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*Article*

# The Potential of Active Compounds from Melinjo Leaves (*Gnetum gnemon*) as an Antihistamine using Molecular Docking Approach for Acetylcholine Muscarinic M3 Receptor Inhibition

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**Nelson Gaspersz[1*], Matthew A.H. Amos[1], Yusthinus T. Male[1], Muh. Daswar A. Baharudin[1], Dean Y.E. Leuwol[1], Rahma El[1], Priska M. Pattiasina[1], Mario R. Sohilait[1], Herland Satriawan[2]**

[1]Department of Chemistry, Faculty of Mathematics and Natural Science (FMIPA), Universitas Pattimura, Indonesia
[2]Institute of Ocean and Earth Sciences, University of Malaya, 50603, Kuala Lumpur, Malaysia

**Abstract.** Melinjo leaves (Gnetum gnemon) have been used traditionally in Maluku to treat spoilage in the fish product. Melinjo leaves contain secondary metabolites with anti-inflammatory and antibacterial biological activities. It has been identified contain gnetumal, callyspinol, cassipuorol, (+)-dehydrovomifoliol, p-coumaric acid, ferulic acid, isovitexin, swersitin, isoswersitin, vicenin 2, swertiajaponin, isoswertiajaponin and ursolic acid. This research aims to predict the potential of the compounds in melinjo leaves as antihistamines by inhibiting the activity of the acetylcholine muscarinic M3 receptor. The research was carried out using an in silico study method using a molecular docking approach. Docking results showed that gnetumal, callyspinol, cassipuorol, (+)-dehydrovomifoliol, p-coumaric acid, ferulic acid, isovitexin, swersitin, isoswersitin, vicenin 2, swertiajaponin, isoswertiajaponin and ursolic acid for the muscarinic acetylcholine receptor M3 had a binding affinity value of -7.4, -8.0, 7.0, 7.0, -6.6, -6.6, -7.4, -5.7, -4.4, -3.7, -6.0, -4.9 and -4.7 kcal/mol, respectively.



***Corresponding Author :***
Nelson Gaspersz
Department of Chemistry, Faculty of Mathematics and Natural Science (FMIPA),
Universitas Pattimura, Indonesia
Email : nelson@fmipa.unpatti.ac.id

## 1. Introduction

Antihistamines are pharmaceutical class of drugs that act to treat histamine-mediated conditions [1]. The term of antihistamine can be used to describe any histamine antagonist, but more often used to refer to classic antihistamines that act on the histamine H1 receptor [2]. The human histamine H1 receptor is a member of the G-protein coupled receptor superfamily. One homology-based model for the histamine H3 receptor was generated using the muscarinic M3 acetylcholine receptor [3].

Indonesia has been known for its biological diversities, were Indonesia has around 30,000 species of plants. However, only around 7,500 species have been used as materials for medicine [4]. Some plants that have been used traditionally in Indonesia are rhizome plants as anticancer [5], kirinyuh leaves as anti-diabetics [6]–[8], betel leaves as skin, cough and mouth ulcers [9].

Melinjo leaves (*Gnetum gnemon*) has been used by local people in Maluku to treat spoilage in fish product. It's contain secondary metabolites that have biological activities such as anti-inflammatory and antibacterial [10]. Phytoconstituents that identified in melinjo leaves are gnetumal, callyspinol, cassipourol, (+)-dehydrovomifoliol, p-coumaric acid, ferulic acid, isovitexin, swersitin, isoswersitin, vicenin 2, swertiajaponin, isoswertiajaponin and ursolic acid [11]–[14]. Research about it's activity as antihistamine using molecular docking has not been done.

*In silico* is a drug design using computational methods. This method done for reducing costs for drug discovery [15]. Molecular docking is a computer modeling of complex structures formed by two or more interacting molecules [16]. The aim of molecular docking is to predict bond conformation and bond affinity [17]. The computer prediction results can be used to predict how a chemical compound can inhibit the action of a receptor or macromolecule.

Based on the explanation above, research was conducted to predict the activity of melinjo leaves as an antihistamine using the *in silico* method, especially the molecular docking approach to predict the potential of active compounds from melinjo leaves in inhibiting M3 muscarinic acetylcholine receptors as antihistamines and to understand the interaction that formed.

## 2. Experimental Section

### 2.1. Materials

The hardware used is a set of Acer One laptops with specifications: Intel Core i3 processor, 4GB RAM and 1TB HDD. The software used is Discovery Studio 2021 Client, Hyperchem, Chemdraw, AutodockTools, and Autodock Vina.

This study used acetylcholine muscarinic M3 receptor (PDB ID: 4DAJ) which was downloaded via the Protein Data Bank (PDB) (https://www.rcsb.org) as the target receptor [18].The ligands used were phytoconstituents compound that has been identified contain in *G. gnemon leaves*. There are gnetumal, callyspinol, cassipourol, (+)-dehydrovomifoliol, p-coumaric acid and ferulic acid [14], isovitexin [12], ursolic acid [11] and swertisin, isoswertisin, vicenin 2, swertiajaponin and isoswertiajaponin [13].

### 2.2. Procedures

The receptors were prepared using Discovery Studio, then optimized by adding polar hydrogen atoms and determining the parameters of the gridbox used in the docking process. Test ligands that have been drawn and geometrically optimized are carried out by adding kollman charges, hydrogen, and to determine the rotational bonds when docked.

The ligand is docked to the active site of the receptor. Docking was done using Autodock Vina with x, y and z centers of the gridbox are -14.106, -6.420 and -42.697 and volume 15 × 15 ×15. This type of docking simulation were using oriented gridbox is called focused docking [19].

The results of the docking were then analyzed for bond affinity and rmsd l.b./u.b. of each ligand. The constant inhibition (Ki) was determine using equation that $Ki_{pred}=e^{(\Delta G/RT)}$. Where $\Delta G$ is the AutoDock Vina binding affinity output results (cal/mol), R (gas constant) is 1.98 cal.(mol.K)$^{-1}$, and T

(room temperature) is 298.15 Kelvin [20]. In addition, other yield parameters analyzed were amino acid residues bound to the tested ligands, bond types and bond lengths.

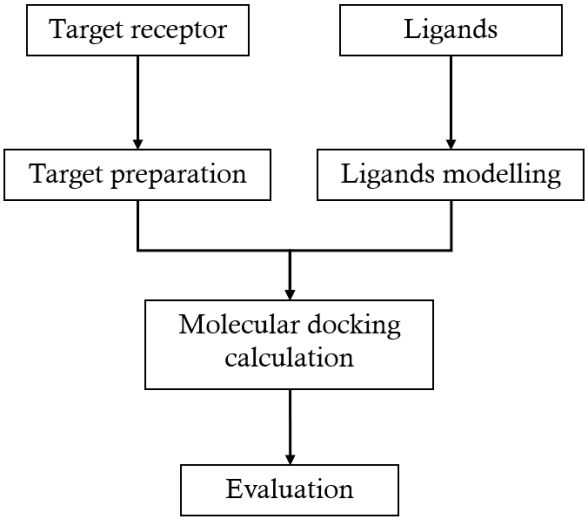



**Figure 1.** Molecular docking procedure

## 3. Results and Discussion

The docking process was validated by redocking tiotropium, the cocrystallized compound with the M3 receptor. Tiotropium was redocked into the same domain of the receptor. Estimation of the RMSD between the native ligand and standard ligand conformation gave 1.161 Å (less than 2.00 Å). The superimposition of these conformation shows in Figure 2. The ligand is almost completely engages in extensive hydrophobic contacts with the receptor. A pair of hydrogen bond bonds are formed with ASP147 and ASN507 as it showns in Table 1.

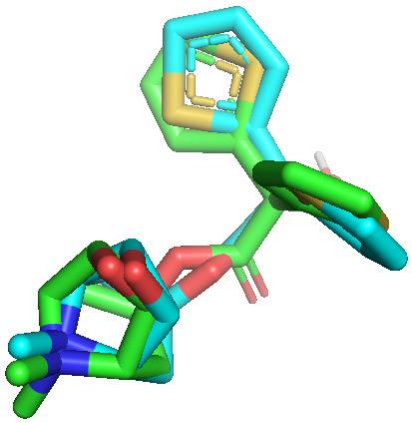


**Figure 2.** The superimposition of native ligand (green) and standard ligand (blue) conformation using PyMol

The result of molecular docking is shown in Table 1. It is shown that 14 compounds that identified in melinjo leaves has potency to inhibit the M3 receptor. There are two different values of RMSD, RMSD l.b. (lower bound) and RMSD u.b. (upper bound). These two differ in how atom are denoted in distance calculation [21].

**Table 1.** AutoDock Vina molecular docking results

| Ligand | Binding affinity (kcal/mol) | Rmsd l.b. (Å) | Rmsd u.b. (Å) | Constant inhibition μM |
|---|---|---|---|---|
| Tiotropium | -8.9 | 0.863 | 1.229 | $2.994\times10^{-7}$ |
| Callyspinol | -8.0 | 0.862 | 1.319 | $1.368\times10^{-6}$ |
| Gnetumal | -7.4 | 0.598 | 1.317 | $3.765\times10^{-6}$ |
| Isovitexin | -7.4 | 1.076 | 2.481 | $3.765\times10^{-6}$ |
| (+)-Dehydrovomifoliol | -7.0 | 1.620 | 3.730 | $7.395\times10^{-6}$ |
| Cassipourol | -7.0 | 1.228 | 2.155 | $7.395\times10^{-6}$ |
| *p*-Coumaric acid | -6.6 | 1.796 | 1.831 | $1.453\times10^{-5}$ |
| Ferulic acid | -6.6 | 1.244 | 1.244 | $1.453\times10^{-5}$ |
| Swertiajaponin | -6.0 | 1.090 | 1.170 | $3.999\times10^{-5}$ |
| Swertisin | -5.7 | 1.117 | 1.315 | $6.635\times10^{-5}$ |
| Isoswertiajaponin | -4.9 | 1.925 | 6.055 | $2.560\times10^{-4}$ |
| Ursolic acid | -4.7 | 1.735 | 3.436 | $3.588\times10^{-4}$ |
| Isoswertisin | -4.4 | 1.967 | 5.055 | $5.953\times10^{-4}$ |
| Vicenin 2 | -3.7 | 1.582 | 2.090 | $1.940\times10^{-3}$ |

Based on the docking results, 14 compounds identified in melinjo leaves using a molecular docking approach had a negative binding affinity. Where the best binding affinity was obtained by callyspinol of -8.0 kcal/mol. Furthermore, gnetumal and isovitexin with a bond affinity of -7.4 kcal/mol. Then (+)-dehidrovomifoliol and cassipourol with a bond affinity value of -7.0 kcal/mol. Ferulic acid and p-coumaric acid, which are phenolic group compounds, have the lowest affinity having an affinity value of -6.6 kcal/mol.

The docking results that are considered good if had value of rmsd l.b./u.b. ≤ 2 Å [22]. The docking results showed that callyspinol, gnetumal, *p*-coumaric acid, and ferulic acid had rmsd l.b./u.b values ≤ 2 Å. This shows that these ligands can form stable complexes with the M3 muscarinic acetylcholine receptor and the conformation of the test ligands as a result of the binding is getting closer to the standard conformation [6], [23].

Interactions that formed between the ligand and receptor can affect the binding affinity [24]. Where the binding affinity indicates the presence and strength of the interaction between the ligand and protein, so that a comparative analysis of the interaction results of the binding between ligand and M3 receptor using the Discovery Studio is shown in Table 2. There are LEU225 and TYR524 that plays important role in this receptor. These residue are positioned near a probable allosteric, they are relating to with the receptor activity [18].

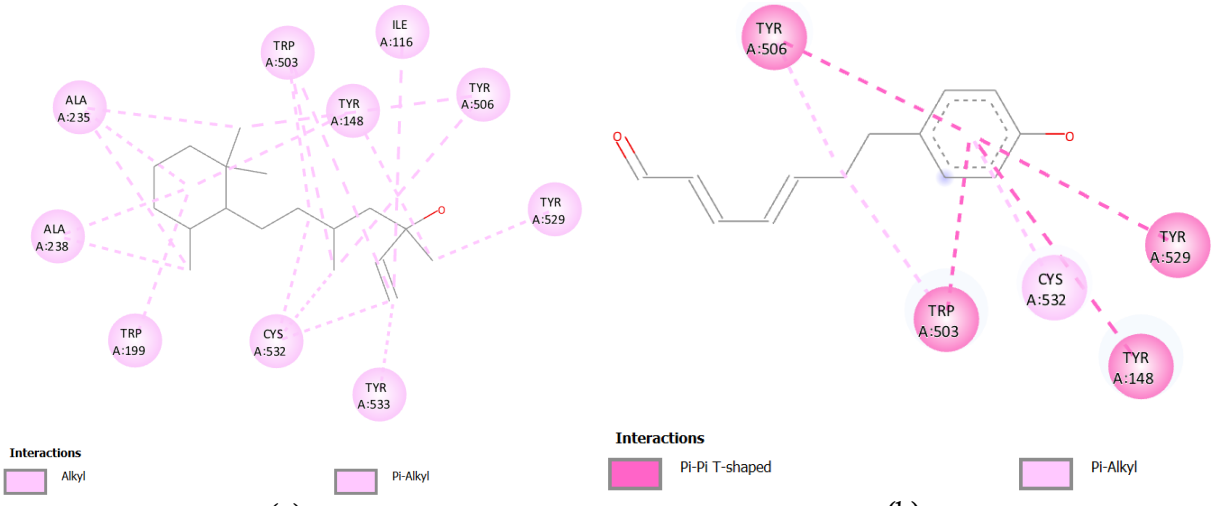
**Table 2**. Comparison of the interaction of the binding results between the standard ligand and the test ligand with the M3 muscarinic acetylcholine receptor

| Ligand | Interaction with amino acid | |
|---|---|---|
| | Hydrogen Bond | Hydrophobic and others interaction |
| Tiotropium | ASP147, ASN507 | ASP147, TYR148, VAL155, ALA235, ALA238, TYR529 |
| Callyspinol | - | ILE116, TYR148, TRP199, ALA235, ALA238, TRP503, TYR506, TYR529, CYS532, TYR533 |
| Gnetumal | - | TYR148, TRP503, TYR506, TYR529, CYS532 |

| | | |
|---|---|---|
| Isovitexin | TYR148, CYS220, ASN526 | TYR148, LEU225, ALA235 |
| (+)-Dehydrovomifoliol | ASN507, CYS532 | TYR506 |
| Cassipourol | TYR148, TYR506 | TYR148, ALA238, TRP503, TYR506. TYR529, CYS532 |
| *p*-Coumaric acid | TYR148, SER151 | TRP503, TYR506, CYS532 |
| Ferulic acid | SER151, ASN507 | TRP199, ALA238, TRP503, CYS532 |
| Swertiajaponin | THR112, ASP178, LYS179, TYR431, HIS450, TYR458 | ASN84, ASP107, TYR108, SER111, TRP428, TYR431, ILE454 |
| Swertisin | THR112, ASP178, LYS179, TYR431, HIS450, TYR458 | ASN84, ASP107, TYR108, TRP428 TYR431, ILE454 |
| Isoswertiajaponin | - | PHE116, PHE119, LEU201, LEU205 |
| Ursolic acid | TYR458 | ASP107, TYR108, TRP428, TYR431, PHE432 |
| Isoswertisin | PRO430, LEU449 | ILE429, PRO430, THR453 |
| Vicenin 2 | ILE425, LEU426 | PRO430 |

Based on the docking results, callyspinol only has hydrophobic interaction with ILE116, TYR148, TRP199, ALA235, ALA238, TRP503, TYR506, TYR529, CYS532 and TYR533. This interaction is an alkyl and π-alkyl interaction. The hydrophobic interaction can stabilize the protein because it produces sufficient Gibbs free energy due to the absence of a desolvation step [25].

Gnetumal is formed by hydrophobic interactions with the interaction center formed on the benzene ring. The hydrophobic interactions are the π-π T-shaped interactions with TYR148, TRP503, TYR506, and TYR529; and π-alkyl interactions with TRP503, TYR506, and CYS532. Gnetumal has benzene ring that could provide the π-orbitals to formed π-π interactions. Gnetumal has more π-π interactions that is a major driving force to stabilizes protein [26]. The interaction of these two ligands are shown in the 2D diagram in Figure 3.

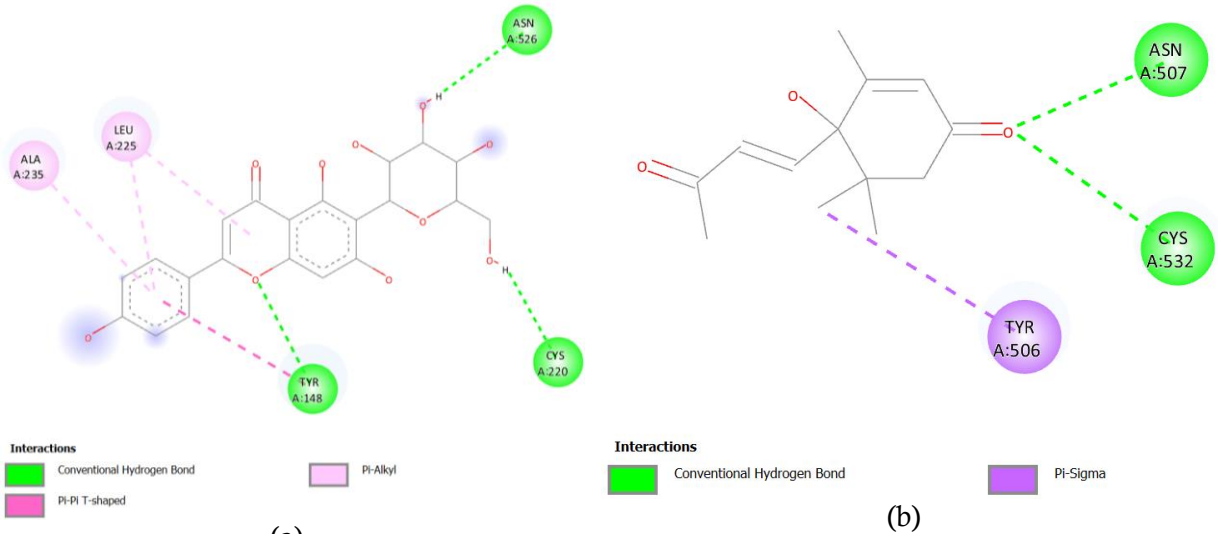



(a) (b)

**Figure 3.** Interaction between amino acid residues and (a) callyspinol and (b) gnetumal

Isovitexin formed hydrogen bonds with TYR148, CYS220, and ASN526; and hydrophobic interactions with TYR148, LEU225 and ALA235. Cassipourol formed hydrogen bonds with ASN507 and CYS532; and hydrophobic interaction with TYR506. The interaction of these two ligands are in Figure 4.

Isovitexin compound has glycone that have important role to binding affinity from docking result, especially for phenolic compound [27].The hydrogen bond can be formed because these two ligands can provide hydrogen bond donor/acceptor to interact with residue from receptor. Hydrogen bonds are the strongest and most important noncovalent interactions in protein-ligand complexes [28], [29].



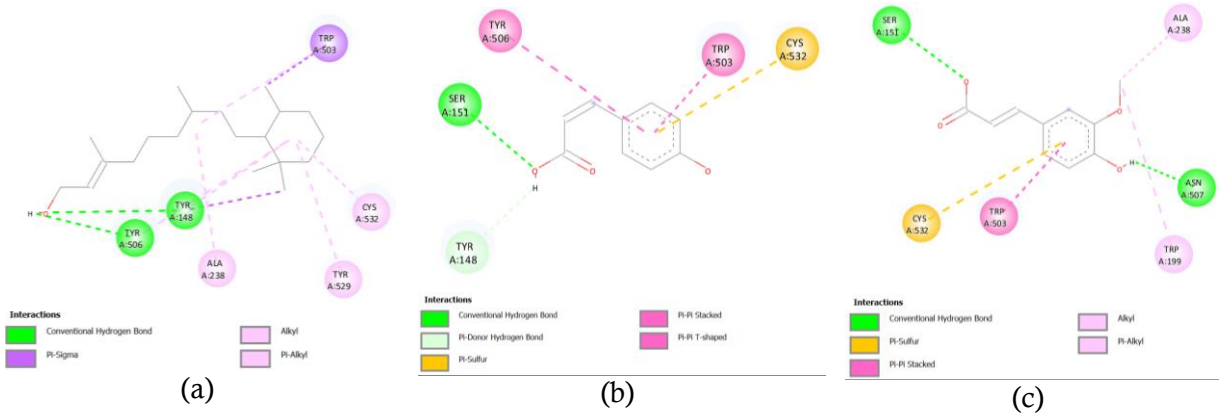

(a) (b)

**Figure 4.** Interaction between amino acid residues and (a) isovitexin and (b) cassipourol

The (+)-dehydromifoliol ligand formed hydrogen bonds with residues TYR148 and TYR506; and hydrophobic interactions with TYR148, ALA238, TRP503, TYR506, TYR529 and CYS532. The p-coumaric acid, formed hydrogen bonds with TYR148 and SER151; hydrophobic interaction with TRP503 and TYR506; and π-sulfur interaction with CYS532. Ferulic acid formed hydrogen

bonds with SER151 and ASN507; hydrophobic interaction with TRP199, ALA238 and TRP503; and π-sulfur interaction with CYS532. The interaction of these three ligands is shown in Figure 5.

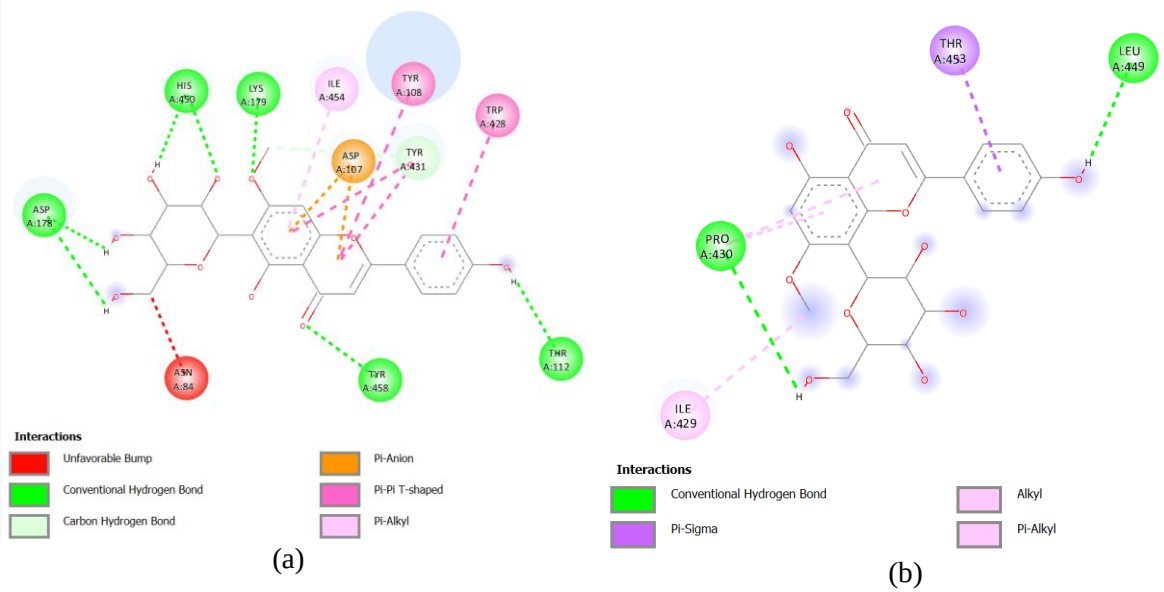
The (+)-dehydromifoliol has no benzene ring that could give π-orbitals to formed π-σ or π-alkyl interactions, it's indicates that the π-orbitals are came from residues to formed hydrophobic interactions. Ferulic acid and p-coumaric acid is the only two ligands that have π-sulfur interaction with the receptor amino acid. The π-sulfur interactions is known as noncovalent interactions between sulfur center and aromatic rings that play important roles in protein folding and stabilization [30], [31].




(a) (b) (c)

**Figure 5.** Interaction between amino acid residues and (a) (+)-dehydrovomifoliol, (b) *p* coumaric acid and (c) ferulic acid

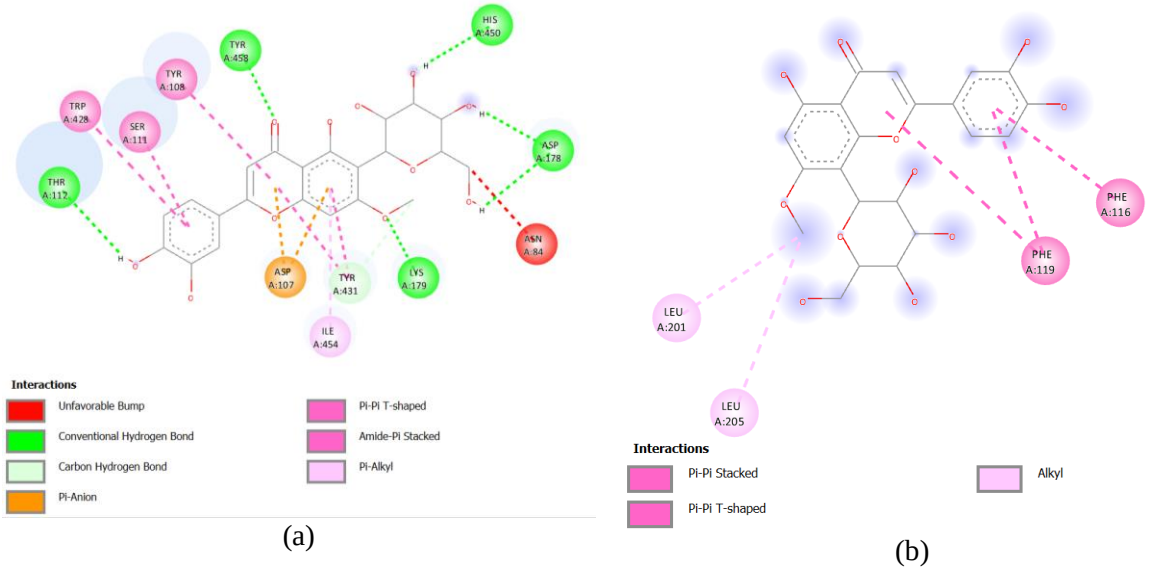
Swertisin formed hydrogen bonds with THR112, ASP178, LYS179, TYR431, HIS450 and TYR458; hydrophobic interactions with TYR108, TRP428, TYR431 and ILE454; electrostatic interactions with ASP107; and unfavorable interactions with ASN84. Isoswertisin formed hydrogen bonds with PRO430 and LEU449; and hydrophobic interactions with ILE429, PRO430 and THR453. These interaction of the two ligands are shown in Figure 6.




(a) (b)

**Figure 6.** Interaction between amino acid residues and (a) swertisin and (b) isoswertisin

Swertiajaponin formed hydrogen bonds with THR112, ASP178, LYS179, TYR431, HIS450 and TYR458; hydrophobic interactions with TYR108, SER111, TRP428, TYR431 and ILE454; electrostatic interactions with ASP107; and unfavorable interactions with ASN84. Isoswertiajaponin formed hydrophobic interactions with PHE116, PHE119, LEU201 and LEU205. These interaction of the two ligands are shown in Figure 7.




(a) (b)

**Figure 7.** Interaction between amino acid residues and (a) swertiajaponin and (b) isoswertiajaponin

Ursolic acid formed hydrogen bonds with TYR458; hydrophobic interactions with TYR108 and TRP428; and unfavorable interactions with ASP107, TYR431 and PHE432. Vicennin 2 formed hydrogen bonds with ILE435 and LEU426; and hydrophobic interactions with PRO430. These interaction of the two ligands are shown in Figure 8.

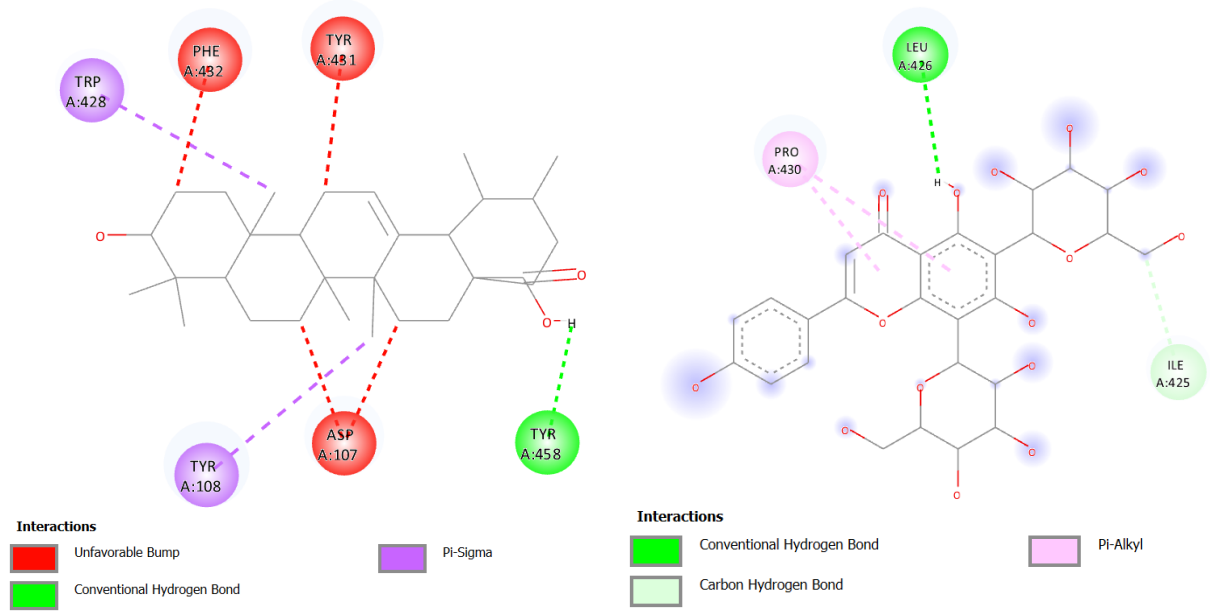



**Figure 8.** Interaction between amino acid residues and (a) swertiajaponin and (b) isoswertiajaponin

The same study using acetylcholine muscarinic M3 receptor (4DAJ) by Fraga and Borges in 2020 also shows the same tendency to formed hydrophobic interactions between benzene rings from test ligand and amino acid residues [32]. Based on Table 2, it's shows that there are more hydrophobic interactions that formed then hydrogen bonds. This predicted to occurs because of the binding sites is more hydrophobic that can provide the hydrophobic interactions to formed and there are less hydrogen donor/acceptor from amino acids to interact with test ligand.

## 4. Conclusion

The results of the molecular docking of compounds from melinjo leaf (*Gnetum gnemon*) namely calispinol, gnetumal, isovitexin, (+)-dehidrovomifoliol, cassipourol, p-coumaric acid, ferulic acid, swertiajaponin, swertisin, isoswertiajaponin, ursolic acid, isoswertisin and vicenin 2 towards the M3 muscarinic acetylcholine receptors have a binding affinity value of - 8.0, -7.4, -7.4, -7.0, -7.0, -6.6, and -6.6 kcal/mol, respectively. The affinities of all compounds indicates that these compounds were predicted to have activity in inhibit the acetylcholine muscarinic M3 receptor. However, further testing is still needed both *in vito* and *in vivo* to prove the ability of melijo leaves as an antihistamine.

## References

[1] F. Monczor and N. Fernandez. (2016). Current Knowledge and Perspectives on Histamine $H_1$ and $H_2$ Receptor Pharmacology: Functional Selectivity, Receptor Crosstalk, and Repositioning of Classic Histaminergic Ligands, *Mol Pharmacol*, vol. 90, no. 5, pp. 640–648.

[2] K. L. Randall and C. A. Hawkins. (2018). Antihistamines and Allergy, *Aust Prescr*, vol. 41, no. 2, pp. 42–45.

[3] N. Ghamari, O. Zarei, D. Reiner, S. Dastmalchi, H. Stark, and M. Hamzeh-Mivehroud. (2019). Histamine $H_3$ Receptor Ligands by Hybrid Virtual Screening, Docking, Molecular Dynamics simulations, and Investigation of their Biological Effects, *Chem Biol Drug Des*, vol. 93, no. 5, pp. 832–843.

[4] A. R. Mugni and A. N. Hasanah. (2018). Artikel Tinjauan: Fitosom sebagai Sistem Penghantar Obat Transdermal Formulasi Baru Obat Herbal untuk Perkembangan Farmasetika di Indonesia, *Farmaka*, vol. 16, no. 1, pp. 61–71.

[5] S. Shavira, A. D. Margaretta, A. D. Sandra, R. U. Sitorus, and F. Fatmaria. (2021). Formulasi dan Uji Stabilitas Sediaan Suspensi Ekstrak Rimpang *Zingiber zerumbet*, *HMJ*, vol. 4, no. 4, pp. 7–13.

[6] N. Gaspersz, E. G. Fransina, and A. R. Ngarbingan. (2022). Uji Aktivitas Penghambatan Enzim α-Amilase dan Glukoamilase dari Ekstrak Etanol Daun Kirinyuh (*Chromolaena odorata* L.), *J. Kim. Mul*, vol. 19, no. 2, pp. 51–57.

[7] N. Gaspersz, M. A. H. Amos, S. H. Kalauw, I. Harjuni, and M. R. Sohilait. (2022). Penambatan Molekuler Penghambatan Aktivitas Enzim α-Amilase dan α-Glukosidase oleh Senyawa Aktif Daun Kirinyuh (*Chromolaena odorata* L.), *Kovalen*, vol. 8, no. 3, pp. 230–237,

[8] N. Gaspersz, A. R. Ngarbingan, W. O. Apriana, and S. L. L. Silayar. (2023). Study of Utilization of Kirinyuh Leaf Extract (*Chromolaena odorata*) as Antidiabetic Plaster: Phytochemical Screening and *in-Vivo* Test, in *The 7th International Conference on Basic Sciences 2021*, Ambon, Indonesia: AIP, Jan. 2023, pp. 1–10.

[9] B. Bustanussalam, D. Apriasi, E. Suhardi, and D. Jaenudin. (2015). Efektivitas Antibakteri Ekstrak Daun Sirih (*Piper betle* Linn) Terhadap *Staphylococcus aureus* ATCC 25923, *JF*, vol. 5, no. 2, pp. 58–64.

[10] E. Kining, D. Firdiani, and S. Asma. (2022). Aktivitas Antibakteri dan Antibiofilm Ekstrak Air Daun Melinjo terhadap Bakteri *Pseudomonas aeruginosa*, *Indonesia Natural Research Pharmaceutical Journal*, vol. 7, no. 1, pp. 19–33.

[11] P. P. Dutta *et al.* (2018). Antiplasmodial Activity of *Gnetum gnemon* Leaves and Compounds Isolated from Them, *Natural Product Communications*, vol. 13, no. 10, pp. 1263–1265.
[12] Y. D. I. Siregar *et al.* (2016). Extraction of Isovitexin from Melinjo (*Gnetum gnemon* L.) Leaves Using Mixtures of Liquid Carbon Dioxide and Ethanol. *International Journal of Biomass & Renewables*, vol. 5, no. 2, pp. 23–30.
[13] J. W. Wallace and G. Morris. (1978). C-Glycosylflavones in *Gnetum gnemon*, *Phytochemistry*, vol. 17, no. 10, pp. 1809–1810.
[14] T. H. Le, T. N. Van Do, H. X. Nguyen, P. H. Dang, N. T. Nguyen, and M. T. T. Nguyen. (2012). A New Phenylheptanoid from the Leaves of *Gnetum gnemon* L. *Natural Product Research*, vol. 35, no. 21, pp. 3999–4004.
[15] L. A. Vu, P. T. C. Quyen, and N. T. Huong. (2015). *In silico* Drug Design: Prospective for Drug Lead Discovery, *IJESI*, vol. 4, no. 10, pp. 60–70.
[16] G. Monika, G. Punam, and S. Sarbjot. (2010). An Overview on Molecular Docking, *IJDDR*, vol. 2, no. 2, pp. 219–231.
[17] M. R. Sohilait, H. D. Pranowo, and W. Haryadi. (2017). Molecular Docking Analysis of Curcumin Analogues with COX-2, *Bioinformation*, vol. 13, no. 11, pp. 356–359.
[18] A. C. Kruse *et al.* (2012). Structure and Dynamics of the M3 Muscarinic Acetylcholine Receptor, *Nature*, vol. 482, no. 7386, pp. 552–556.
[19] D. Ghersi and R. Sanchez. (2009). Improving Accuracy and Efficiency of Blind Protein-Ligand Docking by Focusing on Predicted Binding Sites, *Proteins*, vol. 74, no. 2, pp. 417–424.
[20] S. Shityakov and C. Foerster. (2014). *In Silico* Structure-Based Screening of Versatile P-Glycoprotein Inhibitors Using Polynomial Empirical Scoring Functions, *AABC*, vol. 2014, no. 7, pp. 1–9, Mar.
[21] O. Trott and A. J. Olson. (2010). AutoDock Vina: Improving the Speed and Accuracy of Docking with a New Scoring Function, Efficient Optimization, and Multithreading, *J. Comput. Chem.*, vol. 31, no. 2, pp. 455–461.
[22] K. Shah, S. Mujwar, J. K. Gupta, S. K. Shrivastava, and P. Mishra. (2019). Molecular Docking and *In Silico* Cogitation Validate Mefenamic Acid Prodrugs as Human Cyclooxygenase-2 Inhibitor, *ASSAY and Drug Development Technologies*, vol. 17, no. 6, pp. 285–291.
[23] N. Gaspersz and M. R. Sohilait. (2019). Penambatan Molekuler α, β, dan γ-Mangostin sebagai Inhibitor α-Amylase Pankreas Manusia, *Indo. J. Chem. Res.*, vol. 6, no. 2, pp. 59–66.
[24] M. J. A. Bernaldez, J. B. Billones, and A. Magpantay. (2018). *In Silico* Analysis of Binding Interactions between GSK983 and Human DHODH through Docking and Molecular Dynamics, *AIP Conference Proceedings*, vol. 2045, no. 1, pp. 1–8.
[25] S. G. Krimmer, J. Cramer, J. Schiebel, A. Heine, and G. Klebe. (2017). How Nothing Boosts Affinity: Hydrophobic Ligand Binding to the Virtually Vacated $S_1'$ Pocket of Thermolysin, *J. Am. Chem. Soc.*, vol. 139, no. 30, pp. 10419–10431.
[26] D. Paik, H. Lee, H. Kim, and J.-M. Choi. (2022). Thermodynamics of π–π Interactions of Benzene and Phenol in Water, *IJMS*, vol. 23, no. 17, p. 9811.
[27] S. Chigurupati *et al.* (2022). Molecular Docking of Phenolic Compounds and Screening of Antioxidant and Antidiabetic Potential of *Moringa oleifera* Ethanolic Leaves Extract from Qassim Region, Saudi Arabia, *Saudi Journal of Biological Sciences*, vol. 29, no. 2, pp. 854–859.
[28] G. A. Gyebi *et al.* (2021). Structure-Based Virtual Screening Suggests Inhibitors of 3-Chymotrypsin-Like Protease of SARS-CoV-2 from *Vernonia amygdalina* and *Occinum gratissimum*, *Computers in Biology and Medicine*, vol. 136, no. 1, pp. 1–16.
[29] M. Simončič and T. Urbic. (2018). Hydrogen Bonding Between Hydrides of the Upper-Right Part of the Periodic Table, *Chemical Physics*, vol. 507, no. 1, pp. 34–43.

[30] Y. Jin *et al.* (2023). Sulfur–Arene Interactions: the S···π and S–H···π Interactions in the Dimers of Benzofuran···Sulfur Dioxide and Benzofuran···Hydrogen Sulfide, *Phys. Chem. Chem. Phys.*, vol. 25, no. 17, pp. 12174–12181.

[31] A. L. Ringer, A. Senenko, and C. D. Sherrill. (2007). Models of S/π Interactions in Protein Structures: Comparison of the $H_2S$-Benzene Complex with PDB Data,"*Protein Sci.*, vol. 16, no. 10, pp. 2216–2223.

[32] L. F. Fraga and L. L. Borges. (2020). Busca De Moléculas Com Atividade Broncodilatadora Na Espécie Mikania Glomerata Spreng Empregando Ferramentas *in Silico*, *RBMC*, vol. 6, no. 15, pp. 25–31.