

IDENTIFICATION OF PHYSICOCHEMICAL PROPERTIES, PHARMACOKINETICS AND TOXICITY OF ACTIVE COMPOUNDS OF *Kaempferia galanga* RHIZOME

Laili Nailul Muna¹

¹*Chemistry Education Study Programe, UIN Sunan Kalijaga
Jl. Laksda Adisucipto, Papringan, Caturtunggal, Kec. Depok, Kabupaten Sleman, Daerah
Istimewa Yogyakarta 55281*

**Email Corresponding: lailinailulmuna@gmail.com*

Submitted: January 30, 2024 Revised: March 11, 2024 Accepted: March 17, 2024

ABSTRACT

The Kencur plant has many benefits, is economically valuable, and is widely cultivated by the local community. The development of kencur compounds has not been carried out optimally because of the relatively high cost of identifying the content of each compound. This study aimed to determine the physicochemical properties of kencur compounds and their pharmacokinetics, as well as the toxicity of the herb kencur rhizome in silico. The method used in this study uses a Marvin sketch to draw the structure of the compound, which was then analyzed using Lipinski to determine its physico-chemical properties. Pharmacokinetic and toxicity analyses were performed using the pKCSM software. The results showed that all compounds followed Lipinski's rules; therefore, predictably, they could be developed into drug molecules from these compounds. In addition, all compounds had fairly high Water Solubility (log S). All HIA compounds were >90%, so they were well absorbed in the intestine and showed good permeability to CaCO-2 with a log Papp value of > 0.90. All the above compounds have low permeability to the skin due to the log Kp value > -2.5, so they are not suitable for transdermal preparations.

Keywords: Physicochemistry, Pharmacokinetics, Toxicity, Kencur, In silico

INTRODUCTION

Indonesia is a country with abundant natural resources, including herbal plants that have health maintenance activities (Tarigan et al., 2017). One of the herbal plants with pharmacological activity is the kencur rhizome, which is also widely used by the community in cooking spices (Hakim, 2015). The kencur plant has many benefits, is fairly economical, and is widely cultivated by the local community (Andriyono, 2019). The use of natural ingredients in traditional medicine has many advantages, such as minimizing the risk of side effects of synthetic drugs (Muna, 2022). The plant part used is the rhizome, which contains several compounds including ethyl paramethoxy cinnamate, ethyl cinnamate, borneol, kampheol, sineol, anisic acid, paraeumarin, methyl canyl acid, cinnamic acid, penta decaan (Nugraha et al., 2012). The kencur plant has many pharmacological activities, including analgetic, anti-inflammatory, antimicrobial, antiallergic, antioxidant, and wound healing abilities (Andriyono, 2019). The results of phytochemical screening of secondary metabolites contained in kencur rhizomes include flavonoids, steroids, tannins, sesquiterpenes, and monoterpenes (Soniman et al., 2022).

Compound development has not been optimized because of the high cost of identifying the content of each compound. Thus, a computational method is needed

to identify the content of each compound with pharmacological activity to minimize the cost of developing new drug compounds (Suhud, 2015). Toxicological analysis methods provide an overview of safety decisions in compound development and apply ethical principles to reduce in vivo experiments using test animals and saving funds (Meles, 2010).

The use of computing to find new drug candidates is a trend in the development of chemistry because it is faster and more effective and can determine the physicochemical properties, pharmacokinetics, and toxicity of the active compound content of new drug candidates. After collecting all test data, new compounds can be developed from natural materials to be used as candidates for new drug development (Abdullah et al., 2021). It is hoped that before synthesizing and testing in vitro and in vivo, the targets of new compounds are more specific. The purpose of this research was to develop medicinal compounds by determining their physicochemical properties, pharmacokinetics, and toxicity in kencur rhizome herbal plants.

RESEARCH METHODS

Equipment and Materials

Pure isolates of kencur rhizomes were used in this study, and the compound structure was drawn using Chem Draw or Marvin sketch software.

The tools used in this research are CPU Intel® Core (TM) i5-2450 dan i7-8565U CPU @2.50 GHz, Windows 10 64-Bit Operation System. The software used included Chem Draw or Marvin Sketch to illustrate the structure, PreADMET to determine the pharmacokinetics of the compound, and toxicity of the test compound.

Research Procedure

The structure of the chemical compound was drawn using the Marvin Sketh software, checked for structural suitability and compatibility, and then saved in the .sdf file. The results of the test compound were checked with <https://pubchem.ncbi.nlm.nih.gov/> to find out the IUPAC name.

The file storage format, namely.sdf, was then checked using ChemBioDraw (ChemiCAL Structure Drawing Software) to determine the chemical physical properties of the compound, consisting of Didik Point (K), Melting Point (K), Critical Temperature (K), Critical Press (Bar), Gibbs Energy (kJ/mol), Log P, and Molar Refractivity (MR). Furthermore, analysis was carried out using Lipinski and pKCSM software to determine the pharmacokinetics and toxicity of the test compounds.

RESULT AND DISCUSSION

Physico-chemical characteristics

Table I. Physicochemical Properties of Kencur Rhizome Compounds

	Boiling Point (K)	Melting Point (K)	Critical Temperature (K)	Critical Pressure (Bar)	Gibbs Energy (kJ/mol)	Molar Refractivity (cm ³ /mol)	Molekul Weight (g/mol)	Log P	Number H-bond Acceptors	Number H-bond Donors
Borneol	529,4 7	334,4 6	666,5 9	31,2	-20,5	51,33	154.25 3	2.193 5	1	1

Cinnamate Acid	581,87	372,63	786,49	38,6	-126,38	36,99	148,16	1,7844	2	1
Ethyl Cinnamate	540,34	276,9	742,96	28,1	-76	47,29	176,215	2,2629	2	0

Table I shows the physico-chemical properties of the active compounds contained in galangal rhizomes that can affect adsorption, distribution, metabolism, and excretion (Istnaeny et al., 2013). This is influenced by the hydrophobicity, molecular weight, and structural flexibility of a compound (Abdullah et al., 2021). As shown in **Table I**, the compound with the highest boiling and melting points is cinnamic acid with the chemical formula $C_9H_8O_2$ or $C_6H_5CHCHCOOH$ because it is non-polar and slightly soluble in water (Kiswandono et al., 2016). The melting point is an important factor for determining whether a compound will melt at a certain temperature during the compound preparation process. In addition, its melting point determines its solubility in water. The melting point of a compound increases with increasing molecular weight and boiling point of the compound, followed by its molecular size (Putra et al., 2020), and the melting point of crystalline compounds is determined by two factors: molecular symmetry and intermolecular interactions (Aitipamula et al., 2014). Thus, the structure of cinnamic acid was more symmetrical than that of ethyl semimat and borneol. **Table I** shows the high critical temperature of the cinnamic acid compound because the critical temperature shows the temperature of the compound which causes it not to turn into a liquid even though pressure is applied (Abdullah et al., 2021). Gibbs energy predicts whether a compound can run spontaneously. A compound can run spontaneously if it has a price of $\Delta G < 0$; if $\Delta G > 0$, it cannot react spontaneously. In kencur compounds (borneol, cinnamic acid, and ethyl cinnamate) with $\Delta G < 0$, the reaction can run spontaneously. Molar refractivity (molar refractivity) states the size of the polarization of compounds with units of cm^3/mol influenced by several factors, namely, pressure, temperature, and refractive index. Limitation of molar refractivity according to Lipinski's rule in the range of 40-130 Å so that the compound has good steric properties and easily interacts with the receptor (Annisa Fitriyani Suryana et al., 2022). Kencur compounds (cinnamic acid, ethyl cinnamate, and borneol) have a molar refractivity value in the range, so they can interact well with the receptor (Alfathin et al., 2021). Log P expresses the lipophilicity of a compound, according to Lipinski's rule of limitation $\log p < 5$. All compounds have $\log p$ values < 5 ; therefore, it can be presumed that the compounds are easily absorbed by the body (Kilo et al., 2019). The number of hydrogen bond donors and the number of hydrogen acceptors in the compound follow Lipinski's rule, namely hydrogen bond donors of no more than 5 and acceptor bond donors of no more than 10, so that the compound has conformational stability with the target protein (Annisa Fitriyani Suryana et al., 2022).

Pharmacokinetic Analysis (Absorption, Distribution, Metabolism and Excretion)

Table II. Absorption Profile Analysis Results of Kencur Rhizome Compounds

	Water Solubility (log S)	CaCO ₂ permeability (log Papp in 10 ⁻⁶ cm/s)	Human Intestinal absorption (%)	Skin permeability (log kP)
Borneol	-2,432	1.494	94.357	-2.217
Cinnamate Acid	-1.963	1.723	100	-2.474
Ethyl Cinnamate	-2.437	1.438	97.034	-2.088

Absorption profile analysis can be predicted by several parameters, including Water Solubility (log S), CaCO₂ permeability (log Papp in 10⁻⁶ cm/s), Human Intestinal absorption (%), Skin permeability (log Kp) (Abdullah et al., 2021). The water solubility analysis showed the ability of the compound to dissolve in water at 25°C. Compounds that have high solubility in lipids are absorbed less well than water-soluble compounds, especially if the compound is enteral. Compounds with water solubility values <-6 have low solubility (Abdullah et al., 2021). All compounds contained in kencur (borneol, cinnamic acid, and ethyl cinnamate) had good solubility because they had log S values > -6. CaCO₂ cells can be used to describe the ability of a drug compound to be absorbed into the intestinal mucosa when consumed orally (Hartanti et al., 2022). CaCO₂ cells form a monolayer of epithelial cells that contain physical and biochemical barriers that play a role in the passage of molecules and ions. A compound has good CaCO₂ permeability if it has a predicted value of >0.9 (Apriali et al., 2022). All the above compounds had good CaCO₂ permeability when used orally. The Human Intestinal Absorption (HIA) parameter describes compounds that can be well absorbed in the intestine when used orally. A compound has a poor absorption value if its HIA prediction value is <30% (Abdullah et al., 2021). All of the above compounds had good HIA values because HIA > 90%. Skin permeability is a parameter that describes a compound in its transdermal dosage form. A compound has a relatively small permeability to the skin if it has a logKp > -2.5. All the above compounds have low skin permeability because the log Kp value is > -2.5, which makes them less suitable for transdermal preparations (Gholam & Artika, 2023).

Table III. Distribution Profile Analysis Results of Kencur Rhizome Compounds

	VDss (log L/kg)	BBB (log BB)	CNS permeability (log PS)
Borneol	0.339	0.646	-2.351
Cinnamate Acid	-1.704	0.416	-1.848
Ethyl Cinnamate	0.071	0.269	-1.901

Distribution profile analysis can be performed using several parameters, including VDss (log L/kg), BBB (log BB), and CNS permeability (log PS) (Putra et al., 2020). The volume of distribution is a parameter that describes the total drug dose that circulates at a concentration similar to that in blood plasma. The higher the VDss, the higher is the amount of drug distributed through the blood plasma network, which is affected by renal failure and dehydration. The VDss value parameter is considered low if below 0.71 L/kg (log VDss < -0.15) and high if above 2.81 L/kg (log VDss > 0.45) (Abdullah et al., 2021). The kencur compound in the table above results in the lowest VDss of cinnamic acid compounds, namely -1,704. The human brain is protected from the influence of external compounds through the blood-brain barrier. This parameter is very important for reducing the side effects and toxicity caused by compounds from the outside (Sri et al., 2013). This also affects the efficacy of a compound if it has pharmacological effects on the brain. The blood brain barrier permeability parameter is expressed by logBB, which is the logarithmic ratio of the brain plasma drug concentration. A compound with a logBB > 0.3. can easily penetrate the blood-brain barrier, whereas compounds with a logBB < 1 are poorly distributed in the brain (Sri et al., 2013). All of these compounds have poor permeability distribution to the brain. The logPS (CNS Permeability) indicator can be used to measure cerebral blood permeability (Putra et al., 2020). This measurement method involves the insertion of compound IV into the carotid artery. A log PS value parameter > -2 means that the compound has the ability to penetrate the central nervous system (CNS), whereas a compound with log PS < 3 means that the compound cannot penetrate the CNS (Abdullah et al., 2021). All of the kencur compounds above have a logPS > -2 value, so they have the ability to penetrate the central nervous system.

Table IV. Metabolic Profile Analysis Results of Kencur Rhizome Compounds

	CYP2D6 substrate	CYP3A4 substrate	CYP2C1 9 inhibitor	CYP2C9 inhibitor	CYP2D6 inhibitor	CYP3A4 inhibitor
Borneol	No	No	No	No	No	No
Cinnamate Acid	No	No	No	No	No	No
Ethyl Cinnamate	No	No	No	No	No	No

Metabolic profile analysis can be predicted using the parameters listed in [Table IV](#). Cytochrome P450 is a metabolic enzyme present in the liver. These enzymes convert xenobiotic compounds into inactive metabolites ([Anindyaguna et al., 2022](#)). Cytochrome has an isoform model consisting of several substrate compounds CYP1A2, CYP2C19, CYP2C9, CYP2D6, and CYP3A4. As shown in table above, the kencur compounds (borneol, cinnamic acid, and ethyl cinnamate) contain substrates, so they cannot be metabolized by CYP450. In addition, the compounds listed above do not show being able to be inhibitors so they cannot suppress metabolic activity ([Abdullah et al., 2021](#)).

Table V. Analysis Results of The Exfoliation Profile of Kencur Rhizome Compounds

	Total Clearance (log mL/min/Kg)	Renal OCT2 substrate
Borneol	1.035	No
Cinnamate Acid	0.779	No
Ethyl Cinnamate	0.843	No

Toxicity profile analysis can be predicted by several parameters as shown in [Table V](#). Total clearance indicates the volume of plasma in which a compound dissolves through the renal portion of the glomerulus and is excreted in urine ([Mohammad et al., 2003](#)). The above compounds show that borneol has the highest total clearance; therefore, it is most easily excreted through urine.

Table VI. Toxicity Profile Analysis Results of Kencur Rhizome Compounds

	AM ES toxi city	Max. tolera ted dose (log mg/kg/d ay)	hER G I inhi bito r	hER T II inhi bito r	Oral Rat Acute Toxici ty (mol/k g)	Oral Rat Chronic Toxicity log mg/kg bw/day)	Hep atot oxici ty	Skin Sensiti sation
Borneol	No	0.723	No	No	1.767	1.877	No	Yes
Cinnamate Acid	No	0.664	No	No	2.096	2.441	No	No
Ethyl Cinnamate	No	1.073	No	No	1.977	2.014	No	Yes

The toxicity profile analysis can be predicted using several parameters, as shown in [Table VI](#). All compounds showed sensitive skin responses (except cinnamic acid), and were non-cancer-inducing and non-toxic in the liver.

CONCLUSION

All compounds followed Lipinski's rule, so they could be developed into drug molecules from these compounds. In addition, all compounds had high water solubility (log S). The HIA of all compounds was >90%; therefore, they were well absorbed in the intestine and the permeability to CaCO-2 was good, with a log Papp value > 0.90. All the above compounds

have low permeability to the skin because the log Kp value is >-2.5 , making it unsuitable for transdermal preparation. The lowest VDss value of cinnamic acid compound is -1.704. All compounds have poor permeability to the brain and can penetrate the central nervous system. All compounds at the excretion stage do not contain substrat, so they cannot be metabolized by CYP450. In addition, the compounds did not show the ability to act as inhibitors, so they could not suppress metabolic activity. In the excretion stage, borneol compounds have the highest total clearance; therefore, they are most easily excreted through urine. None of the compounds triggered cancer or caused toxicity in the liver.

REFERENCE

- Abdullah, S. S., Putra, P. P., Antasionasti, I., Rundengan, G., Suoth, E. J., Abdullah, R. P. I., & Abdullah, F. (2021). Analisis Sifat Fisikokimia, Farmakokinetik Dan Toksikologi Pada Pericarpium Pala (*Myristica Fragrans*) Secara Artificial Intelligence. *Chemistry Progress*, 14(2), 81. <https://doi.org/10.35799/cp.14.2.2021.37112>
- Aitipamula, S., Wong, A. B. H., Chow, P. S., & Tan, R. B. H. (2014). Pharmaceutical salts of haloperidol with some carboxylic acids and artificial sweeteners: Hydrate formation, polymorphism, and physicochemical properties. *Crystal Growth and Design*, 14(5), 2542–2556. <https://doi.org/10.1021/cg500245e>
- Alfathin, M. F., Herawati, D., & Faqih, T. M. (2021). Studi In Silico Senyawa Turunan Ftalosianin terhadap Reseptor InhA pada Mycobacterium tuberculosis sebagai Kandidat Senyawa Photosensitizer. *Prosiding Farmasi*, 284–291. <http://dx.doi.org/10.29313/v0i0.28107>
- Andriyono, R. I. (2019). Kaempferia galanga L. sebagai Anti-Inflamasi dan Analgetik. *Jurnal Kesehatan*, 10(3), 495. <https://doi.org/10.26630/jk.v10i3.1458>
- Anindyaguna, A., Mustofa, S., Anggraini, D. I., Oktarlina, R. Z., Biokimia, B., Fakultas, M., Lampung, U., Ilmu, B., Kedokteran, F., & Lampung, U. (2022). Drug-Induced Liver Injury Akibat Penyalahgunaan Parasetamol Drug-induced Liver Injury Due to Paracetamol Abuse. 12, 500–507.
- Annisa Fitriyani Suryana, Hilda Aprilia, & Taufik Muhammad Fakihi. (2022). Uji Aktivitas In Silico Senyawa Amritoside, Tinosporaside dan Turunannya sebagai Kandidat Senyawa. *Bandung Conference Series: Pharmacy*, 2(2), 1–10. <https://doi.org/10.29313/bcsp.v2i2.4369>
- Apriali, K. D., Triana, E., & Farhani, M. indah. (2022). Studi Penambatan Molekul Dan Prediksi Admet Senyawa Metabolit Sekunder Tanaman Kelor (*Moringa Oleifera* L.) Sebagai Inhibitor Bace1. *FITOFARMAKA: Jurnal Ilmiah Farmasi*, 12(1), 12–26.
- Gholam, G. M., & Artika, I. M. (2023). Potensi Terbentuk Interaksi Molekuler pada Fitokimia Alami sebagai Inhibitor Sap 2 *Candida albicans*: Pendekatan In silico. 11(2), 54–62.
- Hakim, L. (2015). *Rempah & Herba Kebun-Pekarangan Rumah Masyarakat* (Issue 164).
- Hartanti, I. R., Putri, A. A., Auliya AS, N. N., Triadenda, A. L., Laelasari, E., Suhandi, C., & Muchtaridi, M. (2022). Molecular Docking Senyawa Xanton, Benzofenon, Dan Triterpenoid Sebagai Antidiabetes Dari Ekstrak Tumbuhan Garcinia Cowa. *Jurnal Kimia*, 16(1), 72. <https://doi.org/10.24843/jchem.2022.v16.i01.p10>
- Istnaeny, H. M., Elvianto Dwi Daryon, & Muyassaroh. (2013). Minyak Kencur dari Rimpang Kencur dengan Variabel Jumlah Pelarut dan Waktu Maserasi. *Jurusan Teknik Kimia, Fakultas Teknologi Industri, Institut Teknologi Nasional (ITN) Malang*, 8(1), 1–7.
- Kilo, A. L., Aman, L., Sabihi, I., & Kilo, J. (2019). Studi potensi prazolin tersubstitusi 1-N dari tiiosemikarbazon sebagai agen antiamuba melalui uji in-silico. *Indonesian Journal of Chemistry*, 7(1), 9–16.
- Kiswandono, A. A., Iswanto, A. H., & Susilowati, A. (2016). Analisis Kandungan Asam Sinamat Dan Skrining Fitokimia Hiliferum) Dari Tapanuli Utara Analysis Of The Content Of Cinnamic Acid And Phytochemical Screening Sap Incense Type Of Bulu (*Styrax Benzoine* Var . Hiliferum) Of North Tapanuli. *Prosiding Seminar Nasional*

- Kimia-Lombok, Xxx*, 10–11.
- Meles, D. K. (2010). Peran Uji Praklinik Dalam Bidang Farmakologi. *Pusat Penerbitan Dan Percetakan Unair (AUP)*, 1–33.
https://simdos.unud.ac.id/uploads/file_penelitian_1_dir/767616f64cd58798f36164d0c9396ffb.pdf
- Mohammad, A., Tan, C. K., & Prayitno, A. (2003). Farmakokinetika Klinis. In *asi Klinis* (pp. 73–99).
- Muna, L. N. (2022). Aktivitas antioksidan ekstrak air daun kelor (*Moringa oleifera*) dengan metode DPPH serta analisis kualitatif kandungan metabolit sekunder. *Sasambo Journal of Pharmacy*, 3(2).
- Nugraha, S. A., Siadi, K., & Sudarmin. (2012). Uji Antimikroba Etil p-Metoksi Sinamat dari Rimpang Kencur Terhadap *Bacillus Subtilis*. *Indonesian Journal of Chemical Science*, 1(2), 147–151.
- Putra, P. P., Fauzana, A., & Lucida, H. (2020). Analisis Sifat Fisika-Kimia, Potensi Target dan Toksikologi Senyawa Isolat Murni dari Bahan Alam dengan Metode In Silico. *Indonesian Journal of Pharmaceutical Science and Technology Journal Homepage*, 7(3), 107–117. <http://jurnal.unpad.ac.id/ijpst/UNPAD>
- Soniman, M., Syaputra, D., & Kurniawa, A. (2022). Effectivity Of Kencur Combination Active Compounds *Kaempferia Galanga* Dan *Andropogon squarrosus* *Cylindrica* In Vitro Against Positive Gram Bacteria And Negative Gram Bacteria. *Journal of Aquatropica Asia*, 7(1), 19–33.
- Sri, A., Ulfa, Y., Gde, T., Mahadewa, B., Kedokteran, F., Udayana, U., & Pusat, S. S. (2013). Sawar Darah Otak. *Fakultas Kedokteran, Universitas Udayana*, 2(9), 1–21.
- Suhud, farida. (2015). Uji Aktivitas in-silico Senyawa baru 1-Benzil-3-benzoylurea induk dan tersubstitusi sebagai agen antiproliferatif. *Jurnal Farmasi Indonesia*, 7(4), 242–251.
- Tarigan, D. M., Alqamari, M., & Alridiwersah. (2017). *Budidaya Tanaman Obat & Rempah*.

