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Targeting Androgen Receptor with *Boesenbergia rotunda* Phytoconstituents: A Computational-based Perspective

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Abstract

Prostate cancer is characterized by abnormal cell proliferation within the prostate gland, driven in part by the activation of androgen receptor (AR) upon binding with testosterone. The receptor, therefore, represents a critical therapeutic target in prostate cancer management. *Boesenbergia rotunda* or fingerroot (or temu kunci in Indonesia), a medicinal plant widely used in traditional medicine, has been reported to exhibit diverse pharmacological activities, including anticancer effects. However, despite these promising bioactivities, no molecular level or computational studies have been conducted to explore its interaction with AR. In this study, the anticancer potential of eighteen secondary metabolites from *B. rotunda* rhizome was investigated *in silico* against the AR to identify new therapeutic candidates. The test ligands were evaluated for their physicochemical properties in accordance with Lipinski's rule of five, ADME/Tox predictions, pharmacophore screening, and molecular docking, in comparison with the reference drug bicalutamide. Among the evaluated compounds, boesenbergin A demonstrated the strongest binding affinity to AR, with a binding energy of –11.89 kcal/mol and an inhibition constant of 1.92 nM. Importantly, boesenbergin A engaged amino acid residues, including TRP: 741, like bicalutamide, indicating comparable binding interactions. These findings suggest that boesenbergin A holds substantial promise as a natural anticancer lead compound targeting the AR and warrants further investigation as a potential therapeutic agent for prostate cancer.

Keywords: *Boesenbergia rotunda, boesenbergin A, prostate cancer, in silico.*

INTRODUCTION

Prostate cancer is the sixth leading cause of death in men, with a global incidence rate of 30.7 per 100,000 men and a mortality rate of 7.7 per 100,000 men (IAUI, 2022), making it an urgent health concern and a priority for developing effective therapies. The disease arises from dysregulated cell proliferation and apoptosis, largely driven by androgen receptor (AR) overexpression upon binding with testosterone ligands (Akhtar, *et al*., 2024). Current pharmacological therapy often relies on bicalutamide, a competitive antagonist of testosterone that prevents ligand binding to AR

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and inhibits downstream signaling (Bulldan, *et al*., 2017). While clinically effective, bicalutamide is associated with adverse side effects, including sunburn, shortness of breath, haematuria, jaundice, and chest pain (Shipley, *et al*., 2017), underscoring the need for safer therapeutic alternatives.

Boesenbergia rotunda (syn *B. pandurata*) or fingerrot (temu kunci in Indonesia), a member of the Zingiberaceae family, has long been used in traditional medicine and is reported to exhibit anticancer activities by inhibiting cancer cell proliferation (Choerunisa, 2018; Nurrachma, *et al*., 2018). Its rhizomes are rich in flavonoids, chalcones, and other secondary metabolites with known antioxidant, antibacterial, anti-inflammatory, and antiviral properties (Lailiyyah, *et al*., 2023). Notably, compounds such as galanal A and galanal B have shown strong antioxidant activity (IC_{50} of 3.57 µg/mL and 7.73 µg/mL, respectively) and are suggested to contribute to anticancer effects (Hop & Son, 2023). Further study in breast cancer also demonstrated its anticancer activity (Nurrachma, *et al*., 2020). Despite these promising bioactivities, no studies to date have explored the molecular interactions between *B. rotunda* secondary metabolites and the AR, leaving a critical gap in understanding its mechanism of action at the molecular level.

To address this gap, the present study investigates the interaction of *B. rotunda* secondary metabolites with the AR using a computational approach. Molecular docking, alongside Lipinski's Rule of Five, ADME/Tox prediction, and pharmacophore modeling, was employed to identify potential compounds with strong binding affinities and favorable drug-like properties. This study aims to evaluate the potency of *B. rotunda* phytoconstituents through molecular interaction simulation and to identify lead compounds that could serve as candidates for the development of safer prostate cancer chemotherapy.

METHODS

Lipinski Rule of Five Prediction

Physicochemical properties were predicted using Lipinski's Rule of Five (RO5) through the https://mcule.com website to determine whether the tested compounds met the requirements for the development of a candidate drug to be administered orally in humans (Qurrotaayun, *et al*., 2024). Based on a review by Hop and Son (2023), 47 secondary metabolites of fingerroot rhizomes were identified through maceration extraction. Out of the numerous compounds, 18 compounds were evaluated based on the abundance within the extract.

ADMET Prediction

The pharmacokinetic profile of secondary metabolites in fingerroot rhizomes was examined based on predictions from absorption, distribution, metabolism, and excretion (ADME) tests, as well as toxicity tests. The tests were conducted by copying the MOL structure of the compounds from PubChem and submitting them to "load molecule" feature on the PreADMET website (https://preadmet.webservice.bmdrc.org). The test compounds were evaluated through this web tool, yielding results from the HIA test, Caco-2 cell test, plasma protein binding fraction test, and blood-brain barrier penetration test. From these toxicity tests, the mutagenic and carcinogenic properties of the compounds were recorded in mouse and rat models (Kyla, *et al*., 2024).

Pharmacophore Modelling and Screening

Pharmacophore modelling began with preparing 100 active compounds and 400 decoys downloaded from https://dude.docking.org/targets and saved in MOL file format. In Ligandscout version 4.4.5, the test compound type is changed to test, while active compounds and decoys are changed to training. The active compound

database file is minimised, and the training type is obtained for each cluster. A pharmacophore model is validated using the ROC (receiver operating characteristic) curve along with the AUC (area under curve) score. The next step involved selecting the pharmacophore model with the highest pharmacophore fit score as an initial filter for the compound hits database (Ismail, *et al*., 2025).

Preparation of Ligands, Receptors, and Validation of Docking Methods

Native ligands and receptors (PDB ID code 2AM9; resolution 1.64 Å) were downloaded from https://rscb.org, and their water molecules were removed using Biovia Discovery Studio. In AutoDock, the native ligands were added to hydrogen atoms, non-polar merging was performed, and Gasteiger charges were assigned. For receptors, only hydrogen atoms and Kollmann charges were assigned. The energy in the test compounds was minimized through Chem3D, with hydrogen atoms and Gasteiger charges assigned in Autodock. The redocking process was applied by interacting the native ligand (testosterone) with the receptor, with a grid box setting of X=26.907, Y=2.557, Z=5.181 was applied during redocking. This grid address was then used to perform docking between the AR and compounds from fingerroot rhizomes as ligands (Ismail, *et al*., 2025).

Molecular Docking

Molecular docking for the test compounds was performed using a Lenovo IdeaPad Flex i5 RAM personal laptop with AutoDock (version 4.2.7), using bicalutamide as the reference drug and AR (2AM9) as the target. AutoDock was performed using the command prompt (CMD) with the commands autogrid4-p, dock.gpf-l, dock. glg on the first line, followed by the commands autodock4-p, dock.dpf-l, dock.dlg on the second line, and the results can be viewed using Notepad. Visualization was performed in Biovia Studio to obtain images of the ligand bonds with amino acids on the receptor (Qurrotaayun, *et al*., 2024).

Table 1. Lipinski's rule RO5 results from *B. rotunda* bioactives.

No	Compound Name	Molecular Weight (<500 Da)	Log *P* (<5)	Hydrogen Binding		Description
				Donor (<5)	Acceptor (<10)	
1	Alpinetin	270.2789 Da	3.1073	1	4	Eligible
2	Pinocembrin	256.2523 Da	2.8043	2	4	Eligible
3	Pinostrobin	270.2789 Da	3.1073	1	4	Eligible
4	Trans-methyl cinnamate	162.1845 Da	1.8728	0	2	Eligible
5	2',4',6'-Trihydroxydihydrochalcone	258.2682 Da	2.3000	3	4	Eligible
6	Helichrysetin	286.2783 Da	2.7081	3	5	Eligible
7	Boesenbergin A	404.50 Da	6.2076	1	4	Eligible Violation=1
8	Cardamonin	270.279 Da	3.0025	2	4	Eligible
9	5-6 Dehydrokawain (demethoxyyangonin)	228.2423 Da	2.8188	0	3	Eligible
10	(+)-Panduratin A	406.51 Da	6.0116	2	4	Eligible Violation=1
11	Geranyl-2,4-dihydroxy-6-phenylbenzoate	366.4488 Da	5.6143	2	4	Eligible Violation=1
12	Nerol	154.2490 Da	2.6714	1	1	Eligible
13	Pinostrobin chalcone	270.2789 Da	3.0025	2	4	Eligible
14	Boesenbergin B	404.4965 Da	6.207S6	1	4	Eligible Violation=1
15	Galanal A	318.4495 Da	3.6943	1	3	Eligible
16	Galanal B	318.4495 Da	3.6943	1	3	Eligible
17	Geraniol	154.249 Da	2.6714	1	1	Eligible
18	Camphor	152.2330 Da	2.4017	0	1	Eligible

RESULTS

The Prediction of Physicochemical Properties of *B. rotunda* Bioactives

The physicochemical properties of *B. rotunda* bioactive compounds were evaluated using Lipinski's rule of five (RO5) to assess their potential as orally active drugs. Fourteen compounds fully complied with RO5 without any violations, while four compounds (boesenbergin A, (+)-panduratin A, geranyl-2,4-dihydroxy-6-phenylbenzoate, and boesenbergin B) showed a single violation of the log *P* criterion, with values greater than five (Table 1). According to RO5 principle, compounds with no more than one violation are still considered suitable for oral administration. Based on this assessment, all of the tested compounds demonstrate favorable oral drug-likeness and can proceed to ADME/Tox prediction analysis.

The Prediction of the Pharmacokinetic Profile and Toxicity of *B. rotunda* Bioactives

ADME/Tox predictions were made to predict the absorption, distribution, metabolism, excretion, and toxicity profiles of the compounds. It was found that all test compounds had good HIA percentages, *i.e.* >70%. The Caco-2 results obtained showed that pinocembrin had a low Caco-2 value, less than 4 nm/sec, while the other seventeen compounds had moderate Caco-2 values, 4–70 nm/sec. In the predicted distribution results, three compounds had PBB values below 90%, and the remaining fifteen compounds were classified as high due to values above 90%. The three compounds with PBB values below 90% are trans-methyl cinnamate, 2',4',6'-trihydroxydihydrochalcone, and 5-6 dehydrokawain. Furthermore, the BBB prediction results for the test compounds showed that six compounds have the ability to

Table 2. ADME/Tox prediction results from *B. rotunda* bioactives.

No	Compound Name	Absorption		Distribution		Toxicity		
		HIA (%)	Caco-2 (nm/sec)	PPB (%)	BBB	Mutagen	Carsinogen	
							Mouse	Rat
1	Alpinetin	95.52	14.68	93.69	0.15	Mutagen	-	+
2	Pinocembrin	92.35	2.47	98.45	0.91	Mutagen	-	-
3	Pinostrobin	95.51	14.68	93.52	0.17	Mutagen	-	+
4	Trans-methyl cinnamate	100.00	24.46	66.95	1.46	Mutagen	-	-
5	2',4',6'-Trihydroxydihydrochalcone	87.89	19.61	81.72	0.45	Mutagen	-	-
6	Helichrysetin	88.52	18.51	93.72	0.69	Mutagen	+	+
7	Boesenbergin A	96.50	46.77	94.29	5.74	Non-mutagen	-	-
8	Cardamonin	92.83	15.98	92.70	0.56	Mutagen	-	+
9	5-6 Dehydrokawain (demethoxyyangonin)	98.87	50.12	87.52	0.09	Mutagen	+	+
10	(+)-Panduratin A	94.94	27.17	100.00	6.12	Mutagen	+	-
11	Geranyl-2,4-dihydroxy-6-phenylbenzoate	94.45	24.28	94.32	6.33	Non-mutagen	-	-
12	Nerol	100.00	8.76	100.00	6.74	Mutagen	+	-
13	Pinostrobin chalcone	92.83	15.01	93.01	0.46	Mutagen	+	-
14	Boesenbergin B	96.49	44.23	93.89	5.69	Non-mutagen	-	-
15	Galanal A	95.53	21.14	98.62	1.87	Non-mutagen	-	+
16	Galanal B	95.53	21.14	98.62	1.87	Non-mutagen	-	+
17	Geraniol	100.00	8.76	100.00	6.74	Mutagen	+	-
18	Camphor	100.00	26.82	100.00	0.87	Mutagen	-	+

penetrate the blood-brain barrier. This is indicated by a BBB value above two. The compound with the smallest BBB value is 5-6 dehydrokawain. In the toxicity prediction results, five compounds were non-mutagenic, and six compounds were non- carcinogenic. The compounds that were o verall non-toxic were boesenbergin A, geranyl-2,4-dihydroxy-6-phenylbenzoate, and boesenbergin B (Table 2).

Pharmacophore Modelling and Screening of *B. rotunda* Bioactives

Based on the screening results, models 3 and 4, each with an AUC value of 0.79, demonstrated good compatibility with the pharmacophore model, indicating a high probability of biological activity (Table 3). An AUC value above 0.7 is generally considered acceptable, reflecting reliable discrimination between active and inactive compounds. Therefore, the values obtained suggest that models 3 and 4 possess strong predictive performance in identifying potential bioactive compounds. This also implies that the structural features represented in these pharmacophore models are relevant to the interaction with the AR and could serve as a robust basis for virtual screening of novel ligands. In this context, the identified models not only confirm the biological potential of *B. rotunda* metabolites but also provide a rational framework for prioritizing compounds for further molecular docking studies.

Table 3. Area under curve (AUC) results of the pharmacophore model.

Model	Hits	True Positive	False Positive	AUC
Model 1	405	95	310	0.78
Model 2	405	95	310	0.78
Model 3	405	95	310	0.79
Model 4	405	95	310	0.79
Model 5	433	95	338	0.72
Model 6	433	95	338	0.72
Model 7	437	97	340	0.70
Model 8	437	97	340	0.70
Model 9	436	97	339	0.70
Model 10	436	97	339	0.70

We then used the model 3 for the subsequent pharmacophore features of the hit compounds and found a total of eleven hit compounds, as seen in Table 4. Following pharmacophore modeling, each constituent of *B. rotunda* was checked to evaluate its compatibility with the pharmacophore features and its potential interaction with the AR. Among these, 2',4',6'-tri-hydroxydihydrochalcone achieved the highest fit score of 34.04, suggesting a strong alignment with the pharmacophore model and a higher probability of favorable binding interactions with the target protein. The presence of key pharmacophoric features (hydrogen bond acceptor, aromatic

hydrophobic, and hydrogen bond donor) further supports its ability to form stable and specific interactions within the receptor binding site. These results highlight 2',4',6'-trihydroxydihydrochalcone as one of the most promising candidates for a subsequent molecular docking study, providing a rationale for prioritizing this compound in the evaluation of *B. rotunda*'s anticancer potential.

Table 4. Pharmacophore features of hit compounds from *B. rotunda*.

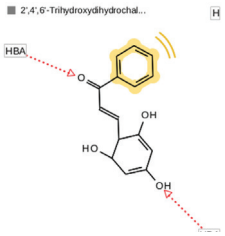
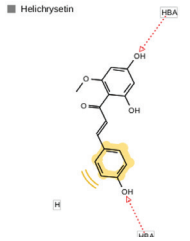
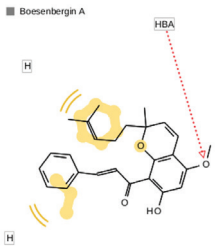
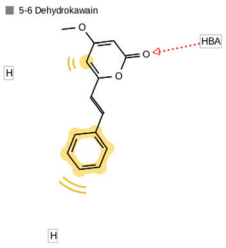
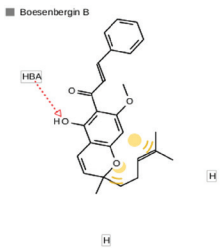
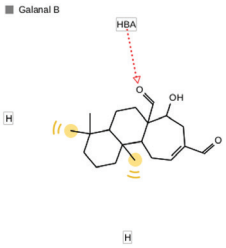
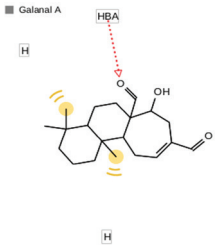
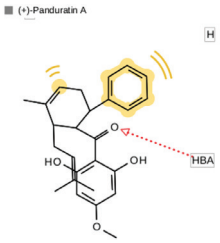
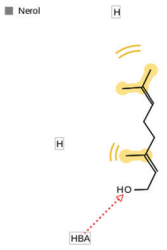
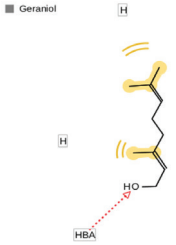
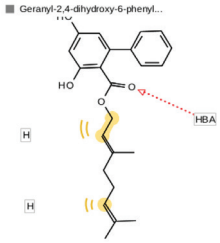
No	Compound Name	Fit Score	Pharmacophore Features
1	2',4',6'-Trihydroxydihydrochalcone	34.04	
2	Helichrysetin	33.44	
3	Boesenbergin A	33.04	
4	5-6D ehydrokawain (demethoxyyangonin)	32.85	

Table 4. Pharmacophore features of hit compounds from *B. rotunda* (continuous).

No	Compound Name	Fit Score	Pharmacophore Features
5	Boesenbergin B	32.84	
6	GalanalB	32.24	
7	GalanalA	32.12	
8	(+)-Panduratin A	31.99	
9	Nerol	31.97	

No	Compound Name	Fit Score	Pharmacophore Features
10	Geraniol	31.97	
11	Geranyl-2,4-dihydroxy-6-phenylbenzoate	31.89	

Molecular Docking of *B. rotunda* Bioactive Compounds

Molecular docking began with the validation of the docking method using the receptor against its native ligand (AR-testosterone). The RMSD value, or a measure of deviation from changes in protein-ligand interactions before and after the redocking process, was 0.79 Å. The redocking method can be applied for docking test compounds if the RMSD value obtained is lower than 2 Å. Hence, our redocking result fulfilled the validation criteria and could be employed in a similar setting for the *B. rotunda* phytoconstituents. Preliminary study reported that the chemotherapy bicalutamide act as an antagonist for AR through π- π binding at PHE A: 746 (Shimmin, *et al*., 2024), along with TRP: 741 (Bassetto, *et al*., 2016). Following the visualization of the interaction (Figure 1), the only test compound that interacts with those amino acid residues was boesenbergin A (Table 5). The test compounds with the lowest binding energy and inhibition constant values, respectively, are boesenbergin A, galanal A, and geranyl-2,4-dihydroxy-6-phenylbenzoate.

Table 5. Molecular docking analysis results from the bioactive compounds of *B. rotunda*.

Compounds B	inding Energy (kcal/mol)	Inhibition Constant	Amino Acid Interactions		
			Hydrogen Binding	Van Der Waals	Other Binding
Testosterone (Native ligand)	-11.99	1.64 nM	Asn A: 705 Gln A: 711	-	Met A: 780 Leu A: 873 Met A: 742 Leu A: 707 Trp A: 741 Met A: 745 Leu A: 704 Phe A: 876

Table 5. Molecular docking analysis results from the bioactive compounds of *B. rotunda* (continuous).

Compounds	Binding Energy (kcal/mol)	Inhibition Constant	Amino Acid Interactions		
			Hydrogen Binding	Van Der Waals	Other Binding
Bicalutamide (Reference drug)	-9,95	51.05 nM	Met A: 742	-	Leu A: 704 Phe A: 764 Met A: 787 Met A: 780 Trp A: 741 Met A: 745 Phe A: 876 Leu A: 873
Alpinetin	-8.60	496.08 nM	Leu A: 873	-	Thr A: 877 Asn A: 705 Met A: 780 Phe A: 764 Met A: 745 Leu A: 701 Phe A: 891 Leu A: 880
Pinocembrin	-8.43	661.40 nM	Asn A: 705 Leu A: 873	-	Met A: 745 Met A: 780 Phe A: 764
Pinostrobin	-8.30	819.23 nM	Asn A: 705 Thr A: 877	-	Met A: 780 Phe A: 876 Leu A: 704 Leu A: 707 Met A: 749 Met A: 745 Phe A: 764 Leu A: 873
Trans-methyl cinnamate	-5.85	51.42 uM	Arg A: 752 Gln A: 711	-	Leu A: 707 Met A: 745 Leu A: 704 Leu A: 873 Phe A: 764 Met A: 780
2',4',6'-Trihydroxydihydrochalcone	-9.10	214.85 nM	Leu A: 704 Arg A: 752 Gln A: 711	-	Leu A: 707 Met A: 745 Phe A: 764 Met A: 780 Leu A: 873
Helichrysetin	-9.58	95.22 nM	Asn A: 705 Leu A: 873 Gln A: 711 Arg A: 752	-	Met A: 780 Met A: 745 Leu A: 707

Table 5. Molecular docking analysis results from the bioactive compounds of *B. rotunda* (continuous).

Compounds	Binding Energy (kcal/mol)	Inhibition Constant	Amino Acid Interactions		
			Hydrogen Binding	Van Der Waals	Other Binding
Boesenbergin A	-11.89	1.92 nM	Arg A: 752 Met A: 787	-	Met A: 895 Phe A: 891 Leu A: 704 Leu A: 707 Trp A: 741 Met A: 745 Met A: 749 Val A: 746 Leu A: 873 Met A: 780 Phe A: 876 Phe A: 764
Cardamonin	-9.21	178.22 nM	Leu A: 704 Gln A: 711 Arg A: 752	-	Leu A: 707 Met A: 745 Val A: 746 Met A: 749 Phe A: 764 Met A: 780 Met A: 742 Leu A: 873
5-6 Dehydrokawain (demethoxyyangonin)	-7.56	2.86 uM	Thr A: 877	-	Phe A: 764 Val A: 746 Met A: 780 Met A: 749 Leu A: 873 Met A: 787
(+)-Panduratin A	-10.42	23.10 nM	-	-	Gln A: 711 Met A: 745 Met A: 749 Leu A: 707 Leu A: 873 Leu A: 880 Leu A: 704 Phe A: 876 Met A: 780 Leu A: 701
Geranyl-2,4-dihydroxy-6-phenylbenzoate	-11.47	3.90 nM	Gln A: 711	-	Phe A: 876 Leu A: 701 Leu A: 873 Leu A: 704 Met A: 745 Leu A: 707 Met A: 780 Met A: 742
Nerol	-6.24	26.59 uM	Arg A: 752 Gln A: 711	-	Met A: 745 Leu A: 704 Phe A: 764 Leu A: 707 Met A: 780 Leu A: 873

Table 5. Molecular docking analysis results from the bioactive compounds of *B. rotunda* (continuous).

Compounds	Binding Energy (kcal/mol)	Inhibition Constant	Amino Acid Interactions		
			Hydrogen Binding	Van Der Waals	Other Binding
Pinostrobin chalcone	-8.04	1.29 uM	Leu A: 704 Gln A: 711 Met A: 742	-	Leu A: 873 Leu A: 707 Met A: 749 Gly A: 708 Phe A: 764 Met A: 745
Boesenbergin B	-8.01	1.34 uM	Gly A: 708	-	Leu A: 880 Met A: 780 Leu A: 704 Met A: 742 Leu A: 873 Val A: 746 Phe A: 764 Met A: 787 Met A: 749 Leu A: 707 Leu A: 701 Asn A: 705 Met A: 745
Galanal A	-11.72	2.57 nM	Leu A: 704 Gln A: 711 Arg A: 752	-	Met A: 745 Leu A: 873 Phe A: 764 Leu A: 701
Galanal B	-11.02	8.39 nM	Arg A: 752 Gln A: 711	-	Leu A: 873 Phe A: 764 Leu A: 704 Leu A: 701 Met A: 745
Geraniol	-6.13	32.10 uM	Arg A: 752 Gln A: 711	-	Met A: 749 Val A: 746 Phe A: 764 Met A: 787 Leu A: 704 Leu A: 707
Camphor	-6.60	14.50 uM	Gln A: 711 Arg A: 752	-	Leu A: 704 Leu A: 707 Phe A: 764 Met A: 745

DISCUSSION

The development of prostate cancer drugs from *B. rotunda* rhizomes can be guided effectively through computational approaches that allow efficient identification of potential drug candidates. The first step was the evaluation of physicochemical properties using Lipinski's Rule of Five (RO5) to predict oral drug-likeness. All 18 test compounds complied with RO5, with only four compounds showing a single violation of the log *P* rule. Compounds with one violation are still

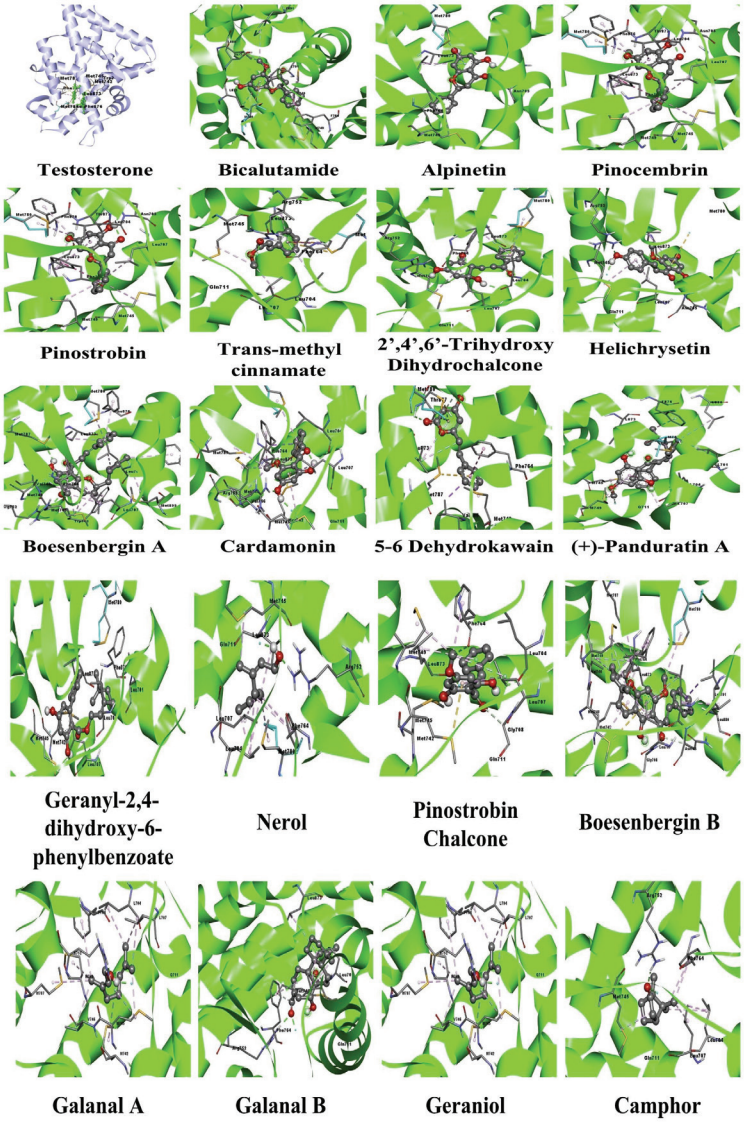


Figure 1. The 3-dimensional docking visualization from molecular docking between native ligand, reference drug, and bioactive compounds in *B. rotunda* toward AR.

considered drug-like (Yang, *et al*., 2020), indicating that all tested metabolites of *B. rotunda* hold potential for oral formulation. To complement these findings, ADME/Tox profiling was performed to assess pharmacokinetic behavior and toxicity risks. Of particular note, boesenbergin A, galanal A, and geranyl-2,4-dihydroxy-6-phenylbenzoate demonstrated favorable ADME/Tox properties, suggesting that these compounds not only fulfill drug-likeness criteria but also possess pharmacokinetic profiles comparable to the reference drug bicalutamide, which is known for its high plasma protein binding (~96%). These results provided a strong basis for advancing these compounds into pharmacophore screening.

Pharmacophore analysis further refined the candidate list by identifying structural features necessary for AR interaction. Eleven hit compounds

were identified across the models, with 2',4',6'-Trihydroxydihydrochalcone achieving the highest fit score (34.04), reflecting strong alignment with the pharmacophore features, particularly hydrogen bond acceptors, hydrogen bond donors, and aromatic hydrophobic interactions. This step not only validated the relevance of *B. rotunda* metabolites as AR modulators but also narrowed the focus for subsequent docking simulations.

Molecular docking then confirmed the interaction profiles of the shortlisted compounds. Boesenbergin A, galanal A, and geranyl-2,4-dihydroxy-6-phenylbenzoate consistently exhibited the lowest binding energies and inhibition constants, signifying stable and efficient interactions with the AR. In the previous study (Shimmin, *et al*., 2024), it was found that interaction occurs between AR and bicalutamide at the amino acid residue TRP A: 741 in the 12th alpha helix position (H12). When antagonists such as bicalutamide bind, H12 is unable close properly, thereby inhibiting AR activation. The only bioactive compound from *B. rotunda* that has this amino acid is boesenbergia A. Interestingly, 2',4',6'-Trihydroxydihydrochalcone, while not replicating bicalutamide's binding residues, emerged as a promising candidate due to its superior fit score, raising the possibility of alternative binding modes, such as interactions with allosteric sites of AR.

When integrating results from drug-likeness, ADME/Tox, pharmacophore screening, and docking, boesenbergin A stands out as the strongest candidate. Although it violates the log *P* rule, its high binding affinity, favorable pharmacokinetics, and pharmacophore compatibility position it as a superior lead compound compared with galanal A and geranyl-2,4-dihydroxy-6-phenylbenzoate. The log *P* violation suggests high lipophilicity, which could impair membrane penetration or lead to accumulation in lipid layers. Nevertheless, this limitation may be addressed through advanced drug delivery strategies, such as nanoparticle systems or lipid-based formulations. Importantly, the anticancer properties of isolated boesenbergin A from *B. rotunda* have been reported, demonstrating cytotoxic effects against HepG2 (hepatoblastoma), HT-29 (colon adenocarcinoma), A549 (non-small cell lung cancer), and PC-3 (prostate adenocarcinoma) cell lines (Isa, *et al*., 2012). These findings highlight the potential of boesenbergin A as a promising natural-derived anticancer candidate for further development.

Despite the promising *in silico* results, several limitations must be considered. First, computational predictions rely on approximations and cannot fully replicate the complexity of biological systems; experimental validation through *in vitro* and *in vivo* studies is essential to confirm bioactivity and safety. Second, docking studies were performed using a single AR structure; receptor flexibility, isoform variation, and potential off-target interactions were not explored. Third, high lipophilicity observed in some compounds (*e.g*., boesenbergin A) may pose formulation challenges that require further optimization. Future studies should therefore focus on experimental validation of the anticancer effects of boesenbergin A, galanal A, and 2',4',6'-trihydroxydihydrochalcone, which are isolated from *B. rotunda*, against prostate cancer cell lines, along with structure–activity relationship (SAR) studies to guide chemical modification of lead compounds. By integrating these approaches, *B. rotunda* metabolites hold strong potential for advancement into novel, plant-derived therapeutics for prostate cancer.

CONCLUSION

Computational prediction of *B. rotunda* metabolites against the AR identified boesenbergin A as the most promising compound. It is the only compound that possesses a similar binding through a crucial amino acid residue to bicalutamide, achieving a high fit score of 33.04, exhibiting

strong binding affinity with a Gibbs free energy change of –11.89 kcal/mol, and showing a low inhibition constant of 1.92 nM. Boesenbergin A also interacted with a key tryptophane residue (TRP A: 741) of the receptor and demonstrated favorable pharmacokinetic and toxicity profiles. These findings suggest that boesenbergin A holds significant potential as a lead compound for further development into an oral therapeutic agent for prostate cancer.

REFERENCES

Akhtar, M., Khaled, A.M., Noheir, M.T., Hussam, T., and Issam, A., 2024, Role of androgen receptor in prostate cancer: A brief update, *Journal on Oncology*, **4**(2), 1142.

Arwansyah, Ambarsari, L., and Sumaryada, T.I., 2014, Simulation of curcumin compound docking and its analogue as androgen receptor inhibitors in prostate cancer, *Current Biochemistry*, **1**(1), 11–19.

Bassetto, M., Ferla, S., Pertusati, F., Kandil, S., Westwell, A.D., Brancale, A., and McGuigan, C., 2016, Design and synthesis of novel bicalutamide and enzalutamide derivatives as antiproliferative agents for the treatment of prostate cancer, *European Journal of Medicinal Chemistry,* **118**, 230-243.

Bulldan, A., Malviya, V.N., Upmanyu, N., Konrad, L. and Scheiner-Bobis, G., 2017, Testosterone/ bicalutamide antagonism at the predicted extracellular androgen binding site of ZIP9, *Biochimica et Biophysica Acta (BBA)-Molecular Cell Research*, **1864**(12), 2402–2414.

Butnasaru, C., Garbera, O.V., Petrini, P., Visal, I. and Visentin, S., 2023, Permeability assessment of a high-throughput mucosal platform pharmaceutics, *Pharmaceutics*, **15**(2), 380.

Choerunisa, R., and Halimah, E., 2018, Review: Anticancer activity of several plants in Indonesia, *Farmaka*, **16**(1), 240–246.

Hess, A.Y., Ramadhani, S.Z., Andhryanti, R.N., Zhafirah, N., Muljono, F.O., Fardhan, F.M., and Novitasari, D., 2024, *In silico* study of bioactive compounds in guava leaves (*Psidium guajava* L.) toward angiotensin converting enzyme (ACE) as a target for hypertension, *Indonesian Journal of Chemical Science*, **13**(3), 209–219.

Ikatan Ahli Urologi Indonesia, 2022, *Procedures for treating prostate cancer*, Jakarta: Kemenkes.

Isa, N.M., Abdelwahab, S.I., Mohan, S., Abdul, A.B., Sukari, M.A., Taha, M.M.E., *et al.*, 2012, *In vitro* anti-inflammatory, cytotoxic, and antioxidant activities of boesenbergin A, a chalcone isolated from *Boesenbergia rotunda* (L.) (fingerroot), *Brazilian Journal of Medical and Biological Research*, **45**, 524-530.

Ismail, D.P.F., Christella, A., Salsabila, N.F., Angelina, A., Muljono, F.O, Fardhan, F.M., and Novitasari, D., 2025, The bioactive compounds in bilimbi leaves (*Averrhoa bilimbi* L.) as natural-based anticancer targeting on VEGFR-2: *In silico* studies, *Notulae Scientia Biologicae*, **7**(1), 12289.

Kyla, F.A.M., Nabilah, R., Sitti, M., Puspita, S.F., Maymuna, F.N., Agus, R., and Novitasari, D., 2024, Studi *in silico* senyawa aktif daun kemangi imbo (Pycnarrhena cauliflora) terhadap reseptor estrogen alfa (ERα) sebagai kandidat antikanker payudara, *Pharmacoscript*, **7**(2), 346–361.

Lailiyyah, H., and Lisdiana, L., 2023, *In silico* testing of the antibacterial activity of active compounds in temu kunci (*Boesenbergia rotunda*) against Mycobacterium tuberculosis, *Lentera Bio.*, **12**(2), 132–149.

Lipinski, C.A., Lombardo, F., Dominy, B.W., and Feeney, P.J., 2012, Experimental and computational approaches to estimate solubility and permeability in drug discovery and development settings, *Advanced Drug Delivery Reviews*, **23**, 3–25.

Nurrachma, M.Y., Fadliyah, H., and Meiyanto, E., 2018, Fingerroot (*Boesenbergia pandurata*): A prospective anticancer therapy, *Indonesian Journal of Cancer Chemoprevention*, **9**(2), 102-

109.

Nurrachma, M.Y., Maran, G.G., Putri, N.B., Esti, Y.F., Hermawan, A., Meiyanto, E., and Jenie, R.I., 2020, Fingerroot (*Boesenbergia pandurata*) extract inhibits proliferation and migration of 4T1 metastatic breast cancer cells, *Indonesian Journal of Cancer Chemoprevention*, **11**(3), 103–114.

Peng, C.C., Chen, C.Y., Cheng, C.R., Chen, C.J., Shen, K.H., Chen, K.C., and Peng, R.Y., 2019, Renal damaging effect elicited by bicalutamide therapy uncovered multiple action mechanisms as evidenced by the cell model, *Scientific Reports*, **9**, 3392.

Praceka, M.S., Yunita, E.N., Semesta, C.D., Putri, R.N., Mikdar, N.N., Sitinjak, E.N., Setyawati, L.U., and Muchtaridi, M., 2022, Molecular docking and toxicity from temulawak rhizome (*Curcuma xanthorrhiza* Roxb.) against COX-2, *Indonesian Journal of Pharmaceutical Science and Technology*, **1**(1), 106–115.

Puspita, P.J., Liliyani, N.P.P., and Ambarsari, L., 2022, *In silico* analysis of active compounds of avocado fruit (*Persea americana* Mill.) as tyrosinase enzyme inhibitors, *Current Biochemistry*, **9**(2), 73–87.

Putri, I.A., Estiningtyas, D.A., Idelia, N.B., Rahmah, S.N.A., Rusdin, A., Fathin, N.M., and Novitasari, D., 2024, Potential bioactive compounds in Java cardamom fruit (Amomum compactum) as candidate COX-2 targeted anti-inflammatory agents: In silico study, *Jurnal Ilmu Farmasi dan Farmasi Klinik*, **21**(2), 198–208.

Qurrotaayun, G.A.P., Sitompul, J.E.N., Fadhilah, N., Pramudita, F.W., Putri, N.S., Muljono, F.O., *et al.*, 2024, Rosmarinic acid from Orthosiphon aristatus potentially targets estrogen receptor-alpha in breast cancer: In-silico study, *Indonesian Journal of Cancer Chemoprevention*, **15**(2), 150–161.

Shimmin, B.A., Haines, L.G., and Shaw, I.C., 2024. *In silico* studies on the molecular interactions of steroid hormones and steroid hormone mimicking drugs in the androgen receptor binding cleft–Implications for prostate cancer treatment, *Steroids*, **208**, 109456.

Shipley, W.U., Seiferheld, W., Lukka, H.R., and Major, N.M., 2017, Radiation with or without antiandrogen therapy in recurrent prostate cancer, *The New England Journal of Medicine*, **376**(5), 417–428.

Yang, W., Gadgil, P., Krishnamurthya, V.R., Landis, M., Mallick, P., Patel, D., *et al.*, 2020, The evolving druggability and developability space: Chemically modified new modalities and emerging small molecules, *The AAPS Journal*, **22**, 21.

Yuliana, A., and Fadhlurrohman, D., 2022, Study molecular docking of pigment compounds derived from Monascus sp. as hepatitis B inhibitors, *Media Ilmu Kesehatan*, **11**(3), 291–299.