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Potential of Bioactive Compounds of *Allium sativum* L. var. solo garlic Extract in Inhibiting InhA Protein in *Mycobacterium tuberculosis*

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Abstract: Tuberculosis (TB) is caused by the bacteria *Mycobacterium tuberculosis*. In general, TB is treated with compounds that inhibit the work of one of the enzymes in the bacteria *Mycobacterium tuberculosis*, namely the InhA enzyme. One of the herbal plants that has the potential to inhibit the InhA protein in the bacteria *Mycobacterium tuberculosis* is. *Allium sativum* L. var. solo garlic. This research aims to determine the bioactive compounds in *Allium sativum* L. var. solo garlic in inhibiting the InhA protein in *Mycobacterium tuberculosis* which can be used as an alternative drug in the treatment of TB through an in silico approach. The method used in this study was qualitative phytochemical screening and GCMS test on *Allium sativum* L. var. solo garlic extract, the bioactive compounds found were tested in silico through a molecular docking approach. The research results obtained that *Allium sativum* L. var. solo garlic contains alkaloids, flavonoids, tannins and steroids. Based on the results of the GCMS test, the bioactive compounds that have an area of more than 5% are Heptadecene-(8)-Carbonic Acid-(1), 3-Deoxy-D-Mannonic Acid, 5-Hydroxymethylfurfural, Melezitose and Oleic acid. The results of molecular docking showed that the compound 3-Deoxy-D-mannonic acid had a binding affinity of -4.9, 5-Hydroxymethylfurfural -4.8, Oleic Acid -6.4, Nicotinamide-Adenine-Dinucleotide (Control) had a binding affinity of -11.4. Low binding affinity indicates that the compound can bind to the protein with little energy. The Gyps energy theory states that the smaller the energy produced from a bond between the ligand and its receptor, the more stable the bond is. The lowest binding energy to the InhA protein is in the control compound Nicotinamide-Adenine-Dinucleotide, Oleic Acid, 3-Deoxy-D-mannonic acid, 5-Hydroxymethylfurfural.

Keywords: *Allium sativum* L. var. solo garlic; bioactive compounds; InhA protein; *Mycobacterium tuberculosis*, molecular docking.

Introduction

Mycobacterium tuberculosis is a bacteria that causes tuberculosis (TB). Indonesia has the second highest ranking in the world, namely 9.2% of total cases (Wahid *et al.*, 2023). In 2022, TB sufferers in Indonesia will reach 969,000 with a death rate of 93,000/year (Fahdhienie *et al.*, 2024). There are several factors that influence the prevalence of TB, including hygiene, age, natural history of TB, gender, socio-economic conditions, smoking status, HIV infection, body mass index and education level (Indarto *et al.*, 2020). In general, TB patients are treated by administering drugs and antibiotics, including rifampin, ethambutol, isoniazid, pyrazinamide, capreomycin,ethionamide (Sharghi *et al.*, 1017). Long-term use of drugs and antibiotics can cause negative effects, including depression, vision impairment, teratogenesis, gastrointestinal symptoms, liver function disorder, cardiac symptoms, hearing impairment, sleep disorder (Arliny *et al.*, 2025). Therefore, alternative treatments are needed, one of which is using herbal medicine.

According to Retno Wardani *et al.*, (2020)

there are several plants that have the potential to be herbal medicines to treat TB infections, namely *Hibiscus rosasinensis* L., *Morindacitrifolia* L., beluntas leaves, Javanese wood bark, *Centellaasiatica*. The contents of this plant are asiatic acid, flavonoids, allicin, asiaticoside, alkaloids, ajoene, saponins, madecasic acid and phenolics which have the ability to kill bacteria *Mycobacterium tuberculosis*. However, this study focused on *Allium sativum* L. var. solo garlic extract.

Allium sativum L. var. solo garlic is garlic which only consists of one siung of single bulb garlic. *Allium sativum* L. var. solo garlic contains allicin and scordinin compounds. Allicin acts as an antibacterial which can inhibit bacterial growth *Mycobacterium tubercolusis*, while scordinin has the ability to increase body resistance (Mardiyah, 2018). According to Hartini *et al.*, (2024) states that *Allium sativum* is known as an anticancer, antiviral, antibacterial, antifungal, antihypertensive and can treat tuberculosis, kidney disease, influenza, acidity and diabetes. According to Fatima & Dwivedi (2020) states that allicin can inhibit the growth of *Mycobacterium tuberculosis* by blocking receptors on the surface. In general, TB is treated with compounds that inhibit the action of one of the enzymes in *Mycobacterium tuberculosis* bacteria, namely the InhA enzyme.

InhA is a target protein in the development of tuberculosis drugs. It is known that inhibition of InhA can inhibit mycolic acid biosynthesis, thereby damaging the integrity of the cell wall and causing cell death (Belete, 2022). InhA is an enyol-acyl carrier protein reductase found in *Mycobacterium tuberculosis* bacteria as a target for drug development in the treatment of TB. This research aims to determine the bioactive compounds in *Allium sativum* L. var. solo garlic in inhibiting InhA protein in *Mycobacterium tuberculosis* which can be used as an alternative drug in the treatment of TB through an in silico approach. Drug discovery with an in silico approach using molecular docking is an effective method in drug development (Ningrat, 2022).

Material and Methods

Materials

The materials used in this research were *Allium sativum* L. var. solo garlic, 96% methanol, and distilled water. Samples of *Allium sativum* L. var. solo garlic were obtained from Pasuruan, East Java.

Preparation and extraction of *Allium sativum* L. var. solo garlic

Allium sativum L. var. solo garlic samples were peeled, then washed with running water until clean, drained and cut into small pieces. After that, the samples were air-dried for 5-7 days. After drying, the samples were ground with a blender until they became powder. *Allium sativum* L. var. solo garlic sample powder was extracted by maceration using 96% methanol solvent. *Allium sativum* L. var. solo garlic powder was soaked for 1x24 hours using 96% methanol solution with a ratio of 1:6 (Alfauzi *et al.*, 2022). After that, the filtering process is carried out to separate the filtrate and dregs. The filtrate is then evaporated using a vacuum rotary evaporator to produce a thick *Allium sativum* L. var. solo garlic extract. The thick extract obtained was stored in a refrigerator to carry out phytochemical tests and Gas Chromatography-Mass Spectrometry (GC-MS) tests.

Phytochemical screening and GC-MS analysis of *Allium sativum* L. var. solo garlic extrract

The qualitative phytochemical screening test of *Allium sativum* L. var. solo garlic extract includes flavonoid, alkaloid, tannin, steroid, triterpenoid and saponin content (Indriaty *et al.*, 2023). Flavonoid test on *Allium sativum* L. var. solo garlic extract is to take 1 mL of extract then add HCl and Magnesium powder (Mg^{2+}) then shake. The sample is indicated to contain flavonoids if there is a color change from light green, red or purple to orange and there is foam (Indriaty *et al.*, 2023). Alkaloid test on *Allium sativum* L. var. solo garlic extract by taking 100 mg of extract then adding 3 mL of NH_3 and 5 mL of chloroform. Then the solution is separated into 3 test tubes and each is added with Mayer, Wagner and Dragendorf reagents. If a white, yellow and reddish brown precipitate is formed, it indicates that the sample is positive for alkaloids.

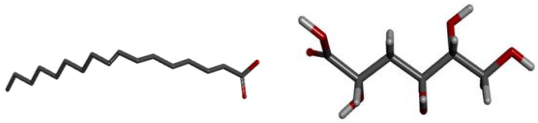
Tannin test by adding 100 mg of extract with 5% FeCl3 as much as 5 drops. The presence of a dark blue or black color change indicates the presence of tannin in the extract. Triterpenoid test, namely weighing 100 mg of extract into a test tube, then

adding Liebermann-Burchad reagent. and shaken. The presence of steroids is indicated by the formation of a green or blue color, while the presence of triterpenoids is indicated by the formation of a purple or red color. Saponin test on *Allium sativum* L. var. solo garlic extract is to weigh 100 mg of extract dissolved in methanol, then heated and shaken vigorously. If the sample forms foam, it indicates the presence of saponin.

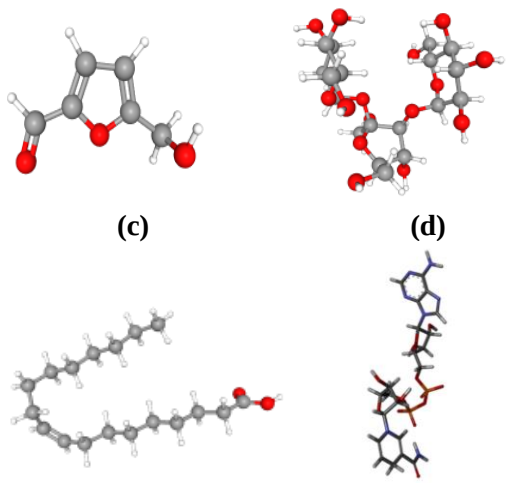
Allium sativum L. var. solo garlic extract was analyzed by GC-MS to determine the percentage of each component by measuring the relative peak area in the chromatogram. *Allium sativum* L. var. solo garlic extract was taken 1 µL injected into the GC-MS and run for 60 minutes. The injector temperature was 250 ^{0}C and set in splitless mode (Masyudi *et al.*, 2022). The obtained chromatograms and MS data were downloaded and further analyzed.

Preparation of Ligand and InhA Protein *Mycobacterium tuberculosis*

The compounds used in this study are five compounds contained in male shallots, namely Heptadecene - (8)- Carbonic Acid-(1), 3-Deoxy – D - Mannonic Acid, 5-Hydroxymethylfurfural, Melezitose and Oleic acid based on the results of the GCMS analysis that has been carried out. These compounds were then prepared using open babel which is integrated into the PyRx 0.8 application to prepare the molecular docking process (Picture 1). This study used *Mycobacterium tuberculosis* InhA protein with PDB ID 2X23 obtained from the RCSB PDB web server (https://www.rcsb.org/). InhA protein preparation was carried out using BIOVIA Discovery Studio to remove water molecules and ligands attached to the protein (Picture 2). Nicotinamide-Adenine-Dinucleotide is a native ligand used as a control in this research (Picture 1).

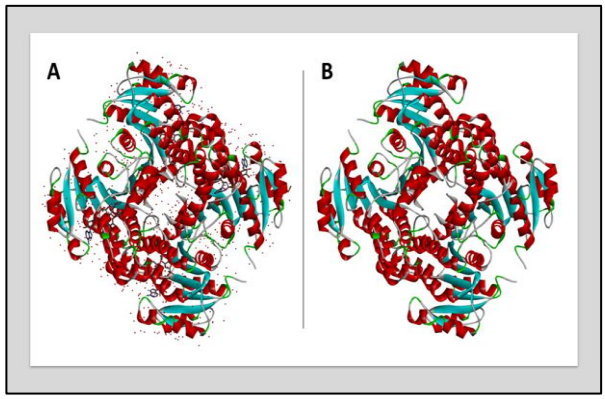


(a) **(b)**



(c) **(d)** **(e)** **(f)**

Picture 1. Active compounds in *Allium sativum* L. var. solo garlic and control compounds (a). Heptadecene-(8)-Carbonic Acid-(1), (b).3-Deoxy-D-Mannonic Acid, (c) 5-Hydroxymethylfurfural, (d). Melezitose, (e). Oleic acid, (f) control compound (Nicotinamide-Adenine-Dinucleotide)



Picture 2. Protein InhA *Mycobacterium tuberculosis* before preparation (A) and after preparation (B)

Data Analysis

Phytochemical screening and GC-MS data of *Allium sativum* L. var. solo garlic extract were analyzed descriptively, while the analysis of the docking results between the ligand and the *Mycobacterium tuberculosis* InhA protein was analyzed in the form of binding positions and amino acid residues formed from the ligand-receptor interaction complex carried out using the PyMol and BIOVIA Discovery Studio applications.

Results and Discussion

Extraction and Phytochemical Screening of *Allium sativum* L. var. solo garlic Extract

In this research, *Allium sativum* L. var. solo garlic used for the extraction process in powder because the interaction between the sample and the solvent is wider so that the compounds in *Allium sativum* L. var. solo garlic can be optimally diffused out of the cell. The extraction process in this study used 96% methanol solvent because methanol is a polar solvent with high solubility so that it is effective in extracting polar and non-polar compounds. (Lee *et al.,* 2024; Nawaz *et al.*, 2020).

Based on the results of qualitative phytochemical screening analysis, it shows that the extract of *Allium sativum* L. var. solo garlic contains alkaloids, flavonoids, tannins, and steroids (Table 1). The presence of alkaloid content can be used as an anti-inflammatory, anesthetic agent, antioxidant, cardioprotective (Ouriagli *et al.,* 2023). According to Sun *et al.,* (2020) stated that alkaloids (pseudopteroxazole and ileabethoxazole) have significant antimicrobial activity against *Mycobacterium tuberculosis*. Alkaloids act as antibacterials because alkaloid compounds inhibit DNA synthesis by inhibiting the topoisomerase enzyme and inhibiting the ATP transport process in cell membranes (Karou *et al.,* 2005). In addition, alkaloids can damage peptidoglycan in bacteria which can cause cell death (Rante Pakadang *et al*., 2021).

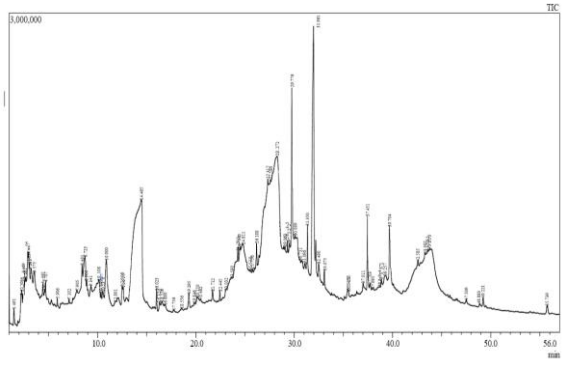
Table 1. Phytochemical Screening Test Results of Methanol Extract of *Allium sativum* L. var. solo garlic

No	Phytochemical Screening Test	Result
1.	Alkaloids	Positive (+)
2.	Flavonoid	Positive (+)
3.	Tannin	Positive (+)
4.	Steroid	Positive (+)
5.	Triterpenoid	Negative (-)
6.	Saponin	Negative (-)

The presence of flavonoid compounds in *Allium sativum* L. var. solo garlic extract can function as antimicrobial, anticancer, antioxidant, antiviral and anti-inflammatory (Koklesova *et al.,* 2021). According to Rabaan *et al.,* (2022) states that flavonoids can inhibit the growth of the microorganism *Mycobacterium tuberculosis*. Tannins also act as antivirus, antioxidants and antibacterials (Kaczmarek, 2020; Sayyah *et al.,* 2004). The presence of steroid content in *Allium sativum* L. var. solo garlic extract also acts as an antibacterial due to the sensitivity of cell lipid membranes which can cause liposome leakage. According to research Gräb *et al.,* (2019) stated that corticosteroids can inhibit cells infected with *Mycobacterium tuberculosis* bacteria.

Gas Chromatography-Mass Spectrometry (GC-MS) Analysis

Analysis of *Allium sativum* L. var. solo garlic extract using GC-MS test contains 82 bioactive compounds (Picture 3). There are 23 compounds that have an area above 0.5% (Table 2).



Picture 3. GC-MS Test Results of Methanol Extract of *Allium Sativum* L. Var. Solo Garlic

Based on the results of GC-MS, the methanol extract of *Allium sativum* L. var. solo garlic has various roles because it contains compounds including 3-Deoxy-d-mannonic acid which acts as an antibacterial (Singh *et al.,* 2023), Heptadecene-(8)-Carbonic Acid acts as an antioxidant, antibacterial and antiproliferative (Saputri & Putri, 2020), The compound 5-Hydroxymethylfurfural functions as anti-inflammatory, antiproliferative, antioxidant and antibacterial (Singh *et al.*, 2023), Melezitose acts as an anticancer and inhibits cell proliferation (Zhou *et al.,* 2024), Oleic acid acts as an antibacterial (Pushparaj Selvadoss *et al.*, 2018), Furaneol functions as an antioxidant anticataract, antimicrobial and antifungal. The compound 1,2,3-Propanetriol found in the methanol extract of *Allium sativum* L. var. solo garlic has a role as antimicrobial, anti-inflammatory,and anticancer,

antibacterial, antiviral, inflammation, antifungal, antidiabetic, antiprotozoal (Leishmania), antipsoriatic, antipruritic non-allergic, antiparkinsonian rigidity relieving, antioxidant, Antineurogenic pain (Casuga *et al.*, 2016; Wasilah *et al.*, 2021).

Table 2. Phytochemical compounds of *Allium sativum* L. var. solo garlic methanol extract by GC-MS

No	Name of Compound	Area%	R. Time	Peak
1	3-Deoxy-d-mannonic acid	30.12	28.272	52
2	Heptadecene-(8)-Carbonic Acid	19.76	31.871	37
3	5-Hydroxymethylfurfural	13.18	14.465	29
4	Melezitose	11.05	43.810	78
5	Oleic Acid	5.84	31.961	62
6	Furaneol	1,02	8.723	18
7	1,2,3-Propanetriol	0.8	10.108	22
8	2,3-Dihydro-3,5-dihydroxy-6-m	1.32	10.900	25
9	2-Cyclopropylbutan-2-ol	1.83	23.590	43
10	Dodecanamide, N, N-bis (2-hydr	2.95	24.290	44
11	1H-Indene, 1-ethylideneoctahy	1.26	24.490	45
12	Cytidine (CAS)	5.11	24.811	46
13	Tetradecanoic acid (CAS)	0.91	27.317	50
14	cis-Vaccenic acid	1.47	29.522	56
15	n-Hexadecanoic acid	4.21	29.776	57
16	Hexadecanoic acid, 2-hydroxy	0.71	37.451	68
17	9,12-Octadecadienoic acid	1.01	39.704	74
18	Hexadecanoic acid, methyl este	0.87	29.325	55
19	CIS-Sabinenehydrate	0.59	25.538	47
20	2-Propanone, 1-hydroxy-	0.65	2.469	4
21	2,3-Butanediol	0.5	2.987	7
22	2-Furancarboxaldehyde	0.79	3.231	9
23	2-Furanmethanol	0.60	3.575	10

The compound 2,3-Dihydro-3,5-dihydroxy acts as an antioxidant (Yu *et al.*, 2013), Cytidine (CAS) acts as anticancer and antimicrobial (Rana *et al.*, 2021), Tetradecanoic acid (CAS) has a role as anticancer and antibakterial (Ghazali *et al.*, 2021), cis-Vaccenic acid functions as antibacterial, antibiofilm and anticancer (Yazıcı, 2024), n-Hexadecanoic acid acts as anti-oxidant, Hypocholesterolemic, Nematicide, Anti-androgenic, Hemolytic, Pesticide, Lubricant, 5-Alpha reductase inhibitor, antipsychotic (Tyagi & Agarwal, 2017).

Senyawa Hexadecanoic acid, 2-hydroxy functions as Hemolytic, pesticide, flavour, antioxidant (Tyagi & Agarwal, 2017), 9,12-Octadecadienoic acid acts as Antimutagenic, Antihypercholesterolemic, Antiseborrhei, Antisecretoric, Antithrombotic, Antiviral (Influenza), Antiinflammatory, Antiulcerative, Antipruritic, Antiinfective, Antiviral (Picornavirus) (Wasilah *et al.*, 2021), Hexadecanoic acid, methyl ester acts as antibacterial, antioxidant, pesticide, nematicide antifungal, antiarthritic, antitumor, hepatoprotective, anticancer, anticoronary, hypocholesterol, anti - inflammatory (Gupta *et al.*, 2023; Shaaban *et al.*, 2021). CIS-Sabinenehydrate acts as antibacterial and antifungal (Judžentienė *et al.*, 2024), 2,3-Butanediol function as anti-inflammatory (Hsieh *et al.*, 2007), 2-Furancarboxaldehyde acts as anti-fungal, anti-diabetic, anti-alzheimer, anti-inflammatory bowel diseases, anticancer, anti-oxidant, anti-aging, and cardio-protective activity (Kumar *et al.*, 2021) and senyawa 2-Furanmethanol has a role as antiviral and antibacterial (Bazaid *et al.*, 2022). In this research, the compounds that will undergo molecular docking are compounds that have an area of more than 5%, namely 3-Deoxy-d-mannonic acid, Heptadecene-(8)-Carbonic Acid, 5-Hydroxymethylfurfural, Melezitose dan Oleic Acid.

Molecular Docking Analysis

Molecular docking is a method based on the recognition process between two or more

molecules which is considered effective and efficient to help reduce costs and time in studying the mechanism of activity of a compound. (Morris & Corte, 2021; Stanzione *et al.*, 2021). This research aims to determine the activity of the compound Heptadecene-(8)-Carbonic Acid-(1), 3-Deoxy-D-Mannonic Acid, 5-Hydroxymethylfurfural, Melezitose and Oleic Acid contained in the extract of *Allium sativum* L. var. solo garlic in inhibiting the InhA protein in bacteria *Mycobacterium tuberculosis*. Nicotinamide–adenine Dinucleotide is a compound that interacts at the active site of the protein and is used as a control in this research.

The results of molecular docking show that the interaction of the compounds Heptadecene-(8)-Carbonic Acid-(1) and 3-Deoxy-D-Mannonic Acid contained in the extract of *Allium sativum* L. var. solo garlic is higher than the control compound. The interaction between Heptadecene-(8)-Carbonic Acid-(1) and the receptor has a lower binding affinity value compared to the interaction between 3-Deoxy-D-Mannonic Acid and the receptor, namely -6.6 kcal/mol and -4.8 kcal/mol respectively. The lower binding affinity value for the interaction between Heptadecene-(8)-Carbonic Acid-(1) and the receptor indicates that the interaction is stronger than the interaction between 3-Deoxy-D-Mannonic Acid and the receptor. The higher binding affinity value in the interaction of the two compounds in *Allium sativum* L. var. solo garlic and the receptor does not completely indicate that the interaction is weaker compared to the control-receptor interaction which has a lower binding affinity value. This is because in molecular docking simulations there are many parameters used to determine the strength and weakness of ligand-receptor interactions, such as binding affinity values, hydrogen interactions, and hydrophobic interactions formed on amino acid residues between ligands and receptors, as well as molecular dynamics results (Pantsar & Poso, 2018).

Table 3. The binding affinity value and amino acid residues formed from ligand-receptor interactions

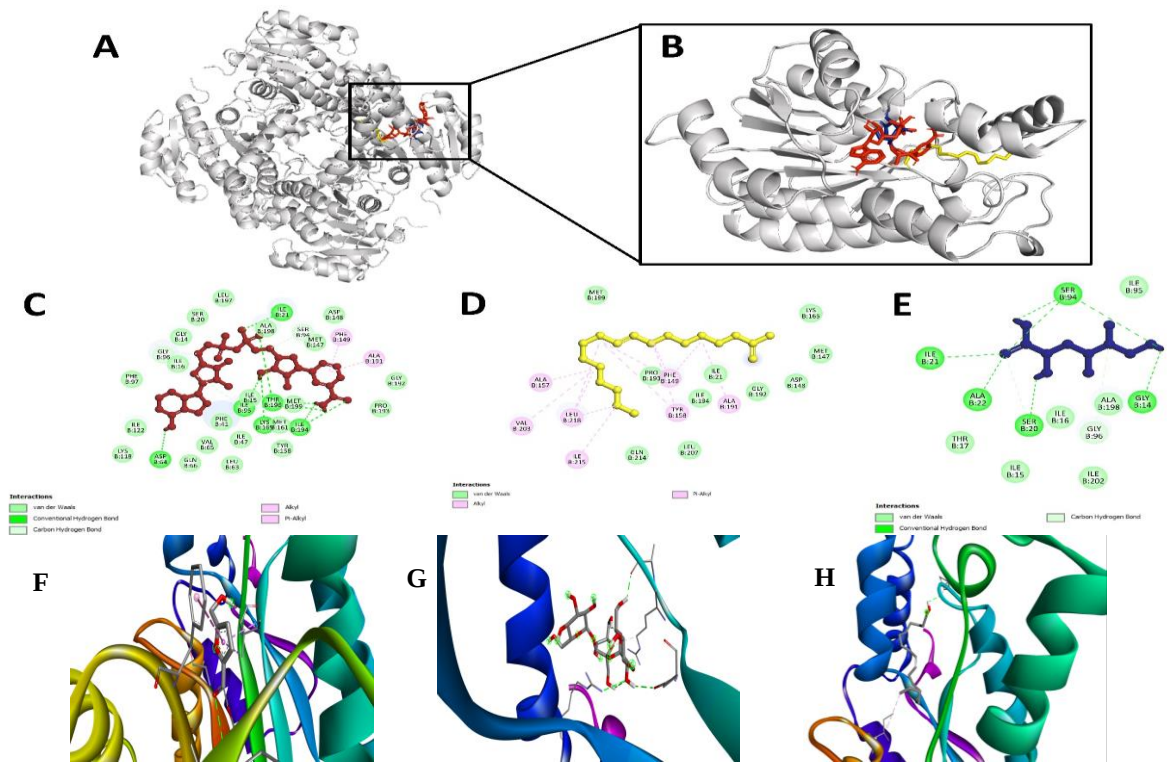
Ligand	**Binding Affinity (kkal/mol)**	**Amino Acid Residues**	
		Hydrogen Interactions	**Hidrofobik Interactions**
Nicotinamide-Adenine-Dinucleotide (Kontrol)	-10,5	Ile21, Asp64, Ile95, Lys165, Ile194, Thr196	Gly14, Ile15, Ile16, Ser20, Phe41, Ile47, Leu63, Val65, Gln66, Ser94, Gly96, Phe97, Lys118, Ile122, Met147, Asp148, Phe149, Tyr158, Met161, Ala191, Gly192, Pro193, Leu197, Ala198, Met199,
Heptadecene-(8)-Carbonic Acid-(1)	-6,6		Ile21, Met147, Asp148, Phe149, Ala157, Tyr158, Lys165, Ala191, Gly192, Pro193, Ile194, Met199, Val203, Leu207, Gln214, Ile215, Leu218,
3-Deoxy-D-Mannonic Acid	-4,8	Gly14, Ser20, Ile21, Ala22, Ser94	Ile15, Ile16, Thr17, Ile95, Gly96, Ala198, Ile202
5-Hydroxymethylfurfural	-4,8	Gly14, Thr39, Leu63	Ile95, Phe41
Melezitose	-6.3	Asp150, Ala154, Arg225, Arg153	
Oleic Acid	-6.4	Pro156	Ile21

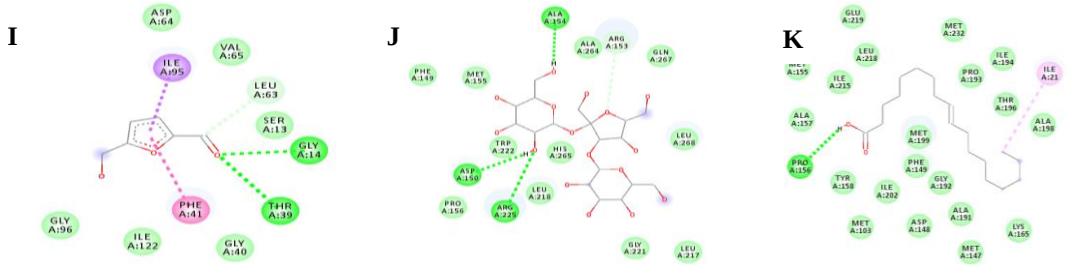
The molecular docking results were visualized with PyMol and Discovery Studio to determine the binding site of the ligand-receptor interaction. The visualization results showed that Heptadecene-(8)-Carbonic Acid-(1) and 3-Deoxy-D-Mannonic Acid have the same binding

site as Nicotinamide-Adenine-Dinucleotide (Control) (Picture 4A and 4B). Visualization is continued by identifying the amino acid residues involved in the ligand-receptor interaction. The interaction of Heptadecene-(8)-Carbonic Acid-(1) and the receptor has 9 amino acid residues in common with the control-receptor interaction. All of these amino acid residues interact with hydrophobic interactions (Table 3 and Picture 4).

Table 3. shows the results of molecular docking of compounds contained in *Allium sativum* L. var solo garlic with InhA, Nicotinamide-Adenine-Dinucleotide as a positive control in conducting this study. The results of molecular docking obtained compounds 3-Deoxy-D-mannonic acid has a binding affinity of -4.9, 5-Hydroxymethylfurfural -4.8, Oleic Acid -6.4, Nicotinamide-Adenine-Dinucleotide (Control) has a binding affinity of -11.4. Low binding affinity indicates that the compound can bind to the protein with little energy. The Gypsum energy theory states that the smaller the energy produced from a bond between the ligand and its receptor, the more stable the bond is. The lowest binding energy to the InhA protein is in the control compound Nicotinamide-Adenine-Dinucleotide, Oleic Acid, 3-Deoxy-D-mannonic acid, 5-Hydroxymethylfurfural.

Based on the visualization, it is known that there are 9 of the 12 same amino acid residues in the interaction of 3-Deoxy-D-Mannonic Acid and the receptor compared to the control-receptor interaction. The nine amino acids involved in the interaction interact hydrogen and hydrophobically (Table 3 and Picture 4). The same amino acid residues in the interactions of Heptadecene-(8)-Carbonic Acid-(1), 3-Deoxy-D-Mannonic Acid, and the control with the receptor indicate that both compounds contained in *Allium sativum* L. var. solo garlic may have the same activity as the control. It is known that the affinity of ligand-receptor binding increases with the presence of amino acid residues involved in the ligand-receptor interaction complex through hydrogen bonds and hydrophobic bonds. It is also known that the hydrophobic interactions formed play an important role in stabilizing the ligand-receptor bond and help increase the affinity of ligand-receptor binding (Chen *et al.*, 2016; Patil *et al.*, 2010).





Picture 4. Visualization of the molecular docking results between Nicotinamide-Adenine-Dinucleotide (Control), Heptadecene-(8)-Carbonic Acid-(1), and 3-Deoxy-D-Mannonic Acid. A & B. Binding positions of both ligands located on the same active site as the control. C-E. Amino acid residues involved in ligand-receptor interaction. *Mycobacterium tuberculosis* InhA protein is shown as a gray ribbon. Ligands are shown as colored sticks, namely Nicotinamide-Adenine-Dinucleotide (red), Heptadecene-(8)-Carbonic Acid-(1) (yellow), and 3-Deoxy-D-Mannonic Acid (blue). F. Molecular docking results of InhA with 5-Hydroxymethylfurfural G. Molecular docking results of InhA with Melezitose H. Molecular docking results of InhA with Oleic Acid I. Amino acid residues of interaction between InhA and 5-Hydroxymethylfurfural J. Amino acid residues of interaction between InhA and Melezitose. K. Amino acid residues of interaction between InhA and Oleic Acid.

Conclusion

Allium sativum L. var. solo garlic contains alkaloids, flavonoids, tannins and steroids. Based on the results of the GCMS test, the bioactive compounds that have an area above 5% are Heptadecene-(8)-Carbonic Acid-(1), 3-Deoxy-D-Mannonic Acid, 5-Hydroxymethylfurfural, Melezitose and Oleic acid. The results of molecular docking showed that the compound 3-Deoxy-D-mannonic acid has a binding affinity of -4.9, 5-Hydroxymethylfurfural -4.8, Oleic Acid -6.4, Nicotinamide-Adenine-Dinucleotide (Control) has a binding affinity of -11.4. Low binding affinity indicates that the compound can bind proteins with little energy. The Gypsum energy theory states that the smaller the energy produced from a bond between a ligand and its receptor, the more stable the bond. The lowest binding energy to the InhA protein is in the control compounds Nicotinamide-Adenine-Dinucleotide, Oleic Acid, 3-Deoxy-D-mannonic acid, 5-Hydroxymethylfurfural.

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