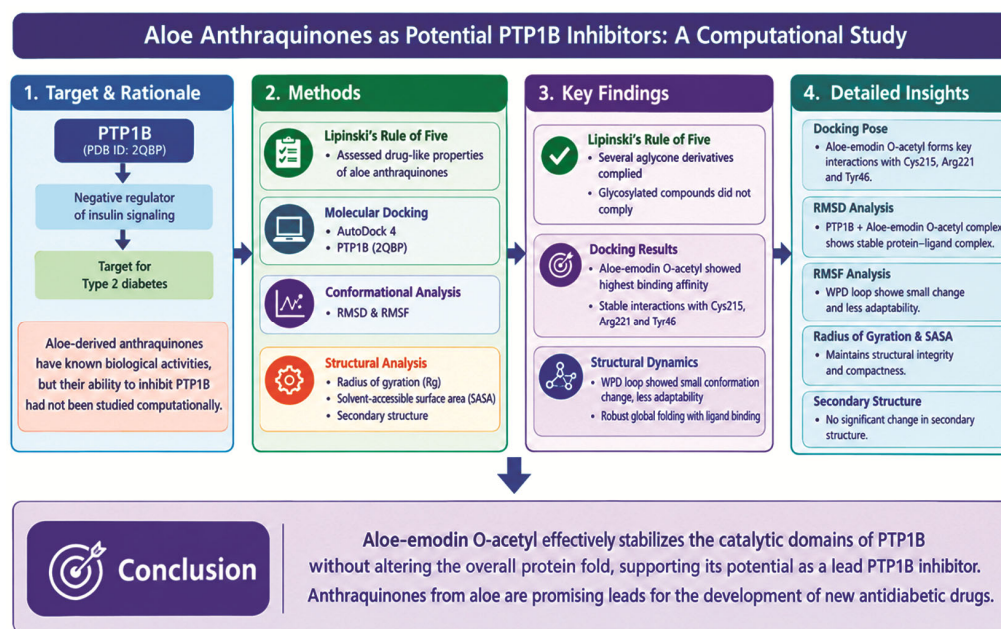




Graphical abstract

Molecular docking

Molecular docking studies were carried out using AutoDock 4⁹ with the target protein PDB ID: 2QBP. The study employed AutoDock 4.9 for molecular docking, focusing on the target protein with PDB ID: 2QBP. The protein's crystal structure was retrieved from the Protein Data Bank. Any ligands attached to protein and water that had crystallized with it were removed. Using PubChem, the structures of aloe anthraquinone were obtained and changed them into PDB format. The protein and ligands were made ready by adding polar hydrogens and Gasteiger charges. Binding affinity and mode of interactions were identified.

RMSD

We used Python-based tools that ran in Google Colaboratory to do the RMSD analysis. We uploaded structural files directly to the Colab environment and used MDAnalysis (v2.10.0) and Biopython (v1.86) 10 to do the analyses¹⁰. We chose heavy atoms of the ligand, leaving out hydrogens, to make sure that the comparisons were chemically meaningful. We calculated normal RMSD without aligning the structures. Kabsch and quaternion RMSD values, on the other hand, were calculated after the best superposition to get rid of rigid-body translations and rotations. We used the MDAnalysis RMSD module to find the Quaternion RMSD and the Biopython Superimposer class to find the Kabsch RMSD. All RMSD values were given in ångströms (Å).

RMSF

Root Mean Square Fluctuation (RMSF) analysis was performed using the CABS-flex 3.0 web server to evaluate protein flexibility¹¹. The PDB structure of the docked protein–ligand complex was submitted as input. CABS-flex simulations were carried out under default parameters, and residue-wise RMSF values were generated to assess ligand-induced changes in protein dynamics and structural stability.

Radius of gyration analysis

Radius of gyration (Rg) analysis was performed using the Sanjeevni Radius of Gyration tool hosted at the Supercomputing Facility for Bioinformatics and Computational Biology (SCFBio), IIT Delhi (<https://scfbio-iitd.res.in/software/proteomics/rgnew.jsp>)⁸. Protein and protein–ligand complex structures in PDB format were uploaded, and Rg was calculated as the root mean square distance of all atoms from the center of mass. The obtained Rg values were evaluated using the empirical relationship $R_g = 0.395 \times N^{3/5} + 7.257$, where N denotes the number of amino acid residues, to assess conformity with globular protein behavior.

Accessible surface area

Accessible Surface Area (ASA) and Relative Accessibility of protein residues were computed using the Accessible Surface Area and Accessibility Calculation for Protein (ver. 1.2) online tool (<http://cib.cf.ocha.ac.jp/bitool/ASA/>). This server

Table 1 — Lipinski rule of 5 for Aloe anthraquinones

Compound Name	Molecular Weight (Da)	H-Bond Donors	H-Bond Acceptors	logP
8-Hydroxy aloe emodin	300.0	4	6	1.11
10-Hydroxy aloe emodin	300.0	4	6	1.11
Aloe emodin	270.0	3	5	1.37
Aloe emodin anthranol	272.0	4	4	1.62
Aloe emodin anthrone	256.0	3	4	1.73
Aloe emodin O-acetyl	326.0	2	6	1.98
Aloin A	434.0	7	9	−0.91
Aloin B	434.0	7	9	−0.91
Iso-aloe emodin	284.0	3	5	1.41

implements a geometric algorithm that estimates the solvent-exposed surface of residues by rolling a virtual solvent probe (typically with a radius of 1.4 Å) over the protein's van der Waals surface. The calculation identifies regions of each amino acid that are accessible to solvent, expressed as absolute ASA (in Å²) and normalized relative accessibility (fraction of maximal exposure), based on reference maximal ASA values for each residue type.

Secondary structure comparison

Secondary structure comparison between the native protein (PDB ID: 2QBP) and the ligand-bound complex was performed using the 2StrucCompare web server (<https://2struccompare.cryst.bbk.ac.uk/>)¹². This tool enables residue-wise and global comparison of protein secondary structure assignments derived from established algorithms.

Results

Lipinski rule of 5

Before performing advanced computational studies (Table 1), Lipinski's Rule of Five was used to determine molecular properties of Aloe anthraquinones. Seven of the nine compounds tested met all of Lipinski's requirements. These included having a molecular weight of less than 500 Da, fewer than five hydrogen bond donors, fewer than ten hydrogen bond acceptors, and logP values of less than five. Aloe emodin, its hydroxy derivatives, anthranol, anthrone, O-acetylated, and iso-forms all had molecular weights between 256 and 326 Da and logP values between 1.11 and 1.98. This means that they are both hydrophilic and lipophilic in the right

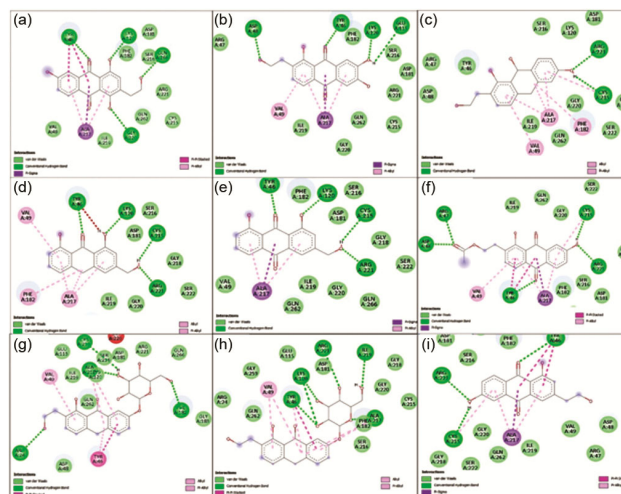


Fig. 1 — Docking interactions of Aloe anthraquinones with PTP1B

amounts. These show that the drug can easily get into the body through the mouth and through membranes. Aloin A and Aloin B, on the other hand, violated Lipinski's rule because they had too many hydrogen bond donors, which was more than the limit. Their molecular weights were still in the range, but their negative logP values showed that they were very polar. This could make it difficult for them to passively diffuse across biological membranes. Most of the compounds had physicochemical profiles that were generally acceptable.

Molecular docking

To determine the degree of binding of aloe-derived compounds' in the PTP1B catalytic pocket and to map their important interactions, molecular docking was performed (Fig. 1 and Table 2). Every ligand got bound at the main active site including TYR46, ASP48, VAL49, LYS120, ASP181, PHE182, CYS215, SER216, ALA217, GLY220, and ARG221, and interacted with important residues. This outcome shows that their alignment within the functional binding site was accurate. Additionally, some functional groups, such as GLN262, GLN266, and ILE219, extended into the secondary pocket, which probably helped to stabilize the structure.

Aloe-derived anthraquinone and anthrone derivatives demonstrated notable binding affinity varied with oxidation state, hydroxyl group placement, glycosylation, and lipophilicity in docking simulations with PTP1B. Every ligand got bound to the catalytic pocket including TYR46, ASP48, VAL49, LYS120, ASP181, PHE182, CYS215, SER216, ALA217, GLY220, and ARG221 and made contact with important

Table 2 — Docking analysis of Aloe anthraquinones

Compound	Binding energy (k Cal/ mol)	Inhibition constant (μ M)	Type of interactions	Interacting residues
8 Hydroxy aloe emodin	-6.58	15.02	Conventional Hydrogen Bond Pi-Pi Stacked Pi-Sigma Pi-Alkyl van der Waals	LYS A:120, SER A:216, GLN A:266, GLY A:220 TYR A:46 ALA A:217 TYR A:46, ALA A:217 ASP A:181, PHE A:182, GLN A:262, ARG A:221, CYS A:215, ILE A:219, VAL A:49
10 Hydroxy aloe emodin	-6.35	22.13	Conventional Hydrogen Bond Pi-Sigma Pi-Alkyl van der Waals	ASP A:48, TYR A:46, LYS A:120, GLU A:115, SER A:216 ALA A:217 VAL A:49, ALA A:217 ARG A:47, PHE A:182, ASP A:181, ARG A:221, GLN A:262, CYS A:215, ILE A:219, GLY A:220
Aloe emodin	-6.85	9.56	Conventional Hydrogen Bond Pi-Sigma Pi-Alkyl van der Waals	TYR A:46, LYS A:120, CYS A:215, ARG A:221 ALA A:217 ALA A:217 PHE A:182, SER A:216, ASP A:181, GLY A:218, SER A:222, VAL A:49, ILE A:219, GLY A:220, GLN A:262, GLN A:266 ARG A:221, CYS A:215
Aloe emodin anthranol	-6.65	13.41	Conventional Hydrogen Bond Pi-Alkyl Alkyl van der Waals	ALA A:217, VAL A:49 ILE A:219 ARG A:47, TYR A:46, ASP A:48, SER A:216, LYS A:120, ASP A:181, GLY A:220, GLY A:218, GLN A:262, PHE A:182, SER A:222
Aloe emodin anthrone	-6.62	14.08	Conventional Hydrogen Bond Pi-Alkyl Alkyl van der Waals	TYR A:46, LYS A:120, CYS A:215, ARG A:221 VAL A:49, PHE A:182, ALA A:217 ALA A:217 SER A:216, ASP A:181, GLY A:218, SER A:222, ILE A:219, GLY A:220
Aloe emodin O acetyl	-8.01	1.35	Conventional Hydrogen Bond Pi-Pi Stacked Pi-Sigma Pi-Alkyl van der Waals	ARG A:47, ASP A:48, TYR A:46, CYS A:215, ARG A:221 TYR A:46 ALA A:217 VAL A:49, ALA A:217 ILE A:219, GLN A:262, GLY A:220, SER A:222, GLN A:266, SER A:216, ASP A:181, PHE A:182
Aloin A	-6.39	20.64	Conventional Hydrogen Bond Pi-Pi Stacked Pi-Alkyl Alkyl van der Waals	ARG A:24, LYS A:120, SER A:216, CYS A:215, ASP A:181, PHE A:182 TYR A:46 VAL A:49, TYR A:46 ILE A:219 GLU A:115, ALA A:217, GLN A:262, GLN A:266, ARG A:221, GLY A:183, ASP A:48
Aloin B	-5.85	50.72	Conventional Hydrogen Bond Pi-Pi Stacked Pi-Alkyl Alkyl van der Waals	ARG A:221, ASP A:181, LYS A:120, TYR A:46, ILE A:219 TYR A:46 VAL A:49, TYR A:46, PHE A:182 ALA A:217 ARG A:24, GLU A:115, GLY A:259, GLN A:262, GLY A:218, GLY A:220, CYS A:215, SER A:216
Iso aloe emodin	-6.99	7.49	Conventional Hydrogen Bond Pi-Pi Stacked Pi-Sigma Pi-Alkyl van der Waals	ARG A:221, CYS A:215, TYR A:46 TYR A:46 ALA A:217 PHE A:182, ILE A:219, VAL A:49 ASP A:181, SER A:216, GLY A:220, GLY A:218, SER A:222, GLN A:262, ASP A:48, ARG A:47

residues. This pattern encourages binding that is probably pertinent to the activity of the enzyme. Our findings indicated that 8-hydroxy-aloe-emodin had a value of -6.10. Hydroxy-aloe-emodin had a weaker binding score in the -6 kcal/mol range, whereas the ligand with a binding energy of -7.58 kcal/mol demonstrated stronger binding. According to this value (35 kcal/mol), the hydrogen-bond geometry and the way the aromatic rings stack are altered when a hydroxyl group is added at particular locations. While retaining π - π stacking with TYR46, an aromatic gate residue that seems crucial for binding, the ligand was probably able to establish hydrogen bonds with LYS120, SER216, and GLN266 due to the additional phenolic groups.

Moderate binding affinities (about -6) were obtained by converting the C-9 carbonyl group into anthrone and anthranol derivatives. The observed values fall between 62 and -6.65 kcal/mol is the estimated energy barrier. Stronger alkyl and hydrophobic interactions with VAL49, ALA217, and ILE219 were demonstrated by these forms. This suggests that the loss of anchoring to carbonyl-mediated interactions typically provide greater flexibility. Aloe-emodin O-acetyl exhibited the highest binding affinity (-8) among the compounds that were tested. The inhibition constant (K_i) was 1, and the calculated binding free energy was 01 kcal/mol at a 35 μ M concentration. The molecule's lipophilicity was enhanced by acetylation, enabling it to further enter the hydrophobic catalytic cleft. In addition, it maintained strong π - π stacking with TYR46 and important hydrogen-bond interactions with ASP48, CYS215, ARG221, and TYR46. Aloin A and Aloin B, the glycosylated forms, exhibited higher inhibition constants and weaker binding. Greater polarity and steric hindrance may be the cause of this, favoring interactions close to the enzyme surface as opposed to deep within the catalytic site.

RMSD

A root mean square deviation (RMSD) analysis was performed to compare with the structures of the docked ligand with enzyme (Table 3). It was found that the normal RMSD was 0.356 Å when we compared coordinates without lining up structures. This determines changes in position and small shape differences between the two sets of coordinates affect each other. The value changes when the structure moves. The Kabsch RMSD went down to 0.290 Å after the best structural superposition. This means that

Table 3 — Root mean square deviation (RMSD) values of the docked Aloe-emodin O-acetyl ligand calculated using different computational approaches

RMSD Type	Methodology	Alignment Applied	RMSD Value (Å)
Normal RMSD	Direct coordinate comparison	No	0.356
Kabsch RMSD	Least-squares optimal superposition	Yes	0.290
Quaternion RMSD	Quaternion-based optimal superposition	Yes	0.291

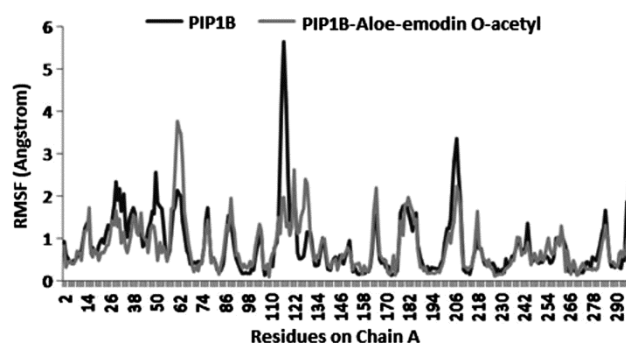


Fig. 2 — RMSF of PTP1B with Aloe emodinO-acetyl

a lot of the differences in normal RMSD were due to changes in how the structure moved and turned, not changes in the structure itself. The quaternion-based RMSD value of 0.291 Å was almost the same as the Kabsch RMSD value. This proves that the two alignment algorithms worked very well together. Throughout the experiments, the RMSD values remain minimal, which suggests that the ligand retains an analogous conformation following docking. The results indicate that, after alignment, the docked conformation is strikingly close to the original structure. This suggests that the docking pose is appropriate for subsequent *in silico* examinations and investigations into ligand-receptor binding.

RMSF

The study conducted root mean square fluctuation (RMSF) analysis to evaluate residue-level flexibility in PTP1B by comparing the apo protein with the Aloe-emodin O-acetyl-bound complex (Fig. 2). In the apo PTP1B system, dynamics were altered throughout multiple loop areas, showing the most significant alterations in regions positioned close to the active site. Residues in the WPD loop showed lower fluctuations in the Aloe-emodin O-acetyl complex than in the apo protein, which suggests that ligand binding limits the conformational movement of this key catalytic loop. Residues within the PTP1B

signature motif, including Cys215 and Arg221, showed lower RMSF values when the ligand was bound. These residues are required for the nucleophilic attack step and for keeping the substrate in place during enzymatic process. In the apo system, the peaks in these regions decreased after ligand binding, which suggests that the ligand stabilizes the structure. Notably, regions distant from the active site showed similar RMSF patterns in both systems, indicating that Aloe-emodin O-acetyl stabilizes specific functional regions without making the entire protein rigid.

Radius of gyration analysis

The radius of gyration (Rg) was determined to identify the compactness of PTP1B (PDB ID: 2QBP) is its original and ligand bound form (Table 4). The native protein had a radius of gyration (Rg) of 18.93 Å. The radius of gyration was 93 Å, which was consistent for protein of this size based on the empirical relation relationship $R_g = 0.395 \cdot N^{3/5} + 7.257$. The radius of gyration changed very little after the ligand got bound. According to the results, the aloe-emodin O-acetyl complex had a Rg of 19.05 Å. The only small rise in Rg when compared to the apo protein indicates that ligand binding does not significantly alter the overall expansion or compaction of the protein structure. This supports the idea that the overall fold integrity preservation. These findings demonstrate that the docked ligands occupy the catalytic pocket without altering the overall structure of PTP1B.

Accessible surface area

ASA was used to compute the absolute accessible surface area and the relative accessibility for each residue in chain A of the Aloe emodin O acetyl structure (Table 5). Absolute ASA values ranged from 0 Å² for residues fully confined in the protein core, such as internal hydrophobic side chains, to more than 180 Å² for residues that are highly exposed on the surface. The residues listed were ARG47 and GLU62. Relative accessibility ranged from 0 to approximately 0.989, supporting the view that surface exposure is spread across the protein. Residues in the predicted ligand-binding regions were less accessible than those in surface loops and other solvent-exposed parts of the protein. For example, the internal core residues LEU69, ILE72, and VAL82 had an ASA of 0, which matches their expected buried location in the structure. By contrast, charged residues on the surface

Table 4 — Radius of gyration for Aloe emodin O acetyl

Compound	Radius of gyration
2QBP	18.9296
2QBP-Aloe emodin O acetyl	19.0469

Table 5 — Accessible Surface Area for Aloe emodin O acetyl

Chain	Res No	Residue	ASA (Å ²)	Rel. Accessibility
A	4	GLU	89.368	0.474
A	5	LYS	148.985	0.697
A	12	LYS	153.959	0.720
A	25	HIS	163.712	0.818
A	29	ASP	135.468	0.863
A	33	ARG	170.264	0.742
A	46	TYR	24.824	0.103
A	47	ARG	169.594	0.739
A	48	ASP	76.345	0.486
A	62	GLU	186.336	0.989
A	75	GLU	152.622	0.810
A	116	LYS	164.676	0.770
A	182	PHE	94.639	0.416
A	285	SER	97.394	0.759
A	289	ASP	89.773	0.572

(e.g., Lys, Arg, Asp, and Glu) tend to interact strongly with the surrounding solvent, which can influence local stability and may affect how the protein associates with other molecules. The residues GLU4, LYS12, and GLU62 showed high relative accessibility values (>0.5), and labeled them as solvent-exposed. Several nearby aromatic and polar residues showed lower solvent accessibility, which suggests they take part in contacts within the binding site. The accompanying data helps identify residues that may interact by comparing their ASA profiles.

Secondary structure comparison

A comparison of secondary structure between the native protein (2QBP) and the Aloe-emodin O-acetyl-bound complex (Table 6) shows that chain A keeps the same secondary structure from residues 2 to 298. The calculated RMSD was 0.0.0 Å, which means that the protein backbones of the two structures fit together perfectly. The consensus secondary structure analysis showed that both structures had the same amounts of helices, β-sheets, and other structural parts. The protein structure in this study had 37.7% residues with α-helical content. With 19.2% β-sheet content, no areas that aren't defined (X = 0.000). The DSSP secondary structure assignments were the same for both the apo and ligand-bound structures, and no residues changed their structure. The analyses of the full sequence and the chosen subset yielded identical results, indicating that the protein chain is consistent throughout. Structural

Table 6 — Secondary structure comparison between native (2QBP) and Aloe-emodin O-acetyl bound protein using 2StrucCompare

Structure	Method	Helix (H)	Sheet (E)	Other (O)
2QBP (apo)	CONSENSUS	0.377	0.192	0.431
2QBP (apo)	DSSP	0.377	0.192	0.431
2QBP -Aloe-emodin O-acetyl complex	CONSENSUS	0.377	0.192	0.431
2QBP -Aloe-emodin O-acetyl complex	DSSP	0.377	0.192	0.431

maps that were aligned by sequence showed that each residue kept the same secondary structure assignment after binding to a ligand. The server didn't find any differences in the distances between C α , the distances between side chains, or the patterns of contact.

Discussion

In this study, we integrated drug-likeness screening with structure-based computational analysis to evaluate Aloe-derived anthraquinones as potential inhibitory agents of PTP1B, a well-established target in diabetes research. The initial Lipinski screening differentiated the aglycone derivatives, which exhibited superior pharmacokinetic properties, from the highly polar glycosides. Following additional assessments, the aloe-emodin O-acetyl stayed in the same place in PTP1B's main pocket. Therefore, after investigating at all the results in particular—including the strength of the interactions, the stability of the structure, and the preservation of the protein's native structure—these results support the hypothesis. There is substantial evidence indicating the reliability of these interactions. It also suggests that aloe anthraquinones may play an important role in halting enzymes from functioning during metabolism.

Lipinski's Rule of Five is a widely accepted parameter for evaluating the 'druggability' of a small molecule intended for oral administration. This is usually employed during the initial phases of study to support in prioritizing and selecting ensuing compounds for exploration. In the present study, the aloe aglycone derivatives were in agreement with Lipinski's rule-of-five, and revealed that their physicochemical properties are compatible with satisfactory drug absorption and distribution. Aloe emodin and associated derivatives possessed a moderate molecular weight and a balanced hydrogen-bonding profile. These properties aid to support acceptable solubility although still permitting membrane permeability. LogP values around 1 indicate a moderate level of lipophilicity, which is often associated with acceptable absorption and a decreases the chance of nonspecific buildup of drugs in the tissues.

With references to Aloin A and Aloin B, these two compounds do not comply with the Rule of Five developed by Lipinski, which shows that the introduction of a sugar group, glycosylation, can make them ineffective pharmacotherapeutic agents. Molecules that have a multitude of hydrogen bond acceptors become highly polar and, therefore, are more likely to suffer loss of permeability along the gastrointestinal epithelium by passive diffusion. This kind of physicochemical profile may hinder oral bioavailability, thus reducing the chances of such candidates being useful oral drugs, although they may be active by other means. Therefore, to rationally design new pharmaceuticals, the aglycone structures are sometimes more appropriate than the glycoside structures. The current investigation contributes significantly to our understanding of the diverse structures of the anthraquinones, which are analogs of the compounds of the anthrax of plants, and the mode of action with the primary pocket of the PTP1B. All the compounds interacted with critical active-site residues (TYR46, ASP48, LYS120, ASP181, PHE182, CYS215, SER 216, ALA217, GLY220, and ARG221) and induced binding arrangements that are associated with biological activity. The oxidation state, hydroxylation pattern, as well as lipophilicity of the ligand became the key factors that controlled the changes in the binding affinity. An example is the potency criterion of acetylation, which was observed to be most important as shown through aloe-emodin O-acetyl which showed the maximum binding affinity and minimum inhibition constant. It was the lipophilic nature of this ligand that made it penetrate further into the hydrophobic cleft and it remained in a constant pose as a result of hydrogen bonds with catalytic residues and π - π stacking with TYR46. The orientation of the hydroxyl substituent was additionally found to be consequential with 8-hydroxy-aloe-emodin exhibiting superior affinity when compared to its 10-hydroxy counterpart, which highlights the role of hydroxyl orientation on the geometry of hydrogen-bonds and aromatic interactions. Substitution of the anthraquinone core to anthrone or anthranol resulted in moderate binding

energies which indicates that the loss of carbonyl based interactions can be to some extent made up by increased flexibility and more hydrophobic contacts. On the other hand, the glycosylated derivatives (Aloin A and B) showed lower binding which is probably due to steric inhibition and increased polarity.

Rootmean square deviation (RMSD) is an omnipresent structural measure that is used to determine whether a computed binding pose is stable over a range of conformations. The RMSD of 0.356 Å used in this analysis is within the range of the small variations with the rigid-body motions, which means that the overall movement of the docked ligand is negligible. Standard RMSD computation does not include an optimal step on superposition that may over contain the distinctions that can be explained by translation and rotation alone. Following a Kabsch alignment, the RMSD of 0.290 Å (computed through least-squares superposition) is a more precise measure of the intrinsic change in conformation of the ligand. The fact that the RMSD calculated by Kabsch is close to that of the quaternions supports the validity of the findings because the two mathematical methods reach close values. Throughout the analysis, the values of RMSD did not exceed 0.4 Å which suggests that the original geometry of the ligand has been preserved to a large degree and that the docked aloec-emodin O-acetyl can be held in a stable position. Conformational rearrangement of the WPD loop of PTP1B is part of the catalytic cycle, and the dynamic is critical in the maintenance of enzymatic turnover rate.

Aloe-emodin O-acetyl binding leads to reduced root-mean-square fluctuation (RMSF) values of residues that make up the WPD loop, indicating that the binding of ligands limits the native flexibility of the loop. This limitation can inhibit the closing step required in catalysis. Docking studies indicate stabilizing interactions with Tyr46, Arg47, Asp48, Val49, and all proximate to the WPD loop, as well as canonical hydrogen bonds and other notable interactions interactions, and hydrophobic interactions. Further interactions between Cys215, Arg221, Ala217 and other surrounding residues also solidify the PTP signature motif. Aloe-emodin O-acetyl is held in the active site through hydrogen bond formation with Cys215 and Arg221, an alkyl contact with Val49 and Ala217 and 8- π interactions with Ala217. The complex is further stabilized by Van der Waals interactions with Ile219, Gly 220, Ser222, Phe182 amidst other residues. The reduction in RMSF that is observed at these positions can be used to support the

strength of the docking interactions. The binding free energy of -8.10 kcal/mol indicates that aloec-emodin O-acetyl inhibits PTP1B by binding to important catalytic motifs and reducing their movement.

The radius of gyration is commonly used to describe protein structural stability due to its ability to quantify the degree of protein denseness yet simultaneously accommodating conformational pliability. In docking and molecular dynamics analyses, large changes in Rg often suggest an implausible binding pose or that the ligand is destabilizing the complex. In this study, native PTP1B and its ligand-bound complexes had nearly the same Rg values, suggesting that Aloe-derived anthraquinones bind without changing the protein's overall fold. This is important for PTP1B because its catalytic activity depends on the exact spatial arrangement of residues in the active site. The small rise in Rg after ligand binding most likely comes from local side-chain shifts within the active-site pocket, rather than from overall protein unfolding. The aloec-emodin O-acetyl complex had higher lipophilicity and reached deeper into the binding pocket, but its Rg was similar to that of the apo protein, suggesting the ligand fits the protein without causing major structural changes. This level of stability enhances assurance in the docking results thereby implying that the anticipated associations are likely to occur under physiological conditions. In this study, Rg analysis supports the binding energy results and the interaction profile via an extensive evaluation of conformational robustness, consequently justifying the preference of the Aloe-derived compounds, with aloec-emodin O-acetyl being particularly prominent.

Accessible Surface Area (ASA) numerically expresses the degree of each residue's solvent accessibility. Within protein structure analysis, it's commonly employed as a surrogate marker to identify potential ligand interaction points and for evaluating structural integrity. In the Aloe emodin O acetyl complex, the ASA values indicate that residue exposure varies across the structure, with some residues buried in the core and others highly exposed on the surface. The ASA values follow the usual pattern seen in protein structure: hydrophobic residues tend to be buried in the core, while charged and polar residues are more exposed to the solvent. Residues with lower ASA than expected for normal solvent exposure, especially near the binding cavity, are likely involved in stabilizing contacts with the docked ligand. Residues such as TYR46 and ARG47 have moderate ASA values, which may reflect that the

ligand partly shields these sites. Exposed charged residues, including GLU62 and LYS12, point to surface regions that may be more mobile, which could affect solvent contact and overall protein flexibility. Since the ASA calculation leaves out explicit solvent and does not sample dynamic conformational shifts, the findings must be viewed as fixed representations. Despite this, the ASA profile operates in parallel with additional structural investigations (*e.g.*, to compare results across methods). This approach combines docking scores with interaction maps to better identify which residues take part in ligand binding. In conjunction with the outcomes from RMSD and interaction energy calculations, ASA contributes to understanding the mechanism of ligand stabilization and the dynamic alterations of the protein surface.

The lack of changes in secondary structure suggests that binding of Aloe-emodin O-acetyl does not alter the protein backbone and that it remains relatively rigid. This pattern is typical of docking-based or rigid-body binding models, in which the ligand is accommodated mainly through local side-chain contacts, with little to no large-scale conformational change. The consistent levels of helices, sheets, and coils indicate that ligand binding does not change the protein's overall fold or its secondary structure. This degree of structural conservation is useful in pharmacology because it suggests that ligand binding is unlikely to disrupt the protein framework. The stabilization probably comes from noncovalent interactions within an already formed binding pocket. Our observations correspond to the insights gained from the accessible surface area and interaction analyses: the variations are primarily confined to solvent exposure and the contact-forming residues, not to changes in the overall backbone geometry. Secondary structure comparisons are based on fixed structures, so they do not show the time-dependent changes that can occur under normal physiological conditions. Despite this, the results support the view that Aloe-emodin O-acetyl binds without causing structural stress.

Conclusion

This study presents a computational evaluation of anthraquinones derived from Aloe as prospective PTP1B inhibitors, utilizing a comprehensive array of *in silico* methodologies. An initial evaluation employing Lipinski's Rule of Five suggested that the majority of aglycone derivatives, such as aloe emodin and various modified variants, meet standard criteria for assessing

drug-like characteristics. The glycosylated compounds Aloin A and B, on the other hand, broke the rules because they were very polar and could form strong hydrogen bonds. This evidence backed up the idea that aglycone molecules should be the focus of the next stage of the analysis. Molecular docking indicates that all evaluated compounds bind within the functional catalytic pocket of PTP1B and engage with residues recognised for their role in enzyme activity. Aloe-emodin O-acetyl was predicted to have the strongest binding affinity of all the compounds tested. This finding is consistent with the compound's lipophilicity, its capacity to maintain crucial hydrogen bonds with catalytic residues, and its role in stabilizing aromatic interactions within the binding site. The RMSD-based structural check showed only small changes in the docked complex's shape, which supports a stable binding pose and suggests that the predicted interaction is reliable. The RMSF analysis shows that important functional areas are less flexible, especially the WPD loop and the PTP signature motif. This suggests that ligand binding stops the catalytic motions that are needed for enzyme activity. Global stability metrics, such as the radius of gyration and the accessible surface area, indicate that ligand binding does not significantly modify the overall protein fold; however, it induces minor localized alterations within the binding pocket. A comparison of the secondary structure confirmed these findings: the helix, sheet, and coil content remained unchanged following ligand binding. These results indicate that aloe-emodin O-acetyl may serve as a promising lead compound for PTP1B inhibition, owing to stable interactions that align with the protein's structural characteristics. This computational study serves as a foundation for subsequent research employing molecular dynamics and laboratory experimentation, highlighting Aloe-derived anthraquinones, particularly aloe-emodin O-acetyl, as potential candidates for antidiabetic drug development.

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Conflict of interest

All authors declare no conflict of interest.

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