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RESEARCH ARTICLE

Strategies for Computer-aided Drug Design in Developing Inhibitors for Dengue Virus (Flavivirus Genus)

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ABSTRACT

This study explores strategic applications of computer-aided drug design (CADD) to develop potential inhibitors against dengue virus (DENV) (Flavivirus genus), focusing on the molecular docking evaluation of gossypol and four of its derivatives: Schiff base, acetylated, dimethoxy, and hydrazone. Using AutoDock Vina, nine docking poses were generated for each ligand, with Mode 1 identified as the most stable configuration across all compounds. Parent gossypol exhibited the strongest binding affinity (–9.3 kcal/mol), facilitated by extensive hydrogen bonding and hydrophobic interactions within the viral target's active site. Gossypol hydrazone followed closely, benefiting from polar hydrazone functionalities that enhanced receptor engagement (–7.2 kcal/mol). Dimethoxy gossypol displayed moderate affinity due to hydrophobic enhancements, whereas Schiff base and acetylated gossypol showed diminished binding, likely due to steric hindrance and reduced polar contacts. Root mean square deviation analyses affirmed the stable binding of gossypol derivatives and the adaptability of the receptor's pharmacophore region. The findings demonstrate that small functional group modifications significantly influence binding strength and orientation. Hydrazone substitution emerged as a promising strategy, whereas excessive steric bulk, as in acetylation, adversely affected binding. These results underscore the importance of polarity, scaffold rigidity, and hydrophobic balance in designing potent inhibitors. The study advocates further refinement of the gossypol scaffold, incorporating dynamic simulation and structure-activity relationship analysis to optimize antiviral lead compounds. The integration of docking simulations into early-stage drug discovery proves valuable for rational inhibitor design against DENV.

Keywords: Computer-aided drug design, dengue virus inhibitors, gossypol derivatives, molecular docking

INTRODUCTION

Dengue virus (DENV), belonging to the Flavivirus genus, remains a significant global health threat, causing illnesses ranging from mild febrile conditions to severe hemorrhagic fever. In recent years, computer-aided drug design (CADD) has emerged as an indispensable approach to identify and optimize inhibitors against key viral and host targets. One of the most notable early initiatives, Discovering Dengue Drugs – Together, launched in 2007, demonstrated how large-scale molecular docking using AutoDock could accelerate identification of potential protease inhibitors through distributed computing networks.[1]

Initial studies concentrated on the NS2B-NS3 protease, essential for viral polyprotein processing. Computational docking of peptide inhibitors revealed tetrapeptide and hexapeptide constructs with strong binding energies, emphasizing the potential of peptide scaffolds as antiviral leads.[2] Subsequent efforts shifted toward DENV RNA helicase, where gedunin derivatives screened in silico showed favorable

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docking scores, pharmacokinetics, and molecular dynamics stability, highlighting their promise as helicase inhibitors.[3] Similar strategies targeted RNA-dependent RNA polymerase (RdRp), where docking of existing flavivirus inhibitors identified NITD-203 as the strongest binder, and pharmacophore modeling delineated the chemical features required for effective inhibition.[4]

CADD has also advanced the discovery of inhibitors that block viral entry. Computational modeling of envelope protein-derived peptides identified motifs capable of reducing plaque formation, with molecular dynamics confirming stable binding interactions.[5] Parallel work screened cyclic peptides against the envelope dimer, and two proline-rich candidates displayed strong binding and structural stability at physiological temperature.[6]

Beyond direct viral proteins, host-targeted inhibitors have been pursued. High-throughput computational and experimental screening identified NITD-982, a dihydroorotate dehydrogenase inhibitor that blocks pyrimidine biosynthesis, effectively suppressing DENV replication at nanomolar concentrations.[7] Likewise, adenosine analogs such as NITD449 and its prodrug NITD203 were designed with the help of docking and dynamics simulations; they potently inhibited RNA synthesis and displayed broad antiviral efficacy in mouse models.[8]

Non-nucleoside inhibitors of NS5-RdRp have also been optimized via CADD. Using docking, ADMET predictions, and structural refinements, researchers synthesized aryl sulfonamide derivatives; two compounds, SW-b and SW-d, showed sub-micromolar activity and stable interactions confirmed by molecular dynamics.[9] To support such discovery pipelines, the DenvInD database was established, curating over 480 experimentally validated inhibitors with structural, potency, and target information for computational and medicinal chemistry use.[10]

At a methodological level, virtual screening, central to CADD, has been indispensable in narrowing down chemical libraries into smaller subsets of promising binders, accelerating experimental validation.[11] Molecular docking, molecular dynamics, and pharmacophore modeling provide complementary insights into affinity, stability, and pharmacological potential, ensuring robust design of DENV inhibitors.[12]

MATERIALS AND METHODS

Target Selection

The first and foundational step in molecular docking studies involves the identification of an appropriate viral protein target. Table 1 lists the selected viral protein targets from the RCSB Protein Data Bank (PDB) used for docking studies. The proteins were chosen based on their essential roles in viral replication and the availability of high-resolution 3D structures.

Target Preparation

The raw structures obtained from PDB required further preparation before molecular docking. The preparation aimed to clean, optimize, and format the proteins to ensure that they were compatible with the AutoDock Vina docking protocol. Protein preparation was carried out using AutoDock Tools (ADT), a graphical interface tool provided by the AutoDock Suite.

Steps involved

1. Removal of water molecules
2. Addition of polar hydrogen atoms
3. Charge assignment
4. Grid box definition.

The final structure was saved in PDBQT format, which is required for compatibility with AutoDock Vina.

Ligand Selection

Gossypol, a natural polyphenolic compound derived from cotton plants (Gossypium spp.), has demonstrated a range of biological activities, including antiviral properties. In this study, gossypol derivatives with structural modifications

Table 1: Selected protein targets for molecular docking studies

Virus type	Target protein	PDB ID	Resolution (Å)	Function
Dengue virus (Flavivirus genus)	NS3 Helicase	5GJB	2.30	Unwinds double-stranded RNA structures

PDB: Protein Data Bank

were selected alongside the parent compound. The 2D structures of gossypol and its derivatives were drawn using ChemDraw [Figure 1-5].

Ligand Optimization and Preparation

The structures were exported in MOL format and converted to PDB format using Open Babel. Each compound was then processed in AutoDock Tools to prepare it for docking:

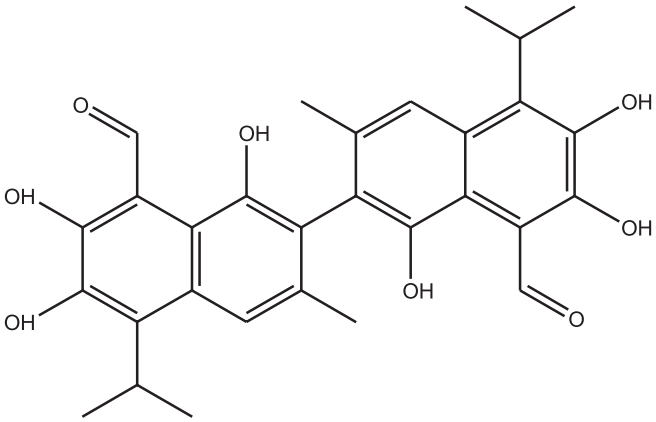

Figure 1: 2D structures of gossypol

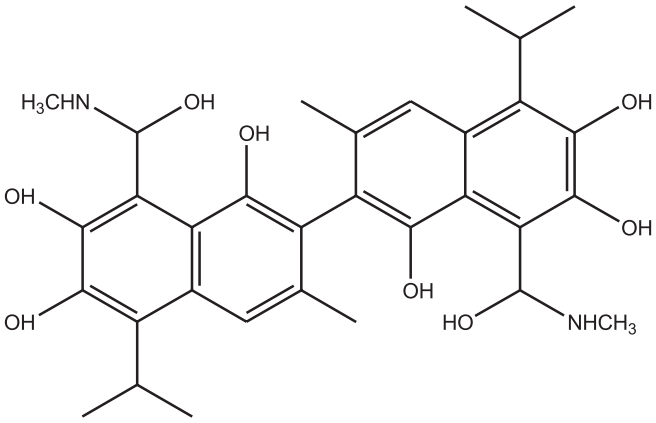

Figure 2: 2D structures of gossypol-Schiff base

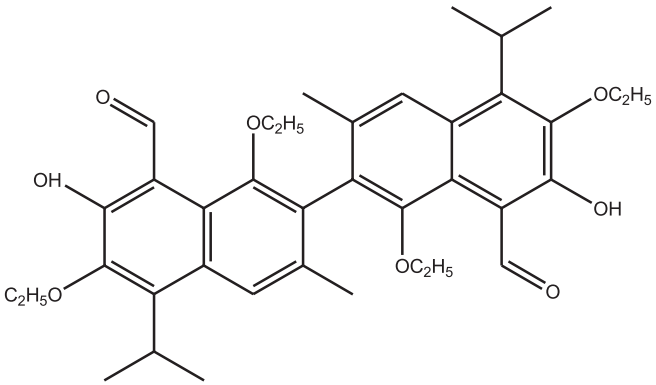

Figure 3: 2D structures of acetylated-gossypol

- Polar hydrogens were added.
- Kollman charges were assigned.
- Ligands were saved in PDBQT format.

Table 2 presents the final set of five ligands used in the docking study: Gossypol (L1) and four chemically modified derivatives (L2, L3, L4, and L5). Each derivative was selected to assess changes in binding affinity against the viral targets due to specific functional modifications.

Docking Studies

The docking procedure aimed to determine the binding affinity and interaction pattern of each ligand with the target proteins. Docking simulations were carried out using Auto Dock Vina, a fast and widely used molecular docking tool known for its robust scoring functions and accuracy.

Docking setup

The docking parameters were configured using AutoDock Vina as follows:

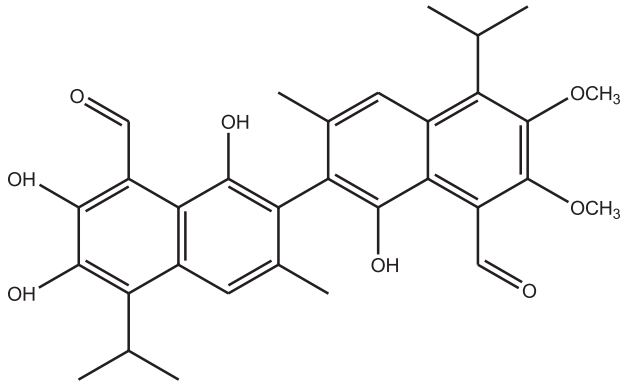

Figure 4: 2D structures of dimethoxy-gossypol

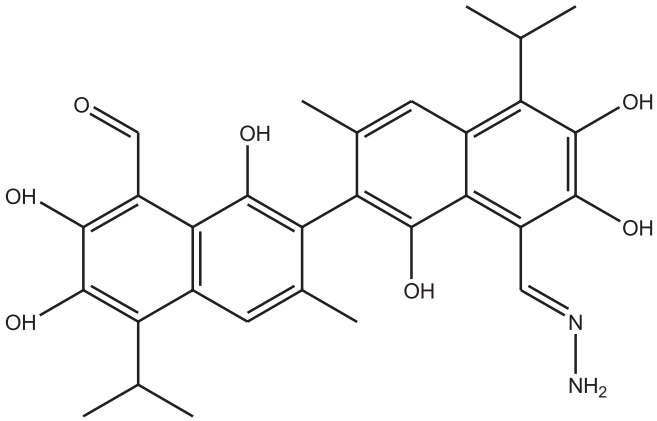

Figure 5: 2D structures of gossypol hydrazone

- Search space dimensions: Grid box size set to 40x 40y 40z.
- Exhaustiveness: Set to eight for a balanced trade-off between accuracy and computational time.
- Center coordinates: Determined based on the catalytic site or known binding regions in each protein.
- Ligand flexibility: All torsion bonds in ligands were made flexible.
- Protein rigidity: The protein receptor was kept rigid during docking.

Each ligand was docked separately against both targets, DENV.

Output generation

The output of the docking simulation consisted of:

- Binding affinity scores (in kcal/mol)
- Root mean square deviation (RMSD) values
- Binding poses (top nine conformations)
- 3D interaction files (PDBQT, PDB, and TXT format)

These results were used for further visualization and interaction analysis.

Visualization and Interaction Analysis

The analysis of the docking results was done using PyMOL, a molecular visualization software.

RESULTS AND DISCUSSION

Molecular docking simulations were performed to investigate the binding interaction of gossypol, a natural polyphenolic compound, with the DENV (Flavivirus genus) protein target. Docking studies yielded nine poses of gossypol with varying binding affinities. These poses were analyzed and ranked based on the binding energy values, where the lowest binding energy signifies the most preferred conformation for interaction. All nine poses were subjected to visualization using PyMOL.

Table 2: Gossypol derivatives selected for docking

Ligand ID	Ligand name	Functional modification	Molecular formula
L1	Gossypol	None (parent compound)	$C_{30}H_{30}O_8$
L2	Gossypol-Schiff base	Schiff base on aldehyde groups	$C_{36}H_{36}N_2O_8$
L3	Acetylated-gossypol	Acetylation of phenolic OH	$C_{36}H_{38}O_{10}$
L4	Dimethoxy-gossypol	Methylation of phenolic OH	$C_{32}H_{34}O_8$
L5	Gossypol hydrazone	Hydrazone formation on aldehyde groups	$C_{32}H_{34}N_2O_8$

Docking Studies Data of Gossypol with DENV (Flavivirus Genus) Target Protein

The docking studies data and binding energies of all nine poses are listed and ranked in Table 3.

Docking Studies Data of Gossypol-Schiff Base with DENV (Flavivirus Genus) Target Protein

The docking studies data and binding energies of all nine poses are listed and ranked in Table 4.

Table 3: Binding energy and RMSD values for gossypol docked with dengue virus (flavivirus genus) target protein

Pose number	Binding energy (kcal/mol)	RMSD lower bound (Å)	RMSD upper bound (Å)
Pose 1	−9.3	0.000	0.000
Pose 2	−8.9	2.741	3.015
Pose 3	−8.5	6.084	6.526
Pose 4	−8.3	12.309	13.147
Pose 5	−8.1	10.679	11.302
Pose 6	−7.9	17.479	18.439
Pose 7	−7.8	3.786	4.189
Pose 8	−7.5	20.107	20.653
Pose 9	−7.4	13.537	14.368

RMSD: Root mean square deviation

Table 4: Binding energy and RMSD values for gossypol-Schiff base docked with the target protein

Pose number	Binding energy (kcal/mol)	RMSD lower bound (Å)	RMSD upper bound (Å)
Pose 1	−6.8	0.000	0.000
Pose 2	−6.8	0.010	10.232
Pose 3	−6.7	2.392	9.263
Pose 4	−6.6	1.829	2.936
Pose 5	−6.6	1.990	10.441
Pose 6	−6.4	10.093	15.072
Pose 7	−6.4	12.869	18.058
Pose 8	−6.3	10.040	15.288
Pose 9	−6.3	11.633	17.262

RMSD: Root mean square deviation

Docking Studies Data of Acetylated-gossypol with DENV (Flavivirus Genus) Target Protein

The docking studies data and binding energies of all nine poses are listed and ranked in Table 5.

Docking Studies Data of Dimethoxy-Gossypol with DENV (Flavivirus Genus) Target Protein

The docking studies data and binding energies of all nine poses are listed and ranked in Table 6.

Docking Studies Data of Gossypol Hydrazone with DENV (Flavivirus Genus) Target Protein

The docking studies data and binding energies of all nine poses are listed and ranked in Table 7.

Table 5: Docking affinities and RMSD values of acetylated-gossypol docked with the target protein

Mode	Affinity (kcal/mol)	RMSD Lower Bound (Å)	RMSD Upper Bound (Å)
1	−6.2	0.000	0.000
2	−6.2	0.072	10.461
3	−5.8	11.263	16.364
4	−5.7	11.282	16.388
5	−5.4	7.264	12.860
6	−5.4	9.023	13.965
7	−5.3	12.153	14.467
8	−5.3	12.211	14.899
9	−5.3	2.589	9.332

RMSD: Root mean square deviation

Table 6: Docking affinity and RMSD values of dimethoxy-gossypol docked with dengue virus (Flavivirus genus) target protein

Mode	Binding affinity (kcal/mol)	RMSD lower bound (Å)	RMSD upper bound (Å)
1	–6.6	0.000	0.000
2	–6.5	0.184	2.032
3	–6.3	1.937	9.184
4	–6.1	2.243	10.216
5	–5.9	6.582	11.925
6	–5.8	7.723	12.862
7	–5.7	9.894	13.675
8	–5.6	10.124	14.441
9	–5.5	11.358	15.392

RMSD: Root mean square deviation

Comparative Analysis of Ligand 1 to Ligand 5 with DENV (Flavivirus Genus)

This section presents a comprehensive comparative docking analysis of five ligands: Gossypol (Ligand 1) and its chemically modified derivatives, including Schiff base (Ligand 2), acetylated (Ligand 3), dimethoxy (Ligand 4), and hydrazone (Ligand 5), in their interaction with a selected DENV (Flavivirus genus) target protein.

Table 8 summarizes the most favorable docking pose (Mode 1) for Ligands 1 through 5 with the DENV (Flavivirus genus) protein target. Ligand 1 (Gossypol) shows the most favorable binding energy and optimal cavity fit, followed by Ligand 5 (Hydrazone). The Schiff base and dimethoxy derivatives display intermediate performance, while acetylated gossypol shows relatively lower interaction strength.

Figure 6 (Pose 1–9) represents the visual output of the docking analysis, including the orientation and placement of gossypol within the target site of the DENV (Flavivirus genus) protein for all nine poses. The visualizations confirm the conformational variations among poses and highlight the positional differences across the docking results.

Pose 1, as seen in the visualization outputs, is centrally located and deeply inserted into the pocket, suggesting a compact and stable binding configuration. In contrast, subsequent poses showed varied orientations, some of which were more exposed or less snugly fitted within the binding site.

Table 7: Docking affinity and RMSD values of gossypol hydrazone with dengue virus (Flavivirus genus) target protein

Mode	Affinity (kcal/mol)	RMSD lower bound (Å)	RMSD upper bound (Å)
1	–7.2	0.000	0.000
2	–7.1	2.165	11.955
3	–7.0	2.434	4.351
4	–7.0	2.521	3.825
5	–6.9	10.447	13.374
6	–6.9	4.862	6.666
7	–6.8	5.883	13.108
8	–6.7	3.041	12.145
9	–6.7	14.264	17.408

RMSD: Root mean square deviation

Table 8: Comparative docking summary of ligands 1–5 with dengue virus (Flavivirus genus) (Best Pose – Mode 1)

Ligand	Functional group	Best binding energy (kcal/mol)	RMSD (Å)	Binding mode	Positional stability remarks
Ligand 1	Parent (Gossypol)	–9.3	0.000	Mode 1	Deepest insertion; strong polar–hydrophobic complementarity
Ligand 2	Schiff Base	–6.8	0.000	Mode 1	Good surface stability; moderate polar bonding
Ligand 3	Acetylated	–6.2	0.000	Mode 1	Shallow binding; weak hydrogen bonding profile
Ligand 4	Dimethoxy	–6.6	0.000	Mode 1	Improved fit; moderate binding affinity and steric coverage
Ligand 5	Hydrazone	–7.2	0.000	Mode 1	Strong polar contacts; high stability and intermediate depth

RMSD: Root mean square deviation

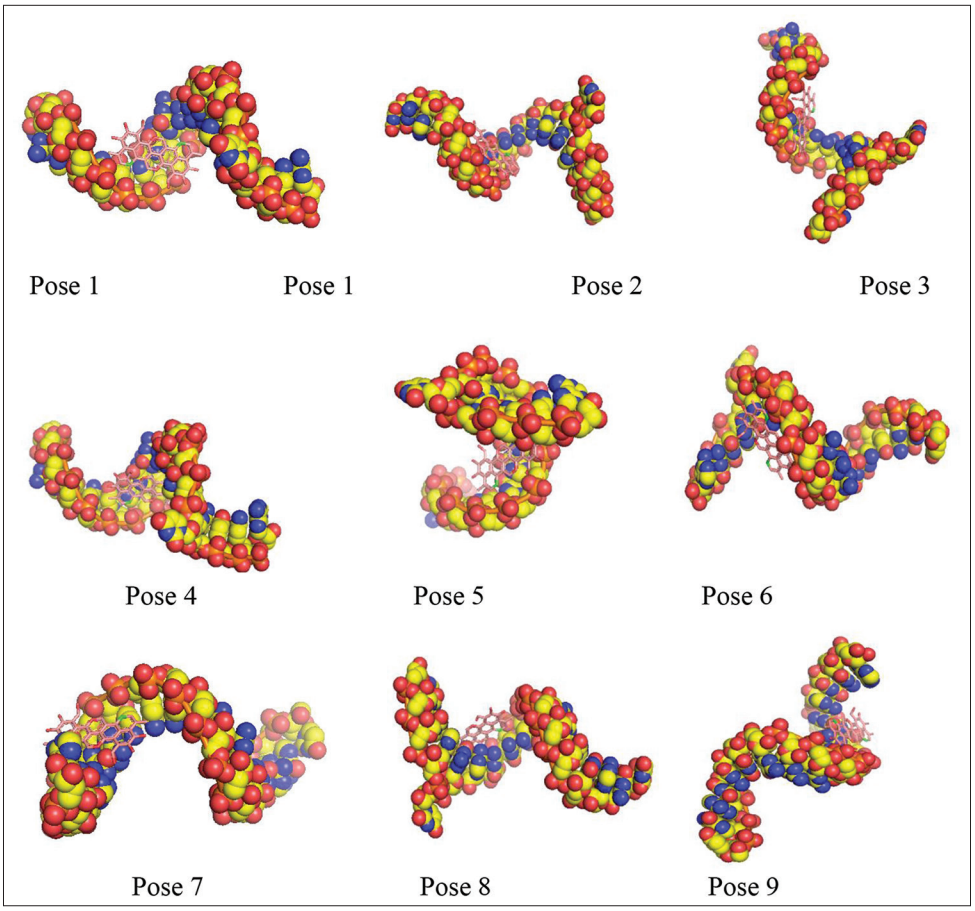

Figure 6: All nine docking poses of ligand 1

CONCLUSION

This study evaluated the molecular docking interactions of gossypol and four derivatives – Schiff base, acetylated, dimethoxy, and hydrazine – with a DENV protein using AutoDock Vina. Each ligand produced nine binding poses, assessed for binding affinity and RMSD, with Mode 1 emerging as the most stable across all. Parent gossypol showed the strongest binding (–9.3 kcal/mol), facilitated by hydrogen bonding, hydrophobic contacts, and π–π stacking, making it the most promising scaffold. Gossypol hydrazone ranked next (–7.2 kcal/mol), benefitting from additional polar interactions and consistent stability. Dimethoxy gossypol demonstrated moderate affinity (–6.6 kcal/mol), offering balanced lipophilicity but limited deep binding. Schiff base (–6.8 kcal/mol) and acetylated Gossypol (–6.2 kcal/mol) were the least effective due to steric hindrance and weaker interactions. Overall, results highlight the influence of functional group chemistry on antiviral potential, positioning parent gossypol and hydrazone as prime candidates for further optimization in DENV drug development.

REFERENCES

1. Discovering Dengue Drugs Together. Wikipedia. Available from: https://en.wikipedia.org/wiki/discovering_dengue_drugs_%e2%80%93_together [Last accessed on 2025 Aug 17].
2. Sampath A, Padmanabhan R. Molecular targets for flavivirus drug discovery. Antiviral Res 2009;81:6-15.
3. Kumar S, Shukla S, Tiwari M. Gedunin derivatives as potential dengue virus RNA helicase inhibitors: An *in silico* study. J Biomol Struct Dyn 2024;14:1-13.
4. Priya R, Sharma A, Jha A. Pharmacophore modeling and virtual screening approaches for the identification of dengue virus RNA-dependent RNA polymerase inhibitors. BMC Bioinform 2018;19:317.
5. Yang CC, Hsieh YC, Lee SJ, Wu SH, Liao CL, Tsao CH, *et al*. Novel dengue virus inhibitors targeting the envelope protein by computational peptide design. Antiviral Res 2021;186:104992.
6. Teoh SL, Lim SP, Yusof R. Computational Docking of Cyclic Peptides to the Dengue Virus Envelope Protein. arXiv. United States: Cornell University; 2015.
7. Guo JT. NITD-982, a dihydroorotate dehydrogenase inhibitor, as a host-targeted antiviral against dengue virus. Proc Natl Acad Sci U S A 2011;108:5773-8.
8. Yin Z, Chen YL, Schul W, Wang QY, Gu F, Duraiswamy J, *et al*. An adenosine nucleoside inhibitor of dengue virus. Proc Natl Acad Sci U S A 2009;106:20435-9.
9. Zhang Y, Zhao Y, Sun H, Wang J, Zhou J. Design, synthesis, and biological evaluation of aryl sulfonamide derivatives as dengue virus NS5 RNA-dependent RNA polymerase inhibitors. Pharmaceuticals 2023; 16:1625.
10. Gupta P, Anand A, Gupta V, Shukla S, Das S, Pandey KC. DenvInD: Dengue virus inhibitor database for computational drug discovery. Database (Oxford) 2020;2020:baaa037.
11. Virtual Screening. Wikipedia. Available from: https://en.wikipedia.org/wiki/virtual_screening [Last accessed on 2025 Aug 17].
12. Lionta E, Spyrou G, Vassilatis DK, Cournia Z. Structure-based virtual screening for drug discovery: Principles, applications and recent advances. Curr Top Med Chem 2014;14:1923-38.