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In Silico Docking Study Of Vani Vallarai Nei Phytocomponents Targeting Gaba-B Receptor For Potential Muscle Relaxant Activity In Spasticity Management

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Abstract

Background: *Cerebral palsy (CP) is a leading cause of childhood motor disability, often associated with spasticity due to impaired inhibitory neurotransmission in the central nervous system. Conventional treatments for spasticity, including baclofen and diazepam, offer limited efficacy and are often accompanied by undesirable side effects. Siddha medicine offers alternative approaches, and the traditional formulation Vani Vallarai Nei is reputed for its neuroprotective and cognitive-enhancing properties.*

Objective: *This study aimed to evaluate the muscle relaxant potential of phytocomponents present in Vani Vallarai Nei by assessing their binding affinity with the GABA-B receptor using molecular docking approaches.*

Materials and Methods: *Nine bioactive compounds—asiatic acid, asiaticoside, humulene, quercetin, myricetin, oleic acid, magnolin, linoleic acid, and picein—derived from the herbal ingredients of Vani Vallarai Nei were retrieved from literature. Molecular docking simulations were carried out using AutoDock 4.2 against the GABA-B receptor (PDB ID: 4MS4), targeting the core functional residues Tyr250 and Trp278. Binding energies, inhibition constants, and interaction profiles were analyzed.*

Results: *Among the compounds tested, linoleic acid (−8.03 kcal/mol) and picein (−8.07 kcal/mol) demonstrated the highest binding affinity with the GABA-B receptor, followed by humulene and asiaticoside. These compounds exhibited stable interactions with key active site residues, including hydrogen bonding with Tyr250 and Trp278, which are essential for receptor activation and inhibitory neurotransmission.*

Conclusion: *The molecular docking results suggest that phytochemicals from Vani Vallarai Nei have the potential to modulate GABA-B receptor activity and may contribute to muscle relaxation. These findings support the traditional use of this Siddha formulation in managing neurological conditions and provide a scientific basis for further in vitro and in vivo validation.*

Keywords: *Molecular docking, GABA-B receptor, Vani Vallarai Nei, muscle relaxant, phytocompounds, Siddha medicine, spasticity.*

1.INTRODUCTION

Cerebral palsy (CP) is a group of non-progressive neurological disorders that affect movement, muscle tone, and posture, primarily resulting from injury to the developing brain before, during, or shortly after birth. It represents the most common cause of childhood physical disability, with a global prevalence ranging from 2 to 3 per 1,000 live births. Among the different types, spastic cerebral palsy accounts for nearly 70–80% of cases and is characterized by increased muscle tone, stiffness, and motor dysfunction [1]. Children affected by CP often face significant challenges in mobility, communication, and daily activities, which persist throughout life and require multidisciplinary care. Despite advances in neonatal care and rehabilitation, current pharmacological interventions for managing spasticity offer limited efficacy and are often associated with adverse effects such as sedation and muscle weakness [2].

Gamma-aminobutyric acid (GABA) is the principal inhibitory neurotransmitter in the central nervous system, playing a crucial role in maintaining neuronal excitability and synaptic balance [3]. In individuals with spastic

cerebral palsy, the inhibitory pathways mediated by GABA are often disrupted due to perinatal brain damage, particularly affecting descending corticospinal tracts and spinal inhibitory interneurons. This disruption leads to a reduction in presynaptic and postsynaptic inhibition, resulting in excessive motor neuron excitability and sustained muscle contraction [4]. The impairment of GABAergic signalling contributes significantly to spasticity and muscle hypertonia, making the GABAergic system a key therapeutic target for spastic disorders.

The GABA-B receptor, a metabotropic G-protein-coupled receptor, modulates neuronal excitability through second messenger systems and is primarily involved in slow and prolonged inhibitory signaling. Activation of GABA-B receptors leads to reduced neurotransmitter release, decreased calcium influx, and enhanced potassium conductance, which together contribute to dampening neural excitability and muscle tone [5,6]. Unlike GABA-A receptors that mediate rapid synaptic inhibition, GABA-B receptors offer a sustained inhibitory effect, making them an attractive target for managing chronic spasticity. Pharmacological agents like baclofen, a GABA-B agonist, are clinically used for spasticity but are limited by systemic side effects [7,8]. Despite the availability of various conventional pharmacological options such as baclofen, diazepam, and dantrolene for the treatment of spasticity, their long-term use is often limited by adverse effects including sedation, drowsiness, tolerance, dependency, and hepatotoxicity [9]. Moreover, these agents offer only symptomatic relief without addressing the underlying neurochemical imbalance or promoting neuronal repair. Surgical interventions and invasive procedures carry inherent risks and may not be feasible in all Paediatric cases. In contrast, Siddha medicine, one of the oldest traditional systems practiced in South India, offers a holistic approach rooted in centuries of empirical knowledge [10]. Siddha formulations, particularly those prepared using ghee-based media like Vani Vallarai Nei, are believed to possess superior bioavailability and the ability to cross the blood–brain barrier. These polyherbal preparations incorporate neuroprotective, anti-inflammatory, and muscle relaxant herbs that act synergistically to restore physiological balance [11]. The integrative nature of Siddha medicine, combining detoxification, rejuvenation, and neurological support, positions it as a promising complementary strategy for managing complex neurodevelopmental conditions such as cerebral palsy.

The Siddha formulation Vani Vallarai Nei, traditionally recommended in classical Siddha literature for neurological health, comprises several medicinal ingredients with documented neuropharmacological properties [12]. Vallarai (Centella asiatica) is recognized for its neuroprotective, anticonvulsant, and cognitive-enhancing effects [13], while Vasambu (Acorus calamus) has been reported to improve memory [14] and exert anticonvulsant activity [15]. Kadugurohini (Picrorhiza kurroa) is known to prevent memory deficits [16], and Sangu Poo (Clitoria ternatea) exhibits central nervous system depressant and anti-stress effects. The use of ghee as a lipid-based medium in Siddha therapeutics is traditionally valued for its ability to enhance bioavailability and facilitate drug delivery across the blood–brain barrier due to its lipophilic nature [17,18]. Phytoconstituents such as asiatic acid, asiaticoside, and humulene are derived from Centella asiatica, while magnolin is sourced from Acorus calamus, linoleic acid from cow ghee, and picein from Picrorhiza kurroa. These compounds were evaluated for their ability to interact with the GABA-B receptor (PDB ID: 4MS4), specifically targeting the functionally critical residues Tyr250 and Trp278 through hydrogen bonding [19]. Such interactions are hypothesized to enhance receptor activation and inhibitory postsynaptic signaling, which could attenuate neural excitability and promote muscle relaxation. The favorable binding of these phytochemicals to the receptor's active site suggests their potential role as natural therapeutic agents in the management of muscle spasm and spasticity [20,21].

Computational drug discovery techniques, particularly molecular docking, serve as vital tools for assessing the binding potential and interaction profiles of bioactive compounds with target proteins. These in silico approaches enable rapid, cost-effective screening of phytochemicals and provide molecular-level insights into their potential therapeutic mechanisms [22,23]. Considering the central role of the GABA-B receptor in regulating neuronal inhibition and muscle tone, this study employed molecular docking to investigate the binding interactions of nine phytocomponents derived from the Siddha formulation Vani Vallarai Nei with the active site residues of the GABA-B receptor. The primary objective of this research was to assess the potential of these phytochemicals to act as natural modulators of GABA-B-mediated inhibitory signaling, thereby offering a rationale for their use in the management of muscle spasticity associated with cerebral palsy.

2.MATERIALS AND METHODS

2.1.Selection of Compounds

Phytochemical constituents present in the Siddha formulation Vani Vallarai Nei were identified through a comprehensive review of published literature. A total of nine bioactive compounds were selected based on their known neuroprotective and muscle-relaxant properties. These included asiatic acid, asiaticoside, humulene, quercetin, myricetin, oleic acid, magnolin, linoleic acid, and picein, derived respectively from Centella asiatica, Acorus calamus, Clitoria ternatea, Picrorhiza kurroa, and cow ghee.

The phytoconstituents selected for molecular docking were derived from medicinal ingredients present in the traditional Siddha formulation Vani Vallarai Nei. Centella asiatica is a well-documented neuroprotective herb containing active constituents such as asiatic acid, asiaticoside, and humulene, which contribute to its cognitive-enhancing and antioxidant effects [13]. From Clitoria ternatea, the key bioactives identified include quercetin, myricetin, and oleic acid—compounds with established CNS depressant, anti-stress, and neuroprotective roles [20]. Acorus calamus, another integral ingredient, contributes magnolin, known for its role in improving memory and cognitive function [24]. Cow ghee, traditionally used as a lipid base in Siddha preparations, provides linoleic acid, which facilitates brain-targeted delivery due to its lipophilic nature [25]. Picrorhiza kurroa yields picein, a compound recognized for its antioxidant and neuroprotective potential [26]. These phytochemicals were selected for their reported neurological benefits and were evaluated for their interaction with the GABA-B receptor to explore their muscle relaxant potential.

2.2.Ligands Structure Generation

The 2D molecular structures of the selected compounds were sketched using ChemDraw Ultra (PerkinElmer, USA), ensuring accurate bond angles and stereochemistry. These structures were then converted into 3D conformers and energy-minimized using Chem3D. The optimized structures were saved in PDB format and further processed for docking studies.

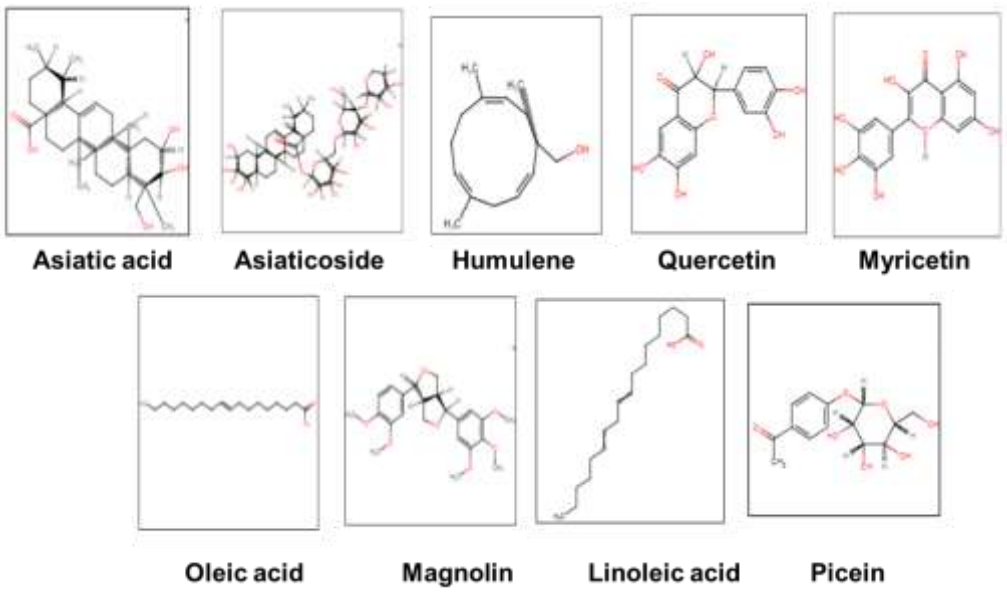

Figure 1. Two-Dimensional Representations of Selected Phytocompounds Used in Docking Studies

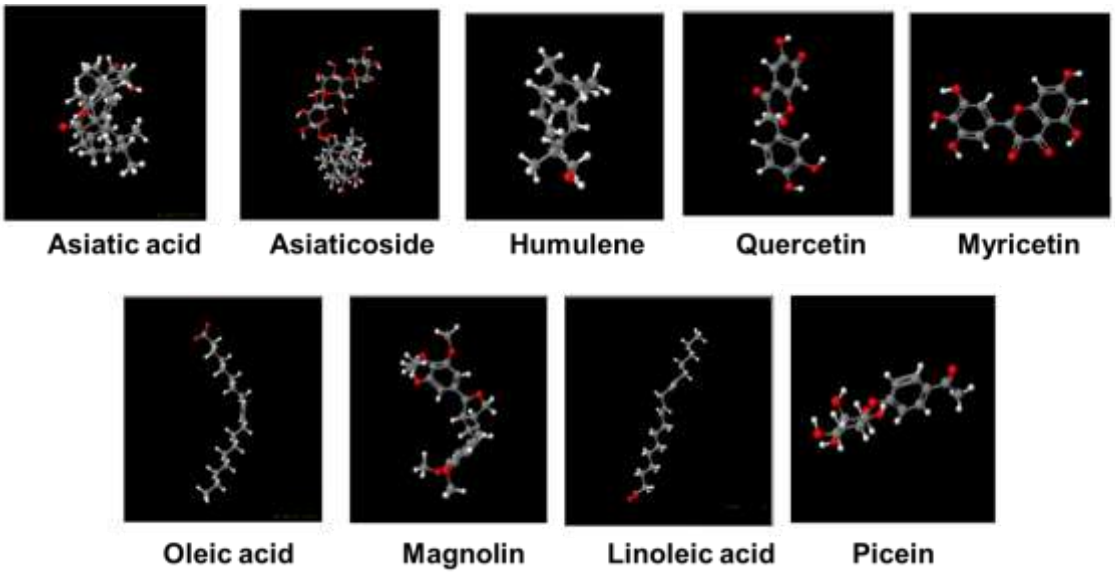

Figure 2. Three-Dimensional Representations of Selected Phytocompounds Used in Docking Studies

2.3.Assessment of Physicochemical Properties

The physicochemical properties of the selected phytocompounds were retrieved and analyzed using the PubChem database (https://pubchem.ncbi.nlm.nih.gov/). For each compound, parameters such as molecular weight, hydrogen bond donors and acceptors, number of rotatable bonds, and topological polar surface area (TPSA) were documented to evaluate their drug-likeness and potential for oral bioavailability. These properties were further compared against Lipinski's Rule of Five to preliminarily assess their suitability as therapeutic agents. The collected data provided insight into the structural and chemical behavior of the ligands, which is essential for interpreting their docking performance and biological relevance.

2.4.Protein Selection and Preparation

The crystal structure of the GABA-B receptor was obtained from the Protein Data Bank (PDB ID: 4MS4). The protein structure was prepared using AutoDock Tools by removing water molecules and heteroatoms, adding polar hydrogen atoms, and computing Gasteiger charges. Non-polar hydrogens were merged, and the structure was saved in PDBQT format for docking analysis [27].

2.5.Identification of the Active Site

The active site of the GABA-B receptor was determined based on prior reports highlighting Tyr250 and Trp278 as critical residues involved in receptor activation [28]. These residues were visually confirmed using PyMOL molecular visualization software and defined within the docking grid box to ensure targeted interaction assessment during simulation.

2.6.Docking Analysis Using AutoDock

Molecular docking simulations were performed using AutoDock 4.2 to evaluate the interaction between selected phytocompounds and the GABA-B receptor (PDB ID: 4MS4). The receptor was prepared by incorporating essential hydrogen atoms, assigning Kollman united atom type charges, and applying solvation parameters using AutoDock Tools. The docking grid was centered around the active site residues Tyr250 and Trp278, with a grid spacing of 0.375 Å, and affinity maps were generated using the Autogrid program. The docking process employed the Lamarckian Genetic Algorithm (LGA) and the Solis and Wets local search method to ensure an efficient exploration of the conformational space [29,30]. Parameters included a population size of 150, a maximum of 250,000 energy evaluations, a mutation rate of 0.02, and a translational step size of 0.2 Å. Quaternion and torsion step sizes were set to 5. For each ligand, ten docking runs were conducted with randomized initial orientations and fully flexible torsions [31]. The most favorable binding pose was selected based on the lowest binding free

energy and interaction stability. Post-docking analysis was carried out to examine hydrogen bonding, van der Waals, and electrostatic interactions using Discovery Studio Visualizer and PyMOL.

Figure 3. Three-Dimensional Crystal Structure of the GABA-B Receptor (PDB ID: 4MS4) Used for Docking Simulation

3.RESULTS

3.1.Physicochemical Profiling of the Phytocompounds

The physicochemical characteristics of the nine phytocompounds selected for molecular docking were assessed to understand their drug-likeness and potential suitability as orally active therapeutic agents. Molecular weight varied significantly across the compounds, with asiaticoside being the heaviest (959.1 g/mol) and humulene the lightest (204.35 g/mol). Notably, asiaticoside exceeded the commonly accepted molecular weight threshold of 500 g/mol, which may affect its membrane permeability and oral bioavailability.

The hydrogen bonding capacity, a key determinant of molecular recognition and interaction with biological targets, ranged from 0 (humulene) to 12 donors and 19 acceptors (asiaticoside). Flavonoids such as quercetin and myricetin exhibited a moderate number of hydrogen bond donors and acceptors, supporting their favorable interaction potential within the receptor's binding pocket. In contrast, humulene, a hydrocarbon sesquiterpene, lacked any hydrogen bond donor or acceptor groups, suggesting its interaction with the GABA-B receptor may primarily occur via hydrophobic forces. Rotatable bonds, influencing molecular flexibility, varied widely—from rigid molecules like humulene (0 rotatable bonds) to highly flexible ones like oleic acid (15) and linoleic acid (14). Higher flexibility may facilitate better accommodation within the receptor's binding site but may also compromise binding specificity. Compounds like asiatic acid and magnolin displayed an optimal balance between rigidity and flexibility, with 2 and 7 rotatable bonds respectively. Overall, several compounds—particularly quercetin, myricetin, magnolin, and picein—met key physicochemical criteria, indicating favorable drug-like properties. While asiaticoside and oleic/linoleic acids showed certain limitations due to size or flexibility, their docking scores suggest that these parameters may be offset by strong receptor interactions, warranting further pharmacokinetic evaluation as shown in Table 1.

Table 1. Ligand Properties of the Compounds Selected for Docking Analysis

Compound	Molar weight g/mol	Molecular Formula	H Bond Donor	H Bond Acceptor	Rotatable bonds
Asiatic acid	488.7 g/mol	C30H48O5	4	5	2
Asiaticoside	959.1 g/mol	C48H78O19	12	19	10
Humulene	204.35 g/mol	C15H24	0	0	0
Quercetin	302.23 g/mol	C15H10O7	5	7	1
Myricetin	318.237g/mol	C15H10O8	6	8	1
Oleic acid	282.5 g/mol	C18H34O2	1	2	15
Magnolin	416.5 g/mol	C23H28O7	0	7	7
Linoleic acid	280.452 g/mol	C18H32O2	1	2	14
Picein	298.29/mol	C14H18O7	4	7	4

3.2.Molecular Docking Analysis of Phytocompounds with GABA-B Receptor

The molecular docking results revealed variable binding affinities of the nine selected phytocompounds towards the GABA-B receptor (PDB ID: 4MS4), as evidenced by their estimated free binding energies and inhibition constants (Ki). Among all compounds, picein and linoleic acid demonstrated the strongest binding affinities, with free binding energies of −8.07 kcal/mol and −8.03 kcal/mol, respectively, and inhibition constants in the low micromolar range (1.21 μM and 1.30 μM). These results suggest a high likelihood of interaction and potential efficacy as GABA-B modulators.Humulene also exhibited a strong binding profile (−7.51 kcal/mol; Ki: 3.11 μM) despite lacking hydrogen bonding capability, indicating its interaction is likely driven by hydrophobic contacts. Similarly, asiatic acid, quercetin, and myricetin showed moderately favorable binding energies ranging from −6.62 to −6.83 kcal/mol, with inhibition constants between 9.83 and 14.04 μM, suggesting consistent, though less potent, interaction profiles.

Asiaticoside displayed a binding energy of −6.18 kcal/mol and a Ki of 29.60 μM, likely influenced by its high molecular weight and polar surface area, which may affect receptor accommodation. Oleic acid had the weakest binding energy (−4.32 kcal/mol) and the highest Ki value (679.77 μM), suggesting limited affinity towards the GABA-B receptor. Interestingly, magnolin, although showing a relatively moderate binding energy (−5.19 kcal/mol), demonstrated a significantly negative total intermolecular energy (−7.27 kcal/mol), supported by the largest interaction surface area (950.545 Å²), implying potential stabilization via non-electrostatic interactions. In summary, picein and linoleic acid emerged as the most promising candidates based on their strong binding energies and low inhibition constants, followed by humulene, asiatic acid, and quercetin. These findings suggest that selected phytocompounds from Vani Vallarai Nei may contribute to GABA-B receptor modulation and hold potential for further pharmacological validation as muscle relaxants as listed in table 2.

Table 2. Summary of the molecular docking studies of compounds against GABA-B With PDB- 4MS4

Compound	Est. Free Energy of Binding	Est. Inhibition Constant, Ki	Electrostatic Energy	Total Intermolecular . Energy	Interaction Surface
Asiatic acid	-6.83 kcal/mol	9.83 uM	-0.31 kcal/mol	-6.93 kcal/mol	852.677
Asiaticoside	-6.18 kcal/mol	29.60 uM	-0.23 kcal/mol	-6.08 kcal/mol	787.257
Humulene	-7.51 kcal/mol	3.11 uM	-0.03 kcal/mol	-7.51 kcal/mol	577.739
Quercetin	-6.64 kcal/mol	13.58 uM	-0.19 kcal/mol	-5.83 kcal/mol	771.288
Myricetin	-6.62 kcal/mol	14.04 uM	-0.03 kcal/mol	-6.17 kcal/mol	778.02
Oleic acid	-4.32 kcal/mol	679.77 uM	-0.16 kcal/mol	-4.62 kcal/mol	403.389
Magnolin	-5.19 kcal/mol	156.83 uM	-0.24 kcal/mol	-7.27 kcal/mol	950.545
Linoleic acid	-8.03 kcal/mol	1.30 uM	-0.16 kcal/mol	-11.53 kcal/mol	842.822
Picein	-8.07 kcal/mol	1.21 uM	-0.05 kcal/mol	-8.28 kcal/mol	695.922

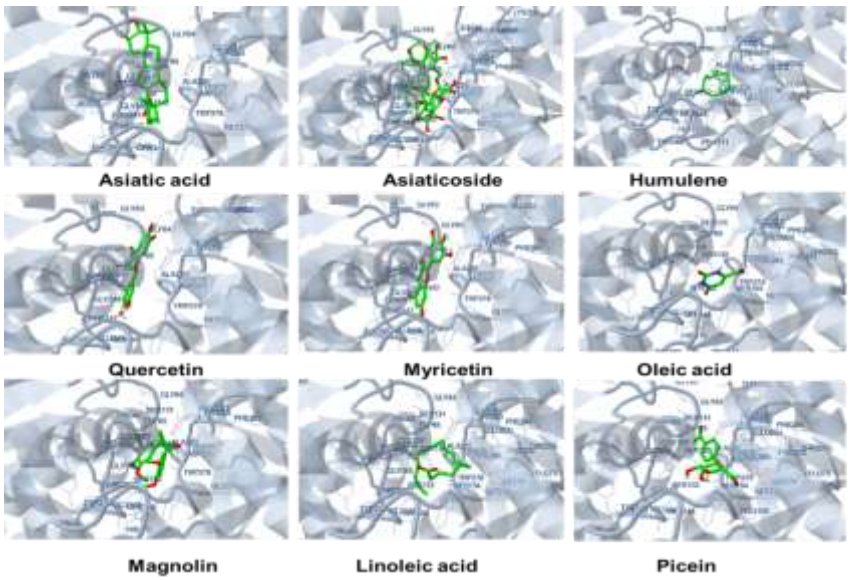

Figure 4.Docking Poses of Selected Phytocompounds within the GABA-B Receptor Binding Site (PDB ID: 4MS4)

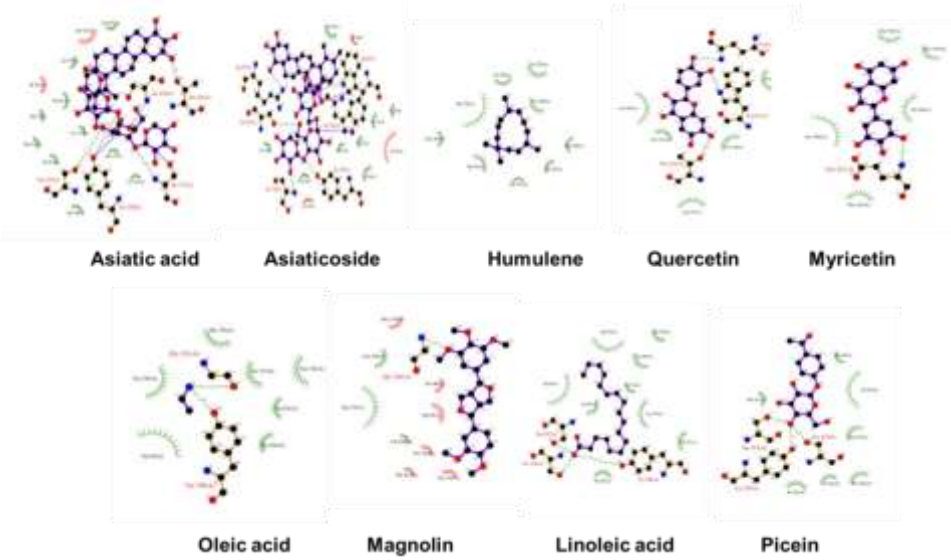

Figure 5. 2D Interaction Diagrams Depicting Key Residue Contacts Between Ligands and the GABA-B Receptor

3.3. Amino Acid Residue Interactions of Lead Compounds with GABA-B Receptor

The docking analysis revealed distinct interaction profiles between the phytocompounds and the active site residues of the GABA-B receptor (PDB ID: 4MS4), with particular emphasis on the core functional residues Tyr250 and Trp278, which are critical for receptor activation. Among the compounds, asiaticoside exhibited the most extensive interaction network, engaging 15 amino acid residues, including Tyr250, Glu251, Thr252, Trp278, and Glu349. The wide range of contacts, including hydrogen bonding and polar interactions, suggests a strong stabilizing effect within the receptor's binding pocket.

Picein, linoleic acid, and magnolin also demonstrated robust interaction profiles, each engaging between 10 to 13 amino acid residues, with consistent binding to both Tyr250 and Trp278. These residues play a central role in modulating receptor function, and their occupation suggests potential for effective GABA-B receptor modulation. Humulene and quercetin showed intermediate interaction patterns, forming contacts with 8 and 7 residues respectively, again including Tyr250 and Trp278, supporting their moderate binding affinities.Asiatic acid displayed interactions with 8 residues, primarily targeting Trp278, Glu251, and Glu349—suggesting stabilizing polar contacts. Myricetin, although forming fewer interactions, still engaged critical residues such as Glu251 and Glu349. In contrast, oleic acid, despite its weak docking score, interacted with a substantial number of residues (11), including a mix of polar and non-polar amino acids, suggesting flexibility in binding, although likely with less specificity.

Table 3. Amino acid Residue Interaction of Lead against GABA-B with PDB- 4MS4

COMPOUND	INTERACTION	AMINO ACID RESIDUE															
ASIATIC ACID	1	65	66	69	251	278	280	348	349								
		TRP	PRO	GLN	GLU	TRP	ALA	GLN	GLU								
ASIATICOSIDE	2	65	66	69	250	251	252	255	278	279	280	281	286	347	348	349	
		TRP	PRO	GLN	TYR	GLU	THR	ARG	TRP	TYR	ALA	ASP	ILE	PHE	GLN	GLU	
HUMULENE	2	65	153	201	202	250	276	278	279	349							
		TRP	SER	VAL	PHE	TYR	ILE	TRP	TYR	GLU							
QUERCETIN	2	65	66	250	251	278	279	280									
		TRP	PRO	TYR	GLU	TRP	TYR	ALA									
MYRICETIN	0	66	251	252	280	348	349										
		PRO	GLU	THR	ALA	GLN	GLU										
OLEIC ACID	2	65	130	151	153	170	201	202	250	278	349						
		TRP	SER	GLY	SER	HIS	VAL	PHE	TYR	TRP	GLU						
MAGNOLIN	2	65	66	170	250	251	278	279	280	346	349						
		TRP	PRO	HIS	TYR	GLU	TRP	TYR	ALA	GLY	GLU						
LINOLEIC ACID	2	65	66	130	153	202	250	251	276	278	279	348	349				
		TRP	PRO	SER	SER	PHE	TYR	GLU	ILE	TRP	TYR	GLN	GLU				
PICEIN	2	65	66	130	153	201	202	250	251	276	278						
		TRP	PRO	SER	SER	VAL	PHE	TYR	GLU	ILE	TRP						

4.DISCUSSION

Cerebral palsy (CP) remains a lifelong neurodevelopmental disorder with profound implications on motor function and muscle tone. Spasticity, one of its most debilitating manifestations, arises due to disrupted inhibitory signaling pathways within the central nervous system, particularly those mediated by gamma-aminobutyric acid (GABA). Although medications such as baclofen and diazepam are widely used, their long-term administration is often hampered by sedative effects, limited efficacy, and risk of tolerance. This therapeutic gap underscores the growing interest in alternative pharmacological interventions derived from traditional systems of medicine, such as Siddha, which offer a multi-targeted and holistic approach [32-35].These practices emphasize the balance of bodily systems through personalized approaches and plant-based formulations. With a long history of safe usage, they continue to serve as valuable sources of bioactive compounds for modern drug discovery. Integrating traditional knowledge with scientific validation can unlock novel treatment strategies for complex diseases [36]. The Siddha formulation Vani Vallarai Nei exemplifies such traditional wisdom. Comprising ghee and neuroactive botanicals like Centella asiatica, Acorus calamus, Clitoria ternatea, and Picrorhiza kurroa, this polyherbal preparation has been traditionally utilized for enhancing memory, reducing stress, and modulating neurological functions [37,38]. The ghee-based matrix not only acts as a bioenhancer but also facilitates the delivery of lipophilic constituents across the blood–brain barrier, improving central nervous system availability [39].

Computational techniques such as molecular docking offer a predictive and efficient approach to identify potential ligands, estimate binding affinities, and understand receptor–ligand interactions at the atomic level. These in silico strategies not only accelerate the early phases of drug discovery but also reduce the cost and time required for downstream experimental validations [40,41].The physicochemical properties of a compound—such as molecular weight, hydrogen bond donors and acceptors, and conformational flexibility—significantly affect its ability to interact with target receptors. These factors determine the ligand's compatibility with the binding site and the stability of receptor–ligand interactions. Well-balanced molecular features improve both binding strength and pharmacological relevance [42]. A preliminary evaluation of the physicochemical parameters revealed that most of the selected phytocompounds fall within acceptable drug-likeness thresholds. Quercetin, myricetin, picein, and magnolin adhered well to Lipinski's rule, indicating their suitability for oral administration and efficient receptor binding. On the other hand, asiaticoside, due to its high molecular weight and polar surface area, may face permeability challenges despite its robust interaction profile. The rotatable bond count, hydrogen bonding capacity, and polar functional groups of the ligands played crucial roles in determining their binding flexibility and specificity.

Computational docking has emerged as a powerful tool in early-stage drug discovery, enabling rapid screening of bioactive compounds against specific targets. It allows precise prediction of binding orientations, interaction energies, and receptor compatibility. This approach significantly reduces the time and cost involved in experimental screening [43]. Docking methods also aid in optimizing lead structures for improved efficacy and selectivity. Molecular docking results provided insights into the interaction strength and orientation of each ligand within the GABA-B receptor binding pocket. Among all compounds, picein and linoleic acid displayed the most favorable binding energies (−8.07 and −8.03 kcal/mol, respectively) and the lowest inhibition constants, suggesting high binding affinity and potential biological activity. Interestingly, humulene, although devoid of hydrogen bonding features, showed a strong affinity likely driven by hydrophobic interactions within the lipophilic core of the receptor. Flavonoids such as quercetin and myricetin also demonstrated meaningful docking scores, attributed to their polyphenolic structures which enable stable interactions through hydrogen bonding and π–π stacking.

The docking strategy focused specifically on the active residues Tyr250 and Trp278, which are critical for receptor activation and the initiation of downstream inhibitory signaling [28]. Compounds that were able to form stable interactions with these residues are likely to mimic the natural agonists of the GABA-B receptor or modulate its activity favorably. This was particularly evident in the case of asiaticoside, which formed hydrogen bonds with a wide array of amino acids including Tyr250, Glu251, Thr252, and Glu349—suggesting a broad stabilizing effect within the receptor cavity. The active site residues of the GABA-B receptor, particularly Tyr250 and Trp278, play a pivotal role in ligand recognition and receptor activation. These residues are crucial for stabilizing ligand binding through hydrogen bonding and hydrophobic interactions. Targeting these specific amino acids enhances

the likelihood of modulating receptor function effectively. Their involvement is essential for triggering the downstream inhibitory signaling associated with muscle relaxation. Analysis of amino acid interactions further substantiated the docking scores, with most lead compounds—especially linoleic acid, picein, and magnolin—showing consistent engagement with Tyr250 and Trp278, in addition to Glu349, Ala280, and other stabilizing residues. These interaction patterns support the hypothesis that the selected phytocompounds can effectively occupy the receptor's active site and potentially elicit muscle relaxant effects via enhancement of GABAergic inhibition. In summary, the study demonstrates the value of integrating traditional medicinal knowledge with modern computational tools to identify novel therapeutic leads. The phytocomponents of Vani Vallarai Nei show promising binding behavior with the GABA-B receptor, aligning with its traditional use in neurological disorders. While computational findings are predictive in nature, they offer a strong foundation for in vitro validation, pharmacodynamic evaluation, and future clinical development. The encouraging docking profiles of picein, linoleic acid, humulene, and flavonoids mark them as priority candidates for further experimental exploration in the context of cerebral palsy-associated spasticity.

5. CONCLUSION

The findings of this study provide promising evidence that phytocompounds derived from the traditional Siddha formulation Vani Vallarai Nei possess favorable binding affinity toward the GABA-B receptor, a key target implicated in the pathophysiology of muscle spasticity in cerebral palsy. Through a robust in silico docking approach, compounds such as picein, linoleic acid, and humulene demonstrated strong interactions with critical active site residues—particularly Tyr250 and Trp278—suggesting their potential to modulate GABA-B-mediated inhibitory signaling. The integration of traditional knowledge with modern computational tools has allowed for the rational prioritization of bioactive leads with desirable physicochemical and receptor-binding characteristics. These phytoconstituents not only align with the historical therapeutic claims of Siddha medicine but also exhibit drug-likeness and receptor engagement comparable to known pharmacological agents. While computational predictions offer valuable early-stage insights, the outcomes of this study lay a strong foundation for further experimental validations, including in vitro receptor assays and in vivo models of spasticity.

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6.REFERENCES

1. Rosenbaum, P., Paneth, N., Leviton, A., Goldstein, M., Bax, M., Damiano, D., Dan, B. & Jacobsson, B. A report: the definition and classification of cerebral palsy. Dev. Med. Child Neurol. Suppl. 2007; 109, 8–14.
2. Oskoui M, Coutinho F, Dykeman J, Jetté N, Pringsheim T. An update on the prevalence of cerebral palsy: a systematic review and meta-analysis. Dev Med Child Neurol. 2013 Jun;55(6):509–519. doi: 10.1111/dmcn.12080.
3. Golan H, Levav T, Mendelsohn A, Huleihel M. Involvement of GABA(A) receptors in the regulation of neuronal cell death and survival during development. Neuroscience. 2004;123(3):585–595. doi: 10.1016/j.neuroscience.2003.10.031. PMID: 14706792.
4. Delgado MR, Hirtz D, Aisen M, Ashwal S, Fehlings DL, McLaughlin J, Morrison L, Shrader MW, Tilton A, Vargus-Adams J. Practice parameter: pharmacologic treatment of spasticity in children and adolescents with cerebral palsy (an evidence-based review): report of the Quality Standards Subcommittee of the American Academy of Neurology and the Practice Committee of the Child Neurology Society. Neurology. 2010 Jan 26;74(4):336–343. doi: 10.1212/WNL.0b013e3181cbcd2f. PMID: 20101040.
5. Bowery NG, Bettler B, Froestl W, Gallagher JP, Marshall F, Raiteri M, Bonner TI, Enna SJ. International Union of Pharmacology. XXXIII. Mammalian gamma-aminobutyric acid(B) receptors: structure and function. Pharmacol Rev. 2002 Dec;54(2):247–264. doi: 10.1124/pr.54.2.247. PMID: 12037137.
6. Gassmann M, Bettler B. Regulation of neuronal GABA(B) receptor functions by subunit composition. Nat Rev Neurosci. 2012 Jun 13;13(6):380–394. doi: 10.1038/nrn3249. PMID: 22595776.
7. Baclofen for chronic spasticity. Drug Ther Bull. 2004 May;42(5):36–38. doi: 10.1136/dtb.2004.42536. PMID: 15155180.

8. Maccarrone M, Battista N, Centonze D. The endocannabinoid pathway in Huntington's disease: a comparison with other neurodegenerative diseases. Prog Neurobiol. 2007 Oct;81(5-6):349–379. doi: 10.1016/j.pneurobio.2007.04.003. PMID: 17544060.
9. Delgado MR, Hirtz D, Aisen M, Ashwal S, Fehlings DL, McLaughlin J, Morrison L, Shrader MW, Tilton A, Vargus-Adams J. Practice parameter: pharmacologic treatment of spasticity in children and adolescents with cerebral palsy (an evidence-based review): report of the Quality Standards Subcommittee of the American Academy of Neurology and the Practice Committee of the Child Neurology Society. Neurology. 2010 Jan 26;74(4):336–343. doi: 10.1212/WNL.0b013e3181cbcd2f. PMID: 20101040.
10. Sivaraman D, Kumar G, Muthu AK, Kumar R, Sankar C. Siddha medicine – An overview of scientific validation and quality control. Int J Pharm Sci Res. 2013;4(4):1287–1295. doi: 10.13040/IJPSR.0975-8232.4(4).1287-95.
11. Krishnan V, Nestler EJ. Linking molecules to mood: new insight into the biology of depression. Am J Psychiatry. 2008 Nov;165(11):1305–1318. doi: 10.1176/appi.ajp.2008.08020273.
12. Sayabu HMMA. Kadukai Vallaraiyin Thani Manbu. Chennai: Indian Medicine and Homeopathy Department; p. 68.
13. Gray NE, Alcazar Magana A, Lak P, Wright KM, Quinn J, Stevens JF, Maier CS, Soumyanath A. Centella asiatica – Phytochemistry and mechanisms of neuroprotection and cognitive enhancement. Phytochem Rev. 2018 Feb;17(1):161–194. doi: 10.1007/s11101-017-9528-y. PMID: 29422917.
14. Pattanaik J, Kumar Y, Khatri RS. Acorus calamus Linn.: A herbal tonic for central nervous system. Int J Pharm Sci Rev Res. 2013;21(1):64–72.
15. Kim N, Do J, Ju IG, Jeon SH, Lee JK, Oh MS. Picrorhiza kurroa prevents memory deficits by inhibiting NLRP3 inflammasome activation and BACE1 expression in 5xFAD mice. Neurotherapeutics. 2020 Jan;17(1):256–269. doi: 10.1007/s13311-019-00786-5. PMID: 31745723.
16. Rana AP, Shinde R, Bodkhe AA. Role of Brahmi Ghrita Lehana in cerebral palsy. World J Pharm Med Res. 2019;5(9):91–93.
17. Gray NE, Alcazar Magana A, Lak P, Wright KM, Quinn J, Stevens JF, Maier CS, Soumyanath A. Centella asiatica - Phytochemistry and mechanisms of neuroprotection and cognitive enhancement. Phytochem Rev. 2018 Feb;17(1):161–194. doi: 10.1007/s11101-017-9528-y. PMID: 29422917.
18. Pattanaik J, Kumar Y, Khatri RS. Acorus calamus Linn.: A herbal tonic for central nervous system. Int J Pharm Sci Rev Res. 2013;21(1):64–72.
19. Kim N, Do J, Ju IG, Jeon SH, Lee JK, Oh MS. Picrorhiza kurroa prevents memory deficits by inhibiting NLRP3 inflammasome activation and BACE1 expression in 5xFAD mice. Neurotherapeutics. 2020 Jan;17(1):256–269. doi: 10.1007/s13311-019-00786-5. PMID: 31745723.
20. Oguis GK, Gilding EK, Jackson MA, Craik DJ. Butterfly Pea (Clitoria ternatea), a cyclotide-bearing plant with applications in agriculture and medicine. Front Plant Sci. 2019 May 28;10:645. doi: 10.3389/fpls.2019.00645. PMID: 31244659.
21. Carolina Peña-Serna, Delgado ME, Restrepo JH. Chemical, physicochemical, microbiological and sensory characterization of cow and buffalo ghee. Food Sci Technol. 2020;40(2):444–450. doi: 10.1590/fst.00919.
22. Choudhury AA, Arumugam M, Ponnusamy N, Sivaraman D, Sertsemariam W, Thiruvengadam M, Pandiaraj S, Rahaman M, Rajeswari VD. Anti-diabetic drug discovery using the bioactive compounds of Momordica charantia by molecular docking and molecular dynamics analysis. J Biomol Struct Dyn. 2024 Feb;42(3):1423–1439. doi: 10.1080/07391102.2024.2313156. PMID: 38330848.
23. 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(16):1923–1938. doi: 10.2174/1568026614666140929124445. PMID: 25262799.
24. Qiao D, Gan LS, Mo JX, Zhou CX. [Chemical constituents of Acorus calamus]. Zhongguo Zhong Yao Za Zhi. 2012 Nov;37(22):3430–3433. PMID: 23427596.
25. Peña-Serna C. Chemical, physicochemical, microbiological and sensory characterization of cow and buffalo ghee. Food Sci Technol. 2020;40(2):444–450. doi: 10.1590/fst.00919.
26. Kant K, Walia M, Agnihotri VK, Pathania V, Singh B. Evaluation of antioxidant activity of Picrorhiza kurroa (leaves) extracts. Indian J Pharm Sci. 2013;75(3):324–329. doi: 10.4103/0250-474X.117438.
27. Berman HM, Westbrook J, Feng Z, Gilliland G, Bhat TN, Weissig H, Shindyalov IN, Bourne PE. The Protein Data Bank. Nucleic Acids Res. 2000 Jan 1;28(1):235–242. doi: 10.1093/nar/28.1.235. PMID: 10592235.
28. Evenseth LM, Warszycki D, Bojarski AJ, Gabrielsen M, Sylte I. In Silico Methods for the Discovery of Orthosteric GABAB Receptor Compounds. Molecules. 2019 Mar 7;24(5):935. doi: 10.3390/molecules24050935. PMID: 30866507; PMCID: PMC6429233.
29. Bikadi Z, Hazai E. Application of the PM6 semi-empirical method to modeling proteins enhances docking accuracy of AutoDock. J Cheminform. 2009 Apr 7;1:15. doi: 10.1186/1758-2946-1-15. PMID: 20150921.

30. Halgren TA. Merck molecular force field. I. Basis, form, scope, parametrization, and performance of MMFF94. J Comput Chem. 1996 Apr;17(5-6):490–519. doi: 10.1002/(SICI)1096-987X(199604)17:5/ 6<490::AID-JCC1>3.0.CO;2-P.
31. Solis FJ, Wets RJ. Minimization by random search techniques. Math Oper Res. 1981 Feb;6(1):19–30. doi: 10.1287/moor.6.1.19.
32. Vitrikas, K., Dalton, H. & Breish, D. Cerebral palsy: an overview. Am. Fam. Physician 101, 213–220 (2020).
33. Krach, L.E. Pharmacotherapy of spasticity: oral medications and intrathecal baclofen. J. Child Neurol. 16, 31–36 (2001). doi:10.1177/088307380101600107
34. Katz, R.T. & Campagnolo, D.I. Pharmacologic management of spasticity. Phys. Med. Rehabil. Clin. N. Am. 12, 747–768 (2001). doi:10.1016/S1047-9651(18)30053-6
35. Sharma, V., Sarkar, I.N. & Bhattacharya, S. Ethics and evidence-based traditional medicine: merging complementary and conventional health care systems. J. Ayurveda Integr. Med. 11, 406–412 (2020). doi:10.1016/j.jaim.2019.09.004
36. Zhang, A., Sun, H., Wang, P., Han, Y. & Wang, X. Recent advances in natural products from plants for treatment of neurodegenerative diseases. Eur. J. Med. Chem. 63, 442–469 (2013). doi:10.1016/j. ejmech.2012.12.062
37. Pattanaik, J., Kumar, Y. & Khatri, R.S. Acorus calamus Linn.: a herbal tonic for the central nervous system. Int. J. Pharm. Sci. Rev. Res. 22, 137–142 (2013).
38. Kim, N. et al. Picrorhiza kurroa prevents memory deficits by inhibiting NLRP3 inflammasome activation and BACE1 expression in 5xFAD mice. Neurotherapeutics 17, 256–269 (2020). doi:10.1007/s13311-019-00786-5. PMID: 31745723
39. Trivedi, M. et al. Ghee – Its properties, importance and health benefits. Int. J. Ayurvedic Med. 9, 146–150 (2018).
40. Sivaraman D, Selvadoss PP, Manoharan SS, Jeyabalan S, Yaraguppi DA, Choudhury AA, Rajeswari VD, Ramanathan G, Thamaraikani T, Sekar M, Subramaniyan V, Shing WL. Regulation of NS5B polymerase activity of hepatitis C virus by target-specific phytotherapeutics: an in-silico molecular dynamics approach. Cell Biochem Biophys. 2024 Jul;82(3):2473–2492. doi: 10.1007/s12013-024-01359-w. PMID: 39042185.
41. Dhanasekaran S, Jeyabalan S, Choudhury AA, Rajeswari VD, Ramanathan G, Thamaraikani T, Sekar M, Subramaniyan V, Shing WL. Harnessing phytochemicals to regulate catalytic residues of alpha-amylase and alpha-glucosidase in type 2 diabetes. Cell Biochem Biophys. 2024 Oct 10. doi: 10.1007/s12013-024-01575-4. Epub ahead of print. PMID: 39384699.
42. Lipinski, C.A., Lombardo, F., Dominy, B.W. & Feeney, P.J. Experimental and computational approaches to estimate solubility and permeability in drug discovery and development settings. Adv. Drug Deliv. Rev. 46, 3–26 (2001). doi:10.1016/S0169-409X(00)00129-0
43. Sivaraman D, Selvadoss PP, Manoharan SS, Jeyabalan S, Laxmi VM, Choudhury AA, Rajeswari VD, Ramanathan G, Thamaraikani T, Subramaniyan V, Sekar M, Shing WL. Targeting Nudix Hydrolase 5 with bioactive flavonoids: molecular dynamics and docking studies for breast cancer therapy. Cell Biochem Biophys. 2024 Dec 5. doi: 10.1007/s12013-024-01609-x. Epub ahead of print. PMID: 39601355.