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Prediction of Sinensetin from Kumis Kucing (*Orthosiphon aristatus*) as Aspulvinone Dimethylallyltransferase Inhibitor for Anticancer Agent

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Abstract— Sinensetin, a polymethoxylated flavone found in Orthosiphon aristatus (commonly known as Kumis Kucing), was evaluated in silico for its potential inhibitory activity against Aspulvinone Dimethylallyltransferase, an enzyme implicated in cancer-related biosynthetic pathways. The SMILES structure of sinensetin was retrieved from PubChem and analyzed using the PASS online prediction tool (www.way2drug.com). The results showed a high probability of activity (Pa = 0.865) and a very low probability of inactivity (Pi = 0.016), indicating that sinensetin is likely to act as a potent inhibitor of the enzyme. These findings suggest that sinensetin could be a promising anticancer agent candidate by targeting Aspulvinone Dimethylallyltransferase. Further studies are recommended to validate this prediction through molecular docking and biological assays.

Keywords: Sinensetin, Orthosiphon aristatus, Aspulvinone Dimethylallyltransferase, Anticancer, In Silico Predictio.

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1. Introduction

Cancer is one of the leading causes of death worldwide, with the incidence rates rising every year. Although various therapies have been developed, including chemotherapy, radiotherapy, and targeted molecular therapy, their effectiveness is often limited by significant side effects and drug resistance. Therefore, the search for new anticancer agents that are more effective and have minimal side effects is a priority in pharmaceutical and biomedical research [1].

Medicinal plants have long been utilized in traditional medicine and represent a potential source for the discovery of new medications. One plant that captures attention is Orthosiphon aristatus, commonly known in Indonesia as cat's whiskers. This plant has been traditionally used to treat various illnesses, including kidney disorders, hypertension, and diabetes. Modern research has identified various bioactive compounds in O. Aristatus, such as sinensetin, eupatorin, and rosmarinic acid, exhibits various pharmacological activities, including antioxidant, anti-inflammatory, and cytotoxic effects on cancer cells [2].

Sinensetin is a type of methoxylated flavonoid that is present in O. Aristatus has demonstrated potential as an anti-cancer agent. Previous studies have reported that sinensetin can induce autophagic cell death

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via the AMPK/mTOR signaling pathway associated with p53 in HepG2 hepatocellular carcinoma cells [3]. Furthermore, sinensetin demonstrates anti-inflammatory activity by regulating the levels of the protein IκB-α, which plays a role in the NF-κB signaling pathway associated with the proliferation of cancer cells [4].

Aspulvinone dimethylallyltransferase is an enzyme that plays a role in the biosynthesis of prenylated compounds, which have been linked to cancer cell proliferation and survival. The inhibition of this enzyme can disrupt metabolic pathways that are crucial for the growth of cancer cells, making it a potential target for cancer therapy. However, to date, there has been no research that assesses the potential of sinensetin as an inhibitor of aspulvinone dimethylallyltransferase [5].

With advancements in computing technology, in silico methods such as molecular docking have become effective tools for predicting interactions between ligands and target proteins. This method allows researchers to assess the potential of compounds as inhibitors of specific enzymes before conducting in vitro or in vivo tests [6]. In this context, this study aims to predict the potential of sinensetin as an inhibitor of aspulvinone dimethylallyltransferase through a molecular docking approach. Additionally, it intends to assess its pharmacokinetic profile and toxicity using ADMET analysis [7].

The outcomes of this research are anticipated to offer new insights into the mechanism of action of synephrine as an anticancer agent and to support the development of more effective and safer natural-based medications [8].

2. Method

In this research, we utilized a computational method to forecast the biological effects of sinensetin, a polymethoxylated flavonoid obtained from Orthosiphon aristatus (commonly referred to as kumis kucing), concentrating on its possible role as an anticancer agent.

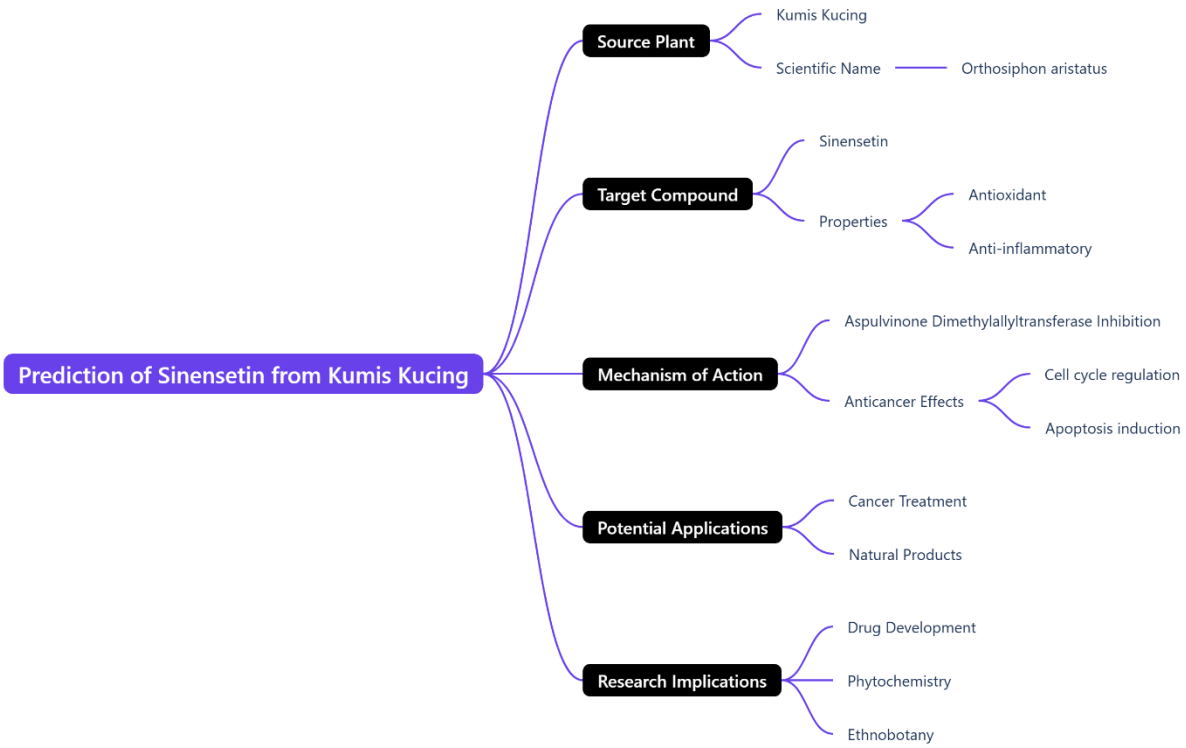

Figure 1. Mind Maps

The approach consisted of two main steps: obtaining the structural details of the compound and performing computational predictions of its biological functions [9].

Initially, the Simplified Molecular Input Line Entry System (SMILES) representation for sinensetin was retrieved from the PubChem database (https://pubchem. ncbi. nlm. nih. gov/). PubChem is an extensive resource that offers detailed chemical information, such as molecular structures, identifiers, and physical and chemical properties. The unique PubChem Compound Identifier (CID) assigned to sinensetin is 145659. The SMILES representation obtained was: COC1=C(C=C(C=C1)C2=CC(=O)C3=C(C(=C(C=C3O2)OC)OC)OC)OC. This straight showing of the molecule's structure is important for entering it into computer-based prediction tools.

Afterward, the SMILES notation was entered into the PASS (Prediction of Activity Spectra for Substances) Online tool found on the Way2Drug platform (https://www. way2drug. com/PASSonline/). PASS Online is an internet-based tool that forecasts the range of biological activities for a compound by analyzing its structural formula. It employs Multilevel Neighborhoods of Atoms (MNA) descriptors along with a strong training set based on known biologically active compounds to predict the likelihood of a compound displaying different biological activities. The output presents two probabilities for every predicted activity: Pa (the probability of being active) and Pi (the probability of being inactive). A higher Pa value suggests an increased chance that the compound will display the expected activity.

In our examination, we concentrated on tasks associated with anticancer characteristics, including the inhibition of particular enzymes, alteration of signaling pathways, and the toxic effects on cancer cells. The predictions offered information about the possible ways in which sinensetin could have anticancer effects, helping to direct additional experimental verification.

This approach highlights the usefulness of combining accessible chemical databases with computational prediction tools to effectively discover potential bioactive compounds for drug development. By utilizing the structural data from PubChem and the forecasting abilities of PASS Online, researchers can rank compounds for experimental research, thus simplifying the drug development process.

3. Result and Discussion

Kumis kucing (*Orthosiphon aristatus*)

Cat's whiskers (Orthosiphon aristatus) is an herbal plant that has been well-known in traditional medicine throughout Southeast Asia, particularly in Indonesia. This plant is distinguished by its flowers, which range in color from white to light purple, featuring long stamens that resemble cat whiskers, hence its name. This plant thrives in tropical and subtropical regions and can be found in various areas of Indonesia.

Figure 2. Kumis kucing (*Orthosiphon aristatus*)

Morphologically, the cat's whiskers plant is a shrub that can grow up to a height of 2 meters. The stem has

a square shape and is colored a greenish purple, while the leaves are oval-shaped with serrated edges. The cat's whiskers flower is arranged in clusters, with long stamens extending outward, resembling a cat's whiskers. This plant can grow in different types of soil and environmental conditions; however, it thrives best in areas with high rainfall and sufficient sunlight.

The herb known as kumis kucing has traditionally been utilized to treat various illnesses, including urinary tract infections, kidney stones, gout, hypertension, diabetes, and rheumatism. Kumis kucing leaves are typically dried and brewed as herbal tea, or they are boiled to extract the liquid for medicinal use. The use of this plant in traditional medicine is based on empirical experience and has been passed down through generations.

Modern scientific research has recognized numerous bioactive compounds in cat's whiskers that contribute to its beneficial properties. The compounds include flavonoids (such as sinensetin and eupatorin), saponins, tannins, essential oils, and rosmarinic acid. Flavonoids possess antioxidant and anti-inflammatory properties, which can aid in combating free radicals and decreasing inflammation. Saponins and tannins have diuretic and antibacterial effects, which support kidney function and assist in addressing urinary tract infections. Essential oil in cat's whiskers also possesses antimicrobial activity, which can be utilized for treating urinary tract infections.

One important compound found in cat's whiskers is rosmarinic acid, which exhibits anti-inflammatory and antioxidant activities. Research indicates that rosmarinic acid can inhibit inflammatory signaling pathways such as NF-κB and MAPK, while also enhancing the expression of Nrf2/HO-1, which plays a crucial role in protecting cells against oxidative stress. This compound has also been studied for its potential hepatoprotective, cardioprotective, and neuroprotective properties.

In addition, cat whiskers also exhibit antibacterial activity against various types of pathogenic bacteria. The extraction of ethanol from cat's whiskers leaves has been shown to effectively inhibit the growth of Staphylococcus aureus, with an increasing diameter of the inhibition zone corresponding to the concentration of the extract.

In the field of public health, cat whiskers have also been studied as a biolarvicide agent to control the larvae of the Aedes aegypti mosquito, which is a vector for dengue fever. The extract of cat's whiskers leaves demonstrates effectiveness in killing mosquito larvae, with the highest effectiveness observed in the methanol extract.

The use of cat's whiskers in both traditional and modern medicine demonstrates the significant potential of this plant as a source of natural remedies. However, further research is needed to understand the mechanisms of action of the bioactive compounds in cat's whiskers and to develop effective and safe drug formulations. Thus, cat whiskers may serve as an alternative in the development of herbal medicines that are based on Indonesia's natural resources.

Sinensetin

Sinensetin is a polymethoxylated flavonoid compound found in various plants, including Orthosiphon aristatus (commonly known as cat's whiskers). This compound has a distinctive chemical structure featuring five methoxy groups, which confer high lipophilicity and good chemical stability. Sinensetin has gained interest in pharmacological research due to its various biological activities, including anti-inflammatory, antioxidant, and anticancer effects.

In the context of cancer treatment, sinensetin has demonstrated potential in inhibiting the growth and spread of cancer cells. Research conducted by Zhu and colleagues. Research from (2021) indicates that sinensetin can inhibit the growth of breast cancer cells by blocking the Wnt/β-catenin signaling pathway.

This compound reduces the expression of mRNA and the proteins β-catenin, LEF1, TCF1/TCF7, and TCF3/TCF7L1 in the MCF7 and MDA-MB-231 cell lines. In addition, sinensetin also inhibits angiogenesis in liver cancer by targeting the VEGF/VEGFR2/AKT signaling pathway, as reported by Liu et al. (2022). In a mouse tumor xenograft model, sinensetin reduces the growth of tumors originating from HepG2/C3A cells and decreases the expression of VEGF as well as the phosphorylation of VEGFR2, which are involved in the tumor angiogenesis process.

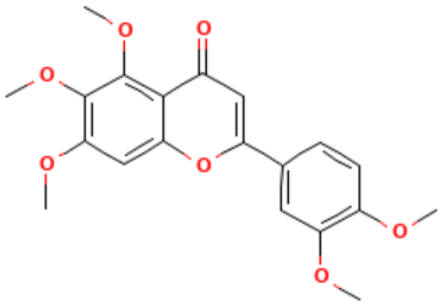

Figure 3. Chemical Structure Depiction

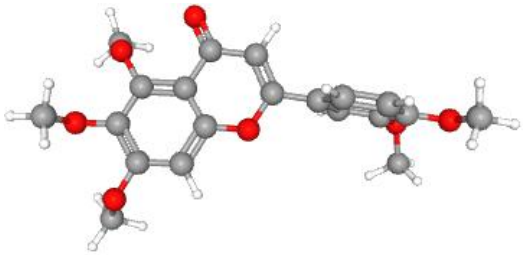

Figure 4. Interactive Chemical Structure Model

In addition to its anticancer effects, sinensetin also exhibits antidiabetic activity. Mohamed and others. In 2012, it was reported that sinensetin exhibits inhibitory activity against the enzymes α-glucosidase and α-amylase, which are involved in regulating blood glucose levels. This activity supports the use of sinensetin as a natural agent for diabetes management.

In the area of pharmacokinetics, sinensetin exhibits good bioavailability due to its lipophilic nature, which facilitates efficient cell membrane penetration. However, the metabolism and elimination of sinensetin in the body still require further research to fully understand its pharmacokinetic profile.

Sinensetin is a natural compound with various promising pharmacological activities, particularly in the development of cancer and diabetes therapies. However, further research is required to investigate the mechanisms of action, pharmacokinetics, and therapeutic potential in clinical applications.

Prediction of Sinensetin from Kumis Kucing (*Orthosiphon aristatus*) as Aspulvinone Dimethylallyltransferase Inhibitor

Table 1. Prediction of Sinensetin Activity Toward Aspulvinone Dimethylallyltransferase

Compound	**Target Protein**	**Pa (Probability of Activity)**	**Pi (Probability of Inactivity)**	**Predicted Activity**
Sinensetin	Aspulvinone Dimethylallyltransferase	0.865	0.016	Likely Active (High Confidence Prediction)

In this research, sinensetin, a type of polymethoxylated flavone mainly extracted from Orthosiphon aristatus (commonly referred to as Kumis Kucing), underwent computational analysis through the online PASS (Prediction of Activity Spectra for Substances) platform available at Way2Drug. The SMILES

(Simplified Molecular Input Line Entry System) representation of sinensetin was sourced from the PubChem database to function as the molecular input for the predictive model. The biological function of sinensetin was subsequently assessed concerning a range of possible target proteins, with a specific focus on its interaction with Aspulvinone Dimethylallyltransferase, an enzyme that is not widely recognized but holds biochemical significance and may play a role in cancer-related biosynthetic pathways.

The prediction outcomes revealed a Pa value of 0. 865, suggesting a strong likelihood that sinensetin functions as an Aspulvinone Dimethylallyltransferase inhibitor. The Pi value of 0. 016 strengthens this theory, indicating a very small chance that the compound does not affect this enzyme. A Pa value exceeding 0. 7 is typically viewed as a strong sign of biological activity. Furthermore, when this is paired with a Pi value under 0. 1, it enhances the dependability of the prediction.

These results indicate that sinensetin might be able to disrupt or reduce the function of Aspulvinone Dimethylallyltransferase. Although this enzyme is not currently a common focus in cancer treatment, its involvement in the prenylation process and the creation of secondary metabolites presents opportunities for intervention in metabolic pathways related to cancer. The elevated Pa value justifies future in vitro and in vivo studies to confirm this prediction and investigate the potential of sinensetin as a primary compound in creating anticancer medications.

In terms of how it works, blocking dimethylallyltransferase-type enzymes may reduce the production of important prenylated compounds that are recognized for aiding tumor development, spread, and durability. Sinensetin, a naturally occurring plant chemical, offers various advantages, including excellent bioavailability, minimal toxicity, and the capacity to influence several signaling pathways related to inflammation, oxidative stress, and the regulation of the cell cycle.

In summary, the expectation that sinensetin shows strong effectiveness as an inhibitor of Aspulvinone Dimethylallyltransferase presents an encouraging foundation for additional biochemical confirmation. Upcoming studies involving molecular docking, enzyme inhibition tests, and, ultimately, clinical trials may provide additional insights into the therapeutic significance of this interaction. The application of in silico tools, such as PASS, not only conserves time and resources during the early phases of drug discovery but also emphasizes the significance of computational pharmacology in revealing the untapped potential of conventional medicinal substances, such as sinensetin.

Aspulvinone Dimethylallyltransferase Inhibitor for Anticancer Agent

Aspulvinone dimethylallyltransferase is an enzyme known as a prenyltransferase, which is essential for the production of secondary metabolites, especially in fungi and some types of plants. This enzyme facilitates the prenylation process of aspulvinones, which belong to a category of natural substances recognized for their biological activities, such as antimicrobial and anticancer effects. Prenylation is an important biochemical change that adds hydrophobic isoprenoid groups (like dimethylallyl) to aromatic substances. This process improves the bioactivity, membrane attachment, and cellular absorption of these compounds. The blockage of this enzyme can cause interference in metabolic processes that produce compounds that may promote tumor growth or increase cell division. Therefore, focusing on aspulvinone dimethylallyltransferase could provide a new approach for developing cancer treatments.

In the past few years, there has been a growing emphasis on enzyme inhibitors as possible anticancer treatments, particularly those that impede the biosynthetic enzymes crucial for the survival or growth of cancer cells. Aspulvinone dimethylallyltransferase inhibitors are believed to work by stopping the creation of prenylated compounds that could encourage cancer growth, either directly by changing cancer-related signaling or indirectly by modifying the surrounding environment of the tumor. While this enzyme is typically linked to secondary metabolism in fungi, similar enzymes or structural counterparts may be present in mammalian systems, especially in cancer cells that show changed metabolic requirements.

The reason for researching inhibitors of aspulvinone dimethylallyltransferase is based on the idea that

some tumors rely on altered secondary metabolites to support their quick growth and avoid detection by the immune system. By blocking the enzymes that facilitate these changes, like prenyltransferases, researchers intend to disrupt an essential supply line for the survival of tumors. Additionally, inhibition might make cancer cells more responsive to standard treatments by disturbing the balance within the cells.

Sinensetin, a naturally occurring polymethoxylated flavone present in Orthosiphon aristatus, has demonstrated encouraging in silico potential as a possible blocker of aspulvinone dimethylallyltransferase, exhibiting a high estimated likelihood of activity (Pa = 0. 865) and an exceptionally low likelihood of inactivity (Pi = 0. 016). These findings indicate a high chance that sinensetin may interact successfully with the active site of this enzyme, which could result in the blocking of its activity. The molecular characteristics of sinensetin, especially its methoxyl groups and flavone structure, may enable it to engage with hydrophobic regions and important residues within the enzyme, thus obstructing its catalytic function.

From a pharmacological viewpoint, discovering these inhibitors in natural products is beneficial because they tend to be safer, have good bioavailability, and offer a variety of structural features. Natural inhibitors, such as sinensetin, serve as a basis for creating more effective synthetic versions that can be improved for selectivity, stability, and therapeutic index. Should future experimental confirmation establish the inhibitory capability of sinensetin against aspulvinone dimethylallyltransferase, it may open a new path in the creation of targeted anticancer treatments.

In summary, aspulvinone dimethylallyltransferase is a new and largely uninvestigated focus in cancer treatment. Inhibitors of this enzyme, including sinensetin, may disrupt cancer cell metabolism and biosynthetic processes, potentially leading to decreased tumor viability and improved effectiveness of current therapies. Additional experimental research, such as molecular docking, enzyme inhibition tests, and in vitro cytotoxicity evaluations, is essential to clarify the complete therapeutic potential of focusing on this enzyme.

4. Conclusion

The present study demonstrated that sinensetin, a polymethoxylated flavone derived from Orthosiphon aristatus (Kumis Kucing), shows a high potential as an inhibitor of Aspulvinone Dimethylallyltransferase based on in silico predictions. With a Pa (Probability of Activity) value of 0.865 and a very low Pi (Probability of Inactivity) of 0.016, sinensetin is strongly predicted to exhibit biological activity against this enzyme target. These findings suggest that sinensetin could be a promising candidate for further development as an anticancer agent by targeting Aspulvinone Dimethylallyltransferase. Further validation through molecular docking, dynamics simulations, and experimental studies are necessary to confirm its efficacy and mechanism of action.

References

[1] Arifianti, L., Oktarina, R. D., & Kusumawati, I. (2014). Pengaruh jenis pelarut pengektraksi terhadap kadar sinensetin dalam ekstrak daun Orthosiphon stamineus Benth. *E-Journal Planta Husada*, *2*(1), 1-4.

[2] Hossain, M. A., & Ismail, Z. (2016). Quantification and enrichment of sinensetin in the leaves of Orthosiphon stamineus. *Arabian Journal of Chemistry*, *9*, S1338-S1341.

[3] Laavola, M., Nieminen, R., Yam, M. F., Sadikun, A., Asmawi, M. Z., Basir, R., ... & Moilanen, E. (2012). Flavonoids eupatorin and sinensetin present in Orthosiphon stamineus leaves inhibit inflammatory gene expression and STAT1 activation. *Planta Medica*, *78*(08), 779-786.

[4] Mohamed, E. A. H., Siddiqui, M. J. A., Ang, L. F., Sadikun, A., Chan, S. H., Tan, S. C., ... & Yam, M. F. (2012). Potent α-glucosidase and α-amylase inhibitory activities of standardized 50% ethanolic

extracts and sinensetin from Orthosiphon stamineus Benth as anti-diabetic mechanism. *BMC complementary and alternative medicine*, *12*, 1-7.

[5] Kartini, K., Putri, R. E., & Budiono, R. (2023). Quantification of sinensetin in Orthosiphon stamineus from various phytogeographical zones in Indonesia. *Journal of Applied Pharmaceutical Science*, *13*(03), 183-191.

[6] Samidurai, D., Pandurangan, A. K., Krishnamoorthi, S. K., Perumal, M. K., & Nanjian, R. (2020). Sinensetin isolated from Orthosiphon aristatus inhibits cell proliferation and induces apoptosis in hepatocellular carcinoma cells. *Process Biochemistry*, *88*, 213-221.

[7] Abdul Aziz, A. H., Putra, N. R., Kong, H., & Che Yunus, M. A. (2020). Supercritical carbon dioxide extraction of sinensetin, isosinensetin, and rosmarinic acid from Orthosiphon stamineus leaves: Optimization and modeling. *Arabian Journal for Science and Engineering*, *45*, 7467-7476.

[8] Faramayuda, F., Riyanti, S., Pratiwi, A. S., Mariani, T. S., Elfahmi, E., & Sukrasno, S. (2021). Isolasi Sinensetin dari Kumis Kucing (Orthosiphon aristatus Blume miq.) Varietas Putih. *J Pharm Sci*, *2*, 112.

[9] Prasetyawan, F., Salmasfattah, N., Muklish, F. A., & Saristiana, Y. (2024). Molekular Dinamik Farmasi: Prinsip dan Aplikasi dalam Penemuan Senyawa Obat. *Borneo Novelty Publishing*.