biomedical Sciences 2018; 197-306.
11. Pathak RK, Singh DB, Sagar M, Baunthiyal M and Kumar A: Computational Approaches in Drug Discovery and Design. In Computer-Aided Drug Design 2020; 1-21.
12. Siebenmorgen T and Zacharias M: Computational prediction of protein–protein binding affinities. Wiley Interdisciplinary Reviews: Computational Molecular Science 2020; 10(3): 1448.
13. Santos LH, Ferreira RS and Caffarena ER: Integrating molecular docking and molecular dynamics simulations. In Docking screens for Drug Discovery, Humana New York NY 2019; 13-34.
14. Abdul WM, Hajrah NH, Sabir JS, Al-Garni SM, Sabir MJ, Kabli SA, Saini KS and Bora RS: Therapeutic role of Ricinus communis L. and its bioactive compounds in disease prevention and treatment. Asian Pacific Journal of Tropical Medicine 2018; 11(3): 177.
15. Ahmed SR, Roy R, Romi IJ, Hasan M, Bhuiyan MK and Khan MM: Phytochemical screening, antioxidant and antibacterial activity of some medicinal plants grown in Sylhet region. IOSR Journal of Pharmacy and Biological Sciences IOSR-JPBS 2019; 14(1): 26-37.
16. Naz R and Bano A: Antimicrobial potential of Ricinus communis leaf extracts in different solvents against pathogenic bacterial and fungal strains. Asian Pacific Journal of Tropical Biomedicine 2012; 2(12): 944-7.
17. Fa O, Et O, Io O and Ef O: Antimicrobial activity and phytochemical screening of leaf extracts of *Catharanthus roseus* against aspergillus Niger. Int J Pure Appl Zool 2019; 7: 12-7.
18. Mboyazi SN, Nqotheni MI, Maliehe TS and Shandu JS: In vitro Antibacterial and In silico Toxicity Properties of Phytocompounds from *Ricinus communis* Leaf Extract. Pharmacognosy Journal 2020; 12(5).
19. Harbone JB: Extraction and Estimation of isolated from the rhizosphere of coconut palms Chlorophylls.
20. Daina A, Michielin O and Zoete V: iLOGP: a simple, robust, and efficient description of n-octanol/water partition coefficient for drug design using the GB/SA approach. Journal of chemical information and modeling. 2014; 54(12): 3284-301.
21. Barreau N, Marsillac S, Bernede JC and Barreau A: Investigation of β-In2S3 growth on different transparent conductive oxides. Applied Sur Sci 2000; 161(1-2): 20-6.
22. Verma A: Lead finding from *Phyllanthus debelis* with hepatoprotective potentials. Asian Pacific Journal of Tropical Biomedicine 2012; 2(3): S1735-7.
23. Nadeem S, Sirajuddin M, Ahmad S, Tirmizi SA, Ali MI and Hameed A: Synthesis, spectral characterization and *in vitro* antibacterial evaluation and Petra/Osiris/ Molinspiration analyses of new Palladium (II) iodide complexes with thioamides. Alexandria Journal of Medicine 2016; 52(3): 279-88.
24. Dong J, Wang NN, Yao ZJ, Zhang L, Cheng Y, Ouyang D, Lu AP and Cao DS: ADMETlab: a platform for systematic ADMET evaluation based on a comprehensively collected ADMET database. Journal of Cheminformatics 2018; 10(1): 29.
25. Acar C, Yalçın G, Ertan-Bolelli T, Onurdağ FK, Ökten S, Şener F and Yıldız İ: Synthesis and molecular docking studies of some novel antimicrobial benzamides. Bioorganic Chemistry 2020; 94: 103368.
26. Ronkin SM, Badia M, Bellon S, Grillot AL, Gross CH, Grossman TH, Mani N, Parsons JD, Stamos D, Trudeau M and Wei Y: Discovery of pyrazolthiazoles as novel and potent inhibitors of bacterial gyrase. Bioorganic & Medicinal Chemistry Letters 2010; 20(9): 2828-31.
27. Trott O and Olson AJ: AutoDock Vina: improving the speed and accuracy of docking with a new scoring function, efficient optimization, and multithreading. Journal of Computational Chemistry 2010; 31(2): 455-61.
28. Suwannakul S, Chaibenjawong P and Suwannakul S: Antioxidant Anti-Cancer and Antimicrobial Activities of Ethanol *Pandanus amaryllifolius* Roxb. leaf extract (*In-*

vitro)-A potential medical application. Journal of Intern Dental & Medical Research 2018 May 1; 11(2).
29. Tsilo PH, Maliehe ST, Shandu JS and Khan R: Chemical composition and some biological activities of the methanolic Encephalartos ferox fruit extract. Pharmacognosy Journal 2020; 12(5).
30. Larayetan R, Ololade ZS, Ogunmola OO and Ladokun A: Phytochemical constituents, antioxidant, cytotoxicity, antimicrobial, antitrypanosomal and antimalarial potentials of the crude extracts of Callistemon citrinus. Evidence-Based Complementary and Alternative Medicine 2019 Aug 28;2019.
31. Rahman MM, Ahmad SH, Mohamed MT and Ab Rahman MZ: Antimicrobial compounds from leaf extracts of Jatropha curcas, Psidium guajava, and Andrographis paniculata. The Scientific World Journal 2014; 2014.
32. Larayetan R, Ololade ZS, Ogunmola OO and Ladokun A: Phytochemical constituents, antioxidant, cytotoxicity, antimicrobial, antitrypanosomal and antimalarial potentials of the crude extracts of *Callistemon citrinus*. Evidence-Based Complementary and Alternative Medicine 2019 Aug 28; 2019.
33. Singh P, Kumawat HL and Agrawal T: GC-MS Analysis of Bio-Active Compounds of *Carica papaya* L. Male Flower. International Journal of Chemical Separation Technology 2020; 6(1): 23-34.
34. Padmashree M, Ashwathanarayana R and Naika RBR: Antioxidant, cytotoxic and nutritive properties of Roem & Schult. *Ipomoea staphylina* plant extracts with preliminary phytochemical and GCMS analysis. Asian Journal of Pharmacy and Pharmacology 2018; 4(4): 473-92.
35. Mboyazi SN, Nqotheni MI, Maliehe TS and Shandu JS: *In-vitro* Antibacterial and *In-silico* Toxicity Properties of Phytocompounds from *Ricinus communis* Leaf Extract. Pharmacognosy Journal 2020; 12(5).
36. Lipinski CA: Drug Discovery Today: Technol 2004; 1: 4.
37. Jagannathan R: Characterization of Drug-like Chemical Space for Cytotoxic Marine Metabolites Using Multivariate Methods. ACS omega 2019; 4(3): 5402-11.
38. Gupta S, Parihar D, Shah M, Yadav S, Managori H, Bhowmick S, Patil PC, Alissa SA, Wabaidur SM and Islam MA: Computational screening of promising beta-secretase 1 inhibitors through multi-step molecular docking and molecular dynamics simulations-Pharmacoinformatics approach. Journal of Molecular Structure 2020; 1205: 127660.
39. Maliehe TS, Tsilo PH and Shandu JS: Computational Evaluation of ADMET Properties and Bioactive Score of Compounds from Encephalartos ferox. Phar J 2020; 12(6).
40. Świacka K, Maculewicz J, Smolarz K, Szaniawska A and Caban M: Mytilidae as model organisms in the marine ecotoxicology of pharmaceuticals-A review. Environmental Pollution. 2019; 254: 113082.
41. Yang X, Wang Y, Byrne R, Schneider G and Yang S: Concepts of artificial intelligence for computer-assisted drug discovery. Chemical Revie 2019; 119(18): 10520-94.
42. Maliehe TS, Tsilo PH and Shandu JS: Computational Evaluation of ADMET Properties and Bioactive Score of Compounds from Encephalartos ferox. Pharmacognosy Journal 2020; 12(6).
43. Nowak M, Helgeson ME and Mitragotri S: Delivery of nanoparticles and macromolecules across the blood–brain barrier. Advanced Therapeutics 2020; 3(1):1900073.
44. Maiuolo J, Gliozzi M, Musolino V, Scicchitano M, Carresi C, Scarano F, Bosco F, Nucera S, Ruga S, Zito MC and Mollace R: The "Frail" brain blood barrier in neurodegenerative diseases: role of early disruption of endothelial cell-to-cell connections. International Journal of Molecular Sciences 2018; 19(9): 2693.
45. Ullah MA, Johora FT, Sarkar B, Araf Y and Rahman MH: Curcumin analogs as the inhibitors of TLR4 pathway in inflammation and their drug like potentialities: a computer-based study. Journal of Receptors and Signal Transduction 2020: 1-5
46. Kou L, Sun R, Bhutia YD, Yao Q and Chen R: Emerging advances in P-glycoprotein inhibitory nanomaterials for drug delivery. Expert Opinion on Drug Delivery 2018; 15(9): 869-79.
47. Hadni H and Elhallaoui M: 3D-QSAR, docking and ADMET properties of aurone analogues as antimalarial agents. Heliyon 2020; 6(4): e03580.
48. Kirchmair J, Göller AH, Lang D, Kunze J, Testa B, Wilson ID, Glen RC and Schneider G: Predicting drug metabolism: experiment and/or computation? Nature Reviews Drug Discovery 2015; (6): 387-404.
49. Shaikh SA, Jain T, Sandhu G, Latha N and Jayaram B: From drug target to leads-sketching a physicochemical pathway for lead molecule design *in silico*. Current pharmaceutical Design 2007; 13(34): 3454-70.
50. Agoni C, Olotu FA, Ramharack P and Soliman ME: Druggability and drug-likeness concepts in drug design: are biomodelling and predictive tools having their say. Journal of Molecular Modeling 2020; 26: 1-1.
51. Jia CY, Li JY, Hao GF and Yang GF: A drug-likeness toolbox facilitates ADMET study in drug discovery. Drug Discovery Today 2020; 25(1): 248-58.
52. Trott O and Olson AJ: AutoDock Vina: improving the speed and accuracy of docking with a new scoring function, efficient optimization, and multithreading. Journal of Computational Chemistry 2010; 31(2): 455-61.
53. Xu JJ, Diaz D and O'Brien PJ: Applications of cytotoxicity assays and pre-lethal mechanistic assays for assessment of human hepatotoxicity potential. Chemico-biological Interactions 2004; 150(1): 115-28.
54. Walker JD, Gerner I, Hulzebos E and Schlegel K: The skin irritation corrosion rules estimation tool (SICRET). QSAR & Combinatorial Science 2005; 3: 378-84.
55. Hariono M, Abdullah N, Damodaran KV, Kamarulzaman EE, Mohamed N, Hassan SS, Shamsuddin S and Wahab HA: Potential new H1N1 neuraminidase inhibitors from ferulic acid and vanillin: molecular modelling, synthesis and *in vitro* assay. Scientific Reports 2016; 6(1): 1-0.
56. Aarjane M, Slassi S, Tazi B, Maouloua M and Amine A: Synthesis, antibacterial evaluation and molecular docking studies of novel series of acridone-1, 2, 3-triazole derivatives. Structural Chemistry 2020; 6:1-9.

How to cite this article:
Maliehe TS, Mboyazi SN, Nqotheni MI, Buthelezi S and Shandu JS: Antibacterial activity, pharmacokinetics and molecular docking of phytochemicals from *Ricinus communis*` leaf extract. Int J Pharm Sci & Res 2021; 12(10): 5308-18. doi: 10.13040/IJPSR.0975-8232.12(10).5308-18.