

Total Synthesis and Potential Bioactivity of Linear Peptide LSAVTPG

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ABSTRACT

The total synthesis of the linear peptide LSAVTPG was successfully carried out using the solid-phase peptide synthesis (SPPS) method. The synthesis employed the Fmoc/tBu protection strategy on a 2-chlorotrityl chloride resin as the solid support. Following chain assembly, the final peptide was cleaved from the resin using 95% trifluoroacetic acid (TFA), affording the target peptide with a yield of 81.6%. Structural confirmation was performed by high-resolution mass spectrometry (HRMS), which showed an observed ion at m/z 644.3621, consistent with the calculated value of m/z 644.3619 for $C_{28}H_{50}N_7O_{10}$. Purity and retention characteristics were further analyzed using analytical HPLC, where the peptide exhibited a retention time of 13.536 minutes. These results demonstrate that the SPPS method with Fmoc/tBu chemistry is an efficient strategy for the synthesis of the LSAVTPG peptide with high yield and analytical confirmation. The potential bioactivity screened computationally and showed some potential such as spanning oncology, cardiometabolic regulation, neurodegeneration, immunology, and inflammation.

Introduction

The LSAVTPG peptide (Figure 1) serves as a precursor to Cyclosenegalina A, which has been isolated from *Annona senegalensis* and *Annona scleroderma*. The cyclic peptide cyclo (–Pro1–Gly2–Leu3–Ser4–Ala5–Val6–Thr7–) was obtained from methanol extracts of both plants. Cyclosenegalina A has demonstrated cytotoxic activity against human prostate cancer DU-145 cells, with an IC_{50} value of $54.9 \pm 2.35 \mu M$ [12]. Despite the cancer potential, the synthesis of Cyclosenegalina A and its analogue showed low antimicrobial properties [3–5].

As LSAVTPG peptide are precursor of derivated peptide from endemic African plants, further exploration of its bioactivity potential is still limited by availability. Linear peptide synthesis is commonly performed using solid-phase peptide synthesis (SPPS). The PTVALSG and SLGPTVA precursor was successfully obtained by this method [3]. SPPS offers significant advantages over classical liquid-phase peptide synthesis (LPPS), including higher product purity and faster reaction times.

The complex isolation procedures required for intermediates in LPPS are replaced in SPPS by simple washing and filtration steps, thereby reducing time and effort [6]. Furthermore, SPPS has been shown to achieve stepwise yields of up to 99.5% or greater, a level of efficiency unattainable with conventional synthetic approaches [7,8].

Bioactivity potential can be obtained through *in silico* prediction [9]. *In silico* target prediction was performed using the SwissTargetPrediction platform, which applies a ligand-based computational approach to estimate the most probable protein targets of small molecules [10]. The method integrates chemical similarity searches against a curated database of known bioactive compounds, combining 2D and 3D similarity metrics to generate ranked target lists across multiple species. By submitting the query molecule into the platform, predicted interactions were obtained for *Homo sapiens*, *Rattus norvegicus*, and *Mus musculus*, enabling comparative analysis of conserved and species-specific targets. This

computational strategy provides an efficient first step in drug discovery, offering mechanistic hypotheses and guiding experimental validation by situating the compound within established therapeutic landscapes [11].

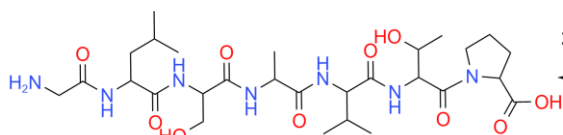


Figure 1. LSAVTPG Peptide

Materials and Methods

The equipment to be used in this study includes standard glassware commonly employed in a synthesis laboratory. In addition, supporting instruments such as an SPPS reactor and a rotary evaporator will be utilized. Compound purification will be carried out using semi-preparative RP-HPLC with a Waters Alliance e2695 system equipped with a PDA (Photodiode Array) 2489 detector and an RP-18e LichroCart column (5 μ m, 4.6 $\times$ 250 mm). For characterization of the purified compound, high-resolution mass spectrometry will be performed using an HR-TOF-MS (High Resolution Time-of-Flight Mass Spectroscopy) Lockspray Waters instrument. Compound purity analysis will be conducted with analytical RP-HPLC using a Waters Alliance 2998 system equipped with a PDA detector and a C-18 Acquity BEH column (1.7 μ m, 2.1 $\times$ 50 mm).

The chemicals used in this study include Fmoc-L-Pro-OH, Fmoc-L-Thr(t-Bu)-OH, Fmoc-L-Val-OH, Fmoc-L-Ala-OH, Fmoc-L-Ser(tBu)-OH, Fmoc-L-Leu-OH, Fmoc-L-Gly-OH, Fmoc-Lys(Boc)-OH, hexafluorophosphate azabenzotriazole tetramethyl uronium (HATU), hexafluorophosphate benzotriazole tetramethyl uronium (HBTU), 1-hydroxy-7-azabenzotriazole (HOAt), 1-hydroxybenzotriazole (HOBt), 2-chlorotriyl chloride (CTC) resin (GLBiochem, China), acetaldehyde, N,N-diisopropylethylamine (DIPEA), dichloromethane (DCM), dimethylformamide (DMF), methanol, propanol, ethyl acetate, n-hexane, piperidine, trifluoroacetic acid (TFA), HPLC-grade acetonitrile, and HPLC-grade methanol.

Synthesis of LSAVTPG

Coupling of the C-terminal amino acid to the resin. The 2-Chlorotriyl chloride resin (capacity 0.99 mmol/g) was weighed (400 mg) and transferred into the reactor. Dichloromethane (DCM, 5 mL) was added, and the mixture was shaken for 15 minutes, filtered, and dried. The dried resin was then treated with a solution of the first amino acid (Fmoc-L-Pro-OH, 0.6 mmol) in DCM (3 mL) and DIPEA (1.2 mmol), and the mixture was shaken overnight at room temperature. After completion, the resin was filtered, washed with DMF and DCM, and dried. The dried resin was subsequently treated with 5 mL of a methanol:DCM:DIPEA mixture (2:7:1) and shaken for 2 $\times$ 10 minutes. The resin was then rinsed with DMF and DCM, dried, and a small portion was taken for resin loading determination.

Fmoc deprotection of the amino acid The dried Resin-Pro-Fmoc was treated with 4 mL of 20% piperidine in DMF. The mixture was shaken for 2 $\times$ 5 minutes, filtered, washed with DMF and DCM, and dried. The dried resin was subjected to the chloranil test. For coupling of the second and subsequent amino acids, Fmoc deprotection was performed using the same procedure with 20% piperidine in DMF.

Coupling of subsequent amino acids to the resin-bound peptide. The resin-bound first amino acid (Resin-Pro-OH) was treated with the next amino acid solution and coupling reagents: DIPEA (3:3:6 equivalents). The mixture was shaken for 4 hours at room temperature. After completion, the resin was filtered, washed with DMF and DCM, dried, and tested with chloranil. The dried resin was then subjected to deprotection. Standard amino acid couplings were carried out using HBTU/HOBt as coupling agents for 4 hours.

Cleavage of the peptide from the resin. The resin containing the linear peptide was treated with 80% trifluoroacetic acid (TFA). The mixture was shaken for 2 hours, filtered, and the filtrate was collected. The filtrate was concentrated using a rotary evaporator. The linear peptide analysed its purity and characterized using HR-TOF-MS.

The *Insilco* prediction was carried out using the [SwissTargetPrediction](#) ([SwissTargetPrediction](#)) operated by the

Molecular Modelling Group of the University of Lausanne and the SIB Swiss Institute of Bioinformatics. The platform applies a ligand-based computational approach to identify potential protein targets of small molecules. The query compound was uploaded in SMILES format, and the system compared its chemical structure against a curated database of bioactive molecules using both 2D and 3D similarity metrics. The output generated ranked lists of predicted targets for *Homo sapiens*, *Rattus norvegicus*, and *Mus musculus*, each accompanied by probability scores and target classifications. This method provides an efficient first step in drug discovery, offering mechanistic hypotheses and guiding experimental validation by situating the compound within established therapeutic pathways [12–14].

Result and Discussion

Synthesis peptide LSAVTPG carried out with SPPS method. The linear peptide was synthesized by initially attaching the first

residue to 2-chlorotrityl chloride resin. Resin loading was determined to quantify the amount of amino acid bound, yielding values of 0.301 mmol/g. This is still an optimum loading resin to process to the next step [15]. Prior to coupling the second residue, the resin was capped with methanol to block unreacted sites. Following capping, the Fmoc protecting group was removed from the resin-bound amino acid to expose the free amine ($-NH_2$), enabling subsequent coupling. Coupling reaction employed HATU/HOAt. Deprotection and coupling cycles were repeated until the complete amino acid sequence was constructed on the resin. The progress of each coupling and deprotection step was qualitatively monitored using the chloranil test. Upon completion of the sequence, the linear peptide was cleaved from the resin with a peptide cleavage cocktail. The filtrate was collected and concentrated under vacuum to afford the heptapeptide. The scheme of LSAVTPG synthesis showed on Figure 2.

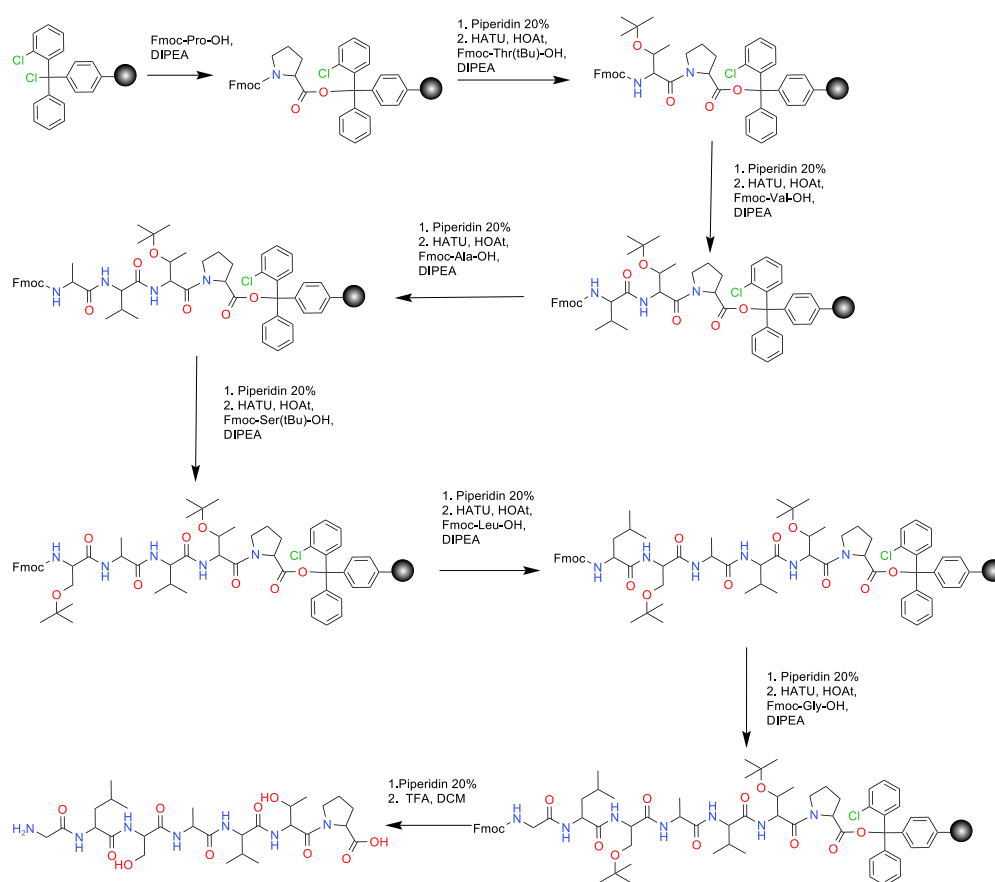


Figure 2. Total synthesis of peptide LSAVTPG

Analytical HPLC revealed retention times of 13.5 minutes for peptide LSAVTPG (Figure 3). Solid-phase synthesis afforded yields of 81.6%. Successful synthesis was confirmed by HR-ToF-MS analysis. The peptide exhibited a molecular ion peak at m/z $[M+H]^+$ 644.3621 (calculated for $C_{28}H_{50}N_7O_{10}$, 644.3619) (Figure 4).

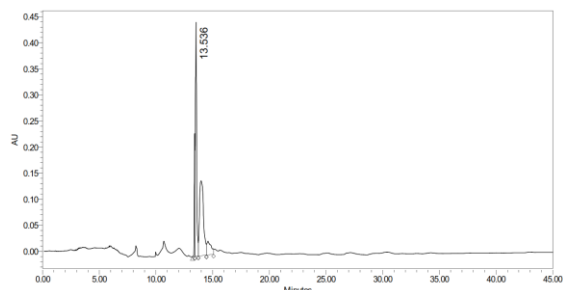


Figure 3. HPLC of peptide LSAVTPG

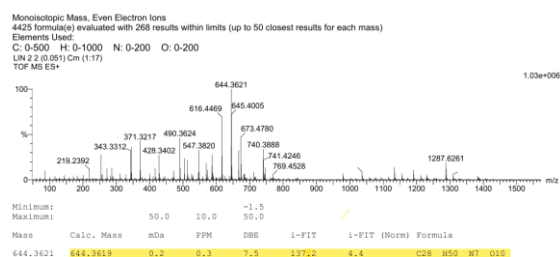


Figure 4. HRMS of peptide LSAVTPG

The successful synthesis of the LSAVTPG peptide provided the foundation for subsequent *in silico* evaluation of its pharmacological potential. Target prediction analysis revealed that this peptide engages multiple protein classes, suggesting a broad spectrum of bioactivity.

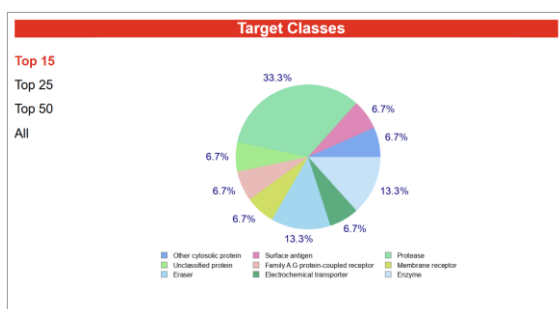


Figure 5. Homo sapiens

In silico target prediction highlights the polypharmacological potential of the LSAVTPG, revealing intersections across oncology, cardiometabolic regulation, neurodegeneration, and immunology (Figure 5). The prioritization of apoptosis regulators such as XIAP, BIRC3, and TRAIL receptor-1 suggests relevance to cancer therapeutics,

where restoration of programmed cell death remains a cornerstone strategy [16,17]. Cardiovascular implications are underscored by predicted interactions with ACE, Renin, and Cathepsin D, aligning with the renin-angiotensin system that underpins hypertension and heart failure management [18]. Metabolic pathways are implicated through DPP4, a clinically validated target in type 2 diabetes, alongside SLC5A1 and SIRT1/2, which are central to glucose homeostasis, aging, and metabolic syndrome. Neurological relevance emerges from BACE1, a pivotal enzyme in amyloid- β generation and Alzheimer's disease pathology, and DLG4, a synaptic scaffolding protein linked to cognition [19,20]. Immunological and inflammatory dimensions are suggested by HLA-A and FPR2, pointing toward potential immunomodulatory activity. Collectively, this constellation of targets situates the compound at the intersection of multiple therapeutic landscapes, offering broad translational promise while emphasizing the necessity of selectivity profiling to delineate its most viable clinical trajectory (Table 1).

Table 1. Top 5 Protein Target in Homo sapiens

No	Target	Common Name	Target class
1	Inhibitor of apoptosis protein 3	XIAP	Other cytosolic protein
2	HLA class I histocompatibility antigen A-3	HLA-A	Surface antigen
3	Dipeptidyl peptidase IV	DPP4	Protease
4	Angiotensin-converting enzyme	ACE	Protease
5	Disks large homolog 4	DLG4	Unclassified protein

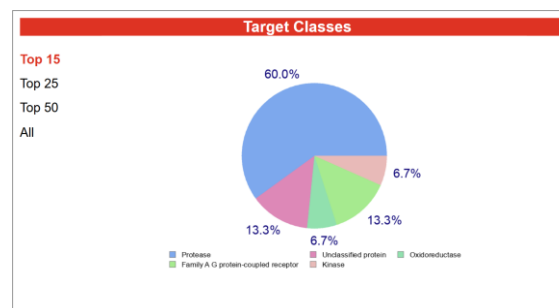


Figure 6. Mus musculus

Computational target prediction in *Mus musculus* provides valuable translational insight into the pharmacological footprint of the LSAVTPG, offering a comparative lens against human targets (Figure 6). The predicted rat proteins span diverse therapeutic landscapes, mirroring pathways of clinical relevance. Apoptosis regulators such as XIAP and BIRC3 suggest conserved mechanisms in cell death modulation, reinforcing their importance in oncology research [21]. Cardiovascular targets, including ACE and Renin, highlight the compound's potential engagement with the renin-angiotensin system, a pathway extensively validated in hypertension and heart failure models. Metabolic implications are evident through DPP4 and SLC5A1, which are central to glucose regulation and have established relevance in diabetes pharmacotherapy [22]. Neurological dimensions emerge from BACE1 and synaptic scaffolding proteins like DLG4, underscoring possible roles in neurodegenerative disease models. Immunological and inflammatory pathways, represented by HLA antigens and FPR2, further broaden the compound's translational scope [23,24]. Taken together, the rat target predictions not only reinforce the compound's multi-pathway potential but also provide a preclinical framework for validating therapeutic hypotheses across oncology, cardiometabolic, neurodegenerative, and immunological domains (Table 2).

Table 2. Top 5 Protein Target in *Mus musculus*

No	Target	Common Name	Target class
1	Dipeptidyl peptidase IV	Dpp4	Protease
2	Angiotensin-converting enzyme	Ace	Protease
3	Formyl peptide receptor-related sequence 1	Fpr-s1	Unclassified protein
4	SGLT1 protein	Slc5a1	Unclassified protein
5	Beta-secretase 1	Bace1	Protease

The successful synthesis of the LSAVTPG peptide enabled subsequent *in silico* evaluation, which revealed a distinct pharmacological footprint across cardiovascular, inflammatory,

and metabolic pathway in *Rattus norvegicus* (Figure 7). Predicted interactions with ACE and REN1 situate the peptide within the renin-angiotensin system, suggesting potential antihypertensive activity and relevance to cardiovascular regulation. The identification of TPP2 highlights possible roles in protein turnover and immune modulation, while PTGS2 (COX-2) implicates the peptide in prostaglandin-mediated inflammatory responses, pointing toward anti-inflammatory or analgesic potential [23]. In addition, the prediction of MME (neprilysin) suggests activity in peptide degradation pathways, with translational implications for both heart failure and amyloid- β metabolism in neurodegeneration [25,26]. (Table 3)

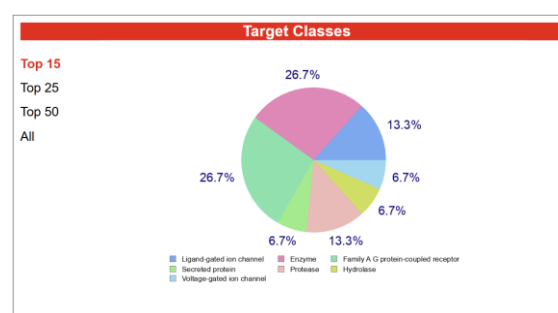


Figure 7. *Rattus norvegicus*

Table 3. Top 5 Protein Target in *Rattus Norvegicus*

No	Target	Common Name	Target class
1	Angiotensin-converting enzyme (by homology)	Ace	Ligand-gated ion channel
2	Tripeptidyl aminopeptidase	Tpp2	Enzyme
3	Renin	Ren1	Family A G protein-coupled receptor
4	Cyclooxygenase-2 (by homology)	Ptgs2	Family A G protein-coupled receptor
5	Neprilysin	Mme	Enzyme

Cross-species target prediction highlights both conserved pharmacological signatures and species-specific nuances, offering a translational framework for assessing the bioactivity of the query compound. In *Homo sapiens*, predicted interactions with apoptosis regulators (XIAP,

BIRC3, TRAIL receptor-1) suggest pro-apoptotic modulation, a mechanism of therapeutic relevance in oncology. Cardiovascular bioactivity is indicated by engagement with ACE and Renin, consistent with modulation of the renin-angiotensin system, while metabolic activity is supported by DPP4, SLC5A1, and SIRT1/2, pointing to potential antidiabetic and metabolic regulatory effects. Neurological bioactivity emerges from BACE1 and DLG4, implicating roles in amyloid processing and synaptic function, respectively. Parallel predictions in *Rattus norvegicus* and *Mus musculus* reinforce these domains, with conserved hits across apoptosis, cardiovascular, and metabolic pathways, thereby strengthening translational confidence. Species-specific predictions, such as Cyclooxygenase-2 (Ptgs2) and Caspase-1 in mouse or Mu opioid receptor (Oprm1) in rat, suggest additional pharmacological axes—anti-inflammatory and analgesic—that may manifest in preclinical models but are less prominent in human predictions. Collectively, the compound demonstrates a polypharmacology profile with potential bioactivity spanning oncology, cardiometabolic regulation, neurodegeneration, immunology, and inflammation. This constellation of targets underscores broad therapeutic promise while emphasizing the importance of selectivity profiling and comparative pharmacology to delineate clinically viable pathways [27–29].

In silico target prediction platforms such as SwissTargetPrediction provide valuable first-line hypotheses for ligand–target interactions; however, their utility is constrained by several inherent limitations. Predictions are primarily driven by chemical similarity to known ligands, which restricts accuracy when compounds fall outside the established chemical space. Moreover, the database coverage is biased toward well-studied proteins, leaving understudied or novel targets underrepresented. The tool's scope is limited to a few model organisms (*Homo sapiens*, *Mus musculus*, and *Rattus norvegicus*), thereby reducing translational relevance across broader biological systems. Importantly, these predictions do not account for physiological complexity, such as tissue-specific expression,

pathway crosstalk, or dynamic regulation, and are therefore prone to both false positives and false negatives. Consequently, while SwissTargetPrediction serves as a useful exploratory resource, experimental validation remains indispensable to confirm biological relevance and therapeutic potential.

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Conclusion

The linear heptapeptide LSAVTPG was successfully synthesized using solid-phase peptide synthesis (SPPS) on 2-chlorotriyl chloride resin. The synthesis yielded 81.6% of the target peptide, confirming the efficiency and accuracy of the SPPS strategy. The cross-species target predictions consistently highlight conserved pathways in apoptosis, cardiovascular regulation, and metabolism, underscoring the compound's broad translational potential. At the same time, species-specific targets reveal additional pharmacological dimensions, reinforcing the importance of comparative validation to guide selective therapeutic development.

Author Contribution

A.A.A - conceptualization, methodology design, investigation, data curation, formal analysis, writing – original draft preparation, and supervision. V.I.P - supporting data collection, conducting the literature review, and contributing to writing – review and editing. S.D - experimental procedures, contributed to validation, and assisted in visualization.

Reference

1. [Cen-Pacheco, F.; Valerio-Alfaro, G.; Santos-Luna, D.; Fernández, J.J. Sclerin, a New Cytotoxic Cyclonapeptide from *Annona Scleroderma*. *Molecules* **2019**, *24*, 554, doi:10.3390/molecules24030554.](#)
2. [Wélé, A.; Zhang, Y.; Caux, C.; Brouard, J.-P.; Dubost, L.; Guette, C.; Poussel, J.-L.; Badiane, M.; Bodo, B. Isolation and Structure of Cycloenegalins A and B.](#)

- Novel Cyclopeptides from the Seeds of *Annona Senegalensis*. *J. Chem. Soc., Perkin Trans.* **1** **2002**, 2712–2718, doi:10.1039/B205035H.
3. Aloanis, A.A.; Herlina, T.; Hardianto, A.; Maharani, R. Total Synthesis of Cyclosenegalinal A. *ChemistryOpen* **2024**, *13*, e202400175, doi:10.1002/open.202400175.
4. Aloanis, A.A.; Herlina, T.; Hardianto, A.; Maharani, R. Structural Design and Synthesis of Cyclosenegalinal A Analogue. *Journal of Heterocyclic Chemistry* **2025**, *62*, 781–790, doi:10.1002/jhet.70020.
5. Paat, V.I.; Aloanis, A.A.; Najohan, J.M.J. Molecular Docking Of Cyclosenegalinal A As Anticancer. *Fullerene Journal of Chemistry* **2025**, *10*, 26–33, doi:10.37033/fjc.v10i1.713.
6. Aloanis, A.A.; Najohan, J.M.J.; Paat, V.I.; Rumengan, S.M.; Rumampuk, R.I. Total Synthesis and Molecular Docking Study of Peptide AWVDY as an Anti-Inflammation Agent. *JKPK (Jurnal Kimia dan Pendidikan Kimia)* **2025**, *10*, 234–254.
7. Aloanis, A.A.; Herlina, T.; Hardianto, A.; Maharani, R. Alanine-Rich Cyclopeptides: Natural Resources, Bioactivity, Total Synthesis, and Spectroscopy Identification. *ChemistrySelect* **2025**, *10*, e202404379, doi:10.1002/slct.202404379.
8. Aloanis, A.A.; Paat, V.I. SINTESIS SIKLOPEPTIDA. *Penerbit Tahta Media* **2024**.
9. Morena, F.; Cencini, C.; Calzoni, E.; Martino, S.; Emiliani, C. A Novel Workflow for In Silico Prediction of Bioactive Peptides: An Exploration of *Solanum Lycopersicum* By-Products. *Biomolecules* **2024**, *14*, doi:10.3390/biom14080930.
10. Daina, A.; Michielin, O.; Zoete, V. SwissTargetPrediction: Updated Data and New Features for Efficient Prediction of Protein Targets of Small Molecules. **2019**, *47*.
11. Daina, A.; Zoete, V. Testing the Predictive Power of Reverse Screening to Infer Drug Targets, with the Help of Machine Learning. *Commun Chem* **2024**, *7*, 105, doi:10.1038/s42004-024-01179-2.
12. Atasever, S. In Silico Drug Discovery: A Machine Learning-Driven Systematic Review. *Med Chem Res* **2024**, *33*, 1465–1490, doi:10.1007/s00044-024-03260-w.
13. Brogi, S.; Ramalho, T.C.; Kuca, K.; Medina-Franco, J.L.; Valko, M. Editorial: In Silico Methods for Drug Design and Discovery. *Front. Chem.* **2020**, *8*, doi:10.3389/fchem.2020.00612.
14. Maia, E.H.B.; Assis, L.C.; de Oliveira, T.A.; da Silva, A.M.; Taranto, A.G. Structure-Based Virtual Screening: From Classical to Artificial Intelligence. *Front. Chem.* **2020**, *8*, doi:10.3389/fchem.2020.00343.
15. Nicolau, I.; Paun, A.; Popescu, C.C.; Hădăde, N.D.; Matache, M. Solid-Phase Peptide Synthesis – Evaluation of Resin Loading and Preparation of an Amide C-Terminal Dipeptide. *J. Chem. Educ.* **2023**, *100*, 2430–2434, doi:10.1021/acs.jchemed.3c00186.
16. Fan, H.; Liu, J.; Hu, X.; Cai, J.; Su, B.; Jiang, J. The Critical Role of X-Linked Inhibitor of Apoptosis Protein (XIAP) in Tumor Development. *Apoptosis* **2025**, *30*, 1202–1215, doi:10.1007/s10495-025-02101-4.
17. Tu, H.; Costa, M. XIAP's Profile in Human Cancer. *Biomolecules* **2020**, *10*, doi:10.3390/biom10111493.
18. Pop, D.; Dădârlat-Pop, A.; Tomoaia, R.; Zdrengea, D.; Caloian, B. Updates on the Renin–Angiotensin–Aldosterone System and the Cardiovascular Continuum. *Biomedicines* **2024**, *12*, doi:10.3390/biomedicines12071582.
19. Vassar, R.; Kandalepas, P.C. The β -Secretase Enzyme BACE1 as a Therapeutic Target for Alzheimer's Disease. *Alz Res Therapy* **2011**, *3*, 20, doi:10.1186/alzrt82.
20. Lee, J.; Cho, Y.; Choi, B.Y.; Kim, H.K.; Lee, Y.; Kim, E.; Han, J.; Sul, J.H.; Kim, J.-S.; Baek, S.H.; et al. HDAC6 Regulates BACE1 Stability and NLRP3 Inflammasome Activation in Alzheimer's Disease. *Brain* **2026**, awag089, doi:10.1093/brain/awag089.
21. Luo, Q.; Zhu, H.; Li, Y.; Wu, Q.; Sun, J.; Zhou, F. Expression Analysis of BIRC3 as One Target Gene of Transcription Factor NF- κ B for Esophageal Cancer. *Processes* **2022**, *10*, doi:10.3390/pr10091673.

22. [Deacon, C.F. Corrigendum: Physiology and Pharmacology of DPP-4 in Glucose Homeostasis and the Treatment of Type 2 Diabetes. *Front. Endocrinol.* **2019**, *10*, doi:10.3389/fendo.2019.00275.](#)
23. [Wang, J.; Miao, Z.; Gao, Y.; Xie, Z.; Liu, M.; Zou, W. Formyl Peptide Receptor 2: A Potential Therapeutic Target for Inflammation-Related Diseases. *Pharmacol. Rep* **2025**, *77*, 593–609, doi:10.1007/s43440-025-00704-x.](#)
24. [Ruiz-Pablos, M.; Paiva, B.; Zabaleta, A. The Origin of Autoimmune Diseases: Is There a Role for Ancestral HLA-II Haplotypes in Immune Hyperactivity. *Front. Immunol.* **2025**, *16*, doi:10.3389/fimmu.2025.1710571.](#)
25. [Richards, A.M.; Pemberton, C.; Yandle, T. How Does Neprilysin Inhibition in Heart Failure Work? *Eur J Heart Fail* **2019**, *21*, 606–608, doi:10.1002/ehf.1454.](#)
26. [Morito, T.; Hashimoto, S.; Takamura, R.; Watamura, N.; Kakiya, N.; Fujioka, R.; Mihara, N.; Sekiguchi, M.; Watanabe-Iwata, K.; Kamano, N.; et al. The Role of Neprilysin and Insulin-Degrading Enzyme in the Etiology of Sporadic Alzheimer's Disease. *J. Neurosci.* **2025**, *45*, doi:10.1523/JNEUROSCI.2152-24.2025.](#)
27. [Friman, T.; Tolvanen, T.; Boström, T.; Sun, Y.; Chernobrovkin, A.; Andersson, Y.; Velasco, C.; Molina, D.M. Abstract 6853: A Standardized Drug Selectivity Profiling Platform with a Proteome Coverage of More than 5,000 Proteins. *Cancer Res* **2025**, *85*, 6853, doi:10.1158/1538-7445.AM2025-6853.](#)
28. [Wang, T.; Pulkkinen, O.I.; Aittokallio, T. Target-Specific Compound Selectivity for Multi-Target Drug Discovery and Repurposing. *Front. Pharmacol.* **2022**, *13*, doi:10.3389/fphar.2022.1003480.](#)
29. [Lica, J.J.; Wieczór, M.; Grabe, G.J.; Heldt, M.; Jancz, M.; Misiak, M.; Gucwa, K.; Brankiewicz, W.; Maciejewska, N.; Stupak, A.; et al. Effective Drug Concentration and Selectivity Depends on Fraction of Primitive Cells. *International Journal of Molecular Sciences* **2021**, *22*, doi:10.3390/ijms22094931.](#)



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