IN SILICO EVALUATION OF THE ANTIOXIDANT ACTIVITY OF 4-MERCAPTOIMIDAZOLE MONOSUBSTITUTED DERIVATIVES

ĐÁNH GIÁ *IN SILICO* HOẠT TÍNH CHỐNG OXY HÓA CỦA CÁC DẪN XUẤT MỘT NHÓM THẾ HỢP CHẤT 4-MERCAPTOIMIDAZOLE

Do Thi Ngoc Hang[1], Nguyen Minh Thong[2]*, Nguyen Quang Trung[2], Nguyen Hong Son[1], Nguyen Thi Trung Chinh[1], Mai Van Bay[2], Quan V. Vo[1]*

[1]The University of Danang - University of Technology and Education, Danang, Vietnam
[2]The University of Danang - University of Science and Education, Danang, Vietnam

* Corresponding author: vvquan@ute.udn.vn; nmthong@ued.udn.vn

(Received: August 30, 2023; Revised: October 06, 2023; Accepted: October 09, 2023)

Abstract - In this study, 54 monoderivatives of 4-mercaptoimidazole (**4MC)** were screened for their antioxidant activity using computational simulations. It was discovered that the presence of electron-absorbing groups, such as NO_2, CN, CF_3, COOH, and $COOCH_3$, increases the BDE (S-H) and IE values of **4MC**, whereas the presence of electron-donating groups, such as Eti, Ph, Ete, Me, Et, OH, OMe, NH_2, NHMe, and NMe_2, decreases these values. 5-NH_2-**4MC** has the lowest BDE value (66.6 kcal/mol), while 5-$NHMe_2$-**4MC** has the lowest IE value (157.1 kcal/mol). Thus, in the gas phase, these derivatives could exhibit the highest radical scavenging activity following the hydrogen transfer and single electron transfer, respectively. The analysis of docking data revealed that 5-Ph-4MC formed strong bonds with the LO and CP450 enzymes, indicating that this compound could inhibit these enzymes.

Key words - 4–mecaptoimidazole; LO enzyme; CP450 enzyme; antioxidant; ADMET.

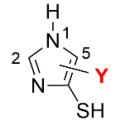
Tóm tắt - Trong nghiên cứu này, các mô phỏng tính toán được sử dụng để nghiên cứu hoạt tính chống oxy hóa của 54 dẫn xuất một nhóm thế của 4-mercaptoimidazole (4MC). Kết quả cho thấy các nhóm thế hút electron (NO_2, CN, CF_3, COOH, và $COOCH_3$) làm tăng giá trị BDE (S-H) và IE của 4MC, ngược lại các nhóm thế đẩy electron (Eti, Ph, Ete, Me, Et, OH, OMe, NH_2, NHMe và NMe_2) làm giảm các giá trị này. Trong đó, 5-NH_2-4MC có giá trị BDE thấp nhất (66,6 kcal/mol), và 5-$NHMe_2$-4MC có giá trị IE thấp nhất (157,1 kcal/mol). Do đó, hai dẫn xuất này thể hiện hoạt tính bắt gốc tự do cao nhất tương ứng với quá trình chuyển hydro và chuyển electron trong pha khí. Ngoài ra, phân tích dữ liệu docking phân tử cho thấy 5-Ph-4MC hình thành liên kết mạnh với các enzyme LO và CP450, do vậy hợp chất này có thể ức chế tốt các enzyme này.

Từ khóa - 4–mecaptoimidazole; enzyme LO; enzyme CP450; chống oxy hóa; ADMET

1. Introduction

A group of heterothiols, specifically the 1-methyl-4-mercaptohistidines (referred to as ovothiols), has been observed to exist in significant quantities within invertebrate eggs [1]. Extensive research has been conducted to investigate the antioxidant properties of these compounds, which have been found to operate in a regenerative cycle. This cycle involves the consumption of hydrogen peroxide followed by regeneration through the action of glutathione [2-4].

The compound known as 4-mercaptoimidazole (**4MC**) exhibits a relationship with ovothiols. The compounds known as **4MC** and its derivatives have demonstrated comparable levels of action to ovothiols, indicating its potential as a class of antioxidants [3, 5, 6]. Based on calculations, it has been suggested that **4MC** exhibits potential as a highly effective scavenger of HO· in both nonpolar and aqueous environments, with overall rate constants of 1.10×10^{10} and 9.58×10^{9} $M^{-1}\,s^{-1}$, respectively. Furthermore, in silico studies have demonstrated its remarkable antiradical activity against $CH_3O^{\bullet}$, $CCl_3O^{\bullet}$, $HOO^{\bullet}$, $CH_3OO^{\bullet}$, $CCl_3OO^{\bullet}$, NO_2, $O_2^{\bullet-}$, $SO_4^{\bullet-}$, DPPH and $ABTS^{\bullet+}$ *in silico* [7]. The examination of monoderivatives holds significant importance in the search for effective antioxidants; nonetheless, this particular matter remains unexplored in current research. The objective of this study is to assess the antioxidant activity of monoderivatives of **4MC** that possess different functional groups, such as NO_2, CN, CF_3, COOH, F, Cl, Br, $COOCH_3$, Ethinyl (Eti), Phenyl(Ph), Ethenyl (Ete), Me, Ethanyl (Et), OH, OMe, NH_2, NHMe, NMe_2. This will be achieved by determining the thermodynamic parameters and conducting molecular docking calculations.



Y = NO_2, CN, CF_3, COOH, F, Cl, Br, $COOCH_3$, Eti, Ph, Ete, Me, Et, OH, OMe, NH_2, NHMe, NMe_2

4-mercaptoimidazole (**4MC**)

Figure 1. *The structure of* ***4MC***

2. Method

2.1. "Drug-likeness" property screening

The SWISS online tool (http://www.swissadme.ch/) [8] was used to screen for drug-likeness compounds that achieve the criteria of all models, including Lipinski [9], Ghose [10], Egan [11], Veber [12], GI (Gastrointestinal) absorption and PAINS (Pan assay interference structures).

2.2. ADMET analysis

The pharmacokinetic properties of potential drug compounds including absorption, distribution, metabolism, excretion, and toxicity (ADMET) were

predicted using the pkCSM online tool (http://biosig.unimelb.edu.au/pkcsm/prediction) [13].

2.3. Molecular docking simulation

Potential drug compounds are converted from optimized chemical structures into *mdb format to create an input database for molecular docking. The structures of pro-oxidant proteins were obtained from the Protein Databank. MOE software [14] was used to simulate the binding of the ligands to the target enzyme. Discovery Studio software was used to visualize the interactions between ligands and receptors [15].

2.4. Thermodynamic calculations

In this work, bond dissociation energies (BDEs) and ionization energies (IEs) of the compounds were calculated with the M06-2X/6-311++G(d,p) method [16-19]. All calculations were carried out using Gaussian 16 software [20].

3. Results and discussion

3.1. Thermodynamic analysis

*3.1.1. The BDE of **4MC** monoderivatives*

Prior research has indicated that the radical scavenging activity of organic compounds, specifically **4MC,** and its derivatives, predominantly adhere to the FHT mechanism in the gas phase. This mechanism is characterized by the BDE value. In the first stage, the BDE of the S-H bond in **4MC** and its monoderivatives with various functional groups including NO_2, CN, CF_3, COOH, F, Cl, Br, $COOCH_3$, Eti, Ph, Ete, Me, Et, OH, OMe, NH_2, NHMe, NMe_2 were calculated. The computed values are presented in Table 1 and Figure 2.

The findings of the research indicate that there exists an inverse relationship between bond dissociation energy and bond stability. Specifically, when the bond dissociation energy decreases, the bond becomes less stable, hence facilitating the separation of the hydrogen atom from the molecule in its free state ($H^{\bullet}$). Consequently, compounds with lower BDE exhibit greater resistance to oxidation by hydrogen atom transfer. Table 1 and Figure 2 provide evidence indicating that electron-absorbing groups, such as NO_2, CN, CF_3, COOH, and $COOCH_3$, predominantly result in an elevation of the BDE value of **4MC** at the S-H bond. The augmentation in BDE of the molecule incorporating the substituent is demonstrated by the NO_2–**4MC** compound at positions 1, 2, and 5. The findings indicate that the BDE value at location 5–NO_2–**4MC** (84.1 kcal/mol) has a greater magnitude compared to the other two sites, with a difference of about 8.26 kcal/mol when compared to the BDE of compound **4MC** (75.9 kcal/mol).

Table 1. *Computed BDE (S–H) and –BDE (S–H) in kcal/mol of **4MC** and its derivatives*

Subsituents	Positions					
	1		2		5	
	BDE	ΔBDE	BDE	ΔBDE	BDE	ΔBDE
NO_2	77.9	2.0	77.8	1.9	84.1	8.2
CN	77.7	1.8	77.6	1.7	79.6	3.7
CF_3	77.4	1.5	77.2	1.3	78.5	2.6
COOH	77.0	1.1	77.9	2.0	81.4	5.5
$COOCH_3$	76.7	0.9	77.6	1.7	81.0	5.2
F	76.5	0.6	75.5	–0.4	74.8	–1.1
Cl	76.2	0.3	75.8	–0.1	75.5	–0.4
Br	76.1	0.2	75.9	–0.03	75.7	–0.2
Eti (C_2H)	76.4	0.5	76.9	1.0	76.6	0.8
Ph (C_6H_5)	75.6	–0.3	75.2	–0.7	75.0	–0.9
Et (C_2H_3)	75.2	–0.7	74.8	–1.1	74.0	–1.9
Me (CH_3)	74.8	–1.1	75.0	–0.9	74.2	–1.7
Ete (C_2H_5)	75.6	–0.3	76.0	0.1	74.3	–1.6
OH	75.9	–0.02	74.0	–1.9	69.8	–6.1
OMe	75.6	–0.3	73.5	–2.4	71.0	–4.9
NH_2	75.5	–0.4	73.6	–2.3	66.6	–9.3
NHMe	75.3	–0.6	73.0	–2.9	68.8	–7.2
NMe_2	75.2	–0.7	72.9	–2.9	69.7	–6.2
4MC (S–H) = 75.9 kcal/mol						

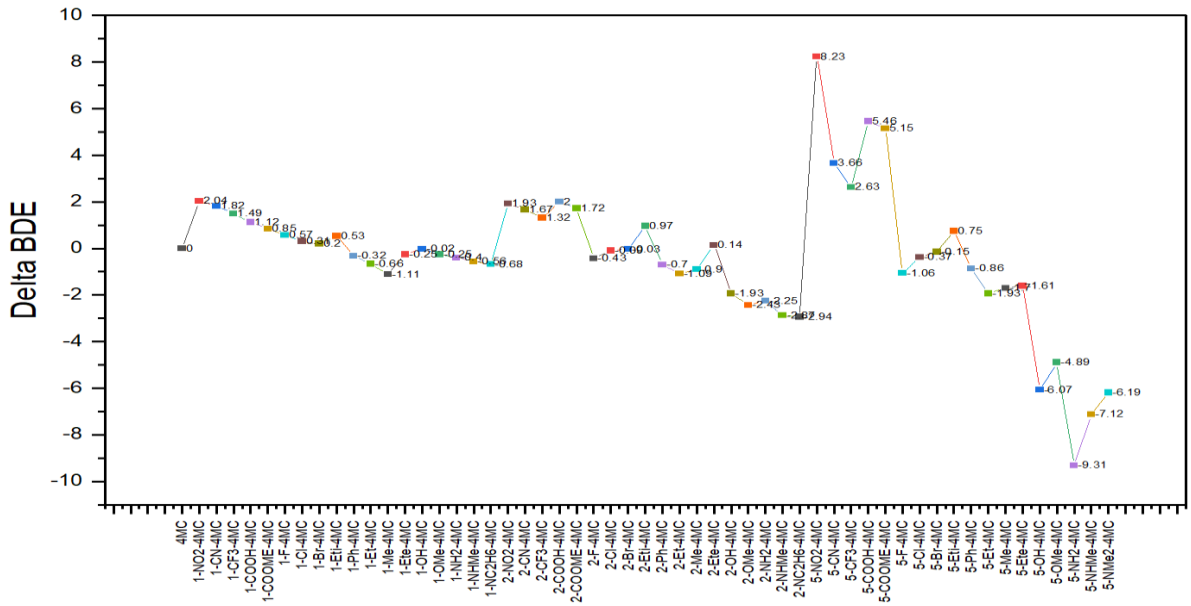


Figure 2. *Calculated ΔBDE of **4MC** and the derivatives*

Certain groups, such as F, Cl, and Br, have the ability to both attract and donate electrons. Additionally, there are electron-donating groups, including Eti, Ph, Ete, Me, Et, OH, OMe, NH_2, NHMe, and NMe_2. These groups significantly decrease the BDE value in comparison to compound **4MC**. When examining the molecule NH_2–**4MC** at locations 1, 2, and 5, it is evident that the most prominent observation is the reduction in BDE value at position 5–NH_2–**4MC** (66.6 kcal/mol) compared to the other two sites. In contrast, the energy reduction is particularly seen to be around 9.3 kcal/mol when compared to compound **4MC**.

When assessing the various positions of compound **4MC** including substituents at the S-H bond, it is seen that the majority of the BDE values decrease at the same place due to the electron-attracting nature of the substituents. In particular, the NO_2 substituent exhibits the maximum value of 77.9 kcal/mol at position 1, while the Me group demonstrates the lowest value with a BDE of 74.8 kcal/mol. In the second position, the COOH group exhibited the highest value of 77.9, while the NMe_2 group had the lowest value of 72.9 kcal/mol for the BDE. In a similar vein, it can be observed that position 5 exhibits the highest BDE when considering NO_2, while the lowest BDE is observed with NH_2, with corresponding values of 84.1 and 66.6 kcal/mol, respectively. Based on the calculated data, 5–NH_2–**4MC** has the lowest BDE value of 66.6 kcal/mol, indicating its comparatively higher propensity for hydrogen atom transfer in comparison to other compounds.

*3.1.2. The IE of **4MC** monoderivatives*

Ionization energy (IE) is one of the essential parameters for measuring antioxidant activity. The calculation of the IE value is based on the SETPT mechanism. The lower the IE value, the more easily electron transfer occurs within the molecule, resulting in greater antioxidant activity. Survey the IE values of all derivatives containing the following substituents at positions 1, 2, and 5: NO_2, CN, CF_3, COOH, F, Cl, Br, $COOCH_3$, Eti, Ph, Ete, Me, Et, OH, OMe, NH_2, NHMe, NMe_2,. The evaluation of IE was performed using the M06-2X/6-311++G(d,p) method, as shown in Table 2 and Figure 3.

Table 2. *Computed IE (S–H) and –BDE (S–H) in kcal/mol of **4MC** and its derivatives*

Subsituents	Positions					
	1		2		5	
	IE	ΔIE	IE	ΔIE	IE	ΔIE
NO_2	201.6	15.3	203.7	17.3	207.8	21.5
CN	202.5	16.2	199.1	12.8	201.5	15.2
CF_3	196.5	10.2	197.0	10.6	198.5	12.1
COOH	191.6	5.2	192.6	6.3	194.5	8.2
$COOCH_3$	188.2	1.8	189.6	3.3	191.0	4.7
F	195.0	8.7	190.2	3.9	188.9	2.6
Cl	190.5	4.2	188.2	1.9	186.9	0.6
Br	188.6	2.3	187.5	1.2	186.0	–0.4
Eti (C_2H)	189.3	2.9	185	–1.4	183.6	–2.8
Ph (C_6H_5)	179.4	–6.9	174.4	–12.0	174.0	–12.4
Et (C_2H_3)	179.5	–6.8	178.4	–8.0	178.1	–8.3
Me (CH_3)	181.3	–5.1	179.6	–6.7	179.5	–6.8
Ete (C_2H_5)	182.9	–3.4	178.3	–8.1	178.4	–7.9
OH	187.9	1.6	179.5	–6.8	181.6	–4.8
OMe (OCH_3)	184.8	–1.5	175.0	–11.3	173.3	–13.0
NH_2	183.5	–2.9	169.2	–17.2	165.5	–20.9
NHMe	181.3	–5.0	164.6	–21.8	160.4	–26.0
NMe_2	179.7	–6.7	160.5	–25.8	157.1	–29.2
4MC = 186.3 kcal/mol						

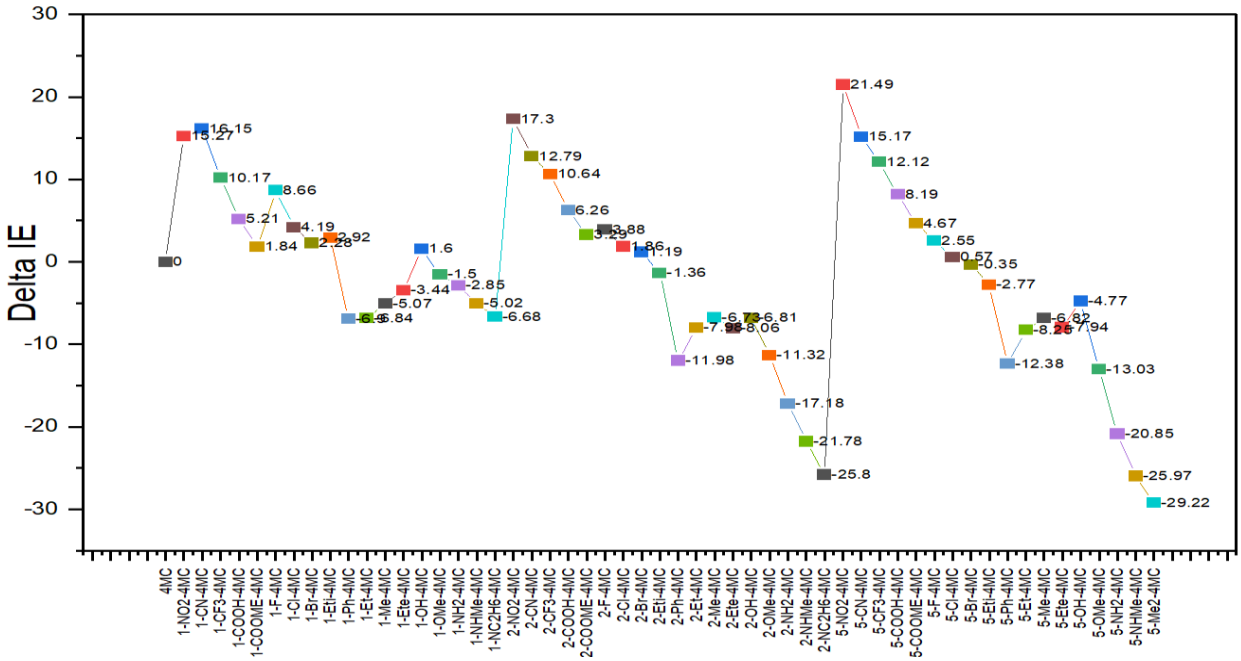


Figure 3. *Calculated ΔIE of **4MC** and the derivatives*

The findings shown in Table 2 and Figure 3 demonstrate that electron-withdrawing groups, such as NO_2, CN, CF_3, COOH, and $COOCH_3$, predominantly result in an increase in the IE value of **4MC**. The molecule NO_2–**4MC** has a noticeable rise in IE at positions 1, 2, and 5, indicating the presence of a substituent. The findings indicate that the IE value at location 5–NO_2–**4MC** (207.8 kcal/mol) has a greater magnitude in comparison to the other two sites, with a difference of about 21.5 kcal/mol when compared to the IE of compound **4MC** (186.3 kcal/mol). Electron-withdrawing substituents, such as Eti, Ph, Ete, Me, Et, OH, OMe, NH_2, NHMe, and NMe_2 significantly decrease the IE value in comparison to compound **4MC**. When examining the molecule $NHMe_2$–**4MC** at locations 1, 2, and 5, it is evident that the most prominent observation is the reduction in IE value at position 5–$NHMe_2$–**4MC** (157.1 kcal/mol) compared to the other two sites. In contrast, the energy value is notably decreased to around 29.2 kcal/mol as compared to compound **4MC**.

The antioxidant capacity of 54 derivatives was investigated in the gas phase using the FHT and SETPT mechanisms, as determined by the computed findings. According to the computed BDE parameters, it was seen that the IE value exceeded the BDE value by about 2.5-fold. The antioxidant activity of the derivatives in nonpolar media can be defined by the FHT mechanism and 5–NH2–**4MC** may be the most potent radical scavenger following the FHT reaction.

3.2. Docking analysis

3.2.1. Analysis of "drug-likeness" properties

Pharmaceutical companies have utilized various screening models to differentiate between drug-like and non-drug molecules in order to reduce drug research and development costs and minimize clinical trial failures. For example, the Lipinski model [9], the Ghose model [10], the Egan model [11], and the Veber model [12]. With great frequency and success, drug designers have employed these techniques.

In this section of the research, we conducted a screening process to identify candidate compounds suitable for oral medication development. These compounds were required to meet the requirements outlined in all the models presented in Table 3. If all the aforementioned requirements are met, substances that have undergone screening are likely to satisfy the criteria for being utilized as oral medications.

Table 3. *Models and criteria for evaluating "drug-likeness" characteristics*

Model	Screening Criteria
GI absorption	High
Lipinski	Number of violations = 0
Ghose	Number of violations = 0
Veber	Number of violations = 0
Egan	Number of violations = 0
PAINS	Number of violations = 0

The outcomes of the drug-likeness screening, as presented in Tables S1 and S2 of the Supplementary Information (SI), indicate that out of the 54 compounds examined, only three meet all the specified requirements and exhibit promise for drug development. These three compounds consist of derivatives of the phenyl substituent at positions 1, 2, and 5 of **4MC**). Hence, in the subsequent phase of the ADMET investigation, our primary emphasis will be on the examination of the pharmacokinetic characteristics shown by these molecules.

3.2.2. Analysis of ADMET parameters

The findings of the ADMET analysis pertaining to the absorption, distribution, metabolism, excretion, and toxicity of three derivatives of the Ph substituent at positions 1, 2, and 5 of **4MC** are presented in Table 4.

Table 4. *ADMET results of three derivatives of 4MC.*

Substituents	Ph (C_6H_5)		
	position 1	position 2	position 5
Absorption			
Caco2 permeability (log Papp in 10^{-6} cm/s)	1.59	1.54	1.42
Intestinal absorption (human) (%)	93.1	91.1	91.2
Distribution			
VDss (human) (log L/kg)	0.72	0.15	0.12
Fraction unbound (human) (Fu)	0.40	0.32	0.32
BBB permeability (log BB)	0.37	0.23	0.27
CNS permeability (log PS)	-1.63	-0.94	-1.12
Metabolism			
CYP2D6 substrate (Yes/No)	No	No	No
CYP3A4 substrate (Yes/No)	No	No	No
CYP2D6 inhibitor (Yes/No)	No	No	No
CYP3A4 inhibitor (Yes/No)	No	No	No
Excretion			
Total Clearance (log ml/min/kg)	0.76	0.88	0.95
Renal OCT2 substrate (Yes/No)	No	No	No
Toxicity			
AMES toxicity (Yes/No)	Yes	Yes	No
hERG I inhibitor (Yes/No)	No	No	No
hERG II inhibito (Yes/No)	No	No	No
Hepatotoxicity (Yes/No)	No	No	No

The absorption capacity of the derivatives was assessed by analyzing the parameters of $CaCO_2$ membrane permeability and intestinal absorption. The assessment of pharmaceutical product permeability often relies on the widely accepted criterion of $CaCO_2$ membrane permeability. A result greater than 0.9 (log Papp in 10^{-6} cm/s) is generally regarded as indicative of high permeability [13]. It was found that all three derivatives are highly permeable through the $CaCO_2$ membrane, with respective values of 1.59, 1.54, and 1.41. Moreover, all derivatives were extremely well absorbed in the human intestine, with 2-Ph-**4MC** exhibiting the lowest absorption rate of 91.1% and 1-Ph-**4MC** exhibiting the highest absorption rate of 93.1 percent. As for distribution,

compounds are thought to be well distributed to tissues if log VDgs > 0.45 and weakly distributed if log VDgs < -0.15 [13]. With a log VDgs value of 0.72, the 1-Ph-**4MC** derivative yields very favorable distributional outcomes. In addition, when evaluating the CNS safety of a drug, two permeability parameters across the blood-brain barrier (log BB) and CNS (log PS) are essential. If the values of log BB and log PS are < -1 and < -3, respectively, the substances are permeable through the blood-brain barrier and central nervous system, and vice versa, they are impermeable or less permeable.

As shown in Table 4, all potential drugs are not substrates of CYP3A4 and CYP2D6, do not inhibit CYP3A4 and CYP2D6, and are thus not metabolized in the liver. Concerning clearance, not all substances are substrates of OCT2 (organic cation transporter 2), which plays an important role in the renal elimination of ionized forms of the drug and endogenous compounds. As the first stage in elimination, the kidney extracts substances from the blood into tubular renal cells. Clearance values for the 1-Ph-**4MC**, 2-Ph-**4MC**, and 5-Ph-**4MC** derivatives were 0.76, 0.88, and 0.95 ml/min/kg, respectively. In terms of toxicity, the 5-Ph-**4MC** derivative is unlikely to induce cardiotoxicity (hERG inhibition), cancer risk (AMES toxicity), hepatotoxicity, or skin irritation.

On the basis of the ADMET analysis of three derivatives of **4MC**, 5-Ph-**4MC** meets the priority criteria of non-toxicity and pharmacokinetic properties and can be developed as a therapeutic. In the subsequent docking investigation, therefore, only the 5-Ph-**4MC** derivative was employed for interaction analysis with the target enzymes.

3.2.3. Molecular docking analysis

Table 5. *Docking score of 5-Ph-4MC and control drugs with target enzymes*

Enzymes	DS (kcal/mol)		
	5-Ph-4MC	Control (neutral)	Control (ionic)
MP (1DNU)	–4.29	–5.21	
LO (1N8Q)	–6.08	–5.03	
CP450 (1OG5)	–5.05	–4.60	–4.44
NO (2CDU)	–5.62	–6.54	
XO (3NRZ)	–4.24	–6.93	–8.62

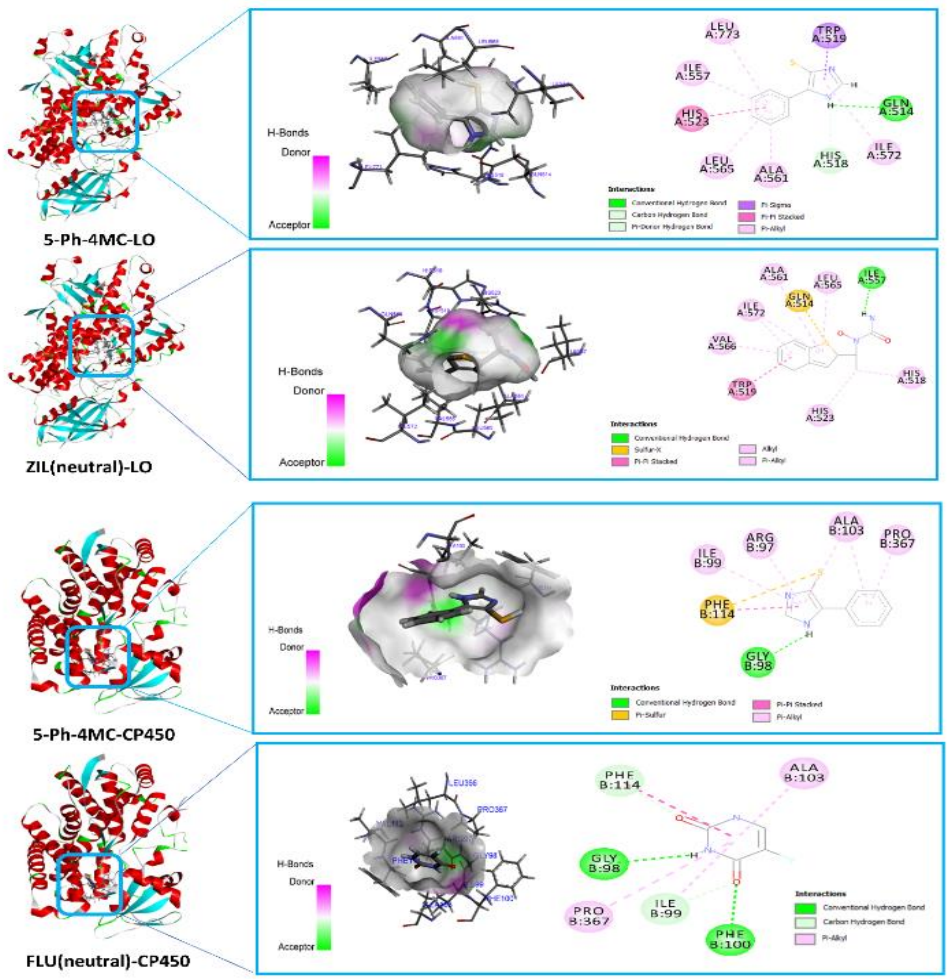


Figure 4*. Interactions between 5-Ph-**4MC** and control drugs with target enzymes*

In docking simulations, the docking score (which corresponds to binding affinity) has been shown to be an important metric for comparing the inhibitory activities of pharmaceuticals on target proteins. The binding affinity between the ligand and the target protein (Table 5) increases as the inhibitor docking score (DS) decreases.

According to the docking study, the 5-Ph-**4MC** derivative has a potent inhibitory effect on both LO and CP450 enzymes, with docking scores (DS) of -6.08 and -5.05 kcal/mol, respectively. These values are greater than the values for the ZIL (DS = -5.03 kcal/mol) and FLU (DS = -4.60 kcal/mol).

Figure 4 illustrates the interactions between ligands and target enzymes at their binding sites. In the case of the LO enzyme, the Zil ligand (control drug) forms a hydrogen bond with amino acid Ile557, a pi-pi interaction with amino acid Trp519, pi-alkyl interactions with amino acids Ala561, Leu565, Val566, and Ile572, and alkyl interactions with amino acids His518 and His523. In a similar manner, 5-Ph-**4MC** derivatives form complexes with LO via hydrogen bonding and pi-sigma, pi-pi, and pi-alkyl interactions with the amino acids Gln514, Trp519, His523, Ile557, Ala561, Leu565, Ile572, Leu773, and His518.

Comparing the interactions of the 5-Ph-**4MC** derivative with FLU for CP450 complexes reveals similarities with FLU. Their association with essential amino acids such as Gly98, Ile99, Ala103, Phe114, and Pro367 in the CP450 active region demonstrates this. According to the analysis of docking data, the 5-Ph-**4MC** derivative bonds well to the LO and CP450 enzymes, indicating that this compound can function as an inhibitor of these enzymes.

4. Conclusion

This work employed computer simulations to evaluate the antioxidant activity of 54 monoderivatives of **4MC**. The investigation revealed that the inclusion of electron-withdrawing groups, such as NO_2, CN, CF_3, COOH, and $COOCH_3$, leads to an augmentation in the BDE and IE values of **4MC**. Conversely, the presence of electron-donating groups, such as Eti, Ph, Ete, Me, Et, OH, OMe, NH_2, NHMe, and NMe_2, results in a reduction of these aforementioned values. The compound 5-NH2-**4MC** exhibits the lowest BDE value, measuring 66.6 kcal/mol. Conversely, 5-NHMe2-**4MC** demonstrates the IE value, which is recorded as 157.1 kcal/mol. The examination of docking data has demonstrated that 5-Ph-**4MC** exhibits robust interactions with the LO and CP450 enzymes, suggesting its potential as an inhibitor for these particular enzymes.

Acknowledgments: This research is funded by Funds for Science and Technology Development of the University of Danang under project number B2021-DN01-01 (N.M.T).

REFERENCES

[1] B. M. Shapiro and P. B. Hopkins, "Ovothiols: biological and chemical perspectives", *Adv Enzymol Relat Areas Mol Biol*, vol. 64, pp. 291-316, 1991. https://doi.org/10.1002/9780470123102.ch6.

[2] E. Turner, L. J. Hager, and B. M. Shapiro, "Ovothiol replaces glutathione peroxidase as a hydrogen peroxide scavenger in sea urchin eggs", *Science*, vol. 242, no. 4880, pp. 939-41, 1988. https://doi.org/10.1126/science.3187533.

[3] K. H. Weaver and D. L. Rabenstein, "Thiol/Disulfide Exchange Reactions of Ovothiol A with Glutathione", *The Journal of Organic Chemistry*, vol. 60, no. 6, pp. 1904-1907, 2002. https://doi.org/10.1021/jo00111a065.

[4] A. Bindoli, J. M. Fukuto, and H. J. Forman, "Thiol chemistry in peroxidase catalysis and redox signaling", *Antioxid Redox Signal*, vol. 10, no. 9, pp. 1549-64, 2008. https://doi.org/10.1089/ars.2008.2063.

[5] V. Zoete, H. Vezin, F. Bailly, G. Vergoten, J. P. Catteau, and J. L. Bernier, "4-Mercaptoimidazoles derived from the naturally occurring antioxidant ovothiols 2. Computational and experimental approach of the radical scavenging mechanism", *Free Radic Res*, vol. 32, no. 6, pp. 525-33, 2000. https://doi.org/10.1080/10715760000300531.

[6] F. Bailly, V. Zoete, J. Vamecq, J. P. Catteau, and J. L. Bernier, "Antioxidant actions of ovothiol-derived 4-mercaptoimidazoles: glutathione peroxidase activity and protection against peroxynitrite-induced damage", *FEBS Lett*, vol. 486, no. 1, pp. 19-22, 2000. https://doi.org/10.1016/s0014-5793(00)02234-1.

[7] Q. V. Vo, D. T. N. Hang, N. T. Hoa, P. C. Nam, T. Q. Duong, and A. Mechler, "The radical scavenging activity of 4-mercaptoimidazole: theoretical insights into the mechanism, kinetics and solvent effects", *New Journal of Chemistry*, vol. 47, no. 21, pp. 10381-10390, 2023. https://doi.org/10.1039/d3nj01800h.

[8] A. Daina, O. Michielin, and V. Zoete, "SwissADME: a free web tool to evaluate pharmacokinetics, drug-likeness and medicinal chemistry friendliness of small molecules", *Sci Rep*, vol. 7, p. 42717, 2017. https://doi.org/10.1038/srep42717.

[9] C. A. Lipinski, "Drug-like properties and the causes of poor solubility and poor permeability", *J Pharmacol Toxicol Methods*, vol. 44, no. 1, pp. 235-49, 2000. https://doi.org/10.1016/s1056-8719(00)00107-6.

[10] A. K. Ghose, V. N. Viswanadhan, and J. J. Wendoloski, "A knowledge-based approach in designing combinatorial or medicinal chemistry libraries for drug discovery. 1. A qualitative and quantitative characterization of known drug databases", *J Comb Chem*, vol. 1, no. 1, pp. 55-68, 1999. https://doi.org/10.1021/cc9800071.

[11] W. J. Egan, K. M. Merz, Jr., and J. J. Baldwin, "Prediction of drug absorption using multivariate statistics", *J Med Chem*, vol. 43, no. 21, pp. 3867-77, 2000. https://doi.org/10.1021/jm000292e.

[12] D. F. Veber, S. R. Johnson, H. Y. Cheng, B. R. Smith, K. W. Ward, and K. D. Kopple, "Molecular properties that influence the oral bioavailability of drug candidates", *J Med Chem*, vol. 45, no. 12, pp. 2615-23, 2002. https://doi.org/10.1021/jm020017n.

[13] D. E. Pires, T. L. Blundell, and D. B. Ascher, "pkCSM: Predicting Small-Molecule Pharmacokinetic and Toxicity Properties Using Graph-Based Signatures", *J Med Chem*, vol. 58, no. 9, pp. 4066-72, 2015. https://doi.org/10.1021/acs.jmedchem.5b00104.

[14] MOE, "The Molecular Operating Environment," ed. software available from Chemical Computing Group Inc., 1010 Sherbrooke Street West, Suite 910, Montreal, Canada H3A 2R7.

[15] D. S. Biovia, "*Discovery studio modeling environ*ment". ed. Dassault Systemes: San Diego, 2015.

[16] A. Galano and J. R. Alvarez-Idaboy, "A computational methodology for accurate predictions of rate constants in solution: application to the assessment of primary antioxidant activity", *J Comput Chem*, vol. 34, no. 28, pp. 2430-45, 2013. https://doi.org/10.1002/jcc.23409.

[17] Y. Zhao, N. E. Schultz, and D. G. Truhlar, "Design of Density Functionals by Combining the Method of Constraint Satisfaction with Parametrization for Thermochemistry, Thermochemical Kinetics, and Noncovalent Interactions", *J Chem Theory Comput*, vol. 2, no. 2, pp. 364-82, 2006. https://doi.org/10.1021/ct0502763.

[18] A. Galano and J. R. Alvarez-Idaboy, "Kinetics of radical-molecule reactions in aqueous solution: a benchmark study of the performance of density functional methods", *J Comput Chem*, vol. 35, no. 28, pp. 2019-26, 2014. https://doi.org/10.1002/jcc.23715.

[19] Y. Zhao and D. G. Truhlar, "The M06 suite of density functionals for main group thermochemistry, thermochemical kinetics, noncovalent interactions, excited states, and transition elements: two new functionals and systematic testing of four M06-class functionals and 12 other functionals", *Theoretical Chemistry Accounts*, vol. 120, no. 1-3, pp. 215-241, 2007. https://doi.org/10.1007/s00214-007-0310-x.

[20] M. J. Frisch *et al.*, ed, *Gaussian 16, Revision A.03*, Gaussian, Inc., Wallingford CT, 2016.

SUPPORTING INFORMATION (SI)

Table S1. *Screening "drug-likeness" properties of 4-mercaptoimidazole derivatives at position 1*

Substituents	GI absorption	Lipinski #violations	Ghose #violations	Veber #violations	Egan #violations	PAINS #alerts
Br	High	0	2	0	0	0
CF_3	High	0	2	0	0	0
Cl	High	0	3	0	0	0
CN	High	0	3	0	0	0
COOH	High	0	3	0	0	0
$COOCH_3$	High	0	3	0	0	0
Ete (C_2H_5)	High	0	3	0	0	0
Et (C_2H_3)	High	0	3	0	0	0
Eti (C_2H)	High	0	3	0	0	0
F	High	0	3	0	0	0
Me (CH_3)	High	0	3	0	0	0
NMe_2	High	0	3	0	0	0
NH_2	High	0	3	0	0	0
NHMe	High	0	3	0	0	0
NO_2	High	0	3	0	0	0
OH	High	0	3	0	0	0
OMe	High	0	3	0	0	0
Ph (C_6H_5)	High	0	0	0	0	0

Table S2. *Screening "drug-likeness" properties of 4-mercaptoimidazole derivatives at position 2*

Substituents	GI absorption	Lipinski #violations	Ghose #violations	Veber #violations	Egan #violations	PAINS #alerts
Br	High	0	2	0	0	0
CF_3	High	0	2	0	0	0
Cl	High	0	3	0	0	0
CN	High	0	3	0	0	0
COOH	High	0	3	0	0	0
$COOCH_3$	High	0	3	0	0	0
Ete (C_2H_5)	High	0	3	0	0	0
Et (C_2H_3)	High	0	3	0	0	0
Eti (C_2H)	High	0	3	0	0	0
F	High	0	3	0	0	0
Me (CH_3)	High	0	3	0	0	0
NMe_2	High	0	2	0	0	0
NH_2	High	0	3	0	0	0
NHMe	High	0	3	0	0	0
NO_2	High	0	3	0	0	0
OH	High	0	3	0	0	0
OMe	High	0	3	0	0	0
Ph (C_6H_5)	High	0	0	0	0	0

Table S3. *Screening "drug-likeness" properties of 4-mercaptoimidazole derivatives at position 5*

Substituents	GI absorption	Lipinski #violations	Ghose #violations	Veber #violations	Egan #violations	PAINS #alerts
Br	High	0	2	0	0	0
CF_3	High	0	2	0	0	0
Cl	High	0	3	0	0	0
CN	High	0	3	0	0	0
COOH	High	0	3	0	0	0
$COOCH_3$	High	0	3	0	0	0
Ete (C_2H_5)	High	0	3	0	0	0
Et (C_2H_3)	High	0	3	0	0	0
Eti (C_2H)	High	0	3	0	0	0
F	High	0	3	0	0	0
Me (CH_3)	High	0	3	0	0	0
NMe_2	High	0	2	0	0	0
NH_2	High	0	3	0	0	0
NHMe	High	0	3	0	0	0
NO_2	High	0	3	0	0	0
OH	High	0	3	0	0	0
OMe	High	0	3	0	0	0
Ph (C_6H_5)	High	0	0	0	0	0