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Original Article

Antiviral potential of selenium complexes from *Brassicaceae* by inhibiting protein bond between MAP4 and the spike of SARS-CoV-2

[Potencial antiviral de complejos de selenio de *Brassicaceae* mediante la inhibición de la unión proteica entre MAP4 y la espiga de SARS-CoV-2]

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Abstract

Context: Coronavirus disease 2019 (COVID-19), a highly contagious viral disease caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), has become a worldwide pandemic. Selenium derived from the plants of the *Brassicaceae* family plays an important role in several biological functions, often as an antioxidant or antiviral. In particular, selenium can be used as an adjuvant in the treatment of various viral infections.

Aims: To determine the potential drug, as well as the affinity and the bond energy associated with chemical interaction bonds between the complex selenium compounds from *Brassicaceae* as inhibitors of the SARS-CoV-2 spike protein and MAP4 through the use of *in silico* studies.

Methods: The methods used in this study consisted of receptor and ligand data collection, ADMET analysis, molecular docking, and molecular dynamics.

Results: The results of this study indicate that the selenium complex compounds have the potential to be used as a spike inhibitor drug for SARS-CoV-2, as they have passed the Lipinski test and showed promise in the pharmacokinetic analysis. The results of the bond docking show that several complex selenium compounds, such as ethaselen, selenomethionine, and selenocystine, have stronger binding affinity values than the controls. This is compared to the control MAP4, which yielded binding affinities of -6.1 kcal/mol and spike protein -7 kcal/mol, respectively. Controlled bisoxatin and estramustine are drugs with mechanisms targeted to MAP4 and spike protein, which are the usual standards used.

Conclusions: The similarity of sites, the binding of several amino acid residues dominated by hydrogen bonds, and the result of molecular dynamic results for the selenium compound derived from *Brassicaceae* showed a stable bond to the spike protein and MAP4 with low fluctuation levels.

Keywords: MAP4; SARS-CoV-2; selenium; spike protein.

Resumen

Contexto: La enfermedad por coronavirus 2019 (COVID-19), una enfermedad vírica altamente contagiosa por coronavirus de tipo 2 causante del síndrome respiratorio agudo severo (SARS-CoV-2), se ha convertido en una pandemia mundial. El selenio derivado de las plantas de la familia *Brassicaceae* desempeña un papel importante en varias funciones biológicas, a menudo como antioxidante o antiviral. En particular, el selenio puede utilizarse como coadyuvante en el tratamiento de diversas infecciones virales.

Objetivos: Determinar el fármaco potencial, así como la afinidad y la energía de enlace asociada a los enlaces de interacción química entre los compuestos complejos de selenio de Brassicaceae como inhibidores de la proteína espiga del SARS-CoV-2 y MAP4 mediante el uso de estudios *in silico*.

Métodos: Los métodos utilizados en este estudio consistieron en la recopilación de datos de receptores y ligandos, análisis ADMET, docking molecular y dinámica molecular.

Resultados: Los resultados de este estudio indican que los compuestos del complejo de selenio tienen potencial para ser utilizados como fármaco inhibidor de picos para el SARS-CoV-2, ya que han superado el test de Lipinski y se han mostrado prometedores en el análisis farmacocinético. Los resultados del acoplamiento de enlaces muestran que varios compuestos complejos de selenio, como el etaseleno, la selenometionina y la selenocisteína, tienen valores de afinidad de enlace más fuertes que los controles. Esto se compara con el MAP4 de control, que arrojó afinidades de unión de -6,1 kcal/mol y proteína pico -7 kcal/mol, respectivamente. La bisoxatina y la estramustina controladas son fármacos con mecanismos dirigidos a MAP4 y spike protein, que son los patrones habituales utilizados.

Conclusiones: La similitud de los sitios, la unión de varios residuos de aminoácidos dominados por enlaces de hidrógeno, y el resultado de los resultados de dinámica molecular para el compuesto de selenio derivado de *Brassicaceae* mostraron una unión estable a la proteína espiga y MAP4 con bajos niveles de fluctuación.

Palabras Clave: MAP4; SARS-CoV-2; selenio; proteína espiga.

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INTRODUCTION

Coronavirus Disease-19 (COVID-19) is an infectious viral disease caused by the SARS-CoV-2 virus and has caused disastrous effects on world demographics, resulting in more than seven million deaths worldwide as the most impactful global health crisis since the influenza pandemic era of 1918. After the first case of this respiratory viral disease was reported in Wuhan, Hubei Province, China, in late December 2019, SARS-CoV-2 rapidly spread worldwide, forcing the World Health Organization (WHO) to declare it a global pandemic on March 11, 2020. Since being declared a global pandemic, SARS-CoV-2 has hit many countries around the world, resulting in the quarantine of millions of residents in affected cities and an explosion of positive cases in healthcare facilities (Lai et al., 2020).

Reinfection cases were noted to be more dangerous, with more deaths than during the first infection. Organ damage can occur as the SARS-CoV-2 virus infection spreads throughout the body, especially in important organ systems such as the respiratory, digestive, and nervous systems, which increase the risk of death (Endharti et al., 2018). SARS-CoV-2 attacks host cells through the spike protein (S), which mediates the entry of the virus through the angiotensin-converting enzyme 2 (ACE2) receptor, which is found on various surfaces of organs and tissues in the human body. Then, the virus fuses with the host cell's membrane, where it releases its genome into the cytoplasm of the cell, utilizing the host cell's organelles to replicate its RNA. After that, the newly synthesized viral RNA and viral proteins are assembled into virions and buds from the cell membrane of the host cells to complete the virus particle (Lai et al., 2020). The spread of a SARS-CoV-2 virus infection from one cell to another is highly dependent on microtubule activity (Yoshimoto, 2020).

Microtubules are an important part of the cytoskeleton, which is one of the elements used by SARS-CoV-2 to facilitate the processes of infection, proliferation, damage of host cells, and spread of infection to other cells. Microtubules have protective proteins around them that maintain the stability of the function of microtubules, where the most abundant type of protein is microtubule-associated protein 4 (MAP4) (Permana et al., 2020). In its process of taking over microtubules, the spike protein part of the SARS-CoV-2 virus will bind to MAP4, which is widely spread around the microtubules. Preventing the SARS-CoV-2 spike protein from entering the microtubule system and spreading to other cells requires an appropriate inhibitor in its prevention mechanism (Simpson and Yamauchi, 2020).

Previous research by Rakib et al. (2021) used a molecular modeling approach to identify several antiviral compounds that have the potential to inhibit the main proteases of SARS-CoV-2; one of these compounds is selenium. Selenium (Se) is a chemical element and a micronutrient that the body needs in small amounts, but it plays an important role in several biological functions, especially as an antioxidant and antiviral. The potential utility of Se is reaching, especially in the context of nutritional supplements. One of the high levels of selenium is found in plants of the *Brassicaceae* family, known as Se accumulators. The current treatment option for SARS-CoV-2 is not sufficient to avoid the risk of target organ damage due to infection with the SARS-CoV-2 virus. Several studies have suggested that selenium complexes may represent a powerful antiviral agent. In particular, selenium can be used as an adjuvant for the treatment of viral infections (Boopathi et al., 2019).

Selenium intake can help prevent several metabolic disorders and provide protection against viral infections. Currently, the epidemic caused by SARS-CoV-2 threatens human health in several countries and has an impact on the global economy. Therefore, Se supplementation could be a complementary treatment to vaccines and pharmacological drugs to reduce viral load, mutation frequency, and improve the immune system of populations with a low intake of Se in the diet (Aminpour et al., 2022). Selenium in plants is extraordinary, especially for the purposes of phytoremediation and Se nutritional supplementation. *Brassica juncea* (Indian mustard) is known as a Se accumulator. Apart from Indian mustard greens, the unique plant *Brassicaceae* is also consumed in Japan. *Brassica rapa* var. hakabura, commonly known as nozawana, and *Brassica rapa* var. peruviridis, commonly known as komatsuna, is a typical *Brassicaceae* plant (Gosh et al., 2020; Kim et al., 2020).

The purpose of this study was to determine the potential of the complex selenium compound found in the plants of the family *Brassicaceae* as a good drug candidate based on Lipinski and pharmacokinetic tests, and to determine the affinity and fluctuation value formed of the chemical interaction bond between the selenium compound and the SARS-CoV-2 spike protein and MAP4. This research is expected to contribute to the development of science on the benefits of potential bioactive selenium compounds found in the plants of the *Brassicaceae* of plants, aid in empowering natural diversity, and can serve as a basis for further research and development related to future COVID-19 drug candidates (Uddin et al., 2020).

Based on the description above, bioactive selenium compounds from the *Brassicaceae* family are used. This research is a way to empower natural diversity that has the potential to inhibit the spread of viruses through MAP4 inhibition at the microtubules. Information should be available to the public. Based on this background, it decided to carry out *in silico* research to determine the potential of the selenium compounds complex found in *Brassicaceae* as a drug candidate as well as the mechanism of its inhibitory effect in the spread of infection with SARS-CoV-2 related to its binding to MAP4 and spike protein.

MATERIAL AND METHODS

Data mining

Data mining of complex selenium compounds was carried out in the *Brassicaceae* family (Table 1). Data for the compounds (PubChem ID, canonical smiles, and two-dimensional structures) were taken from the PubChem database (https://pubchem.ncbi.nlm.nih.gov).

Table 1. Data mining of complex selenium compounds.

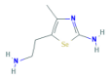
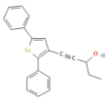
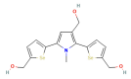
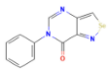
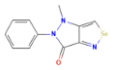
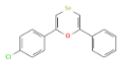
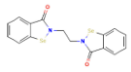
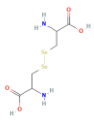
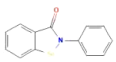
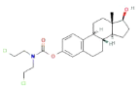
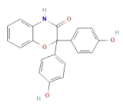
Selenium compound	Plant	Molecular formula	Pubchem ID	Chemical 2D structure
Amselamine	*Brassica oleracea*	$C_6H_{11}N_3Se$	9990475	
1-(2,5-Diphenylselenophen-3-yl)pent-1-yn-3-ol	*Brassica oleracea* var. acephala	$C_{21}H_{18}OSe$	24786497	
1H-Pyrrole-3-methanol, 2,5-bis(5hydroxymethyl) selenophene-2-yl)	*Brassica oleracea* var. gemmifera	$C_{16}H_{17}NO_3Se_2$	11539212	
6-Phenyl-7(6H)-isoselenazolo(4,3-d)pyrimidone	*Brassica napus*	$C_{11}H_7N_3OSe$	134741	
4,5-Dihydro-4-methyl-6-oxo-5-phenyl-6H-pyrazolo(4,5-c)isoselenazole	*Brassica nigra*	$C_{11}H_9N_3OSe$	128911	
2-(4-Chlorophenyl)-6-phenyl-1,4-oxaselenin	*Brassica napus* var. napobrassica	$C_{16}H_{11}ClOSe$	10544652	
Ethaselen	*Brassica oleracea* var. ramosa	$C_{16}H_{12}N_2O_2Se_2$	10387485	
Selenomethionine	*Brassica oleracea* var. botrytis	$C_5H_{11}NO_2Se$	15103	
Selenocystine	*Brassica oleracea* var. gemmifera	$C_6H_{12}N_2O_4Se_2$	15104	
Se-Methylselenocystine	*Brassica oleracea*	$C_4H_9NO_2Se$	147004	
Ebselen	*Brassica juncea*	$C_{13}H_9NOSe$	3194	
Estramustine	Drug (MAP4 control)	$C_{23}H_{31}Cl_2NO_3$	259331	
Bisoxatin	Drug (spike control)	$C_{20}H_{15}NO_4$	28689	

Table 2. Specific active sites of MAP4.

Protein	Docking coordinates	
	Center	Dimension
MAP4	X: 207.6483	X: 48.2653
	Y: 175.6844	Y: 36.8113
	Z: 239.8342	Z: 65.0024
Spike	X: 155.7121	X: 17.8571
	Y: 206.0474	Y: 11.7994
	Z: 219.9416	Z: 18.1501

ADMET analysis

ADMET analysis was carried out using the SwissADME web server to predict whether the compound is feasible as a drug candidate. Selenium compounds from plants of the *Brassicaceae* family were then checked for their potential as drug candidates using the SwissADME web server (http://www.swissadme.ch/) to check the physicochemistry, pharmacokinetics, solubility, lipophilicity, and drug-likeness of the selenium compounds. Compounds that are good drug candidates should not violate the Lipinski rule, have five or fewer hydrogen bond donors, have ten or fewer hydrogen bond acceptors, and have a molecular mass of 500 kg/mol or under (Hollingsworth and Dror, 2019). The pharmacological properties of the bioactive compounds were analyzed using the SwissADME web server (http://www.swissadme.ch). The analysis was performed using each selenium complex canonical smile from the PubChem database and then sent to SwissADME, which is used to display the results of the characteristics and pharmacological properties. In predicting the potential of a ligand to be used as a drug candidate, the drug-likeness method is needed to determine the ability of the ligand to absorb.

Molecular docking

The docking process begins with the preparation of the ligands and the receptor proteins. The preparation was done with the application of PyRx to minimize the binding energy (Gudimchuk and McIntosh, 2021). Ligands were prepared with the OpenBabel menu in the PyRx software, while proteins were prepared with the Biovia Discovery Studio Visualizer 2019 software in order to remove water molecules and pollutant ligands (Mawaddani et al., 2020). The docking process was carried out with the center of the MAP4 protein at the coordinates (Å) X= 207.6483 Y= 175.6844 Z= 239.8342 and dimension (Å) X = 48.2653 Y = 36.8113 Z = 65.0024 and center coordinates of X: 155.7121 Y: 206.0474 Z: 219.9416 and dimension (Å) X: 17.8571 Y: 11.7994 Z: 18.1501 with spike protein at docking coordinates (Table 2). The Vina Wizard menu in PyRx software is used to obtain binding affinity values for MAP4 and spike bonds with selenium complex compounds or controls. After docking, the docked file is saved in PDB format and then opened using the Biovia Discovery Studio Visualizer 2019 software to see the bond resulting from the docking, the 2D interaction structure formed, and the visualization of the MAP4 and spike protein bond when it binds to complex compounds or control (estramustine and bisoxatin).

Visualization

In addition to these previous assays, a PASS test was carried out to predict the biological activity of molecular docking after the compounds were tested using Lipinski's rules. The molecular docking was carried out using Vina Wizard in the PyRx software in order to determine the bond strength of complex selenium compounds to MAP4 and spike protein. The results of these bonds are then compared with the bonds of estramustine and bisoxatin as control ligands. Bonding affinity is defined as the average value of the bond-free energy formed from the bonding conformations of the molecules involved in the docking site. Docking is also useful in the process of predicting the pose of a ligand when it attaches to a target protein (Mawaddani et al., 2020).

Molecular dynamics

Molecular dynamics were carried out to estimate the stability of the bonding of the selenium compounds, which were exhibited as if the process was taking part in the cell and compared to the controls estramustine and bisoxatin. Each selenium compound was combined with MAP4 and spike protein using the PyMOL software and saved in PDB format. Data is then entered into the web CABS-flex 2.0 (http://biocomp.chem.uw.edu.pl/CABSflex2) with 100 cycles; the selection chain, in this case, is a chain. Other settings for this process follow the default web server settings (Fedorov et al., 2019). The highest root-mean-square fluctuation (RMSF) value indicates more flexibility, while the lower values suggest limited

system motion during simulation (Hollingsworth and Dror, 2019). The result is the residual fluctuation of plot data stored in CSV and graphic formats.

Data analysis

The data obtained were analyzed at each stage, including the validation of the docking method, the predicted value of the Lipinski rule, the prediction of absorption and distribution, the prediction of toxicity, the chemical interaction of the ligand and target protein, the binding affinity interaction and molecular dynamics interaction of the ligand and target protein. The ligand interactions of MAP4 and spike proteins were compared to those of native ligands on target proteins.

RESULTS

ADMET analysis

Analysis of the selenium compounds as drug potency drug-likeness test was carried out using SwissADME. Compounds that are good drug candidates do not have violations of Lipinski's rule, MLogP values have hydrogen bond donors less than equal to 5, MLogP values less than equal to 5, have hydrogen bond acceptors less than equal to 10, and molecular mass must be less than equal with 500 (Chinnasamy et al., 2019). Based on the observations at SwissADME, it appears that selenium complex compounds have the potential to become drugs because these compounds do not violate the Lipinski rules. Selenium complex compounds have an average molecular weight below 500 g/mol (328.75 g/mol), have an average MLogP value below 5 (-82.1), the number of hydrogen bond acceptors is below 10 (2.85), and the average number of hydrogen bond donors is below 5 (Table 3).

The pharmacokinetic analysis of selenium compounds involved water solubility tests, gastrointestinal absorption assays, P-gp analysis, testing of their ability to pass through the hematoencephalic barrier, and topological polar surface area (TPSA) analysis. The solubility of the compounds is an important factor in drug development, especially for drugs intended for oral and inhalation administration. Drug candidates should have a high aqueous solubility so the compounds can work in the body effectively. The solubility value can predict the absorption rate and ease of distribution of the drug in the body. Predictions using SwissADME will produce a value in LogS, the decimal logarithmic value of molar solubility in water in units of mol/L or mg/mL. Based on ESOL (Estimated SOLubility) rules, LogS values can be interpreted qualitatively, where LogS values close to 0 indicate a high solubility, values between -4 to -6 indicate moderate solubility levels, though all values above -6 indicate a general solubility. The selenium compounds have a LogS value of -2.66, indicating that this compound is soluble (Table 4). The pharmacokinetics of this compound suggest that it has a high absorption rate in the intestine, cannot be pumped out by P-gp, cannot enter through the hematoencephalic barrier, and has a TPSA value below 140 Å² (127.45 Å²) (Sethi et al., 2019).

Table 3. Lipinski rule analysis on complex selenium compounds.

Compounds (formula)	MW (g/mol)	MLogP	HBA	HBD	Lipinski
	≤500 g/mol	≤5	≤10	≤5	
Selenocystine ($C_6H_{12}N_2O_4Se_2$)	334.09	-5.83	6	4	Yes, 0 violation
Selenomethionine ($C_5H_{11}NO_2Se$)	196.11	-2.20	3	2	Yes, 0 violation
Ethaselen ($C_{16}H_{12}N_2O_2Se_2$)	422.20	2.50	2	0	Yes, 0 violation
$C_{21}H_{18}OSe$	365.33	4.35	1	1	Yes, 0 violation
$C_{16}H_{17}NO_3Se_2$	429.23	0.68	3	3	Yes, 0 violation
$C_{11}H_7N_3OSe$	276.15	0.86	3	0	Yes, 0 violation
$C_{11}H_9N_3OSe$	278.17	1.71	2	0	Yes, 0 violation
Estramustine (MAP4 control drug)	440.40	4.47	3	1	Yes, 1 violation
Bisoxatin (spike control drug)	333.34	2.08	3	4	Yes, 0 violation

MW: molecular weight; HBA: hydrogen bond acceptor; HBD: hydrogen bond donor.

Table 4. Pharmacokinetics of complex selenium compounds.

Compound (formula)	Water solubility (ESOL)			Pharmacokinetics		
	LogS	Solubility	Class	GI absorption	P-gp	TPSA ≤140 Å²
$C_6H_{12}N_2O_4Se_2$	-2.48	6.14e+06 mg/mL; 1.84e+04 mol/L	Highly soluble	Low	No	126.64 Å²
$C_5H_{11}NO_2Se$	0.54	6.76e+02 mg/mL; 3.45e+00 mol/L	Highly soluble	High	No	63.32 Å²
$C_{16}H_{12}N_2O_2Se_2$	-4.26	2.33e-02 mg/mL; 5.53e-05 mol/L	Moderately soluble	High	No	44.00 Å²
$C_{21}H_{18}OSe$	-4.92	4.36e-03 mg/mL; 1.19e-05 mol/L	Moderately soluble	High	Yes	20.23 Å²
$C_{16}H_{17}NO_3Se_2$	-1.95	4.80e+00 mg/mL; 1.12e-02 mol/L	Very soluble	Low	Yes	65.62 Å²
$C_{11}H_7N_3OSe$	-3.01	2.69e-01 mg/mL; 9.74e-04 mol/L	Soluble	High	No	47.78 Å²
$C_{11}H_9N_3OSe$	-3.35	1.24e-01 mg/mL; 4.47e-04 mol/L	Soluble	High	No	39.82 Å²
Estramustine (MAP4 control)	-4.78	7.28e-03 mg/mL; 1.65e-05 mol/L	Moderately soluble	High	Yes	49.77 Å²
Bisoxatin (spike control)	-4.41	7.28e-03 mg/mL; 1.65e-05 mol/L	Moderately soluble	High	Yes	49.77 Å²

GI: gastrointestinal; P-gp: P-glycoprotein; TPSA: topological polar surface area.

Table 5. The results of docking control estramustine and selenium complex compounds with MAP4 protein.

Selenium complex name	Molecular formula	Binding affinity	Hydrophobic force	Hydrogen bonds
Estramustine (control)	$C_{23}H_{31}Cl_2NO_3$	-6.1	PHE122, LEU142, ILE62, LEU70	LYS140
Selenomethionine	$C_5H_{11}NO_2Se$	-6.3	LEU60, LEU70, ILE62	GLN73, SER138, LYS140
Ethaselen	$C_{16}H_{12}N_2O_2Se_2$	-6.2	LEU60, LEU70, ILE62	GLN73, SER138, LYS140
Selenocystine	$C_6H_{12}N_2O_4Se_2$	-6.1	LEU60, LEU70, ILE62	GLN73, SER138, LYS140
Isoselenazolo	$C_{11}H_7N_3OSe$	-6.1	LEU60, LEU70, ILE62	GLN73, SER138, LYS140
1-(2,5-Diphenylselenophen-3-yl)pent-1-yn-3-ol	$C_{21}H_{18}OSe$	-6.0	LEU60, LEU70, ILE62	GLN73, SER138, LYS140
1H-Pyrrole-3-methanol, 2,5-bis(5-(hydroxymethyl)selenophene-2-yl)-1-methyl-	$C_{16}H_{17}NO_3Se_2$	-5.9	LEU60, LEU70, ILE62	GLN73, SER138, LYS140
4,5-Dihydro-4-methyl-6-oxo-5-phenyl-6H-pyrazole(4,5-c)isoselenazole	$C_{11}H_9N_3OSe$	-5.9	LEU60, LEU70, ILE62	SER138, LYS140

*Similar ties are marked with an underline.

Docking of MAP4 with selenium and control compounds

Based on the docking results of the experiments conducted, it was found that several complex selenium compound bonds were lower than the MAP4 bonds to estramustine (control) of -6.1 Kcal/mol (Table 5). These include selenomethionine, ethaselen, and isoselenazolo. The binding affinity values of proteins and ligands can be interpreted as the average binding free energy of the conformations of all interacting molecules. The lower the bond affinity value, the stronger the resulting bonds between molecules (Uddin et al., 2020). When compared to estramustine, the lower binding affinity values of several complex selenium compounds with MAP4 indicate that these compounds can potentially act as SARS-CoV-2 inhibitors in microtubules.

The molecular docking of MAP4 with the complex selenium compounds that have met the criteria for *in silico* drug testing was carried out at the microtubule-

binding domain (MTB) section, namely at the C-terminal. The precise results of the docking position between the protein and the ligand were determined by comparing the RMSD superimposed values during docking to the estramustine control. The superimpose RMSD value for the docking results is 3.4 Å. In this context, the closer the value is to 3, the more similar the location is (Ghost et al., 2020). These results show a similarity in the location of the bond docking positions between the ligand complex of the selenium compound-MAP4 and the estramustine-MAP4 control in the dockings (Fig. 1).

The docking results were carried out on the estramustine control ligand with the domain of MAP4, where hydrophobic bonds were obtained with amino acid residues that interacted with Phe122, Leu142, Ile62, Leu70, and hydrogen bonds with interacting amino acid residues, namely Lys140. The two categories of hydrogen and hydrophobic bonds that are produced contribute to the bonds that occur, which will strengthen the bonds that are formed. This can happen because hydrogen and hydrophobic bonds are the most important bonds in the interaction of proteins and ligands because they have a function in the structure of a bond and increase the stability of the bond (Collier et al., 2019). The residue from the blind docking of the control compounds determines the specific docking active site of each selenium complex compound, with the MAP4 domain indicating its binding potential. The types of bonds formed from each compound as well as the binding variations shown, are shown in the 2D visualization by Discovery Studio (Fig. 2).

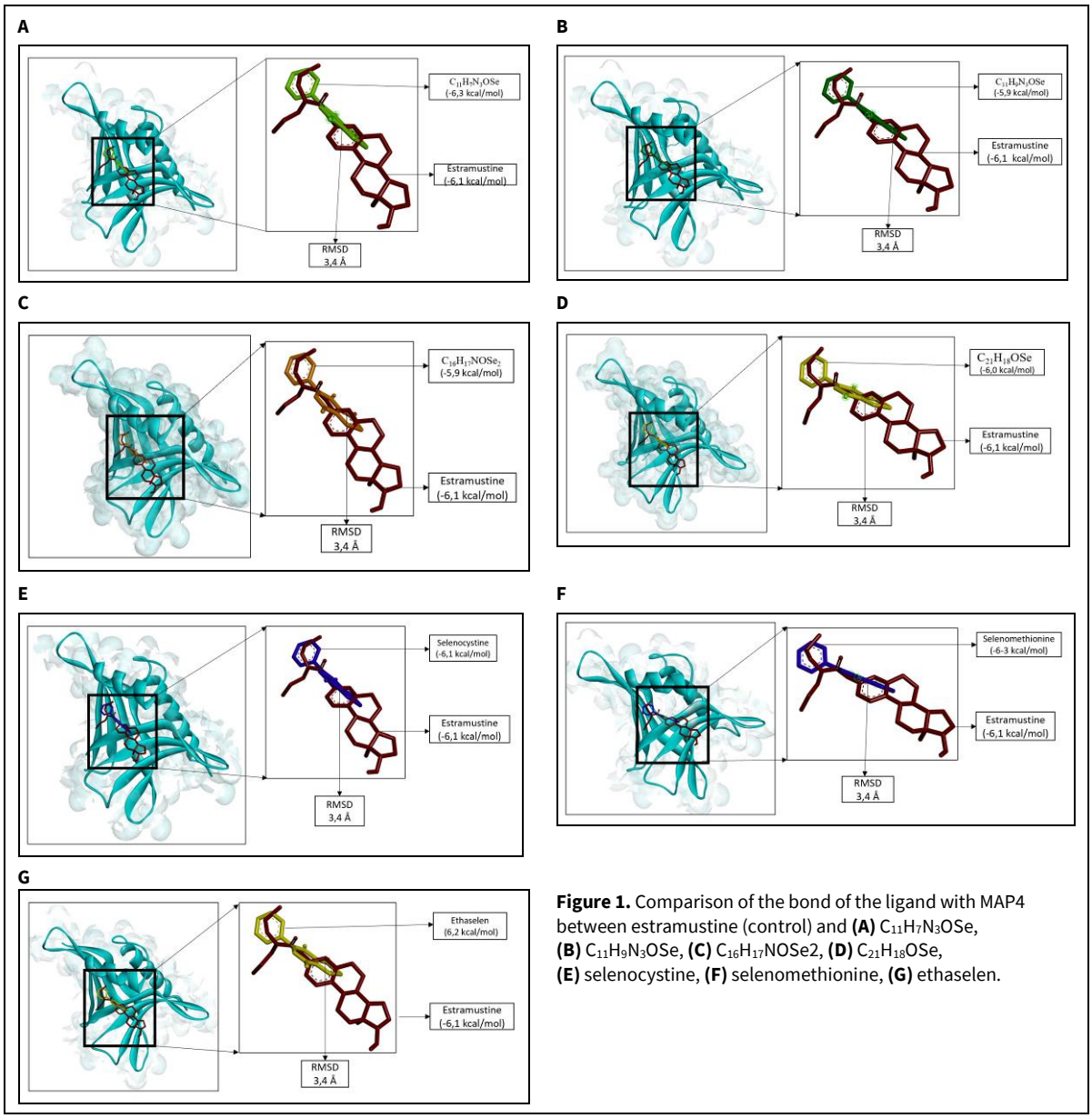


Figure 1. Comparison of the bond of the ligand with MAP4 between estramustine (control) and **(A)** $C_{11}H_7N_3OSe$, **(B)** $C_{11}H_9N_3OSe$, **(C)** $C_{16}H_{17}NOSe2$, **(D)** $C_{21}H_{18}OSe$, **(E)** selenocystine, **(F)** selenomethionine, **(G)** ethaselen.

Docking spike protein with selenium and control compounds

The results of the docking based on the experiments carried out showed that several complex selenium compound bonds were lower than spike protein bonds to bisoxatin (control) of -7 Kcal/mol (Table 6), namely ethaselen and selenophene compounds. The binding affinity values of proteins and ligands can be interpreted as the average binding free energy of the conformations of all interacting molecules. The lower the bond affinity value, the stronger the resulting bond between molecules (Kim et al., 2020). The low bonding of several complex selenium compounds with spike protein compared to the control drug bisoxitin indicates that this compound has the potential to act as a SARS-CoV-2 inhibitor in microtubules.

The process of molecular docking of the SARS-CoV-2 spike protein with selenium complex compounds that meet the *in silico* drug criteria was carried out in the receptor binding domain (RBD) section of the SARS-CoV-2 spike. The precision of the results of the docking position between the protein and the ligand was carried out by comparing the RMSD superimposed values during docking compared to the bisoxatin control. The superimpose RMSD average value on the docking results shows a value of 2.7 Å, where the closer to 3, the more similar the location (Chauhan et al., 2019). This shows a similarity in the location of the bond docking positions between the ligand complex of the selenium-protein spike complex and the bisoxatin-spike control ligand in each docking compound that overlaps or intersects (Fig. 3).

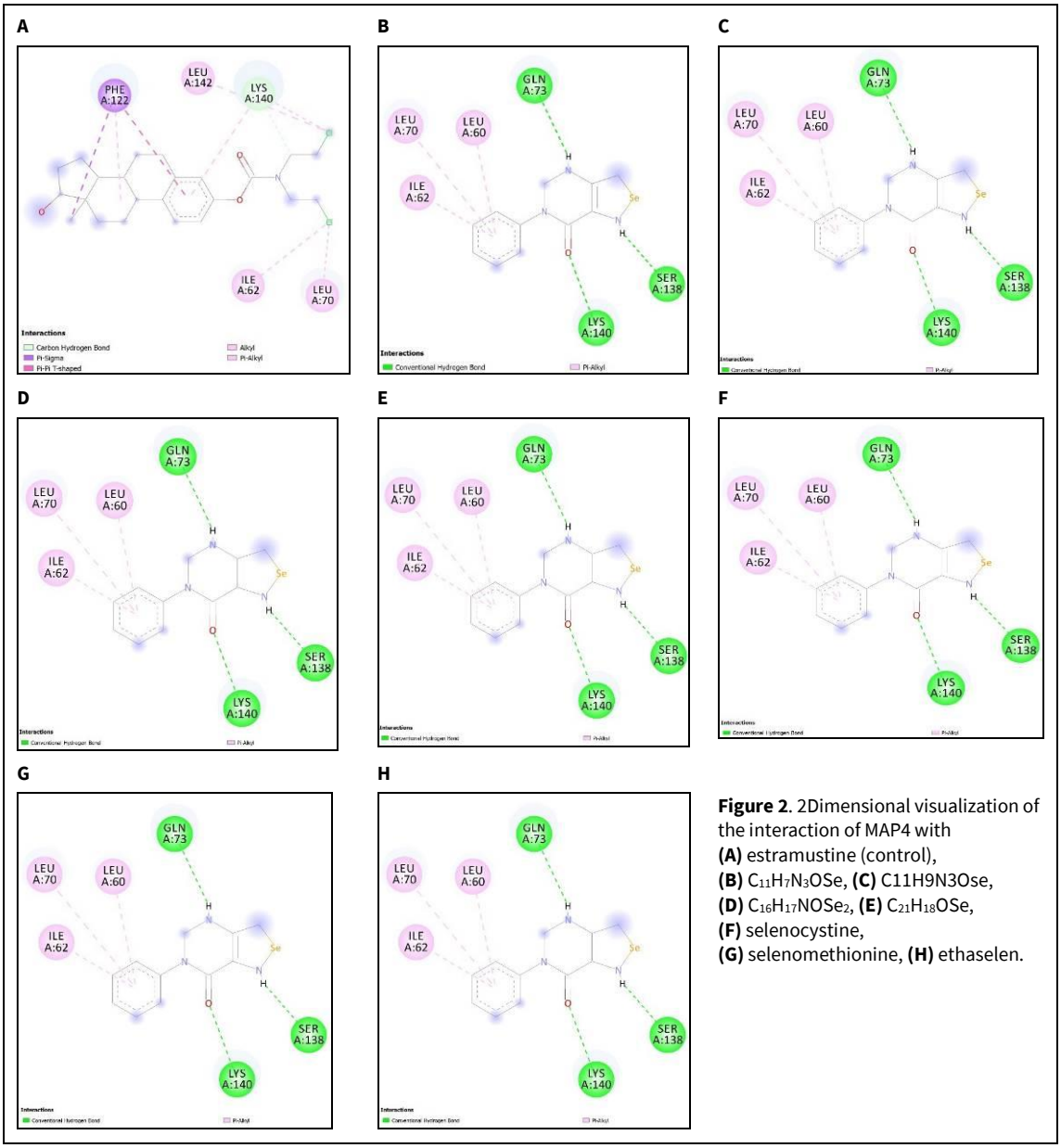


Figure 2. 2Dimensional visualization of the interaction of MAP4 with **(A)** estramustine (control), **(B)** $C_{11}H_7N_3OSe$, **(C)** C11H9N3Ose, **(D)** $C_{16}H_{17}NOSe_2$, **(E)** $C_{21}H_{18}OSe$, **(F)** selenocystine, **(G)** selenomethionine, **(H)** ethaselen.

Table 6. Results of docking control bisoxatin and selenium complex compounds with spike protein.

Selenium complex name	Molecular formula	Binding affinity	Hydrophobic force	Hydrogen bonds
Bisoxatine (control)	$C_{23}H_{31}Cl_2NO_3$	-7.0	LEU368, VAL364	ASP361, ASN340
Ethaselen	$C_{16}H_{12}N_2O_2Se_2$	-7.3	PHE339, ASP361, LEU365	ASP336
Selenocystine	$C_6H_{12}N_2O_4Se_2$	-7.2	LEU368	ASP361, VAL364, SER363
Selenomethionine	$C_5H_{11}NO_2Se$	-7.1	PHE339, LEU365, LEU368	CYS333, PHE335, ASP336, ASP361
H-Pyrrole 3-methanol, (hydroxymethyl) selenophene-2-yl)-1-methyl	$C_{16}H_{17}NO_3Se_2$	-7.0	LEU365, LEU368	CYS333, PHE339, ASN340
Diphenylselenophen-3-yl)pent-1-yn-3-ol	$C_{21}H_{18}OSe$	-6.9	PHE339, LEU365, LEU368, PRO370, TRP433	ASN340
Phenyl-isoselenazolo(4d) pyrimidone	$C_{11}H_7N_3OSe$	-6.8	LEU332, PHE339, LEU365	VAL364, SER363
Dihydro-4-methyl-6-oxo-5-phenyl-6H-pyrazole isoselenazole	$C_{11}H_9N_3OSe$	-6.8	LEU332, PHE339, LEU365	VAL364, SER363

*The similarity of bonds is marked with an underline (underline).

The docking results were carried out on the bisoxatin control ligand with the domain of the SARS-CoV-2 spike protein, which resulted in hydrophobic bonds with interacting amino acid residues LEU368, VAL364, and hydrogen bonds with interacting amino acid residues, namely ASP361, ASN340. The two categories of hydrogen and hydrophobic bonds that are produced contribute to the bonds that occur, which will strengthen the bonds that are formed. This can happen because hydrogen and hydrophobic bonds are the most important bonds in the interaction of proteins and ligands because they have a function in the structure of a bond and increase the stability of the bond (Van et al., 2021). The residue from the blind docking of the control compound determines the specific docking active site on each selenium complex compound with the spike SARS-CoV-2 protein domain, which shows its potential to bind. The types of bonds formed from each compound as well as the binding variations, are shown in the 2D visualization appearance by discovery studio (Fig. 4) (Wang et al., 2020).

Molecular dynamic analysis

The molecular dynamic results show that most of the compounds used in this study from stable bonds to the receptor proteins (in this case, MAP4). Based on the plot, it can be seen that several regions experienced fluctuations. The structure can be said to be stable if the RMSF value is in the range of 1-3 Å. Higher peaks can indicate high fluctuation rates and significant conformational changes. If most of the peaks are in the normal range, this indicates a "successful" computational docking process (Tian et al., 2022). Some regions show mild fluctuations, indicating uninterrupted interaction between MAP4 and spike and their ligands. The interaction of selenium complex compounds with each ligand compared to the drug control was the best, namely the ethaselen, selenocystin, and selenomethionine compounds with the lowest fluctuation values, which were lower than the control (Fig. 5A-B). Based on the overall results of fluctuations, it can be concluded that the selenium complex compounds have stable interactions, as these compounds have an overall average amino acid position with an RMSF value in the range of 1-3 Å (Fig. 5A-B) (Ghosh et al., 2020).

CONCLUSION

Complex selenium compounds found in the *Brassicaceae* family of plants proved to be safe to be used in medicinal preparations, as they did not show any violations of the Lipinski test or pharmacokinetic analysis. Selenium compounds such as ethaselen, selenomethionine, and selenocystine show better binding affinity values when compared to controls on spike protein (-6.1 kcal/mol) and MAP4 (-7 kcal/mol). This is supported by the similarity of binding sites on several amino acid residues dominated by hydrogen bonds. The molecular dynamic results of the RMSF of complex selenium compounds derived from Brassicaceae show bond stability and low fluctuation levels in said bonds, namely 1-3 Å. So, it can be concluded that the complex selenium compounds found in the *Brassicaceae* family of plants have the

potential to be a drug candidate to prevent SARS-COV-2 by inhibiting MAP4 and the spike protein.

CONFLICT OF INTEREST

The authors declare no conflicts of interest.

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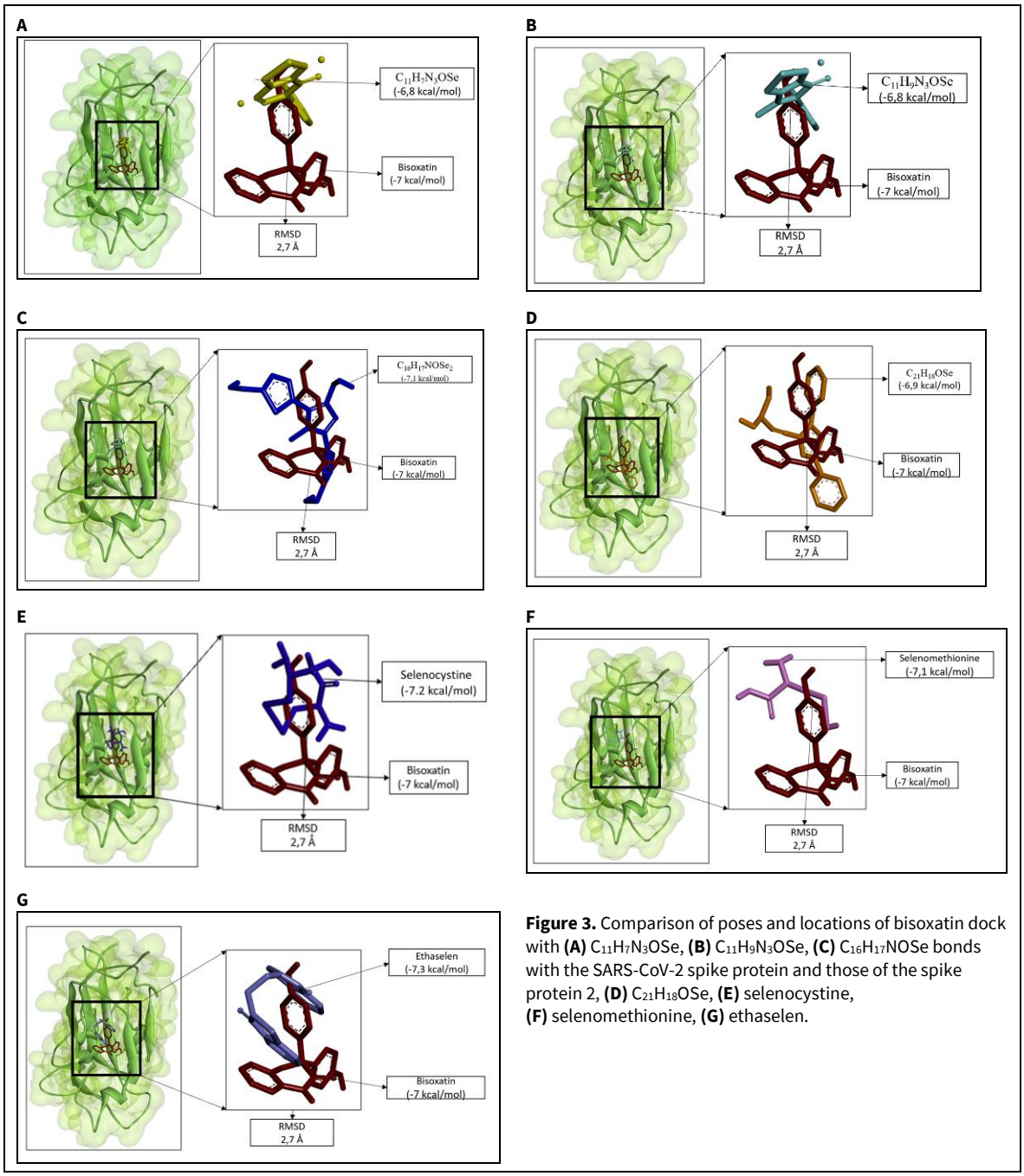

Figure 3. Comparison of poses and locations of bisoxatin dock with **(A)** $C_{11}H_7N_3OSe$, **(B)** $C_{11}H_9N_3OSe$, **(C)** $C_{16}H_{17}NOSe$ bonds with the SARS-CoV-2 spike protein and those of the spike protein 2, **(D)** $C_{21}H_{18}OSe$, **(E)** selenocystine, **(F)** selenomethionine, **(G)** ethaselen.

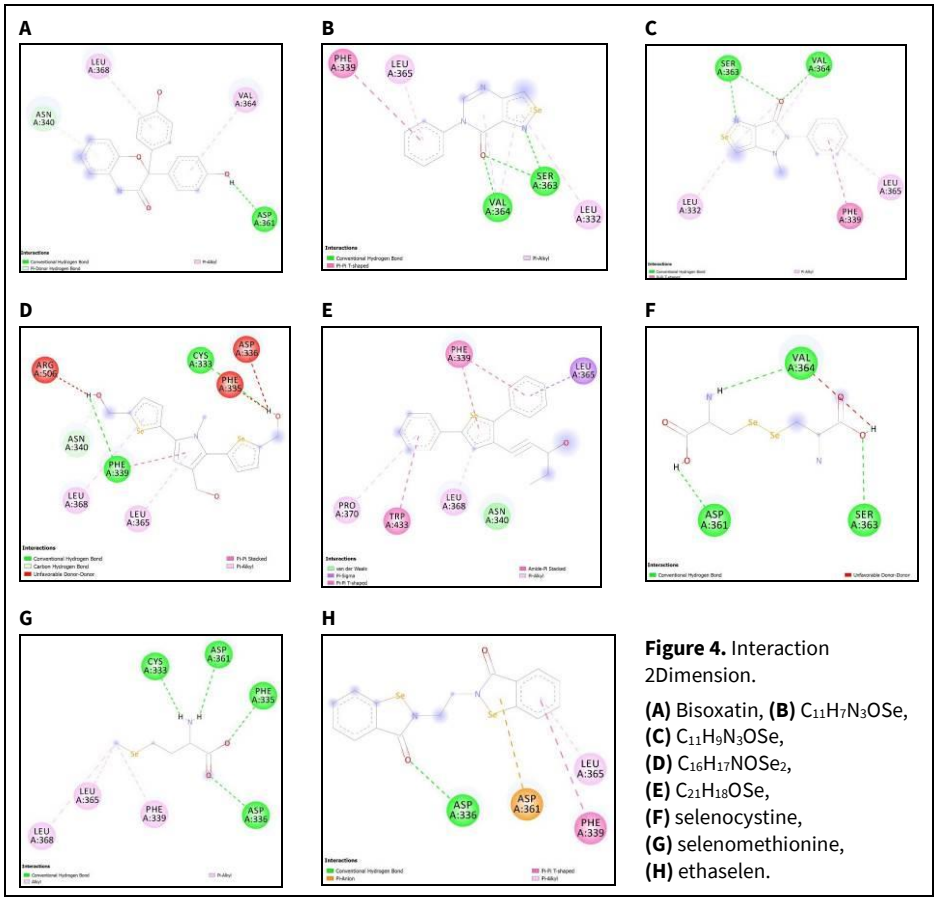

Figure 4. Interaction 2Dimension.
(A) Bisoxatin, **(B)** $C_{11}H_7N_3OSe$, **(C)** $C_{11}H_9N_3OSe$, **(D)** $C_{16}H_{17}NOSe_2$, **(E)** $C_{21}H_{18}OSe$, **(F)** selenocystine, **(G)** selenomethionine, **(H)** ethaselen.

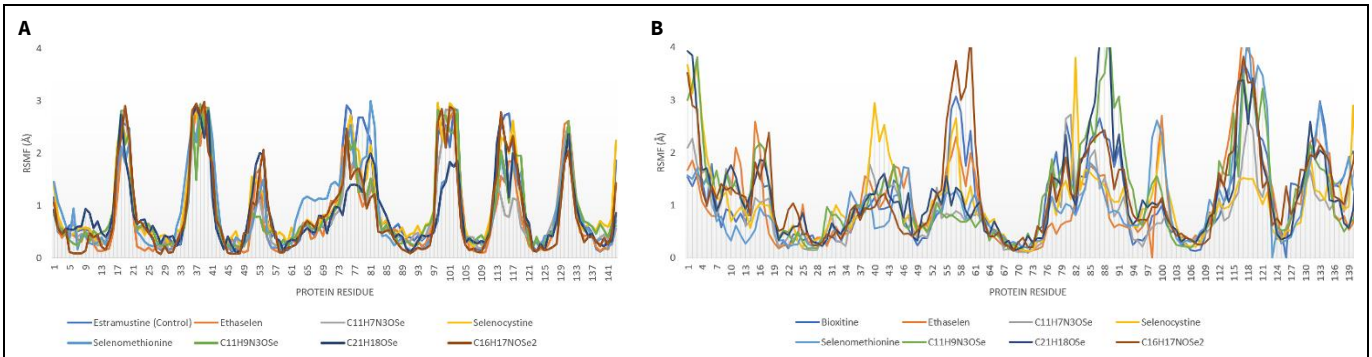

Figure 5. (A) Root mean square fluctuation (RMSF) plot of MAP4 docking with seven complex selenium compounds and estramustine as a control. **(B)** RMSF plot of spike protein docking with seven complex selenium compounds and bisoxatin as a control.

REFERENCES

Aminpour M, Hameroff S, Tuszynski JA (2022) How COVID-19 hijacks the cytoskeleton: Therapeutic implications. Life (Basel) 12(6): 814. https://doi.org/10.3390/life12060814

Boopathi S, Poma AB, Kolandaivel P, Novel (2019) coronavirus structure, mechanism of action, antiviral drug promises and rule out against its treatment. J Biomol Struct Dyn 39(9): 3409–3418. https://doi.org/10.1080/07391102.2020.1758788

Chauhan R, Awasthi S, Srivastava S, Dwivedi S, Pilon-Smits EAH, Dhankher OP, Rudra D (2019) Understanding selenium metabolism in plants and its role as a beneficial element. Crit Rev Environ Sci Tech 49(21): 1937–1958. http://dx.doi.org/10.1080/10643389.2019.1598240

Chinnasamy SG, Selvaraj AC, Kaushik S, Kaliamurthi C, Selvaraj SK, Singh R, Thirugnanasambandam K, Gu A, Wei DQ (2019) Molecular docking and molecular dynamics simulation studies to identify potent AURKA inhibitors: Assessing the performance of density functional theory, MM-GBSA and mass action kinetics calculations. J Biomol Struct Dyn 38(14): 4325–4335. https://doi.org/10.1080/07391102.2019.1674695

Collier TA, Piggot TJ, Allison JR (2019) Molecular dynamics simulation of protein. In: Gerrard J, Domigan L (eds) Protein Nanotechnology. Methods in Molecular Biology, vol 2073. New York, NY: Humana, pp. 311–327. https://doi.org/10.1007/978-1-4939-9869-2_17

Endharti AT, Wahyuningtyas TE, Hardini KH, Widjajanto E, Permana S (2018) *Dendrophthoe pentandra* leaves extract promotes apoptotic effects of doxorubicin in human breast cancer cell via modulation of intracellular calcium and survivin. J Appl Pharm Sci 8(8): 39–43. http://dx.doi.org/10.7324/JAPS.2018.8806

Fedorov VA, Orekhov PS, Kholina EG, Gudimchuk NB (2019) Mechanical properties of tubulin intra- and inter-dimer interfaces and their implications for microtubule dynamic instability. PloS Comput Biol 18: 14–21. https://doi.org/10.1371/journal.pcbi.1007327

Ghosh S, Dellibovi TA, Kerviel A, Pak E, Qiu Q, Fisher M, Takvorian PM, Bleck C, Hsu VW, Fehr AR (2020) β-Coronaviruses use lysosomes for egress instead of the biosynthetic secretory pathway. Cell 83: 1520–1535.e14. https://doi.org/10.1016/j.cell.2020.10.039

Gudimchuk NB, McIntosh JR (2021) Regulation of microtubule dynamics, mechanics and function through the growing tip. Nat Rev Mol Cell Biol 22: 777–795. https://doi.org/10.1038/s41580-021-00399-x

Hollingsworth SA, Dror RO (2019) Molecular dynamics simulation for all. Neuron 99(6): 1129–1143. https://doi.org/10.1016/j.neuron.2018.08.011

Kim D, Lee JY, Yang JS, Kim JW, Kim VN, Chang H (2020) The architecture of SARS-CoV-2 transcriptome. Cell 181: 914–921. https://doi.org/10.1016/j.cell.2020.04.011

Lai CC, Shih TP, Ko WC, Tang HJ, Hsueh PR (2020) Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) and coronavirus disease-2019 (COVID-19): The epidemic and the challenges. Int J Antimicrob Agents 55(3): 105924. https://doi.org/10.1016/j.ijantimicag.2020.105924

Mawaddani N, Wibowo NRK, Nadhira QHH, Pramifta RA (2020) *In silico* study of *Centella asiatica* compund as BACE1 inhibitor in Alzheimer's disease. J Smart Bioprosp Technol 1(2): 36–40. https://doi.org/10.21776/ub.jsmartech.2020.001.02.3

Permana S, Fityanti PR, Norahmawati E, Iskandar A, Mulyadi AED, Endharti TA (2020) Coelomic fluid of *Eisenia fetida* ameliorates cetuximab to reduce K-Ras and vimentin expression through promoting RUNX3 in an AOM/DSS-induced colitis associated colon cancer. Evid Based Complement Alternat Med 2020: 9418520. https://doi.org/10.1155/2020/9418520

Rakib A, Nain Z, Sami SA, Mahmud S, Islam A, Ahmed S, Siddiqui ABF, Babu SMOF, Hossain P, Shahriar A, Nainu F, Emran TB, Simal-Gandara J (2021) A molecular modelling approach for identifying antiviral selenium-containing heterocyclic compounds that inhibit the main protease of SARS-CoV-2: an in silico investigation. Brief Bioinform 22(2): 1476-1498. https://doi.org/10.1093/bib/bbab045

Sethi A, Joshi K, Sasikala K, Alvala M (2019) Molecular docking in modern drug discovery: Principles and recent applications. In: Drug Discovery and Development - New Advances. Gaitonde V, Karmakar P, Trivedi A (eds.). Intechopen. https://doi.org/10.5772/intechopen.85991

Simpson C, Yamauchi Y (2020) Microtubules in influenza virus entry and egress. Viruses 12: 117. https://doi.org/10.3390/v12010117

Tian L, Pang Z, Li M, Lou F, An X, Zhu S, Fan J (2022) Molnupiravir and its antiviral activity against COVID-19. Front Immunol 13: 855496. https://doi.org/10.3389/fimmu.2022.855496

Uddin M, Mustafa F, Rizvi TA, Loney TA, Suwaidi H, Senok AC (2020) SARS-CoV-2/COVID-19: Viral genomics, epidemiology, vaccines, and therapeutic interventions. Viruses 12: 526. https://doi.org/10.3390/v12050526

Van ELE, Binkhorst M, Bourgonje AR, Offringa AK, Mulder DJ, Bos EM, Kolundzic N, Abdulle AE, Van D, Voort PH, Olde RMG, Hoeven JG, Dunnen WF, Hillebrands JL, Goor VH (2021) COVID-19: Immunopathology, pathophysiological mechanisms, and treatment options. J Pathol 254: 307–331. https://doi.org/10.1002/path.5642

Wang Q, Wu J, Wang H, Gao Y, Liu Q, Mu A, Ji W, Yan L, Rao Z (2020) Structural basis for RNA replication by the SARS-CoV-2 polymerase. Cell 182: 417–428. https://doi.org/10.1016/j.cell.2020.05.034

Yoshimoto FK (2020) The proteins of severe acute respiratory syndrome coronavirus-2 (SARS CoV-2 or n-COV19), the cause of COVID-19. Protein J 39(3): 198–216. https://doi.org/10.1007/s10930-020-09901-4

AUTHOR CONTRIBUTION:

Contribution	Ilmiyah SZ	Fakurazi S	Endharti AT	Permana S
Concepts or ideas	x		x	x
Design	x			x
Definition of intellectual content	x	x	x	x
Literature search	x	x	x	x
Experimental studies	x		x	x
Data acquisition	x	x	x	x
Data analysis	x	x	x	x
Statistical analysis	x			
Manuscript preparation	x	x	x	x
Manuscript editing	x	x	x	x
Manuscript review	x	x	x	x

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