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Boiling water extraction of mangrove *Sonneratia Alba* fruit as an antioxidant functional food: combined *in vitro* and pharmacoinformatics studies

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ABSTRACT

Introduction: Extraction of antioxidants from mangrove fruit powder *Sonneratia alba* using boiling water as a solvent is very beneficial, because in addition to using a solvent that is safe for consumers, the results of this study can also be developed in the future as antioxidant functional foods.

Aims and Methods: The purpose of this study was to study the antioxidant activity of boiling water extract of mangrove fruit *S. alba* kindly *in-vitro* nor *in-silico* pharmacoinformatics.

Results: The best results in this first stage were found in the treatment of mangrove fruit *S. alba* Sun drying extracted with boiling water for 5 minutes had the best antioxidant activity, namely IC_{50}DPPH = 2.69 ± 0.32 µg/ml which is smaller than the positive control, namely vitamin C, which has IC_{50}DPPH= 5.04 ± 0.16µg/ml. The best treatment in the first stage is continued in the second stage, namely GC-MS analysis to determine the type of compounds in the extract, and then do the analysis *in-silico* by means of molecular docking. The results of the GC-MS characterization showed 39 compounds that were in the category to be continued in the pharmacoinformatics via molecular docking process, with the docking results showing the bioactive compound Ergosterol; Estra-1,3,5(10)-trien-17ß-ol; Ergosta-5,8,22-trien-3-ol, (3ß,22E)-; 9(11)-Dehydroergosterol tosylate; Dasycarpidan-1-methanol, acetate (ester) shows a very promising value of binding affinity towards 4 protein targets, namely Cytochrome c peroxidase, Fibroblast collagenase, Human ROS1 Kinase Domain and Hyaluronidase with vitamin C as a control.

Conclusion: Based on these findings, boiling water extract of mangrove fruit *S. alba* this can provide a good potential in the discovery and development of candidate new antioxidant compounds.

KEYWORDS

Sonneratia alba fruit, boiling water, DPPH, Molecular Docking, In Silico, nutrition, technology, food technology.

INTRODUCTION

Mangrove development system *Sonneratia alba* happen throughout the year, this type of mangrove is able to survive in coastal locations with the influence of tides and salinity. Mangroves *S.alba* can grow in sandy and muddy areas, high salinity, but still in areas affected by sea tides[1]. In Indonesia, especially North Minahasa Regency, North Sulawesi Province, the mangrove species that grow in several mangrove forest locations are *Bruguiera gymnorhiza, Rhizophora mucronata and Sonneratia alba*. One of *mangrove species* Which Dominantly grow in Wori sub-district, Province North Sulawesi Indonesia which is the place of sampling in this study is *Sonneratia alba.*

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Mangrove *S.alba* potential as a source of natural antioxidants and has been used as a traditional medicine by coastal communities in Indonesia including the coastal community of Kuala Bubon, it is further reported that leaf methanol extract *S.alba* of this area has activity the strongest antioxidant with an IC50 value of 26.68 ± 2.2 mg/L, while the ethyl acetate extract was 33.37 ± 3.4 mg/L and the n-hexane extract had antioxidant activity with an IC value$_{50}$ of 35.37 ± 2.5 mg/L. Mangrove leaf methanol extract *S alba* from Tagulandang District, North Sulawesi, has very strong antioxidant activity with an IC50 DPPH value of 39.5µg/ml[2], ethanol extract of mangrove leaves and stems *S.alba*f rom Tanjung Carat, South Sumatra have antioxidant activity i.e. value IC_{50} DPPH amounts to 18.62 µg/ml and 22.96 µg/ml[3].

Especially mangroves *S.alba* from the coast of Wori Village, North Sulawesi, Indonesia according to several research results it has been reported to have very strong antioxidant activity. Some of the results of these studies include:methanol extract of mangrove fruit flour *S.alba* has an IC value$_{50}$ DPPH is 4.65 µg/ml[4], mangrove young leaf extract *S.alba* obtained by maceration using ethanol has IC_{50} DPPH of 5,01µg/ml and soxletation method using methanol has IC_{50} DPPH of 5.16µg/ml, this result is better than the IC value$_{50}$ The DPPH of vitamin C was 5.21µg/ml[5], also reported that antioxidants (IC_{50} DPPH) in the ethyl acetate fraction of young mangrove leaves *S.alba* classified as very strong at 3.37 µg/ml. The research data that has been found above shows that parts of the mangrove plant *S.alba* both stems and leaves fruit has antioxidant activity that is classified as very strong. Data the research results above also show that the solvent used in the extraction process is an organic solvent that when followed consumed will be harmful to the health of consumers.

Nowadays functional food/beverage products are starting to be in great demand by consumers because Awareness of the importance of healthy living is increasing. one of a kind Food that has been widely developed and researched is health food contains antioxidants. Antioxidants themselves are chemical compounds related to free radicals where these chemical compounds can counteract free radicals. Free radicals cause premature aging as well as various diseases including cancer, respiratory and cardiovascular. For the utilization of mangrove fruit as a functional antioxidant drink, it is necessary to pay attention to the selection of appropriate and safe solvents for consumers, one of the safest solvents to use to extract antioxidants is boiling water. In general, high temperatures can damage the antioxidant chemical components so that it can reduce the antioxidant activity in an extract[6] reported that mangroves *Rhizophora mucronata* extracted at evaporation temperature solvent 70^{O}C has antioxidant activity, namely IC_{50} The DPPH was 0.7021 ppm, after the evaporation temperature was increased to 80^{O}C value IC_{50} DPPH became 1.4152 ppm.

The purpose of this research is studied the antioxidant activity of boiling water extract of mangrove fruit *S. alba* kindly *in-vitro* nor *in-silica* which at the same time can prove the antioxidant bioactive component in mangrove fruit *S. alba* resistant to high temperatures characterized by very strong antioxidant activity. Besides that results of research on the extraction of antioxidants from mangrove fruit powder *S. alba* using boiling water as a solvent is very beneficial, because in addition to using a solvent that is safe for consumers, the results of this research can also be in the future developed as an antioxidant functional drink.

MATERIALS AND METHODS

Materials and tools

The material used is fruit *S. alba.* Chemical material The materials used were methanol (Merck), Folin Ciocalteau (Sigma Aldrich), gallic acid (Merc), Na2CO3 7.5% (Merck), gallic acid (Merck),*1,1-diphenyl-2-picrylhydracyl/DPPH* (Sigma Aldrich), buffer fosfat (Merck), K3Fe (CN)6 (Merck), TCA (Merck), FeCl3 1% dan 5% (Merck), HCL 2 N (Merck) dan akuabides. The tools used were a vacuum evaporator (Buchi, UK), UV-Visible spectrophotometer (Shimadzu type 1240, Japan), 1 mL and 0.5 mL micropipette (Germany) and glassware (Pyrex). Oven, water bath, micropipette.GCMS thermos scientific Trace 1310 Gas Chromatograph dan thermos scientific ISQLT single quadropole Mass Spektrofotometer.

Research methods

This research begins with picking the fruit *S. alba*in the coastal location of the village of Wori, Wori District, North Minahasa Regency, North Sulawesi Province of Indonesia. Plant *S. alba*This was identified in the Jatinangor Herbarium, Laboratory of Plant Taxonomy, Department of Biology FMIPA Padjadjaran University. Mangroves *S. alba* (fruit diameter ≥ 3 cm) washed, thinly sliced, portioned dried in the sun and some moreair-dried indoors. After drying, which is marked with slices of mangrove fruit, it is easy to break with your fingers, then blend it finely so that it forms a powder.

Extraction process

Extraction of fruit powder samples *S. alba*by infusion (using boiling water at 96-98°C) for 5, 10 and 15 minutes, the liquid extract was filtered using Whatman paper No. 1. The liquid extract was evaporated by means of evaporation in a water bath at 70-80°C until a thick extract was obtained, the thick extract was dried in an oven at 70°C to obtain dry extract characterized by a constant weight at 3 times weighing. Then the yield was calculated by comparing the weight of the extract with the weight of the dry sample and multiplied by 100. The results of the extraction were analyzed for phytochemicals referring to[7] with slight modification, the total phenol content was determined using the spectrophotometric method[8], and the antioxidant activity of the DPPH method (*1,1-diphenyl-2-picrylhydracyl*) using the spectrophotometric method referring to[6].

Phytochemical analysis

Alkaloid test

50-100 mg of extract sample added with sufficient chloroform, then 10 mL of ammonia and 10 mL of chloroform were added, then the solution was filtered into a test tube and the filtrate was added with 10 drops of 2N H2SO4. The mixture is shaken regularly, left for a few minutes until 2 layers are formed. The top layer was transferred into three test tubes of 1 mL each. Then each tube was added a few drops of Mayer's, Wagner's and Dragendorff's reagent. If a precipitate forms, it indicates that the sample contains alkaloids, with Mayer's reagent it gives a white precipitate, with Wagner's reagent it gives a brown precipitate and Dragendorff's reagent gives an orange precipitate.

Test triterpenoids and steroids

50-100 mg of the extract sample was added to glacial acetic acid until all samples were submerged, left for 15 minutes then 6 drops of the solution were transferred to a test tube and 2-3 drops of concentrated sulfuric acid were added. The presence of triterpenoids is indicated by the appearance of red, orange or purple, while steroids are indicated by the formation of blue.

Tanin test

50 mg of the extract sample was added with ethanol until the sample was completely submerged, then 2-3 drops of 1% FeCl3 solution were added. A positive result is indicated by the formation of a bluish black or green color.

Flavonoid test

50 mg of extract sample added with a few drops of concentrated HCL. Then added 0.2 g of Mg powder. A positive result is indicated by the appearance of a dark red color for 3 minutes.

Saponin test

50 mg of extract sample was put into a test tube, then added distilled water until all samples were submerged, boiled for 2-3 minutes, then cooled, then shaken vigorously. A positive result is indicated by the formation of stable foam.

Phenolic test

Before identifying phenolic compounds, continuous extraction was carried out using a Soxhlet apparatus with ether solvents to dissolve fat and chlorophyll. The results of the ether extract are blackish green, when reacted with 5% FeCl3 it cannot react so that the ether extract does not contain phenolic compounds. After being extracted with ether then extracted with 90% methanol and continued with 50% methanol to bind polar components. 1 mL of methanol extract added with 5% FeCl3, a color change from brownish yellow to orange brown indicated the presence of phenolic compounds.

Total phenol content

The total phenol content was measured by a spectrophotometer using a reagent *Folin-Ciocalteau*. 0.1 g of the extract was dissolved in 10 ml of methanol Pa in a test tube, 0.1 ml of the extract solution was taken and added 1 ml of the solution *Folin Ciocalteau* 1:2 in distilled water and let stand 5 minutes. Then 1 ml of 7% sodium carbonate solution was added, homogenized and incubated at room temperature for 30 minutes in the absence of light (dark). Gallic acid standard curve was made following the procedure above, only the sample was replaced with gallic acid. The total phenolic content was measured using a spectrophotometer at a wavelength of 750 nm and interpreted as µg gallic acid equivalent/g extract (µg GAE/g extract).

Antioxidant activity of DPPH free radical inhibitor

This analysis is based on the ability of the sample to reduce DPPH free radicals. Samples with a concentration of 25 to 125 ppm as much as 2 ml were put into a test tube and added DPPH solution (1×10^{-4}M) as much as 1 ml, the mixture was homogenized and incubated at room temperature for 30 minutes. Absorbance was measured at a wavelength of 517 nm. Controls were made following the procedure above, only the samples were replaced with methanol. The antioxidant activity of DPPH free radical scavenging was determined as the percentage of inhibition calculated based on the equation:

$$\% \text{ Inhibition} = \frac{\text{Absorbance control - Absorbance of the sample}}{\text{Absorbance control x 100}}$$

The absorbance value of each concentration variation is plotted on the inhibition curve and the IC value$_{50}$ determined.

GC-MS analysis

The GC-MS analysis is as follows: Dissolving the sample with ethanol (EtOH) in the microtube. Then it is homogenized in the following waydivortek, if the sample appears to be not homogeneous, then the sample solution is centrifuged at 9500 rpm for 3 minutes. The supernatant is taken and put in a GC vial, the sample solution is ready for injection.

Molecular docking analysis

Receptor and ligand preparation

Receptor macromolecule download from Protein Data Bank with sitehttp://www.rcsb.org/pdb/. Macromolecular data is downloaded in .pdb format. The protein macromolecules are separated from the solvent and the ligands or non-standard

residues. Separation of macromolecules from unnecessary molecules was carried out using the Discovery Studio 3.5 Visualizer program. The results of the separation will be used for belays which are stored in .pdb format.The structure of the identified bioactive compounds that will be used as ligands is then identified with the SMILES identity of each target ligand using PubChem after that it is compiled in a Notepad++ file with the .smi format.

Validation of molecular docking

Prior to molecular docking, each receptor was validated using the application with *Grid Box* which is adjusted to the distance of 5 Armstrong positions from *native ligand* each receptor. After obtaining a valid method, proceed with the docking process using YASARA-Structure.

Molecular docking target receptors and ligands

The resulting .smi file will be formatted to .sdf to make the tethering process easier *macro* smi2sdf. mcr. The resulting format is a combined format of the SMILES data-based ligand in the .sdf format. The resulting files are carried out using the docking process *virtual screening* on the macros available in the default file YASARA-*Structure*[9] on a validated virtual target.

Analysis and visualization of results from molecular docking

The results of the calculation of the belay are seen in output in notepad format. Determination of the ligand conformation resulting from the binding is carried out by selecting the ligand conformation that has the lowest binding energy (best pose). The ligand with the lowest binding energy value 3 will be seen from the position and orientation of the ligand in the macromolecule, and the amino acids bound to the ligand will be visualized with the Biovia tool to see the shape and volume match of the ligands and their mooring sites.

Profile determination molecular properties

Determination of molecular properties profiles including pharmacokinetic profiles, physicochemical properties and water solubility using the SWISS ADME website (swissadme.ch/index.php) using the SMILE format of compounds resulting from GC-MS characterization using the PubChem data bank (https://pubchem.ncbi.nlm.nih.gov/).

Treatment, analysis and presentation of data

In this study, the treatment involves two main processes: drying mangrove fruit, which is done either by sun drying or air drying indoors, and extracting the fruit in boiling water for varying durations of 5, 10, and 15 minutes. The data analysis is descriptive, using Microsoft Excel 2010 to calculate the average of three independent variables along with their standard deviations (SD). The results are presented in the form of tables and histograms for easy interpretation.

RESULTS AND DISCUSSION

Yield

The yield is calculated based on the ratio of the weight of the resulting extract to the weight of the sample used multiplied by 100%. The yield was obtained through extraction of mangrove fruit samples *S. alba* dried in the sun and mangrove fruit *S. alba* dried in the room using boiling water solvent for 5, 10 and 15 minutes. This yield data serves to determine the value of the active chemical compound components contained in the sample. Data on the yield of boiling water extract of mangrove fruit powder using extraction times of 5, 10 and 15 minutes can be seen in Figure 1.

The data in Figure 1 shows that the highest yield was found in samples dried in the sun with an extraction time of 15 minutes. This was due to the higher temperature in this drying method and the longer extraction time causing the cell walls to become brittle and the components easier to extract inside the cell.[10] reported that at the highest extraction temperature and the longest extraction time of 70°C for 20 minutes produced the highest yield of 70% ethanol extract of leaves.

Phytochemicals

Phytochemical screening data on mangrove fruit powder *S. alba* can be seen in Tables 1 and 2. The phytochemical components studied were alkaloids, tannins, saponins, steroids, triterpenoids and phenolics. Analysis was carried out qualitatively to determine whether or not these components were present in the extract. The data generated in the qualitative phytochemical analysis are only presented in two tables (Tables 1 and 2) because the extraction times of 5, 10 and 15 minutes contain the same phytochemical components. These data show that both samples dried in the sun and samples dried indoors for extraction times of 5, 10 and 15 minutes with boiling water contained the same phytochemical components, namely alkaloids, flavonoids, tannins, saponins and phenolics. Phytochemical content data showed that all extracts of boiling water of mangrove fruit *S. alba* Rich in secondary metabolites that function as antioxidants.

Phenolic compounds which is found on mangrove plants can protect this plant from damage from ultraviolet radiation. This is supported by the statement[11], that the trend of increasing production of phenolic compounds in mangrove plants occurs when the mangrove grows and survives under pressure. Flavonoids in plants also act as a protector against pressures originating from the environment. Furthermore according to[12], Flavonoids are the most important phenolic compounds, in which these compounds have a broad spectrum of chemical and biological activity including the scavenging of

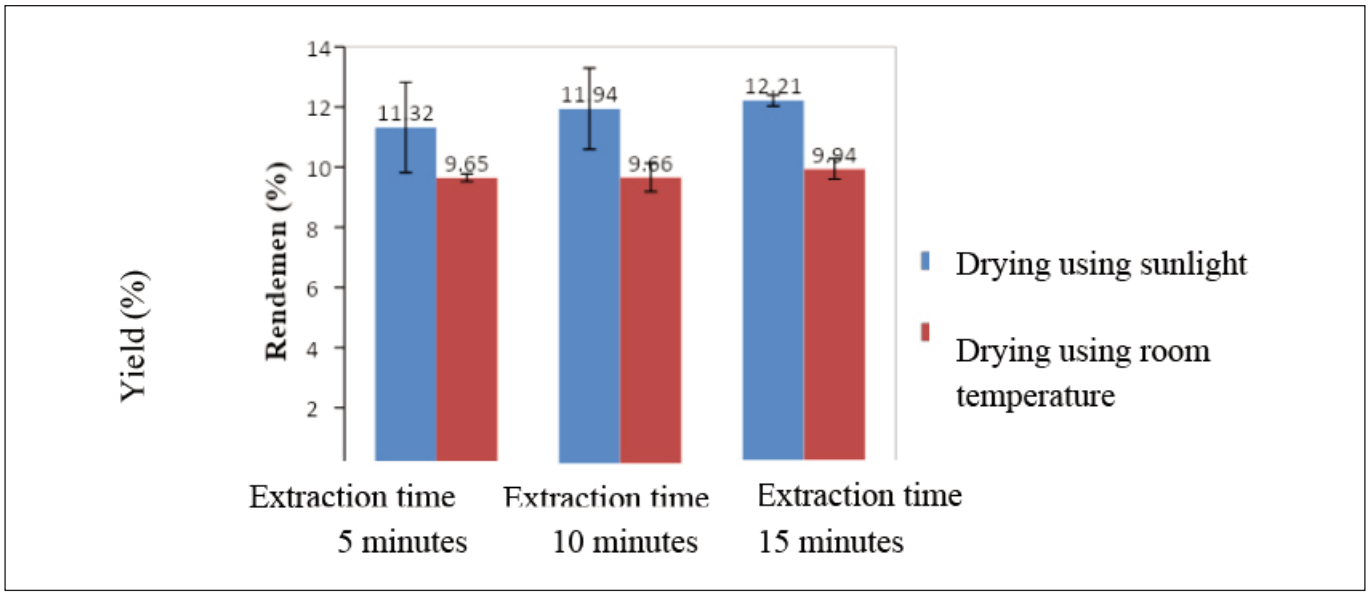

Figure 1. Yield of boiling water extract of mangrove fruit *Sonneratia alba*

Table 1. Phytochemicals of mangrove fruit extract dried in the sun with an extraction time of 5, 10 and 15 minutes with boiling water

Compound class	Results of analysis of phytochemical components qualitatively			Detectable results
	Connected1	Connected 2	Connected 3	
Alkaloid (Dragendorf, Wagner, Meyer)	+++	+++	+++	Dragendorf: Orange colorWagner: Brown colorMeyer: White precipitate
Flavonoid	+	+	+	Red
Tannin	+	+	+	Green color
Saponin	+	+	+	Bubbles / bubbles
Steroid	-	-	-	No discoloration
Triterpenoids	-	-	-	No discoloration
Phenolic	+	+	+	Orange brown color

free radicals which function as antioxidants. From the chemical structure, tannins and alkaloids also function as secondary metabolites that are able to scavenge free radicals so that they can function as antioxidants.

Total phenol content

Determination of total phenol in the sample extract using the method *Folin-Ciocalteu*. The principle of measurement of total phenol with reagents *Folin-Ciocalteu* namely based on the reduction of the hydroxy phenol group which is characterized by the formation of a blue complex compounds[13]. Determination of total phenol in sample extracts was carried out by measuring the absorbance of the extract diluted up to 1000 ppm at a wavelength of 750 nm with a UV-VIS spectrophotometer. The average total phenolic extract of mangrove fruit flour obtained can be seen in Figure 2.

The data in Figure 2 shows that the total phenol content for the three extraction times (5, 10 and 15 minutes) with boiling water did not show a significant difference, while the method of drying mangrove fruit produced a different total phenol content, where the method of drying mangrove fruit with sunlight The sun has a total phenol content in the extract higher than the sample drying method in the room. This is probably due to the shorter drying time in the sun than drying in the

Table 2. Phytochemicals of mangrove fruit extract which were dried in a room with an extraction time of 5, 10 and 15 minutes with boiling water.

Compound class	Results of analysis of phytochemical components qualitatively			Result of change
	Connected 1	Connected 2	Connected 3	
Alkaloid (Dragendorf, Wagner, Meyer)	+++	+++	+++	Dragendorf: Orange colorWagner: Brown colorMeyer: White precipitate
Flavonoid	+	+	+	Red
Tannin	+	+	+	Green color
Saponin	+	+	+	Bubbles
Steroid	-	-	-	No discoloration
Triterpenoids	-	-	-	No discoloration
Phenolic	+	+	+	Orange brown color

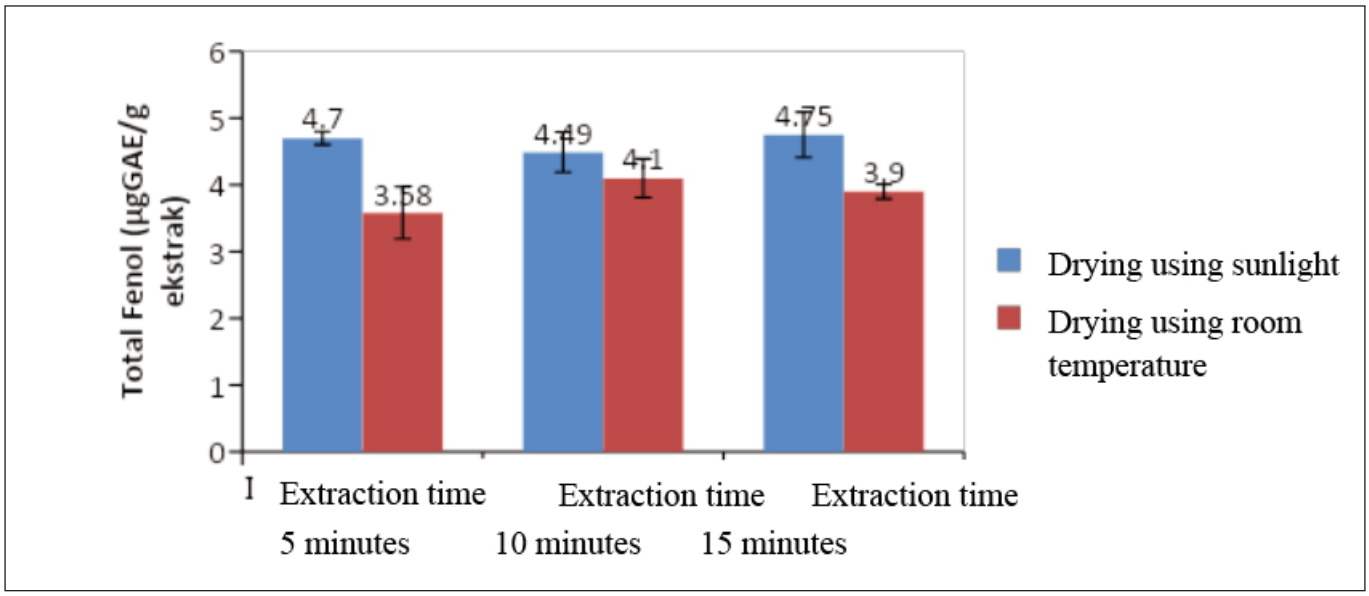

Figure 2. Total phenol content of boiling water extract of mangrove fruit *S. alba*

air in the room[14] stated that drying the sample for too long could destroy some of the phenols.

The data in Figure 2 shows that the total phenol content in the boiling water extract of mangrove fruit *S. alba* dried in the sun ranged from 4.49 ± 0.30 to 4.75 ± 0.34 µgGAE/g much higher than some research results, among others: methanol extract *A. Marina* and *R. apiculata* of 66.55mgGAE and 82.52 mgGAE/g[15], methanol extract of mangrove leaves *E. agolocha* of 60 mgGAE/g[16], methanol extract of fruit seeds *S. hospital* of 221 mgGAE/g[25], methanol extract of raw fruit *Pinch furicans*of 56.393 mgGAE/g[17], fruit methanol extract *S. alba* of 1508mgGAE/g[18], leaf methanol extract *S. alba*of 34.2 mgGAE/g[19], ethanol extract of mangrove leaves *S. alba* of 352 ± 9.77 mgGAE/g. The results of this study prove that boiling water is very potential as a solvent to extract phenolic components from mangrove plants.

Antioxidant activity of DPPH free radical inhibitors

Data on the antioxidant activity of 50% free radical inhibition of DPPH or *Inhibition Concentration 50 %* (IC_{50} DPPH) from mangrove fruit extract *S. alba* dried in the sun and dried

indoors and for a long time extraction 5, 10 and 15 minutes can be seen Figure 3.

The data in Figure 3 shows that the boiling water extract of mangrove fruit *S. alba* Sun drying has stronger antioxidant activity (IC_{50} DPPH is smaller) than boiling water extract of mangrove fruit *S. alba* indoor drying. Mangroves boiling water extract *S. alba* Sun drying for 5 minutes has an IC $value_{50}$ = 2.69 ± 0.32µg/ml better than control, namely Vitamin C which has IC_{50} = 5.04 ± 0.16µg/ml. The data in Figure 3 also shows a relationship between total phenol content and antioxidant activity, where boiling water extract of sun-dried mangrove fruit has a higher total phenol content than boiling water extract of dried mangrove fruit indoors[20,21] stated that the higher the total phenolic content of a material, the greater its antioxidant activity.

Antioxidant activity of boiling water extract of mangrove fruit *S. alba* better than mangrove plant extracts with other organic solvents, including IC_{50}DPPH leaf ethanol extract *S. alba* is 39.95 µg/ml[5], ethylacet extract of mangrove leaves *marine avicenia* have niali IC_{50} DPPH = 182.33 µg/ml[22], leaf methanol extract *A. marina* have an IC_{50}DPPH 248 µg/ml[29], skin methanol extract *R. mucronata* have an IC_{50}DPPH 483.8 µg/ml[23]. Antioxidant activity of boiling water extract of mangrove fruit *S. alba* the results of this study indicate a very strong antioxidant potential that is value IC_{50} less than 50 µg/ml[24] states that a material with an IC $value_{50}$<50 µg/ml is categorized as a very strong antioxidant.

GC-MS and Molecular Docking

Extraction results for 5 minutes in boiling water from mangrove fruit *S. alba* Sun drying has the best antioxidant activity, namely IC_{50} DPPH=2.69 ± 0.32. Based on these results, an in-silico test was carried out to strengthen the findings using the method *molecular docking.* Prior to molecular docking, it is necessary to know the various structures of the chemical compounds contained in the extract which are responsible for antioxidant activity. In this study, the structural characterization of organic compounds was carried out using the GC-MS method (*Gas Chromatography Mass Spectrometry*)[25] stated that the GC-MS method could be used as a reference for determining the molecular structure before doing so *molecular docking*. Figure 4 shows the GC-MS chromatogram of boiling water extract of mangrove fruit *S.alba* which has the best antioxidant activity.

The data in Figure 4, shows the results of reading 22 peaks with the results of compounds that meet the criteria for molecular docking, namely 39 compounds which have been summarized in Table 3. The compounds summarized include compounds that have been sorted against duplicated GC-MS results readings and compound containing silicon atoms. After obtaining the compounds from the results of the GC-MS above, is then carried out *molecular docking.* In this study, the binding of molecules to 4 target proteins was carried out, namely *Cytochrome c peroxidase* (PDB ID: 2X08), *Fibroblast collagenase* (PDB ID: 1CGL), *Human ROS1 Kinase Domain* (PDB ID: 3ZBF) and *Hyaluronidase* (PDB ID: 1FCV). The purpose of this molecule docking is to achieve optimal protein and ligand (target chemical compound) conformation so that the free energy of the system as a whole can be minimized and also the results of this molecule docking can describe the position and orientation of chemical compounds in the protein bond pocket by forming chemical interactions non covalent[26].

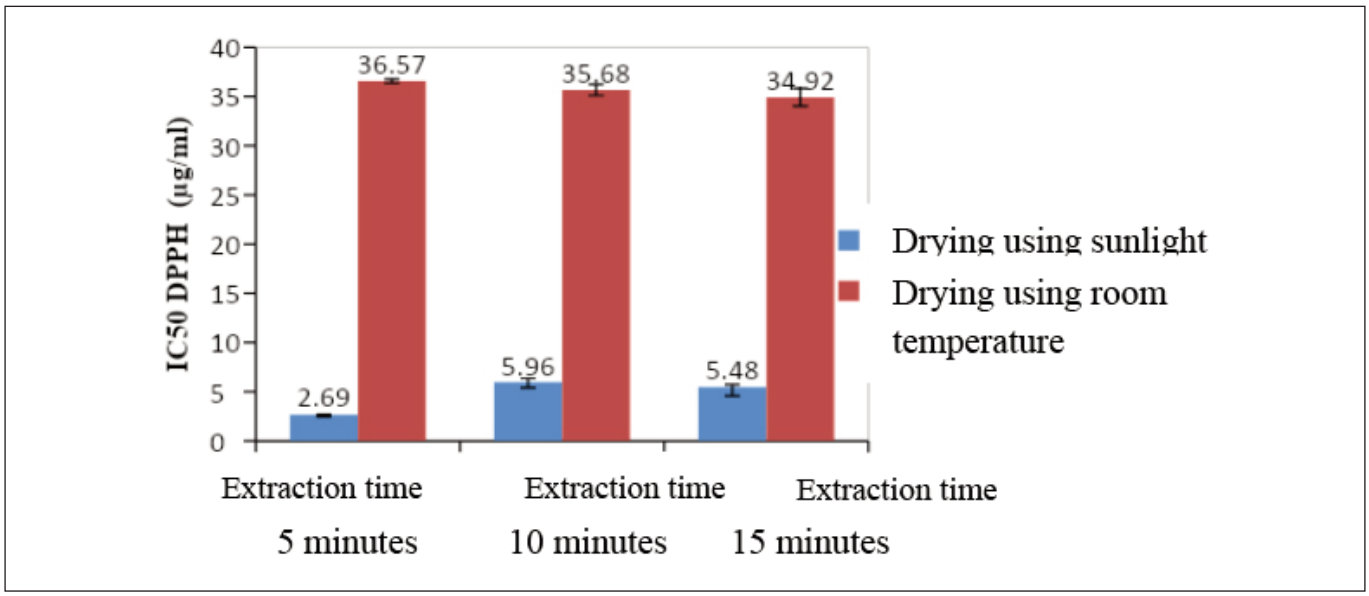

Figure 3. Antioxidant activity of boiling water extract of mangrove fruit *S. alba*

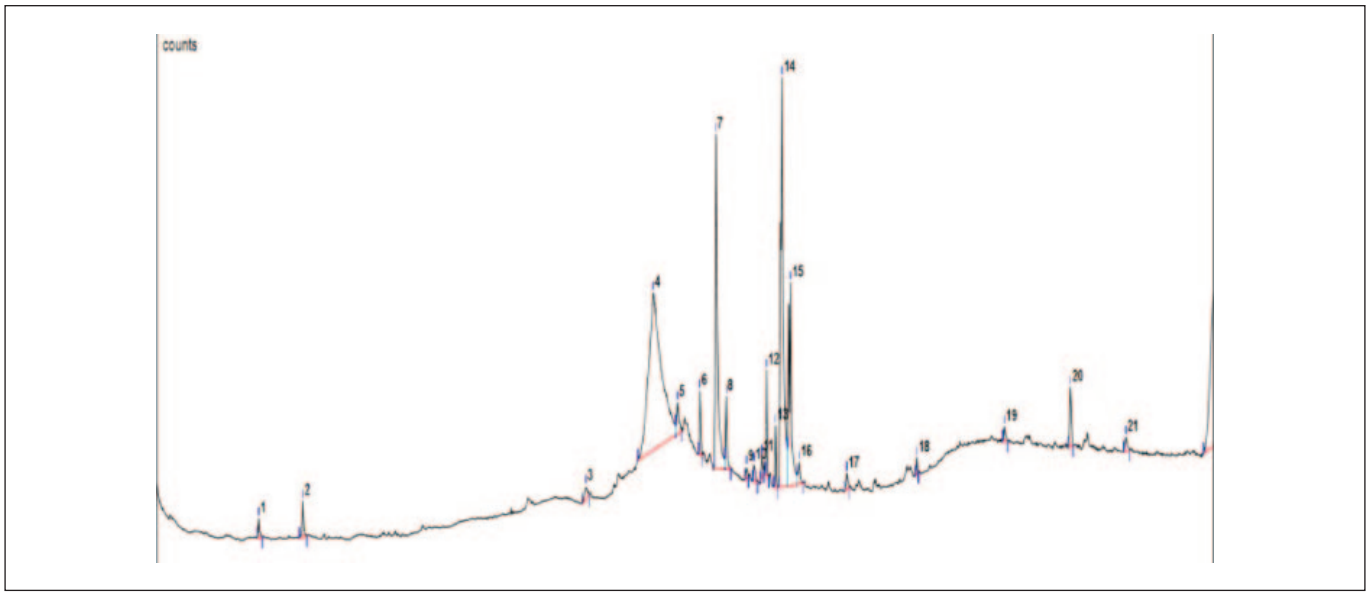

Figure 4. Chromatogram of GC-MS results of boiling water extract for 5 minutes from mangrove fruit *S. alba* sun drying

Cytochrome c peroxidase is one of the enzymes that plays a role in the initiation of cell death in the process of peroxidation of cardiolipin in mitochondria. When there is overproduction of *reactive oxygen species* (ROS) produced by free radicals, *Cytochrome c peroxidase* can release *haemoprotein* contained therein into the cytosol and can trigger the process of apoptosis[27]. Another target used in this research is *Fibroblast collagenase, Human ROS1 Kinase Domain* and *Hyaluronidase* in which the activation process of each of these proteins functions in the aging mechanism (*aging*), in this case ROS also plays an active role in inducing the activation of the 3 enzymes[28]. The results of molecular docking of the target proteins can describe the best candidates from the results of the virtual screening of compounds resulting from the characterization of boiling water extracts of mangrove fruit *S. alba* in assuming an inhibitory pose to the 4 target proteins.

Molecular docking begins with a validation process. The docking method was validated by analyzing the conformation of the original ligand. The docking results obtained were compared with the original crystallographic ligand conformations, which served as a reference for the validation and standardization of inhibitors[29]. The results of the validation of the belay method for this study can be seen in Figure 5, which shows that the belay protocol is valid with reference to the RMSD value (*root mean square deviation*) should be < 2Å[30]. Ascorbic acid was taken as a control to represent the control used in the DPPH antioxidant test.

The results of the validated protocol became input for molecular anchoring of 39 compounds resulting from structural characterization in boiling water extract of mangrove fruit *S. alba*. Table 3 shows the value of the binding affinity (kcal/mol) of the binding results for the target compound, *native ligand* and positive control, namely ascorbic acid.

The assessment of bond energy is an important parameter in evaluating the strength of the molecular interaction between the ligand and the receptor, where a lower or more negative binding affinity value indicates a stronger and more effective interaction between the ligand and the receptor[27]. Based on this, it can be seen that three compounds from each of the results of binding to the 4 target proteins, namely compounds **Ergosterol, Estra-1,3,5(10)-trien-17ß-ol gives Ergosta-5,8,22-trien-3-ol, (3ß,22E)-** which have binding affinity values of -9.238 kcal/mol, -8.676 kcal/mol and -7.882 kcal/mol for protein respectively **Cytochrome c peroxidase**; Compound **9(11)-Dehydroergosterol tosylate, Ergosta-5,8,22-trien-3-ol, (3ß,22E)- dan Ergosterol** has a binding value of -11,824 kcal/mol, - 9,318 kcal/mol and - 8,932 kcal/mol to protein **Fibroblast collagenase**; compound **Estra-1,3,5(10)-trien-17ß-ol, 9(11)-Dehydroergosterol tosylate, dan Dasycarpidan-1-methanol, acetate (ester)** with affinity values of -8,593 kcal/mol, -8,516 kcal/mol and -8,093 kcal/mol for protein **Human ROS1 Kinase Domain**; **9(11)-Dehydroergosterol tosylate, Ergosterol dan Ergosta-5,8,22-trien-3-ol, (3ß,22E)-** sequentially have a binding affinity of -8,398 kcal/mol, -7,458 kcal/mol and -7,270 kcal/mol for protein **Hyaluronidase**. All compounds compared with the results of the binding energy between the positive control and each target protein showed smaller results so that it could show that the ligand with the lowest binding energy showed the formation of the strongest complex with the highest receptor affinity compared to the bioactive compound ascorbic acid[41].

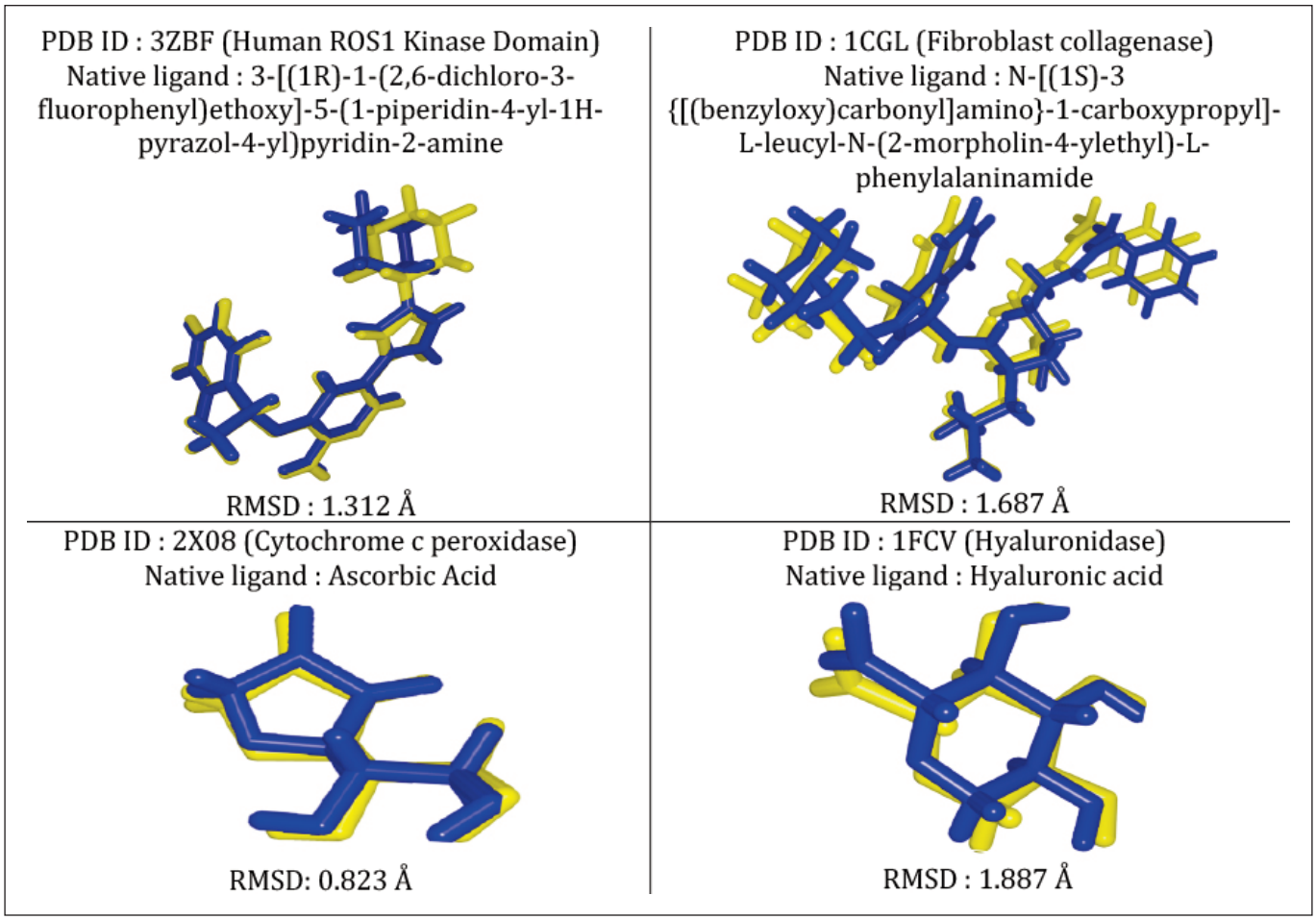

Figure 5. Molecular docking validation (Blue: Co-crystal ligand, Yellow: Re-docking ligand)

Table 3. Molecular docking results

No	Compounds	Molecular Formula	Molecular Weight	PubChem ID	Molecular Bonding Results (kcal/mol)			
					2X08 Cytochrome c peroxidase	1CGL Fibroblast collagenase	3ZBF Human ROS1 Kinase Domain	1FCV Hyaluronidase
1	(E)-9-Octadecenoic acid ethyl ester	$C_{20}H_{38}O_2$	310	5364430	-5.568	- 5.638	-4.971	-5.496
2	.psi.,.psi.-Carotene, 1,1',2,2'-tetrahydro-1,1'-dimethoxy-	$C_{42}H_{64}O_2$	600	5366411	-5.843	- 7.515	-6.627	-6.954
3	[1,1'-Bicyclopropyl]-2-octanoic acid, 2'-hexyl-, methyl ester	$C_{21}H_{38}O_2$	322	552098	-6.229	- 6.832	-5.797	-5.912
4	16-Octadecenoic acid, methyl ester	$C_{19}H_{36}O_2$	296	5364513	-5.722	- 5.378	-5.034	-5.312
5	2-Acetoxy-1,1,10-trimethyl-6,9-epidioxydecalin	$C_{15}H_{24}O_4$	268	538309	-5.920	- 6.822	-6.251	-5.341
6	3,7-Dimethyl-6-nonen-1-ol acetate	$C_{13}H_{24}O_2$	212	5363308	-6.384	- 5.642	-5.420	-5.452
7	3-O-Methyl-d-glucose	$C_7H_{14}O_6$	194	8973	-5.364	- 5.658	-3.818	-3.906
8	5,7,9(11)-Androstatriene, 3-hydroxy-17-oxo-	$C_{19}H_{24}O_2$	284	540760	-7.652	- 8.898	-7.896	-6.893

Table 3 continuation. Molecular docking results

No	Compounds	Molecular Formula	Molecular Weight	PubChem ID	Molecular Bonding Results (kcal/mol)			
					2X08 Cytochrome c peroxidase	1CGL Fibroblast collagenase	3ZBF Human ROS1 Kinase Domain	1FCV Hyaluronidase
9	7-Hydroxy-6-methyl-oct-3-enoic acid	$C_9H_{16}O_3$	172	536827	-5.913	- 6.205	-5.356	-4.825
10	7-Methyl-Z-tetradecen-1-ol acetate	$C_{17}H_{32}O_2$	268	5363222	-6.166	- 5.870	-5.528	-5.293
11	9(11)-Dehydroergosterol tosylate	$C_{35}H_{48}O_3S$	548	91691728	-7.444	-11.824	-8.516	-8.398
12	9-Octadecenoic acid, (2-phenyl-1,3-dioxolan-4-yl)methyl ester, cis-	$C_{28}H_{44}O_4$	444	5366356	-6.225	- 7.633	-6.498	-6.191
13	a-D-Mannofuranoside, 1-O-decyl-	$C_{16}H_{32}O_6$	320	552217	-6.370	- 6.162	-5.235	-5.704
14	cis-13-Octadecenoic acid	$C_{18}H_{34}O_2$	282	5312441	-5.786	- 5.944	-4.917	-5.150
15	cis-Vaccenic acid	$C_{18}H_{34}O_2$	282	5282761	-5.697	- 6.054	-4.778	-5.471
16	Cyclohexene, 1,5,5-trimethyl-6-acetylmethyl-	$C_{12}H_{20}O$	180	579163	-5.759	- 5.958	-5.610	-5.195
17	Cyclopropanetetradecanoic acid, 2-octyl-, methyl ester	$C_{26}H_{50}O_2$	394	552099	-5.278	- 5.933	-4.937	-5.592
18	Dasycarpidan-1-methanol, acetate (ester)	$C_{20}H_{26}N_2O2$	326	550072	-5.996	- 8.570	-8.093	-6.943
19	Desulphosinigrin	$C_{10}H_{17}NO_6S$	279	9601716	-6.767	- 7.036	-5.319	-4.925
20	Dodecanoic acid, 2,3-bis(acetyloxy)propyl ester	$C_{19}H_{34}O_6$	358	169212	-6.743	- 6.449	-5.330	-5.870
21	Ergosta-5,8,22-trien-3-ol, (3ß,22E)-	$C_{28}H_{44}O$	396	91743901	-7.882	- 9.318	-7.709	-7.270
22	Ergosterol	$C_{28}H_{44}O$	396	444679	-9.238	- 8.932	-8.014	-7.458
23	Estra-1,3,5(10)-trien-17ß-ol	$C_{18}H_{24}O$	256	9811784	-8.676	- 8.441	-8.593	-6.821
24	Heptadecanoic acid, 16-methyl-, methyl ester	$C_{19}H_{38}O_2$	298	110444	-5.452	- 5.264	-5.037	-5.264
25	Heptadecanoic acid, 9-methyl-, methyl ester	$C_{19}H_{38}O_2$	298	554038	-5.595	- 5.907	-4.593	-4.868
26	Hexadecanoic acid, 1-(hydroxymethyl)-1,2-ethanediyl ester	$C_{35}H_{68}O_5$	568	99931	-4.987	- 5.941	-5.369	-5.302
27	Hexadecanoic acid, 14-methyl-, methyl ester	$C_{18}H_{36}O_2$	284	520159	-6.008	- 5.940	-5.200	-4.512
28	Hexadecanoic acid, 1a,2,5,5a,6,9,10,10a-octahydro-5,5a-dihydroxy-4-(hydroxymethyl)-1,1,7,9-tetramethyl-11-oxo-1H-2,8a-methanocyclopenta[a]cyclopropa[e] cyclodecen-6-yl ester, [1aR-(1aa,2a,5ß,5aß,6ß,8aa,9a,10aa)]-	$C_{36}H_{58}O_6$	586	622252	-6.429	-8.705	-6.886	-6.454
29	Hexadecanoic acid, ethyl ester	$C_{18}H_{36}O_2$	284	12366	-5.706	- 5.521	-5.038	-5.322
30	Hexadecanoic acid, methyl ester	$C_{17}H_{34}O_2$	270	8181	-5.509	- 5.396	-5.479	-4.950
31	Isopropyl palmitate	$C_{19}H_{38}O_2$	298	8907	-5.438	- 5.479	-5.262	-4.974

Table 3 continuation. Molecular docking results

No	Compounds	Molecular Formula	Molecular Weight	PubChem ID	Molecular Bonding Results (kcal/mol)			
					2X08 Cytochrome c peroxidase	1CGL Fibroblast collagenase	3ZBF Human ROS1 Kinase Domain	1FCV Hyaluronidase
32	l-(+)-Ascorbic acid 2,6-dihexadecanoate	$C_{38}H_{68}O_8$	652	54722209	-4.840	- 7.431	-5.952	-5.498
33	Methyl stearate	$C_{19}H_{38}O_2$	298	8201	-5.527	- 5.825	-4.889	-4.828
34	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	256	985	-5.674	- 5.758	-4.800	-4.804
35	Octadecanoic acid	$C_{18}H_{36}O_2$	284	5281	-5.753	- 5.781	-4.938	-4.677
36	Paromomycin	$C_{23}H_{45}N_5O_{14}$	615	165580	-6.413	- 8.544	-7.130	-5.810
37	Pentadecanoic acid, 13-methyl-, methyl ester	$C_{17}H_{34}O_2$	270	554151	-5.758	- 5.380	-4.989	-5.383
38	ß-d-Mannofuranoside, methyl	$C_7H_{14}O_6$	194	6420200	-5.739	- 5.913	-4.331	-4.508
39	trans-13-Octadecenoic acid	$C_{18}H_{34}O_2$	282	6161490	-5.793	- 5.944	-4.917	-5.003
40	Ascorbic Acid	$C_6H_8O_6$	Native ligand	-5.316	-	-	-	
41	N-[(1S)-3-{[(benzyloxy)carbonyl]amino}-1-carboxypropyl]-L-leucyl-N-(2-morpholin-4-ylethyl)-L-phenylalaninamide	$C_{33}H_{47}N_5O_7$		-	-9.862	-	-	
42	3-[(1R)-1-(2,6-dichloro-3-fluorophenyl)ethoxy]-5-(1-piperidin-4-yl-1H-pyrazol-4-yl)pyridin-2-amine	$C_{21}H_{22}Cl_2FN_5O$		-	-	-9.576	-	
43	Hyaluronic acid	$(C_{14}H_{21}NO_{11})_1$		-	-	-	-4.574	
44	Ascorbic Acid (Kontrol +)	$C_6H_8O_6$	176	54670067	-5.316	-6.083	-4.410	-4.537

Figure 6 shows the binding pose between Ergosterol compounds, Estra-1,3,5(10)-trien-17ß-ol and Ergosta-5,8,22-trien-3-ol, (3ß,22E)- to Cytochrome protein c peroxidase. The three ligands form bonds with the amino acids His181, Val45, and Arg184 where these three amino acids become binding targets for γ-heme in cytochrome c peroxidase[29]. So it can be assumed that these three compounds can stabilize the binding bag of the cytochrome c peroxidase enzyme.

The binding to the Fibroblast collagenase protein in Figure 7 shows that the compound 9(11)-Dehydroergosterol tosylate appears to stabilize the collagenase protein with various chemical bonds, this is consistent with the very low binding value of -11,824 kcal/mol. It is also interesting when compared between the three compounds resulting from docking with collagenase protein, these three compounds form hydrogen bonds and hydrophobic bonds with the amino acid Leu 181. These findings form similarities with research conducted by[30].

Many Alkyl and Pi-Alkyl bonds are formed in Figure 8, which is an interaction between Estra-1,3,5(10)-trien-17ß-ol, 9(11)-Dehydroergosterol tosylate and Dasycarpidan-1-methanol, acetate (ester) and the Human ROS1 Protein Kinase Domain demonstrated that nonpolar (hydrophobic) amino acid residues tend to cluster together within the interior of the protein, preventing or minimizing contact with water molecules where hydrophobic interactions can take the form of Alkyl and Pi-Alkyl bonds[27]. So it can be concluded that the hydrophobic interaction between the ligand and the receptor can reduce the interaction between amino acid residues and water molecules.

Figure 9 shows the interaction between the Hyaluronidase protein and the target ligand. Interesting things were found

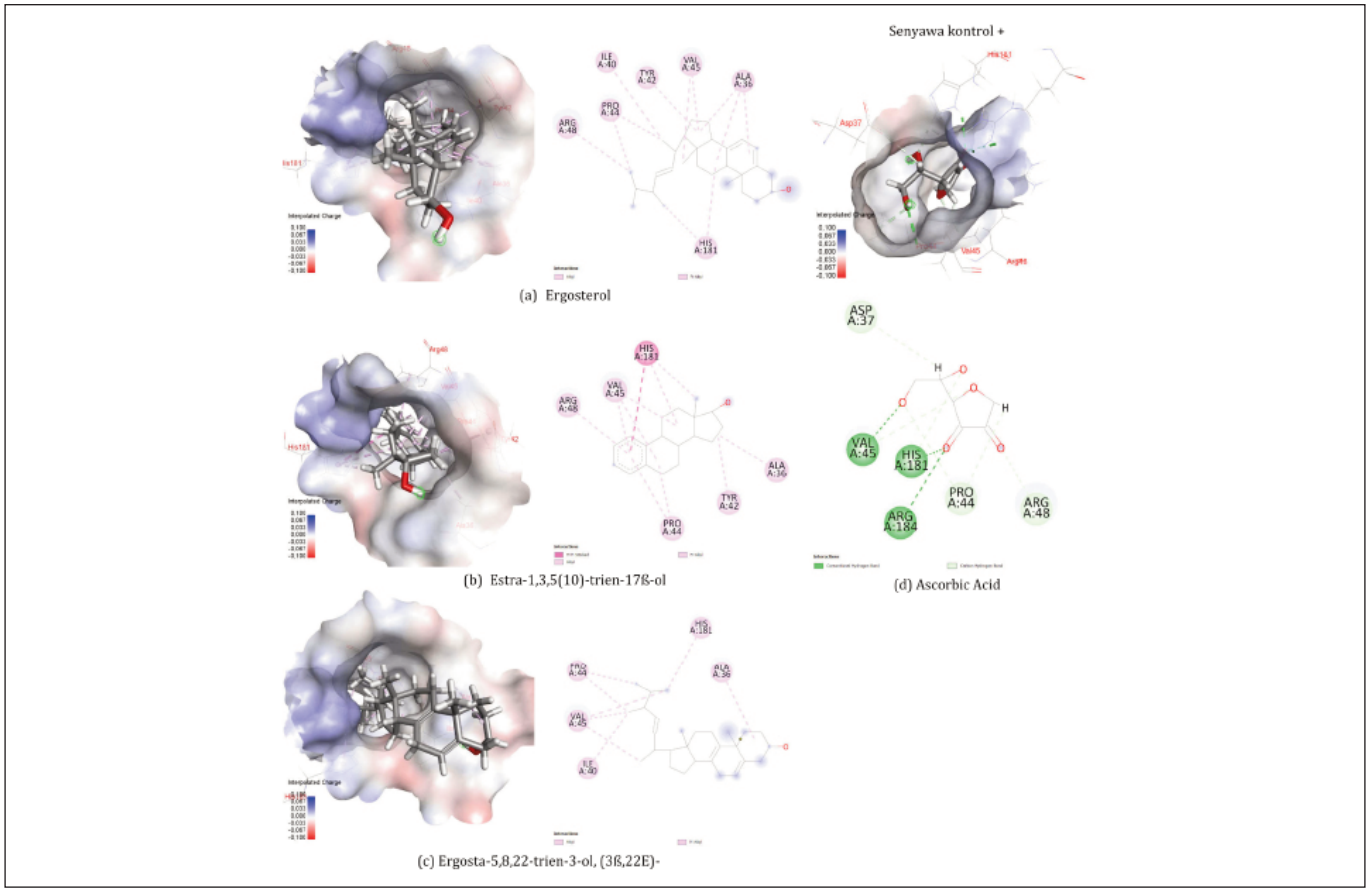

Figure 6. Interactions and binding poses with the target protein Cytochrome c peroxidase

in the interaction of the Ergosta-5,8,22-trien-3-ol, (3ß,22E)-compound which showed hydrogen interactions at the Glu 113 amino acid which showed the same thing as the ascorbic acid control. The hydrogen bonds that are formed may reflect the fact that the unique quantum effects of these bonds are required to build conformational dynamic systems that fluctuate in function of the interaction between the protein and the ligand compound. So it can be concluded that hydrogen bonds represent the universal language of life molecular interactions[27].

Table 4 below shows a summary of the bonds and interactions created between the 3 target compounds with the lowest binding values with each target receptor. Table 4 shows the results of the analysis *Molecular properties* compounds resulting from GC-MS characterization of boiling water extracts of mangrove fruit*S.alba*. The table shows the analysis of the pharmacokinetic profile with parameters*human gastrointestinal absorption* (HIA) (said to be good if >90%), *Blood-brain barrier* (BBB) *permeation, Permeability glycoprotein* (P-gp substrate) for which knowledge of compounds that are or are not substrates of glycoproteins is key to assessing active efflux through biological membranes, for example from the wall of the digestive tract into the lumen or from the brain[28]. It is known that inhibition of cytochromes P450 isoenzymes is one of the main causes of drug interactions related to pharmacokinetics. This may cause toxic effects or other unwanted side effects due to reduced clarification and accumulation of the drug or its metabolites[30]. *Skin permeability coefficient* (K_p) determines the more minus, the less the compound will permeate into the skin). Lipinski parameterization aims to describe the extent of the physicochemical activity of the target compound by relying on 5 standards. Molecular mass must be less than 500 Daltons, Lipophilicity (assumed with LogP less than 5), less than 5 *hydrogen bond donors*, less than 10*hydrogen bond acceptors*, and*Molar refractivity* with parameter values between 40-130.

Table 5 also presents the Drug-likeness score, indicating the bioavailability of a compound. The Water Solubility, measured on the Log S (Ali) scale, reflects the compound's ability to dissolve in water, which is crucial for developing health-related materials. The results show that the characteristics of each compound are determined through computational methods.

Table 4. Interaction of chemical bonds to ligands and controls on each target protein

Receptors	In the league		Interacting amino acids
	PubChem ID/Control	Compound	
PDB ID: 2X08 (Cytochrome c peroxidase)	444679	Ergosterol	Alkyl & Pi-Alkyl: HIS A:181, ARG A:48, PRO A:44, ILE A:40, TYR A:42, VAL A:45, ALA A:36
	9811784	Estra-1,3,5(10)-trien-17ß-ol	Pi-Pi Stacked: HIS A:181, VAL A:45, ARG A:48, PRO A:44, TYR A:42, ALA A:36
	91743901	Ergosta-5,8,22-trien-3-ol, (3ß,22E)-	Alkyl & Pi-Alkyl: PRO A:44, VAL A:45, ILE A:40, HIS A:181, ALA A:36
	control +	Ascorbic Acid	Conventional hydrogen bond: VAL A:45, HIS A:181, ARG A:184; Carbon Hydrogen Bond: PRO A:44, ARG A:48, ASP A:37
PDB ID: 1CGL (Fibroblast collagenase)	91691728	9(11)-Dehydroergosterol tosylate	Van der Waals: TYR B:240; Conventional hydrogen bond: LEU B:181; Carbon Hydrogen Bond: ASN B:180; Pi-Anion: GLU B:219; Pi-Pi T-shaped & Amide Pi-Stacked: SER B:239, HIS B:218; Alkyl & Pi-Alkyl: HIS B:222, HIS B:228, VAL B:215
	91743901	Ergosta-5,8,22-trien-3-ol, (3ß,22E)-	Alkyl & Pi-Alkyl: VAL A:215, HIS A:222, LEU A:181, TYR A:240, ALA A:182, HIS A:218, PRO A:238, HIS A:228
	444679	Ergosterol	Conventional hydrogen bond: LEU A:181; Alkyl & Pi-Alkyl: PRO B:238, ALA A:182, HIS A:218, HIS A:183, HIS A:228, HIS A:222
	control +	Ascorbic Acid	Conventional hydrogen bond: ARG A:214, HIS A:218, GLU A:219, ALA A:182; Carbon Hydrogen Bond: SER A:239, VAL A:215
PDB ID: 3ZBF (Human ROS1 Kinase Domain)	9811784	Estra-1,3,5(10)-trien-17ß-ol	Pi-Sigma: LEU A:1951; Alkyl: ALA A:1978, VAL A:1959, LEU A:2026, LYS A:1980, LEU A:2086.
	91691728	9(11)-Dehydroergosterol tosylate	Alkyl: LEU A:2035, LEU A:1951, LEU A:2086, ALA A:1978, LEU A:2010, LEU A:2026, VAL A:1959, LYS A:1980
	550072	Dasycarpidan-1-methanol, acetate (ester)	Alkyl & Pi-Alkyl: LEU A:2026, ALA A:1978, LEU A:2086, LEU A:1951, VAL A:1959
	control +	Ascorbic Acid	Carbon Hydrogen Bond: MET A:2029
PDB ID: 1FCV (Hyaluronidase)	91691728	9(11)-Dehydroergosterol tosylate	Pi-Sigma: TRP A:301; Pi-Pi Stacked: TYR A:184; Alkyl & Pi-Alkyl: TYR A:55, PRO A:18, PHE A:20, PHE A:46
	444679	Ergosterol	Pi-Alkyl: TRP A:267, TYR A:227, TRP A:301, TYR A:55
	91743901	Ergosta-5,8,22-trien-3-ol, (3ß,22E)-	Conventional hydrogen bond: GLU A:113,; Pi-Alkyl: TYR A:55, TRP A:301, TRP A:267
	control +	Ascorbic Acid	Conventional hydrogen bond: ASP A:111, GLU A:113, SER A:114; Carbon Hydrogen Bond: PHE A:112

CONCLUSION

The best antioxidant activity was found in the treatment of mangrove fruit *S. alba* Sun drying extracted with boiling water for 5 minutes, this treatment has antioxidant activity, namely IC_{50}DPPH= 2.69 ± 0.32 µg/ml better than the positive control, namely vitamin C, which has IC_{50}DPPH= 5.04±0.16µg/ml. The GC_MS characterization results of extracts that have the best antioxidant activity show 39 compounds that are in the category to be continued in the molecular docking process. The docking results showed Ergosterol bioactive compounds; Estra-1,3,5(10)-trien-17ß-ol; Ergosta-5,8,22-trien-3-ol, (3ß,22E)-; 9(11)-Dehydroergosterol tosylate; Dasycarpidan-1-methanol, acetate (ester) shows a very promising value of binding affinity towards 4 protein targets, namely *Cytochrome c peroxidase* (PDB ID: 2X08), *Fibroblast collagenase* (PDB ID: 1CGL), *Human ROS1 Kinase Domain*

Table 5. Molecular properties compounds resulting from GC-MS characterization of boiling water extracts of mangrove fruit *S. alba*.

No	Pharmacokinetics									Druglikeness		Water Solubility - Log *S* (wings) scales
	GI absorption	BBB permeant	P-gp substrate	CYP1A2 inhibitor	CYP2C19 inhibitor	CYP2C9 inhibitor	CYP2D6 inhibitor	CYP3A4 inhibitor	Log Kp (skin permeation)	Lipinski	He was Availability Score	
1	Low	No	No	Yes	No	No	No	No	-2.49 cm/s	Yes; 1 violation: MLOGP>4.15	0.55	Poorly soluble
2	Low	No	Yes	No	No	No	No	No	-0.03 cm/s	No; 2 violations: MW>500, MLOGP>4.15	0.17	Insoluble
3	High	No	No	Yes	No	Yes	No	No	-2.57 cm/s	Yes; 1 violation: MLOGP>4.15	0.55	Poorly soluble
4	High	No	No	Yes	No	No	No	No	-2.95 cm/s	Yes; 1 violation: MLOGP>4.15	0.55	Poorly soluble
5	High	Yes	No	No	No	No	No	No	-5.82 cm/s	Yes; 0 violation	0.55	Soluble
6	High	Yes	No	No	No	No	No	No	-4.61 cm/s	Yes; 0 violation	0.55	Moderately soluble
7	Low	No	No	No	No	No	No	No	-9.54 cm/s	Yes; 0 violation	0.55	Highly soluble
8	High	Yes	Yes	No	Yes	No	No	No	-6.56 cm/s	Yes; 0 violation	0.55	Soluble
9	High	Yes	No	No	No	No	No	No	-6.50 cm/s	Yes; 0 violation	0.85	Soluble
10	High	Yes	No	No	No	Yes	Yes	Yes	-3.55 cm/s	Yes; 1 violation: MLOGP>4.15	0.55	Poorly soluble
11	Low	No	Yes	No	No	Yes	No	Yes	-3.34 cm/s	No; 2 violations: MW>500, MLOGP>4.15	0.17	Poorly soluble
12	Low	No	Yes	Yes	No	Yes	No	Yes	-2.79 cm/s	Yes; 1 violation: MLOGP>4.15	0.55	Poorly soluble
13	High	No	Yes	No	No	No	No	No	-6.45 cm/s	Yes; 0 violation	0.55	Moderately soluble
14	High	No	No	Yes	No	Yes	No	No	-2.60 cm/s	Yes; 1 violation: MLOGP>4.15	0.85	Poorly soluble
15	High	No	No	Yes	No	Yes	No	No	-2.60 cm/s	Yes; 1 violation: MLOGP>4.15	0.85	Poorly soluble
16	High	Yes	No	No	No	No	No	No	-5.65 cm/s	Yes; 0 violation	0.55	Soluble
17	Low	No	Yes	Yes	No	No	No	No	-0.75 cm/s	Yes; 1 violation: MLOGP>4.15	0.55	Insoluble
18	High	Yes	Yes	No	No	No	Yes	No	-6.01 cm/s	Yes; 0 violation	0.55	Soluble

Table 5 continuation. Molecular properties compounds resulting from GC-MS characterization of boiling water extracts of mangrove fruit *S. alba*.

No	Pharmacokinetics									Druglikeness		Water Solubility - Log *S* (wings) scales
	GI absorption	BBB permeant	P-gp substrate	CYP1A2 inhibitor	CYP2C19 inhibitor	CYP2C9 inhibitor	CYP2D6 inhibitor	CYP3A4 inhibitor	Log Kp (skin permeation)	Lipinski	He was Availability Score	
19	Low	No	Yes	No	No	No	No	No	-8.91 cm/s	Yes; 0 violation	0.55	Very soluble
20	High	No	Yes	No	Yes	Yes	No	Yes	-4.65 cm/s	Yes; 0 violation	0.55	Poorly soluble
21	Low	No	No	No	No	Yes	No	No	-3.81 cm/s	Yes; 1 violation: MLOGP>4.15	0.55	Poorly soluble
22	Low	No	No	No	No	Yes	No	No	-3.44 cm/s	Yes; 1 violation: MLOGP>4.15	0.55	Poorly soluble
23	High	Yes	Yes	No	No	No	Yes	No	-4.76 cm/s	Yes; 1 violation: MLOGP>4.15	0.55	Moderately soluble
24	High	No	No	Yes	No	No	No	No	-2.32 cm/s	Yes; 1 violation: MLOGP>4.15	0.55	Poorly soluble
25	High	No	No	Yes	No	Yes	No	No	-2.23 cm/s	Yes; 1 violation: MLOGP>4.15	0.55	Poorly soluble
26	Low	No	Yes	No	No	No	No	No	0.20 cm/s	No; 2 violations: MW>500, MLOGP>4.15	0.17	Insoluble
27	High	No	No	No	No	Yes	No	No	-2.53 cm/s	Yes; 1 violation: MLOGP>4.15	0.55	Poorly soluble
28	Low	No	Yes	No	No	No	No	Yes	-4.12 cm/s	No; 2 violations: MW>500, MLOGP>4.15	0.17	Insoluble
29	High	No	No	Yes	No	No	No	No	-2.44 cm/s	Yes; 1 violation: MLOGP>4.15	0.55	Poorly soluble
30	High	Yes	No	Yes	No	No	No	No	-2.71 cm/s	Yes; 1 violation: MLOGP>4.15	0.55	Poorly soluble
31	High	No	No	Yes	No	No	No	No	-2.31 cm/s	Yes; 1 violation: MLOGP>4.15	0.55	Poorly soluble
32	Low	No	Yes	No	No	No	No	No	-0.30 cm/s	No; 2 violations: MW>500, MLOGP>4.15	0.56	Insoluble
33	High	No	No	Yes	No	No	No	No	-2.19 cm/s	Yes; 1 violation: MLOGP>4.15	0.55	Poorly soluble

Table 5 continuation. Molecular properties compounds resulting from GC-MS characterization of boiling water extracts of mangrove fruit *S. alba*.

No	Pharmacokinetics									Druglikeness		Water Solubility - Log *S* (wings) scales
	GI absorption	BBB permeant	P-gp substrate	CYP1A2 inhibitor	CYP2C19 inhibitor	CYP2C9 inhibitor	CYP2D6 inhibitor	CYP3A4 inhibitor	Log Kp (skin permeation)	Lipinski	He was Availability Score	
34	High	Yes	No	Yes	No	Yes	No	No	-2.77 cm/s	Yes; 1 violation: MLOGP>4.15	0.85	Poorly soluble
35	High	No	No	Yes	No	No	No	No	-2.19 cm/s	Yes; 1 violation: MLOGP>4.15	0.85	Poorly soluble
35	Low	No	Yes	No	No	No	No	No	-16.25 cm/s	When; 3 violations: MW>500, NorO>10, NHorOH>5	0.17	Highly soluble
37	High	Yes	No	No	No	Yes	No	No	-2.84 cm/s	Yes; 1 violation: MLOGP>4.15	0.55	Poorly soluble
38	Low	No	Yes	No	No	No	No	No	-9.37 cm/s	Yes; 0 violation	0.55	Highly soluble
39	High	No	No	Yes	No	Yes	No	No	-2.60 cm/s	Yes; 1 violation: MLOGP>4.15	0.85	Poorly soluble

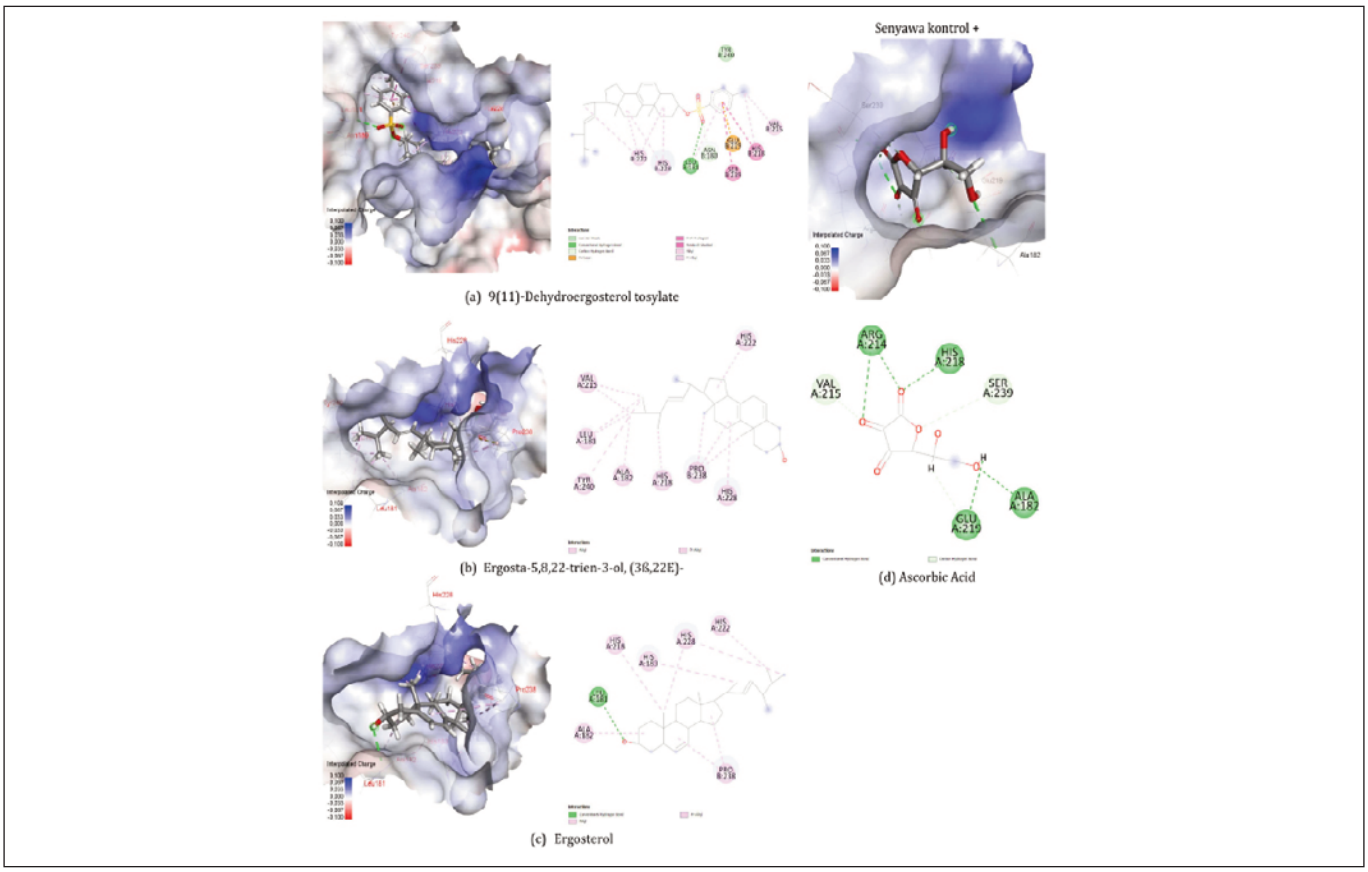

Figure 7. Interactions and binding poses with the target protein Fibroblast collagenase

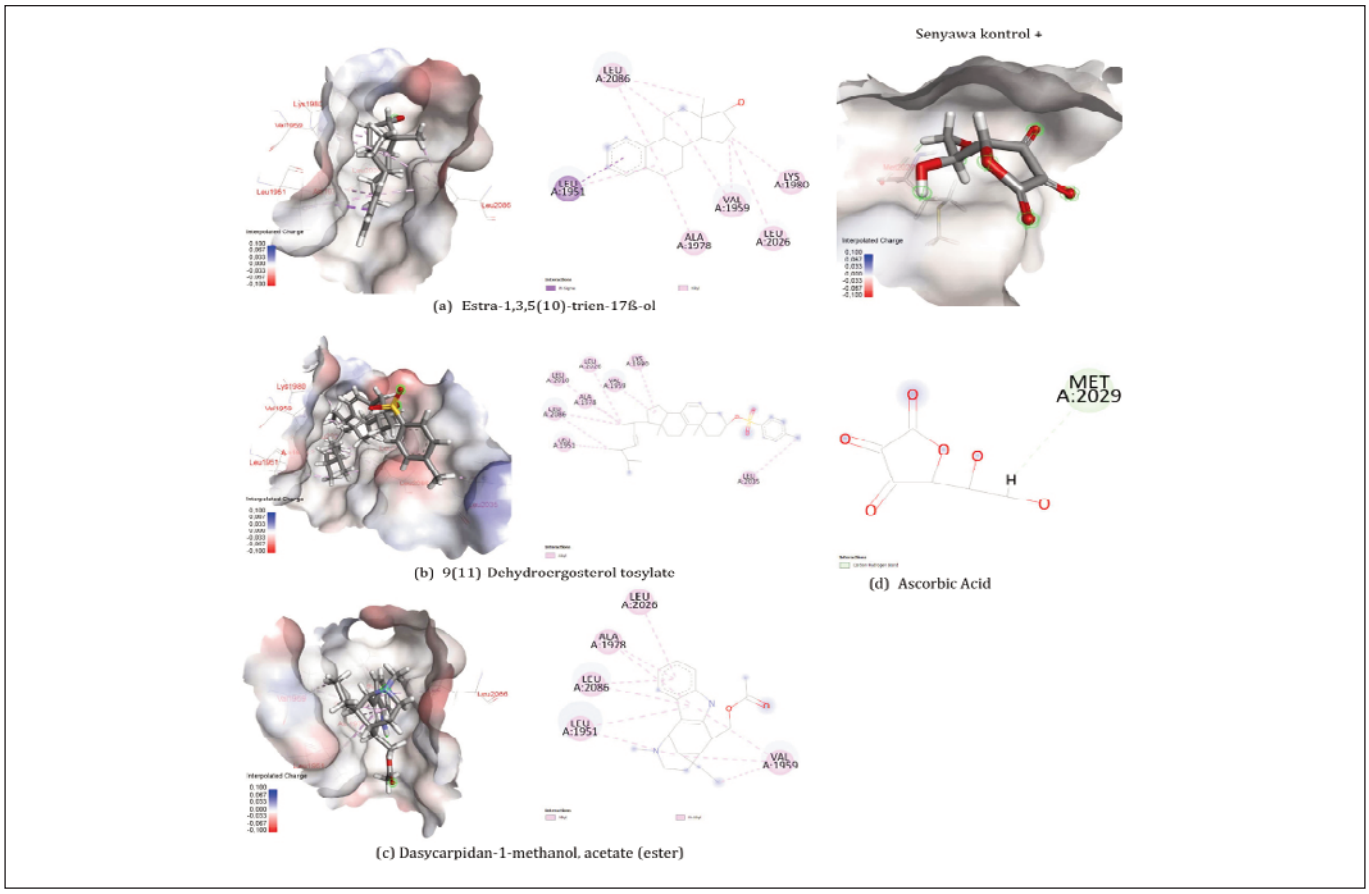

Figure 8. Interactions and binding poses with the target protein human ROS1 kinase domain

(PDB ID: 3ZBF) and *Hyaluronidase* (PDB ID: 1FCV) with vitamin C as control. Based on these findings, boiling water extract of mangrove fruit *S. alba* this can provide a good potential in the discovery and development of candidate new antioxidant compounds, so that further testing is needed to determine the bioactivity of these new antioxidant compounds in real terms, so that the boiling water extract of mangrove fruit *S. alba* It is acceptable as an antioxidant functional food.

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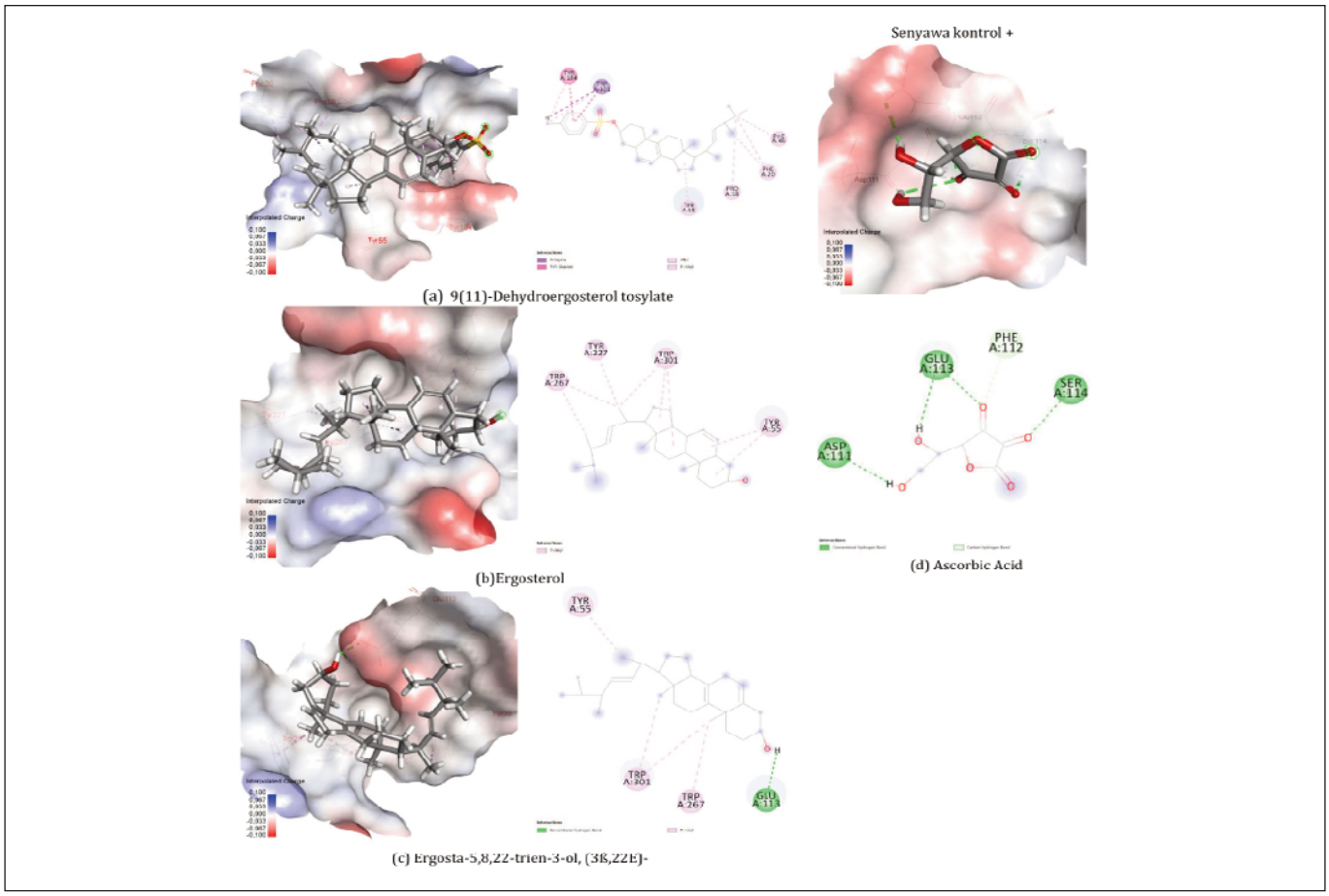

Figure 9. Interactions and binding poses with Hyaluronidase target proteins

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