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# Evaluation of Antioxidant Activity and *In Silico* Prediction of Bioactive Compounds from the n-Hexane Extract of *Gracilaria compressa* (C. Agardh) Greville

Syafrizayanti[1]*, Andre Priandoko[1], Mai Efdi[1]

[1]*Department of Chemistry, Universitas Andalas, Padang 25175, Indonesia*

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***Abstract***— This study evaluated the antioxidant potential and in silico pharmacological properties of the *n*-hexane extract of *Gracilaria compressa* (C. Agardh) Greville. Antioxidant assays using DPPH and ABTS radicals demonstrated moderate to strong scavenging activities, with $IC_{50}$ values of 74.51 mg/L and 58.81 mg/L, respectively. LC–MS/MS profiling identified several major lipophilic compounds, including uvaretin, mangostanol, and palmitic acid. Molecular docking simulations against Cyclooxygenase-2 (COX-2) revealed that uvaretin exhibited the strongest binding affinity (–9.0 kcal/mol), supported by hydrogen bonding and hydrophobic interactions with key active-site residues. Pharmacokinetic evaluation based on Lipinski's Rule of Five indicated favorable drug-likeness and bioavailability properties for the identified compounds. These findings suggest that *G. compressa n*-hexane extract contains bioactive constituents with promising antioxidant and anti-inflammatory potential for further pharmacological development of marine-derived compounds as candidate therapeutics agents, particularly in the design of natural COX-2 inhibitors, and supports their potential application in pharmaceutical and nutraceutical formulations.

***Keywords***— *Antioxidant; COX-2; Gracilaria compressa; LC-MS/MS; Molecular docking; N-hexane extract*

## 1. INTRODUCTION

Marine red algae (Rhodophyta) are rich source of bioactive compounds with a wide range of pharmacological activities, including antioxidant, antimicrobial, and anticancer effects. Several studies report that red algae produce phenolics, flavonoids, terpenoids, and pigments such as carotenoids and phycobiliproteins that may have scavenge free radicals and protect biomolecule from oxidative damage [1-2].

The genus Gracilaria has been widely studied for its biological properties, and multiple species have shown measurable activity of antioxidant in different extracts. However, most published work focused on polar extracts such as methanol and ethanol, and polysaccharide fractions, while studies on nonpolar (lipophilic) extracts are less common. Lipophilic fractions may contain bioactive compounds that can contributed to antioxidant or radical scavenging activity, so investigating n-hexane extracts may reveal different classes of bioactive compounds than polar solvents [1-3].

Experimental comparison across extraction solvents have shown solvent-dependent differences in antioxidant yield and activity. For example, in Gracilaria and related red algae the antioxidant activity and metabolic profiles vary significantly between n-hexane, ethyl acetate, and polar extracts. These differences justify specific evaluation of n-hexane extracts to detect lipophilic antioxidants that may be overlooked in conventional polar extraction [4].

Oxidative stress is closely linked to inflammation through the overproduction of reactive oxygen species (ROS), which can activate inflammatory signaling pathways, including the upregulation of cyclooxygenase-2 (COX-2) [5-6]. COX-2 plays a key role in the biosynthesis of pro-inflammatory prostaglandins and is widely recognized as a therapeutic target for inflammation-related conditions [7-8]. Therefore, evaluating the interaction between identified compounds and COX-2 provides a relevant molecular basis for exploring the potential anti-inflammatory effects associated with antioxidant activity. Although other targets such as matrix metalloproteinases (e.g., MMP9) are also involved in inflammatory processes, COX-2 was selected in this study due to its direct association with oxidative stress-mediated inflammation and its established use as a primary target in anti-inflammatory drug discovery [9].

In addition, *in silico* approaches such as molecular docking and computational profiling are increasingly used to predict interaction between identified metabolites and oxidative stress related target. These methods speed up hypothesis about possibility of molecular mechanism and structure-activity relationship, and its complement to *in*

***Corresponding author**.
Email address: syafrizayanti@sci.unand.ac.id
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*vitro* antioxidant assay by prioritizing candidate compounds for follow-up testing [10-11].

Therefore, this study aim to assess the antioxidant potential of the n-hexane extract of one of the Gracilaria species, *Gracilaria compressa* (C. Agardh) Greville using standard chemical assay (e.g., DPPH and ABTS), and to perfom in silico analysis of the main compounds detected in n-hexane extract toward oxidative stress related target. Combining chemical assay, compound identification using LC-MS/MS, and computational approaches will provide preliminary, complementary evidence about which lipophilic constituents might contribute to the observed antioxidant activity.

## 2. EXPERIMENTAL SECTION

### 2.1. Materials

Sample of red algae *G. compressa* (C. Agardh) Greville were collected from Purus, Padang, West Sumatra, Indonesia. The specimens were identified in Herbarium ANDA Laboratory, Department of Biology, Faculty of Mathematics and Natural Sciences, Universitas Andalas. N-hexane, methanol, DPPH (2,2-diphenyl-1-picrylhydarzyl), ascorbic acid, ABTS (2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid)), potassium persulfate ($K_2S_2O_8$), and Trolox used in antioxidant were obtained from Sigma-Aldrich. For in silico analysis, the three-dimensional structure of the target protein cyclooxygenase-2 (COX-2) (PDB ID: 6COX) was retrieved from the Protein Data Bank. The native ligand (S58) was used as a reference compound to validate the docking protocol, while the identified compounds from LC-MS/MS analysis were employed as test ligands for molecular docking studies.

### 2.2. Instrumentations

The instruments used in this study included a rotary vacuum evaporator (IKA), ultraviolet–visible (UV–Vis) spectrophotometer (Shimadzu), and Liquid Chromatography Tandem Mass Spectroscopy (LC-MS/MS) system (Advanced Laboratory, IPB University, Indonesia. In silico analysis was performed using a personal computer with the following specifications: Desktop-FRS8P4L, Windows 10 Pro 64-bit (10.0, Build 19045), Lenovo Intel(R) Core(TM) i5-5200U CPU 2.20GHz (4 CPUs), and 8GB RAM. The three-dimensional structure of the target protein was obtained from the Protein Data Bank (https://www.rcsb.org). Molecular docking simulations were carried out using AutoDock Vina, with ligand and protein preparation conducted using AutoDockTools 1.5.6. Visualization and analysis of docking results were performed using Discovery Studio Visualizer 2025 and PyMOL 3.1.8.

### 2.3. Preparation of *G. compressa* extract

A total of 5 kg of fresh sample *G. compressa* were dried for 15 days in an open area without any direct exposure to sunlight. The dried sample were grounded using grinder to obtain a dry powder [12]. The extraction was carried out through maceration method using n-hexane as a solvent. The powdered samples were macerated with 1 L n-hexane for 3 days at a room temperature with triple repetition. The n-hexane filtrate was concentrated using a rotary evaporator at 40°C to obtain extract [13].

### 2.4. Antioxidant Activity via DPPH Radical Scavenging Assay

The antioxidant activity of *G. compressa* n-hexane extract was evaluated using the DPPH (2,2-diphenil-1-picrylhydrazyl) radical scavenging method [14]. A DPPH stock solution was prepared by dissolving 4 mg of DPPH in methanol up to a total volume of 100 mL. The control solution consisted of 2 mL methanol and 3 mL of 0.1 mM DPPH solution in a vial. For sample preparation, 25 mg of the *G. compressa* extract was dissolved in methanol in a 25 mL volumetric flask, then serially diluted to obtain concentration of 0, 5, 10, 20, 25, 35, 40, 50, 75, 100 mg/L. Each concentration (2 mL) was transferred into a vial and mixed with 3 mL of DPPH solution. Ascorbic acid was used as a positive control and prepared at a stock concentration of 100 mg/L in a 25 mL volumetric flask. It was then serially diluted to concentration of 0, 5, 10, 20, 35, 40, 50, 75, and 100 mg/L. Each concentration (2 mL) was also mixed with 3 mL of DPPH solution in a vial. All solutions were homogenized, and the absorbance was measured using a UV-Vis spectrophotometer at a wavelength 517 nm [15], with methanol as the blank. The percentage of DPPH inhibition was calculated for each concentration, and the $IC_{50}$ value was determined based on the linear regression equation of the inhibition curve [16].

### 2.5. Antioxidant Activity via ABTS Radical Scavenging Assay

The ABTS radical scavenging activity was evaluated using the ABTS (2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic-acid) assay [17-18]. To prepare the ABTS stock solution, 0.036 g of ABTS was dissolved in 10 mL of deionized water up to 25 mL. Equal volume (1:1 v/v) of the ABTS and potassium persulfate solutions were mixed and incubated in the dark for 16 hours at 25 °C, resulting in the formation of a dark blue $ABTS^{\bullet+}$ solution. The *G. compressa* extract was prepared by dissolving 25 mg of dried extract 25 mL deionized water. This solution was serially diluted to obtain final concentration 0,5,10,20,25,35,40,50,75, and 100 mg/L. Each sample solution (1 mL) was mixed with 5 mL of ABTS stock solution and incubated for 6 minutes in the dark. Trolox was used as a positive control. A stock solution of 100 mg/L Trolox was prepared in a 25 mL volumetric flask using deionized water and serially diluted to final concentrations of 0,5,10,20,25,35,40,50,75, and 100 mg/L. Each solution (1 mL) was mixed with 5 mL of the ABTS stock solution in test tube and homogenized. The absorbance of each sample, control, and positive control was measured using a UV-Vis spectrophotometer at 734 nm [19-20]. The percentage of ABTS radical inhibition

was calculated, and an inhibition curve generated to determine the $IC_{50}$ value based on the regression equation obtained.

### 2.6. Phytochemical Profiling via LC-MS/MS

Phytochemical profiling and molecular weight determination of *G. compressa* (C. Agardh) Greville extract were carried out using LC-MS/MS at the Advanced Laboratory, IPB University. The extract was first diluted and filtered through a 0.45 µm miliphore membrane filter. The analysis employed an HPLC system coupled to an ESI-QTOF, following established metabolomics approaches [21]. The mobile phase consisted solvent A (0.1% formic acid in acetonitrile) and solvent B (5 mM ammonium formate in water), delivered at a flow rate 0.4 mL/min. Separation process was achieved on an HSS C18 column (1.8 µm, 2.1 x 150 mm) using a linear gradients solvent A from 95% to 5% over 15 minutes. The QTOF detector operated in positive electrospray ionization (ESI) mode with source voltage +2.9 kV, gas flow 50 L/h, source temperature 41°C, and desolvation temperature 120°C. Mass spectra were recorded across an m/z range of 100-500 Da, and the detected metabolites were identified by matching m/z values with entries in a natural product database [22].

### 2.7. *In Silico* Study

The *in silico* study was performed to assess the interactions between bioactive compounds and the target protein using molecular docking. The three-dimensional structure of Cyclooxygenase-2 (COX-2), which serves as the target protein, was retrieved from Protein Data Bank (PDB) database (https://www.rcsb.org). Both ligand and receptor structure were prepared using PyMOL software. Molecular docking simulations were performed using AutoDock Vina. The grid box was defined based on the active site of the COX-2 enzyme, which was determined from the position of the co-crystallized native ligand in the PDB structure. The grid box was centered on the binding pocket coordinates of the native ligand (S58) to ensure accurate targeting of the catalytic site. The grid dimensions were set to 20 × 20 × 20 Å to adequately cover the active site region and allow flexibility for ligand interaction within the binding pocket. The binding affinity values were calculated to estimate the interaction strength between each ligand and the target protein. Visualization and analysis of docking poses were conducted using Biovia Discovery Studio Visualizer 2025, enabling interpretation of the binding modes and involved interaction such as hydrogen binding and hydrophobic contacts.

## 3. RESULT AND DISCUSSION

### 3.1. Antioxidant DPPH Result

The n-hexane extract of *G. compressa* (C. Agardh) Greville showed strong DPPH radical scavenging activity with an $IC_{50}$ of 74.51 ± 0.32 mg/L, calculated from the linear regression of percent inhibition versus concentration. By comparison, the standard antioxidant ascorbic acid (vitamin C) exhibited an $IC_{50}$ OF 28.59 ± 0.30 mg/L, indicating that while the n-hexane extract is less potent than ascorbic acid, but it still demonstrates a meaningful antioxidant effect. This suggests that lipophilic constituents in the n-hexane extract (such as fatty acids, sterols, and terpenoids) may contribute to radical neutralization via electron transfer or hydrogen donation pathways. Fatty acids and other nonpolar metabolites from red algae have been reported to possess measurable DPPH scavenging activity, supporting the notion that hexane-soluble components can be antioxidant contributors [23-24].

Although many seaweed studies report higher DPPH activity in polar extracts (methanolic or ethanolic) because these solvents efficiently extract hydrophilic phenolics, there are multiple examples where nonpolar fraction exhibit notewhorthy activity or contain antioxidant lipophilic molecules (e.g., carotenoids, lipophilic phenolics, and fatty acids). Thus, the comparatively high activity observed for the n-hexane extract of *G. compressa* (C. Agardh) Greville likely reflects an enrichment of these lipophilic antioxidants that are not recovered by polar solvents [25-26]

Beyond radical scavenging, polyphenol-rich seaweed extracts have been linked to antiproliferative and anticancer effects in several studies, likely through a combination of antioxidant activity, modulation of endogenous cellular defense systems, and induction of apoptosis. Thus, the antioxidant potency of the n-hexane fraction of *G. compressa* (C. Agardh) Greville supports further evaluation of its bioactive constituents for possible cytoprotective and antiproliferative properties representative work linking polyphenol-rich seaweed extract to anticancer activity includes studies on *Euchema cottonii* and *Sargassum* species [27-28].

### 3.2. Antioxidant ABTS Result

The n-hexane extract of *G. compressa* (C. Agardh) Greville displayed a moderate ABTS radical cation scavenging capacity, with an $IC_{50}$ of 58.81 ± O,72 mg/L. In contrast, the standard antioxidant Trolox achieved a markedly stronger performance, with an $IC_{50}$ of 21.41 ± 0.16 mg/L. The lower $IC_{50}$ value of Trolox highlights that while the n-hexane extract does scavenge ABTS radicals, its effectiveness is lower than that of this well-established standard.

The ABTS assay is especially useful in evaluating both hydrophilic and lpophilic antoxidant activity, because the ABTS radical cation is solube in both aqueous and organic media. This makes it a better match for assessing antioxidant capacity of nonpolar or semi-polar extracts than assays limited to aqueous environtments.

In literature, ABTS scavenging activities for seaweed extracts vary widely depending on species, extraction solvent, and assay conditions. For example, some studies report ABTS radical inhibition by seaweed extracts with $IC_{50}$ values in the tens of micrograms to low milligram per liter ranges [29].

### 3.3. Phytochemical Profiling Result

The chemical profiling of secondary metabolites in the n-hexane extract of *G. compressa* (C. Agardh) Greville was performed using LC-MS/MS, with compound identification based on retention time (RT) and mass spectral data. Analysis revealed the presence of 10 major compounds exhibiting the highest peak intensities. These metabolites represented various chemical classes, predominantly fatty acids, sterols, terpenoids, and phenolic derivates, which are commonly associated with antioxidant and other bioactive properties in marine algae.

The LC-MS/MS chromatogram of the n-hexane extract of *G. compressa* (C. Agardh) Greville (**Fig. 1.**) revealed 23 distinct peaks, corresponding to various secondary metabolites identified based on their retention time (RT) and mass-to-charge ratio (m/z). Among these, 10 major compounds showing the highest peak intensities were selected for further discussion with molecular docking simulation.

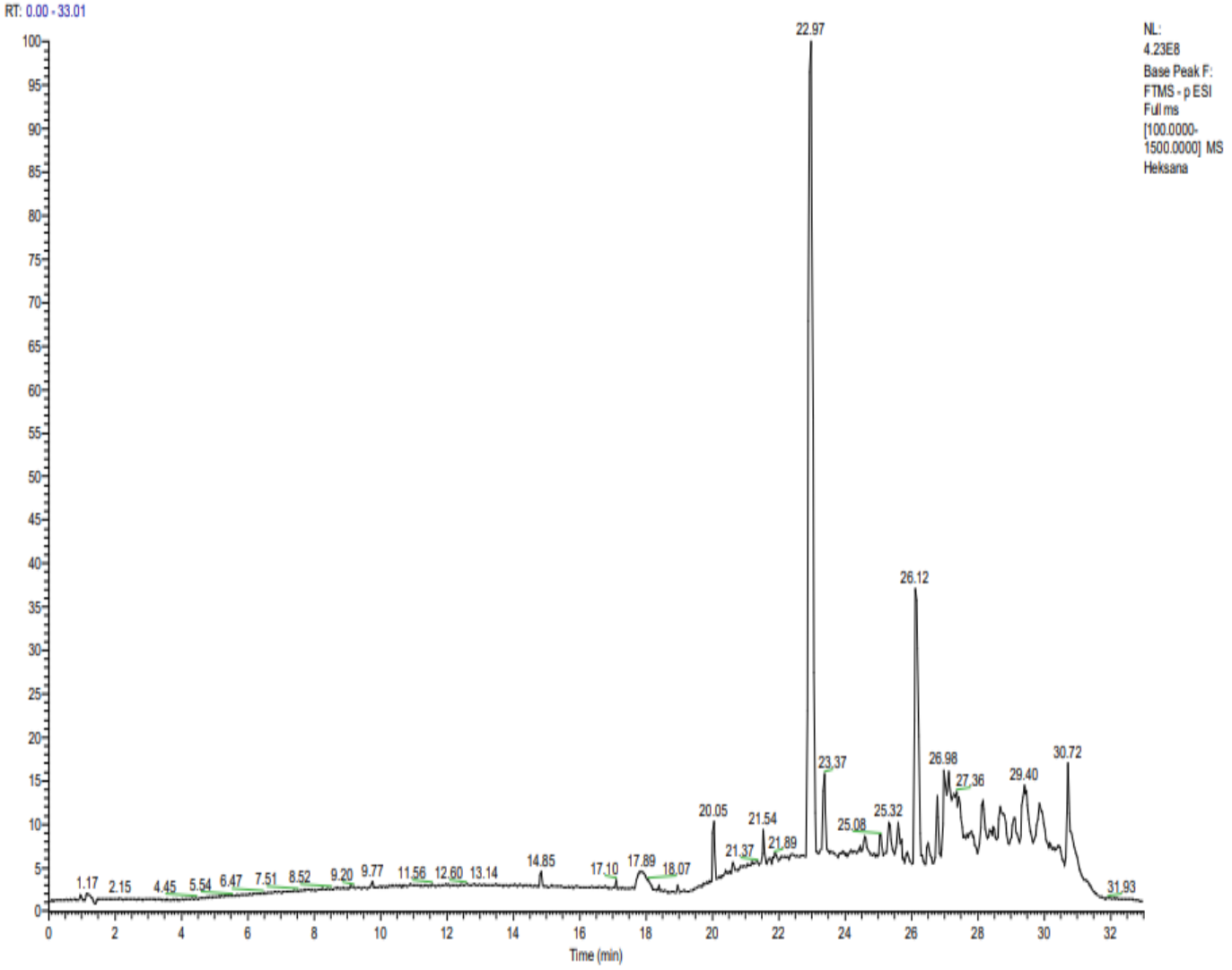
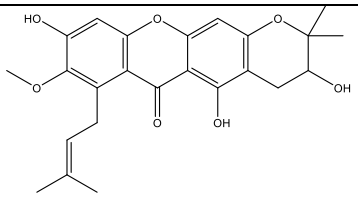
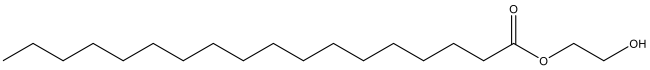
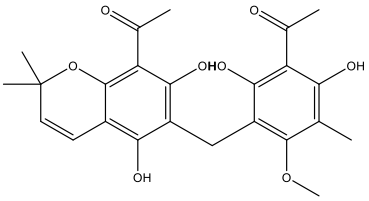
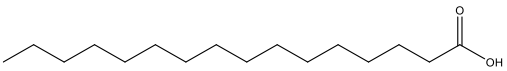
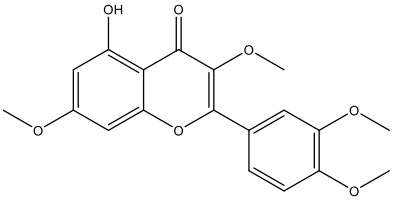
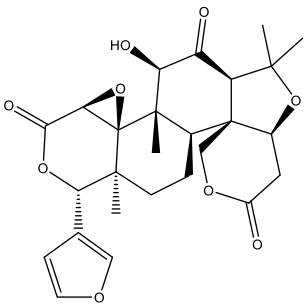
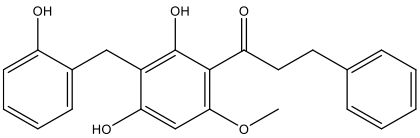
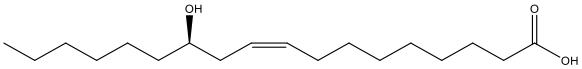
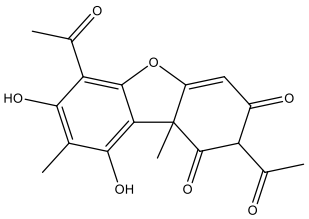
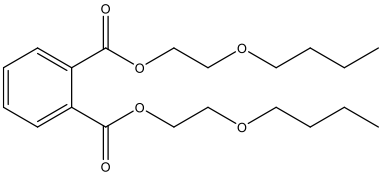



**Fig. 1.** LC-MS/MS chromatogram n-hexane extract of *G. compressa* (C. Agardh) Greville

Among these 10 most abundance compound of *G. compressa* (C. Agardh) Greville in **Table 1.**, usnic acid stands out for its well documented antioxidant and radical scavenging capacity. Usnic acid exhibited strong activity against several radicals, for example ABTS•$^{+}$ scavenging with an $IC_{50}$ 10.41 µg/mL [30]**.**

Regarding fatty acids, palmitic acid and ricinoleic acid (a hydroxylated C18 fatty acid) are known to contribute to antioxidant behaviour although not primarily as top radical scavengers. Studies on palmitic acid show it can induce oxidative stress in high concentrations, but some co-treatments (or use in combinations) suggest a modulatory role in oxidative balance. For instance, Elham in his study (2016) shown that palmitoleic acid reduced the oxidative damage induced by palmitic acid [31]. Ricinoleic acid with similar structure to fatty acid with hydroxyl groups suggest potential hydrogen donating or membrane protecting roles. Some flavonoids such as retusin and rutaevin generally donate hydrogen atoms or electrons to neutralize radicals.

**Table 1.** Top 10 major compounds observed in n-hexane of *G. compressa* (C. Agardh) Greville

| Compounds | Formula | Retention Time (RT) | Molecular Weight (Da) | Structure |
|---|---|---|---|---|
| Mangostanol | $C_{24}H_{26}O_7$ | 22.97 | 426.1675 |  |
| Glycol stearate | $C_{20}H_{40}O_3$ | 26.78 | 368.4622 |  |
| Mallotochromene | $C_{24}H_{26}O_8$ | 20.05 | 442.1633 |  |
| Palmitic acid | $C_{16}H_{32}O_2$ | 27.05 | 256.6231 |  |
| Retusin | $C_{19}H_{18}O_7$ | 18.14 | 358.3479 |  |
| Rutaevin | $C_{26}H_{30}O_9$ | 16.25 | 486.1889 |  |
| Uvaretin | $C_{23}H_{22}O_5$ | 17.10 | 378.1467 |  |
| Ricinoleic acid | $C_{18}H_{34}O_3$ | 20.95 | 298.4690 |  |
| Usnic acid | $C_{18}H_{16}O_7$ | 14.52 | 344.0895 |  |
| Dibutoxyethyl phtalate | $C_{20}H_{30}O_6$ | 14.71 | 366.2102 |  |

### 3.4. Molecular Docking Result

In this study, Cyclooxygenase-2 (COX-2) was selected as the target for molecular docking simulation due to its recognize as a key enzyme in the inflammatory pathway and also exploring for antioxidant potential. In silico docking studies often adopt COX-2 to assess the binding affinity of natural products, since inhibition of COX-2 can reduce prostaglandin synthesis and mitigate oxidative stress mediated inflammation [32]. To ensure the reliability of the docking protocol, method validation was performed through a re-docking procedure. The co-crystallized native ligand was extracted from the COX-2 structure and re-docked into the active site using the same docking parameters. The accuracy of the docking method was evaluated by calculating the root-mean-square deviation (RMSD) between the predicted ligand pose and the original crystallographic position. An RMSD value of less than 2.0 Å was considered acceptable, indicating that the docking protocol was valid and capable of reproducing the experimentally observed binding mode.

The docking simulation were performed using the AutoDock Vina 1.5.6 and the result were analyzed based on binding affinity, interaction types, and visualization. The COX-2 structure was obtained from the Protein Data Bank (PDB ID: 6COX). A total 10 major compounds from n-hexane of *G. compressa* (C. Agardh) Greville were docked to COX-2, and their binding affinities were calculated.

**Table 2.** Binding free energy of 10 major compounds of n-hexane extract in *G. compressa* (C. Agardh) Greville and reference drug with target protein Cyclooxygenase-2 (COX-2/6COX), retrieved by website PubChem

| PubChem CID | Ligands | Binding free energy (Kcal/mol) | Hydrogen Bonds |
|---|---|---|---|
| 10048103 | Mangostanol | -6.6 | HIS:90, PHE:518 |
| 24762 | Glycol stearate | -5.6 | - |
| 126969 | Mallotochromene | -6.1 | - |
| 985 | Palmitic acid | -6.5 | TYR:355 |
| 5352005 | Retusin | -6.4 | TYR:385 |
| 441805 | Rutaevin | -5.5 | ARG:120, TYR:348 |
| 73447 | Uvaretin | -9.0 | ARG:120, TYR:385 |
| 643684 | Ricinoleic acid | -6.0 | - |
| 5646 | Usnic acid | -5.8 | ARG:120, TYR:385 |
| 8345 | Dibutoxyethyl phthalate | -5.4 | - |
| 2662 | Celecoxib | -11.2 | HIS:90, ARG:120, GLN:192, SER:353 PHE:518 |

This docking study (**Table 2.**) shows that among the ten major compounds from the n-hexane extract of *G. compressa*, Uvaretin exhibited a binding free energy of –9.0 kcal/mol, while Celecoxib (used here as a reference drug) displayed a substantially stronger binding at –11.2 kcal/mol. Other ligands such as Mangostanol (–6.6 kcal/mol) and Palmitic Acid (–6.5 kcal/mol) also showed moderate to good affinity. These results suggest that while some of the natural compounds have measurable interaction potentials with the target Cyclooxygenase-2 (COX-2), their in-silico affinities are still weaker compared to celecoxib, a well-established COX-2 inhibitor. For example, previous studies report celecoxib binding energies in the range of approximately –8 to 10 kcal/mol in similar setups [33-35].

Diagram of palmitic acid with COX-2 (**Fig. 2.**) shows that the ligand forms a conventional hydrogen bond with Tyr355, contributing to its stability within the binding pocket. An unfavorable donor–donor interaction is detected with Arg120, which may slightly reduce binding affinity. Several alkyl and π-alkyl interactions with hydrophobic residues such as Leu352, Val349, Ala523, and Trp387 help stabilize the long aliphatic chain of palmitic acid in the enzyme's hydrophobic channel.

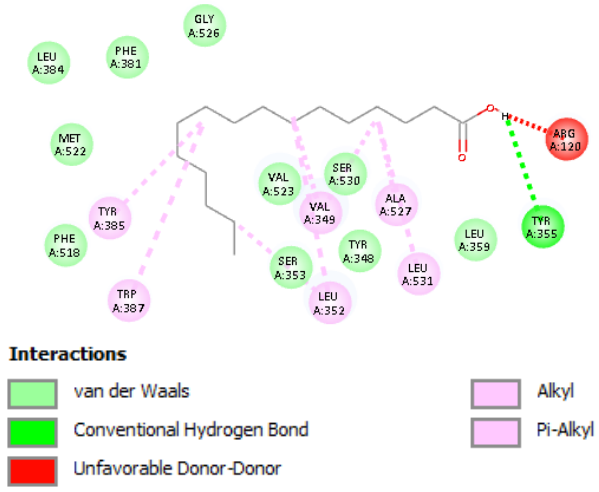



**Fig. 2.** 2D interaction of palmitic acid with COX-2

The 2D interaction diagram of the mangostanol–COX-2 complex (**Fig. 3.**) reveals that mangostanol binds within the active pocket of COX-2 through a network of hydrogen bonds and hydrophobic interactions. A hydrogen bond is formed between the hydroxyl group of mangostanol and Phe518, while additional stabilizing hydrophobic contacts are observed with residues such as Leu352, Leu384, Tyr385, Trp387, and Val349. The presence of His90 and Ser353 near the ligand suggests possible polar interactions that may contribute to binding orientation and stability. These amino acid residues have been previously reported as critical sites in the COX-2 active pocket responsible for substrate and inhibitor recognition [36].

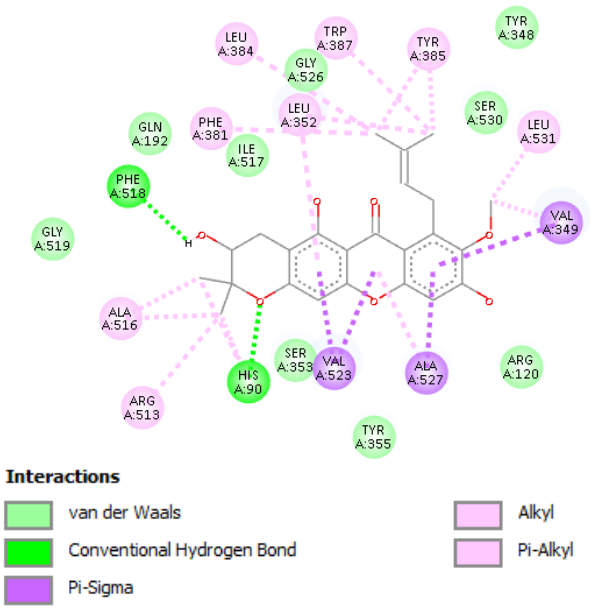



**Fig. 3.** 2D interaction of mangostanol with COX-2

Uvaretin forms a series of stabilizing interactions within the enzyme's active pocket (**Fig. 4.**). A conventional hydrogen bond is observed with Tyr385 and Phe518, which are known catalytic residues essential for COX-2 activity. Additionally, the ligand engages in π–π T-shaped and π-alkyl interactions with aromatic residues such as Trp387, Phe518, and Tyr355, further stabilizing its position through hydrophobic stacking. A π-sulfur interaction is also formed with Met522.

The presence of both polar and hydrophobic contacts indicates that uvaretin is well accommodated in the COX-2 hydrophobic channel. The unfavorable acceptor–acceptor contact with Tyr355 may slightly weaken the binding orientation; however, the overall network of interactions suggests a strong affinity, consistent with its docking score of –9.0 kcal/mol. This value is comparable to those reported for other natural xanthone derivatives showing inhibitory potential toward COX-2.

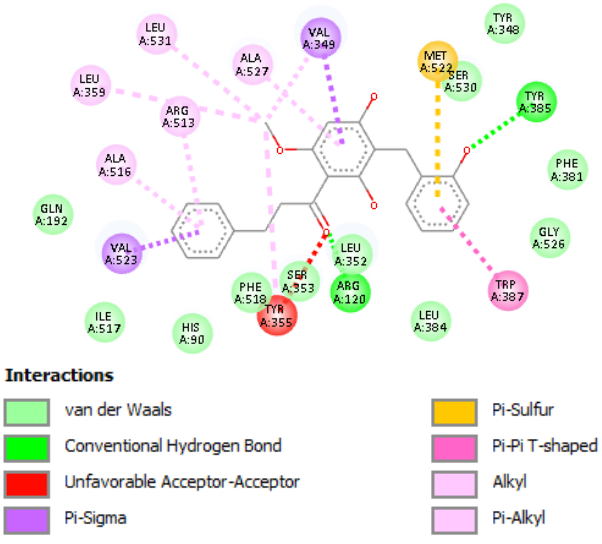



**Fig. 4.** 2D interaction of uvaretin with COX-2

Conventional hydrogen bonds are observed with residues His90, Gln92, and Ser353, suggesting strong polar interactions that contribute to binding stability (**Fig. 5.**) Additional carbon–hydrogen bonds with Arg353 and Val116 further enhance interaction specificity. Several hydrophobic interactions, including Pi–Sigma, Pi–Alkyl, and Alkyl contacts, are formed with residues such as Tyr355, Leu359, Trp387, Met522, and Ala527, indicating the importance of nonpolar stabilization within the binding pocket. These combined hydrogen bonding and hydrophobic interactions suggest a well-oriented ligand conformation, promoting strong and stable complex formation within the protein's active site.

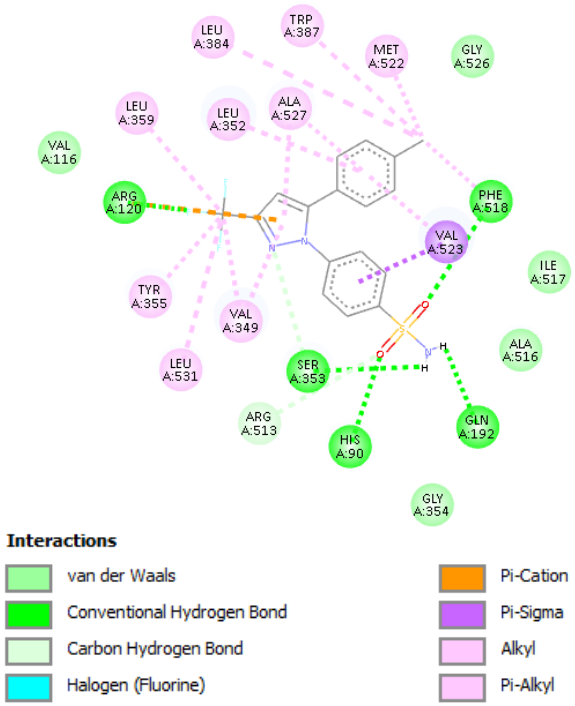



**Fig. 5.** 2D interaction of celecoxib (drug) with COX-2

The pharmacokinetic evaluation of compounds identified from the *n*-hexane extract of *Gracilaria compressa* revealed that all ligands exhibited physicochemical characteristics within or close to the acceptable range defined by Lipinski's Rule of Five (**Table 3.**). Uvaretin (LogP = 5.61) slightly exceeds the lipophilicity limit (LogP < 5), suggesting a strong hydrophobic nature that may enhance membrane permeability but could potentially reduce solubility. Mangostanol and Palmitic acid demonstrate favorable molecular weights and hydrogen-bonding profiles, indicating good oral bioavailability. Celecoxib, used as the reference drug also complies with Lipinski's criteria, supporting the reliability of this comparison. The evaluated parameters suggest that the natural compounds, particularly mangostanol and palmitic acid, possess suitable pharmacokinetic properties for drug-likeness, while uvaretin shows strong lipophilicity that may favor passive absorption through biological membranes. These findings align with the general rule that compounds obeying Lipinski's parameters tend to

have favorable absorption and permeation properties in biological systems.

**Table 3.** Pharmacokinetics properties of identified compounds from n-hexane extract of *G. compressa* (C. Agardh) Greville

| Ligand | Molecular Weight (Da) | Hydrogen Donor <5 | Hydrogen Acceptor <10 | LogP <5 |
|---|---|---|---|---|
| Uvaretin | 378.1467 | 3 | 5 | 5.61 |
| Mangostanol | 426.1675 | 3 | 7 | 4.02 |
| Palmitic Acid | 256.6231 | 1 | 2 | 3.85 |
| Celecoxib (Drug) | 381.3722 | 2 | 5 | 4.21 |

## 4. CONCLUSION

The *n*-hexane extract of *Gracilaria compressa* (C. Agardh) Greville exhibited notable antioxidant activity, as demonstrated by its DPPH and ABTS radical scavenging capacities. LC–MS/MS profiling revealed the presence of several lipophilic bioactive compounds, including uvaretin, mangostanol, and palmitic acid, which are known for their antioxidant and pharmacological potential. Molecular docking analysis indicated that these compounds interact favorably with the COX-2 enzyme through a combination of hydrogen bonding and hydrophobic interactions, with uvaretin showing the strongest affinity (–9.0 kcal/mol). The pharmacokinetic prediction further confirmed that most compounds fulfilled or closely approached Lipinski's drug-likeness criteria, suggesting good absorption and bioavailability potential. Collectively, these findings highlight the *n*-hexane extract of *G. compressa* as a promising natural source of antioxidant and bioactive molecules with potential therapeutic relevance, warranting further investigation through in vitro and in vivo validation studies.

## 5. AUTHOR'S DECLARATION

### 5.1. Supporting Information

There is no supporting information in this paper.

### 5.2. Acknowledgements

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### 5.3. Conflict of Interest

The authors declared that no conflicts of interest regarding the publication of this article.

### 5.4. Author Contributions

All authors are the main contributors to this paper. The authorship contributions such as AP: data collection, analysis of data, and drafting of the manuscript. S and ME: helped with the conceptualization of the study and supervision.

### 5.5. AI Statement

The authors used ChatGPT (OpenAI) to assist with language editing and improving the clarity of the manuscript. No AI tool was used to generate, analyze, or interpret the scientific data or results. The authors reviewed all AI-generated content and assume full responsibility for the final version of the manuscript.

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