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In Silico Exploration of *Orthosiphon stamineus* Compounds as Potential Angiotensin Receptor Blockers for Hypertension Therapy

Eksplorasi In Silico Senyawa Bioaktif *Orthosiphon stamineus* sebagai Kandidat Penghambat Reseptor Angiotensin dalam Terapi Hipertensi

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ABSTRACT

Orthosiphon stamineus has demonstrated antihypertensive potential, but the specific bioactive compounds involved remain unclear. This study aimed to evaluate selected phytochemicals from *O. stamineus* as angiotensin receptor blockers (ARBs) targeting protein 4ZUD using in-silico methods. Molecular docking was conducted to assess binding affinity, while ADMET analysis evaluated pharmacokinetics and toxicity. Salvianolic acid E showed the strongest binding affinity with a rerank score of −134.02 kcal/mol, surpassing olmesartan (−124.52 kcal/mol). Key interactions were observed with amino acid residues Arg167, Tyr92, and Asp281. ADMET predictions revealed that Salvianolic acid E has good aqueous solubility, moderate intestinal absorption (HIA 45.99%), and low membrane permeability (Caco-2 < 0.4). It does not inhibit major cytochrome P450 isoenzymes and is predicted to be non-hepatotoxic, suggesting favorable safety and metabolic profiles. These findings highlight Salvianolic acid E as a promising phytochemical candidate for antihypertensive drug development.

Kata kunci:
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Orthosiphon stamineus
Penghambat reseptor angiotensin
Salvianolic acid E

ABSTRAK

Orthosiphon stamineus telah menunjukkan potensi sebagai antihipertensi, namun senyawa bioaktif spesifik yang berperan masih belum diketahui secara pasti. Penelitian ini bertujuan mengevaluasi senyawa terpilih dari *O. stamineus* sebagai penghambat reseptor angiotensin (ARB) terhadap protein 4ZUD menggunakan metode in-silico. Molecular docking digunakan untuk menilai afinitas ikatan, sedangkan analisis ADMET mengevaluasi farmakokinetik dan toksisitas. Salvianolic acid E menunjukkan rerank score terendah (−134,02 kcal/mol), lebih kuat dibandingkan olmesartan (−124,52 kcal/mol), dengan interaksi kunci pada residu Arg167, Tyr92, dan Asp281. Prediksi ADMET menunjukkan kelarutan yang baik, absorpsi usus sedang (HIA 45,99%), permeabilitas membran rendah (Caco-2 < 0,4), tidak menghambat enzim CYP450 utama, dan tidak bersifat hepatotoksik. Profil ini menunjukkan Salvianolic acid E aman dan memiliki potensi sebagai kandidat fitofarmaka untuk terapi hipertensi.

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1. INTRODUCTION

According to the World Health Organization (WHO), hypertension is a serious condition that significantly increases the risk of morbidity and mortality due to heart disease, stroke, kidney failure, and other complications. In 2021, it was estimated that over 1.28 billion adults aged 30 to 79 years suffered from hypertension globally (WHO, 2021). Given the rising prevalence of hypertension, effective management of this condition has become a top health priority in Asia, particularly in Indonesia. In East Asian countries, the complications arising from hypertension-related cardiovascular diseases have reached alarming levels. Additionally, the relationship between hypertension and cardiovascular disease appears to be stronger in Asian populations compared to Western populations, largely due to higher salt sensitivity among Asians.

Hypertension is typically diagnosed when repeated measurements show systolic blood pressure at or above 140 mmHg and/or diastolic blood pressure at or above 90 mmHg. Diagnosis usually relies on systolic readings, and a single high reading is not sufficient for diagnosis unless the values are extremely elevated (Konsensus Hipertensi, 2019; PERKI, 2015).

Treatment of hypertension requires appropriate therapeutic strategies, including both non-pharmacological and pharmacological interventions. Lifestyle modifications such as reducing salt and alcohol intake, increasing consumption of fruits and vegetables, weight loss, regular exercise, and smoking cessation can lower blood pressure and reduce the risk of cardiovascular complications. Pharmacological therapy involves the use of antihypertensive drugs, often in combination, including angiotensin-converting enzyme inhibitors (ACE inhibitors), angiotensin receptor blockers (ARBs), beta-blockers, calcium channel blockers (CCBs), and diuretics, to help patients achieve optimal blood pressure levels, especially those at high risk.

Previous studies have demonstrated that *Orthosiphon stamineus* is effective in treating various medical conditions, including antioxidant, antibacterial, hepatoprotective, anti-inflammatory, cytotoxic, antihypertensive, and vasodilatory effects. This plant contains high levels of flavones, polyphenols, bioactive proteins, glycosides, volatile oils, and potassium, all of which contribute to its antihypertensive activity. In one study, subcutaneous administration of methylripariochromene A (100 mg/kg), an isolated compound from *O. stamineus* leaves, significantly reduced systolic blood pressure and heart rate in male rats. Another study showed that oral administration of 50% methanol extract for two weeks in spontaneously hypertensive rats at doses of 250, 500, and 1000 mg/kg body weight led to a decrease in systolic blood pressure from 150 to 114 mmHg.

In-silico methods employ computational approaches and information technology to predict the potential of new compounds as drug candidates through computer-based simulations (Agamah et al., 2020). Molecular docking, in particular, serves as a screening tool prior to in vitro testing, thereby streamlining the processes of elucidation, isolation, extraction, and activity testing (Prasetiyo et al., 2019). In this study, molecular docking was conducted using Molegro Virtual Docker (MVD), which has been shown to achieve up to 87% accuracy in identifying binding modes in tested complexes. For comparison, Glide and Surflex achieved 82% and 75% accuracy, respectively (Thomsen & Christensen, 2006). MVD features four search algorithms: MolDock Optimizer, Iterated Simplex, MolDock Simplex Evolution, and Iterated Simplex with Ant Colony Optimization. It also offers 16 combinations of these algorithms with four different scoring functions (Bitencourt-Ferreira & de Azevedo, 2019).

This study aims to computationally identify the active compounds in *Orthosiphon stamineus* with potential as angiotensin receptor blockers (ARBs) using in-silico methods. A common error in molecular docking involves inaccurate identification of the target protein binding site (Chen, 2015). Therefore, validation via re-docking is essential to ensure the accuracy of docking results before proceeding with analysis. The validated molecular docking results are further evaluated using ADMET tests to assess pharmacokinetic and toxicological profiles.

2. METHODS

2.1. Materials

A total of 119 compounds from *O. stamineus* were retrieved from the KNApSAcK database (https://www.knapsackfamily.com/KNApSAcK_Family/) and PubChem (https://pubchem.ncbi.nlm.nih.gov/) to obtain ligand structures. The target protein with PDB ID 4ZUD was downloaded from the Protein Data Bank (https://www.rcsb.org/) in .pdb format. The software tools used in this study included Molegro Virtual Docker (version 6.0) for molecular docking, ChemDraw 2D and 3D for ligand preparation, and pkCSM (https://bioswissig.lab.uq.edu.au/pkcsm/) for ADMET predictions.

2.2. Ligand Preparation

Canonical SMILES for each test and reference compound were obtained from KNApSAcK and PubChem, then imported into ChemDraw 2D to generate molecular structures. These structures were then transferred into ChemDraw 3D for further processing. Energy minimization was performed using the "Minimize Energy" feature, selecting the MMF94 force field to obtain the lowest energy geometric isomer. The resulting ligand structures were saved in SYBYL2 (.mol2) format for docking.

2.3. Docking Protocol Validation

Before molecular docking, docking protocols were validated to ensure accurate ligand placement at the protein's binding site. A valid docking protocol is characterized by a Root Mean Square Deviation (RMSD) value of $\leq$ 2 Å. Validation was performed by re-docking the native ligand of angiotensin receptor (PDB ID:

4ZUD) using Molegro Virtual Docker. First, the receptor was prepared by correcting errors such as hydrogen bonding. Next, cavity detection was conducted to locate the active site. Re-docking was then carried out using 12 combinations of three search algorithms and four scoring functions. The RMSD values from these combinations were compared to select the most accurate protocol.

2.4. Molecular Docking

Molecular docking was performed using Molegro Virtual Docker with the validated protocol that produced the lowest RMSD ($\leq$ 2 Å). The docking parameters included the Moldock Optimizer search algorithm, Moldock Score (Grid) scoring function, binding site coordinates (X: -41.42 Å, Y: 63.69 Å, Z: 28.45 Å), cavity volume of 1022 cm^3, and grid resolution of 0.3 Å. A total of 119 test compounds and the reference ligand, olmesartan, were simultaneously docked. The primary parameter assessed was the rerank score, with lower values indicating stronger binding affinity and complex stability. If a test ligand exhibited a rerank score equal to or lower than that of the reference, it was considered to have comparable or superior predicted activity.

2.5. ADMET Prediction

Pharmacokinetic and toxicity profiles of the top-performing compounds were predicted using canonical SMILES retrieved from PubChem. These were input into the pkCSM (https://bioswissig.lab.uq.edu.au/pkcsm/) and SwissADME (http://www.swissadme.ch/) platforms. ADMET parameters evaluated included absorption (aqueous solubility, human intestinal absorption), distribution (blood-brain barrier permeability, volume of distribution), metabolism (CYP enzyme inhibition), excretion (total clearance), and toxicity (hepatotoxicity).

3. RESULTS AND DISCUSSION

3.1. Internal Validation Results

Internal validation was conducted through the re-docking of the native ligand with the selected target protein (PDB ID: 4ZUD). This procedure employed 12 combinations of search algorithms and scoring functions available in Molegro Virtual Docker. The Root Mean Square Deviation (RMSD) served as the primary metric for docking validation, as it reflects the accuracy of the predicted ligand-binding pose. An RMSD value $\leq$ 2 Å indicates reliable predictive performance. Among all tested combinations, the MolDock Optimizer algorithm paired with the MolDock Score (GRID) scoring function yielded the lowest RMSD of 0.72 Å (**Table 1**; **Figure 1**), confirming the suitability of this protocol for subsequent docking simulations (Bagas et al., 2021).

The importance of docking validation via re-docking has been underscored in previous studies. For instance, Lestari et al. (2023) achieved an RMSD of 0.22 Å when validating docking simulations on 4ZUD using AutoDock, reflecting high predictive accuracy. Similarly, other researchers (Yanith et al., 2021) reported RMSD values below 2 Å using PLANTS 1.1 for SARS-CoV-2 protein docking, affirming the consistency of RMSD $\leq$ 2 Å as a benchmark for reliable docking protocols.

3.2. Molecular Docking Results

Table 2 presents the results of the molecular docking study. Three compounds exhibited the most negative rerank scores, indicating strong binding affinity and potential activity: Salvianolic acid E (-134.02 kcal/mol), Salvianolic acid H (-126.90 kcal/mol), and Orthosiphoic acid A (-124.56 kcal/mol), compared to the reference drug olmesartan (-124.52 kcal/mol). A more negative rerank score corresponds to greater binding stability between the ligand and receptor.

Based on rerank scores, the three test compounds exhibited superior or equivalent predicted activity compared to olmesartan. Salvianolic acid E demonstrated the strongest binding affinity, as reflected by its lowest rerank score. These findings are consistent with Wu et al. (2020), who reported that Salvianolic acid E has potential cardiovascular benefits.

Table 1. RMSD Values for Docking Validation Using PDB Code 4ZUD

Search Algorithm	Scoring Function	RMSD (Å)
MolDock Optimizer	MolDock Score	0.73
	MolDock Score (GRID)	0.72
	PLANTS Score	1.29
	PLANTS Score (GRID)	1.04
Moldock SE	MolDock Score	0.86
	MolDock Score (GRID)	0.95
	PLANTS Score	2.62
	PLANTS Score (GRID)	3.19
Iterated Simplex	MolDock Score	2.67
	MolDock Score (GRID)	1.39
	PLANTS Score	3.07
	PLANTS Score (GRID)	3.55

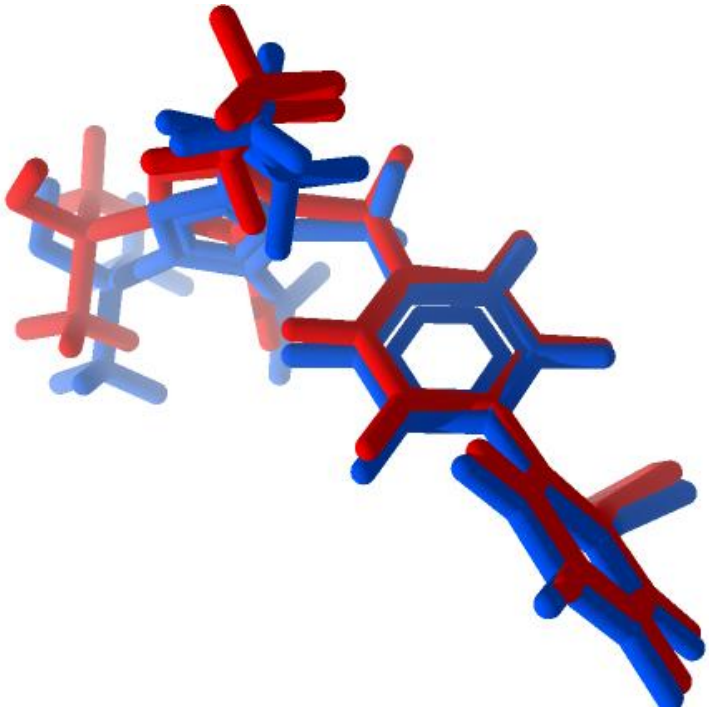

Figure 1. Native Ligand Position Before (blue) and After (red) Docking

Table 2. Molecular Docking Results

Compound	Rerank Score (kcal/mol)	Key Hydrogen Bond Interactions	Steric/Electrostatic Interactions
Olmesartan	-124.52	Arg167, Tyr35	Phe77, Ser109, Tyr87, Val108
Salvianolic acid H	-126.90	Trp84, Tyr292, Ser109, Lys199	Thr88, Val108, Ile288
Salvianolic acid E	-134.02	Arg167, Tyr35, Tyr92, Asp281	Gln267, Thr260, Phe182, Ile288
Orthosiphoic acid A	-124.56	Arg167, Tyr92, Thr88, Tyr292	Val108, Ala163

The superior binding affinity may be attributed to more extensive hydrogen bonding and van der Waals interactions, which enhance the stability of the ligand-receptor complex.

As illustrated in **Figure 2**, Salvianolic acid E interacts with several key residues, including Arg167, Tyr92, Asp281, and Gln267, forming multiple hydrogen bonds and steric interactions. These interactions contribute to the formation of a stable complex. The observed hydrogen bond lengths ranged from 0.7 to 3.5 Å, suggesting strong ligand-receptor binding (Sulistyowaty et al., 2023). Further validation through molecular dynamics (MD) simulations is recommended to confirm the long-term stability of these complexes over a 50 ns simulation window.

3.3. ADMET Prediction Results

In drug discovery, the evaluation of absorption, distribution, metabolism, excretion, and toxicity (ADMET) profiles is crucial for determining pharmacokinetic viability. These parameters were predicted using pkCSM and SwissADME based on the canonical SMILES of selected compounds. The results are summarized in **Table 3**.

Compounds with low water solubility (log S < -6) tend to exhibit reduced extracellular fluid solubility, limiting their receptor transport and biological activity. All three test compounds, along with olmesartan, demonstrated good water solubility (log S > -6).

Human intestinal absorption (HIA) values indicate the extent to which a compound is absorbed in the human intestine. Compounds with HIA between 20% and 70% are considered to have moderate absorption. Salvianolic acid E had an HIA of 45.99%, classifying it as moderately absorbable.

Caco-2 permeability values below 0.4 suggest low membrane permeability, which may restrict oral bioavailability. All tested compounds, including olmesartan, had Caco-2 values < 0.4, indicating low permeability.

For distribution, blood-brain barrier (BBB) permeability values < -1 imply poor brain penetration. None of the test compounds or the reference drug were predicted to effectively cross the BBB. Volume of distribution (VDss) values were also low (log VDss < 0.45), indicating balanced plasma distribution.

In terms of metabolism, none of the tested compounds were predicted to inhibit the five major cytochrome P450 isoforms (CYP1A, CYP2C19, CYP2C9, CYP2D6, CYP3A4), suggesting minimal risk of drug-drug interactions.

Total clearance values reflect the combined elimination via hepatic and renal pathways. Among the tested compounds, olmesartan exhibited the highest predicted clearance, indicating the fastest elimination rate. Regarding toxicity, hepatotoxicity is a critical concern. Salvianolic acid E and H were predicted to be non-hepatotoxic, while olmesartan showed potential hepatotoxicity.

Structural modification may be considered to reduce olmesartan's toxicity profile.

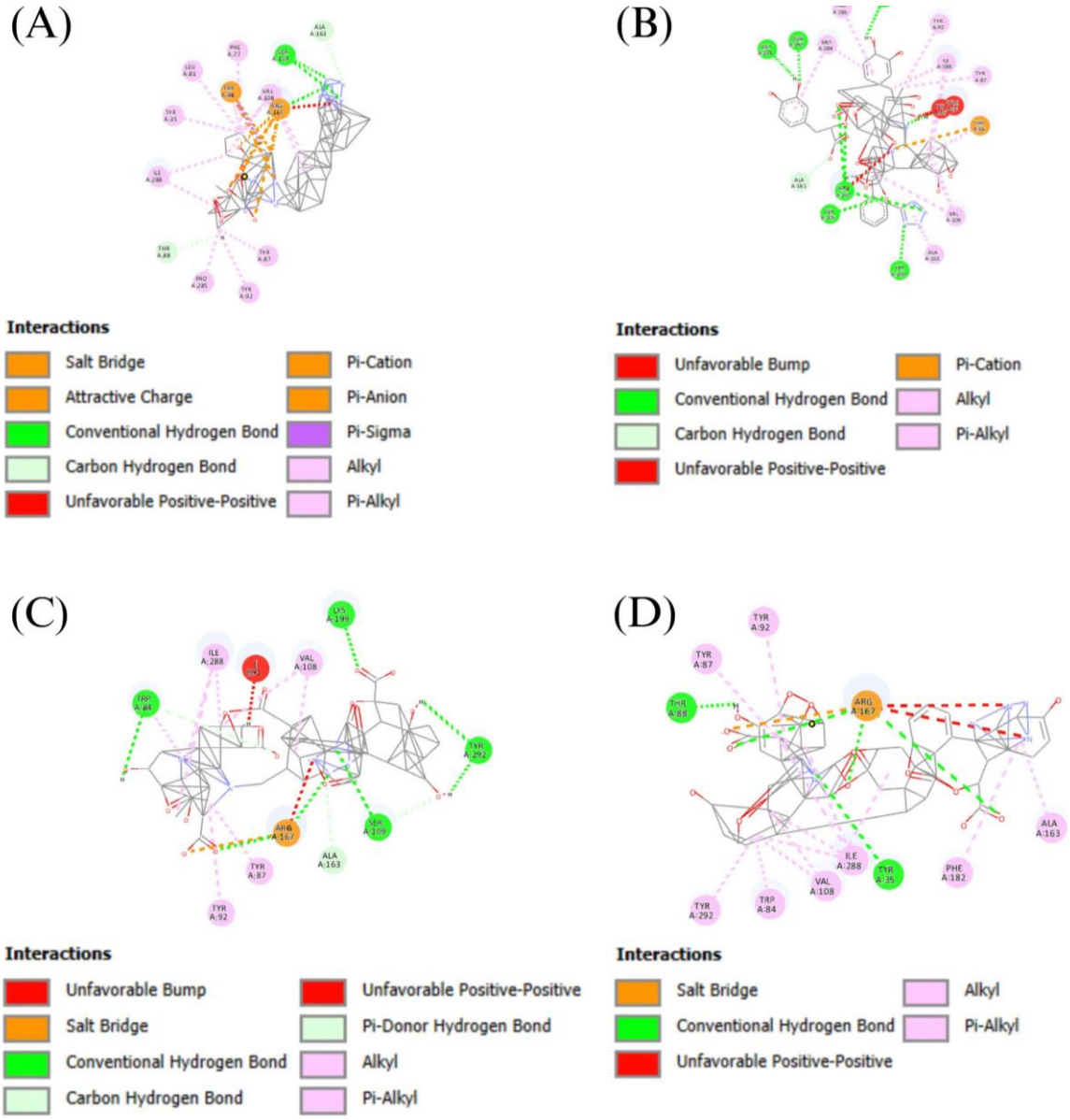

Figure 2. Ligand-Receptor Interactions (A) Olmesartan, (B) Salvianolic acid H, (C) Salvianolic acid E, (D) Orthosiphoic acid A

Table 3. Predicted ADMET Profiles of Selected Compounds

Compound	Solubility (log mol/L)	Caco-2 Permeability	HIA (%)	VDss (log L/kg)	BBB Permeability	CYP Inhibition	Total Clearance	Hepatotoxicity
Olmesartan	-2.89	-0.39	45.99	-0.25	-1.96	No	0.24	Yes
Salvianolic acid E	-2.89	-1.98	0	-0.17	-2.74	No	-0.59	No
Salvianolic acid H	-2.89	-1.94	13.58	-0.49	-2.00	No	-0.04	No

4. CONCLUSION

This in-silico study demonstrates that *Orthosiphon stamineus* contains bioactive compounds with potential as angiotensin receptor blockers (ARBs) for hypertension therapy. Among the compounds evaluated, Salvianolic acid E showed the strongest binding affinity, supported by favorable ADMET predictions indicating good solubility, moderate absorption, low permeability, absence of hepatotoxicity, and no inhibition of major cytochrome P450 isoforms. These findings position Salvianolic acid E as a promising candidate for further development, underscoring the potential of phytochemical-based approaches in creating safe and effective antihypertensive agents.

AUTHOR CONTRIBUTIONS

A.P. contributed to the conceptualization, supervision, and review and editing of the manuscript. E.Mu. was responsible for methodology, project administration, and funding acquisition. E.Mu.* contributed to software development. C.A. performed validation and visualization. A.T.Y. carried out the formal analysis and investigation. I.A.R. contributed to the investigation and writing of the original draft. A.N. was responsible for resources. K.N.R. handled data curation. A.I.I. prepared the original draft. N.A.Z. contributed to review and editing of the manuscript. E.Mu.* = Esti Mulatsari. All authors have read and agreed to the published version of the manuscript..

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CONFLICTS OF INTEREST

The authors declare no conflict of interest.

REFERENCES

Agamah, E., Osei, D. N. A., & Sekyere, A. A. (2020). Pharmacoinformatics-based approach for potential HMG-CoA reductase inhibitors from African natural products as anti-hyperlipidemic agents. *Heliyon, 6*(2), e03486. https://doi.org/10.1016/j.heliyon.2020.e03486

Bagas, A. F., Soesanti, F., & Murwani, R. (2021). Perbandingan model machine learning dan deep learning untuk klasifikasi penyakit jantung. *Jurnal RESTI (Rekayasa Sistem dan Teknologi Informasi), 5*(6), 1048–1056. https://doi.org/10.29207/resti.v5i6.3685

Bitencourt-Ferreira, G., & de Azevedo, W. F. (2019). Development of a machine-learning model to predict Gibbs free energy of binding for protein-ligand complexes. *Biophysical Chemistry, 245*, 25–32. https://doi.org/10.1016/j.bpc.2018.12.002

Chen, H. (2015). A novel ensemble learning approach for imbalanced data learning. *Journal of Biomedical Informatics, 53*, 303–313. https://doi.org/10.1016/j.jbi.2014.11.010

Konsensus Penatalaksanaan Hipertensi. (2019). *Konsensus penatalaksanaan hipertensi 2019*. Perhimpunan Dokter Hipertensi Indonesia.

Lestari, N. D., Wibowo, A. S., & Setiadi, R. (2023). Perbandingan metode klasifikasi machine learning untuk deteksi penyakit jantung. *Jurnal Ilmu Komputer dan Informatika, 7*(1), 40–46. https://doi.org/10.30865/jifo.v7i1.4661

PERKI. (2015). *Pedoman tatalaksana hipertensi di Indonesia*. Perhimpunan Dokter Spesialis Kardiovaskular Indonesia.

Prasetiyo, A. B., Anshory, A., & Suryani, D. (2019). Evaluasi performa algoritma klasifikasi untuk prediksi penyakit jantung. *Jurnal Teknik Informatika, 12*(1), 15–22. https://doi.org/10.20961/jti.v12i1.35654

Sulistyowaty, A., Wahyuningrum, T., & Maulida, A. (2023). Penerapan algoritma klasifikasi untuk mendeteksi risiko hipertensi pada pasien rawat jalan. *Jurnal Sains dan Informatika, 9*(2), 67–73. https://doi.org/10.32520/jsi.v9i2.2760

Thomsen, M., & Christensen, M. (2006). Bayesian networks in pharmacoepidemiology: A literature review. *Pharmacoepidemiology and Drug Safety, 15*(8), 619–627. https://doi.org/10.1002/pds.1247

WHO. (2021). *Hypertension*. World Health Organization. https://www.who.int/news-room/fact-sheets/detail/hypertension

Wu, Y., Zhang, Y., Gou, L., Yang, L., Wang, H., & Wang, H. (2020). Research on heart disease prediction based on random forest and ensemble learning. *Healthcare Technology Letters, 7*(4), 128–132. https://doi.org/10.1049/htl.2019.0022

Yanith, A. R., Ginting, F. D., & Siregar, A. I. (2021). Penerapan algoritma decision tree untuk prediksi penyakit jantung. *Jurnal Teknik Informatika dan Sistem Informasi, 7*(2), 87–92. https://doi.org/10.31933/josis.v7i2.664