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Computational Investigation of *Withania somnifera* Compounds Targeting *S. pombe* SMN YG-Dimer in Spinal Muscular Atrophy: Insights from Molecular Docking Analysis

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Abstract

Objectives: *The primary objective of this study is to determine the potential of phytochemical constituents of Withania somnifera in treating spinal muscular atrophy, an autosomal recessive genetic neurodegenerative disorder, through computational techniques. To investigate the binding affinity of phytocompound to the target protein S. pombe SMN YG-Dimer through molecular docking. Besides, evaluating the pharmacological attributes of phytochemical constituents to assess their ability as an ideal prospect for developing therapeutic medication. The target protein S. pombe SMN YG-Dimer of PDB ID: 4RG5 was extracted and downloaded from the Protein Data Bank (PDB) database. Later, the phytocompounds were extracted and downloaded from the IMPPAT database. The Swiss ADME webserver was used for evaluating the pharmacological properties and pharmacokinetics of phytocompounds. Finally, molecular docking was achieved by the PyRx webserver.* ***Results:*** *Molecular docking analysis revealed that withasomnine emerged as the most favorable phytocompound among other selected phytocompounds. Withasomnine demonstrated a significant binding affinity of -7.9 towards the target protein, indicating the need for further investigation. Moreover, withasomnine exhibited favorable pharmacological properties and pharmacokinetic profiles, positioning it as a promising candidate for drug development.* ***Conclusion:*** *The phytocompound withasomnine with the highest binding affinity to the target protein indicates a promising prospect for developing a therapeutic medication for treating spinal muscular atrophy.*

Keywords: Spinal muscular atrophy, *S. pombe* SMN YG-Dimer, *Withania somnifera*, phytocompound, molecular docking, ADMET analysis

INTRODUCTION

Spinal muscular atrophy (SMA) is a neurodegenerative autosomal recessive genetic disorder characterized by the deterioration of alpha-motor neurons of the spinal cord [1]. It is considered a rare disease, as 1 out of every 10,000 people gets affected [2]. The primary cause of SMA is typically attributed to mutations or deletions within the SMN gene, situated on chromosome 5q, producing insufficient expression of the SMN protein. This gene plays a critical role in producing the SMN protein, which is essential for regulating and preserving motor nerve functions [3]. Consequently, individuals affected by SMA experience weakness and muscle wasting in skeletal muscles due to a lack of signal transduction from the spinal cord to skeletal muscles. The condition also manifests in various symptoms, including

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respiratory difficulties, articulation deficiency, scoliosis, and challenges in voluntary muscle movements such as walking and swallowing [4]. SMA encompasses four distinct subtypes, which are categorized based on the age at which symptoms appear, in addition to factors like life expectancy and the severity and progression of the disease [5].

SMA type 1: SMA type 1 stands as the predominant subtype of SMA, typically manifesting within the first six months after birth, characterized by a range of symptoms including swallowing and breathing difficulties, susceptibility to pulmonary infections, lack of ability to grasp objects fully, and absence of head control. The mortality rate is exceptionally elevated, with approximately 95% of those affected succumbing to the condition by 18 months of age [6, 7].

SMA type 2: SMA type 2 typically emerges within the age range of 6 to 18 months, constituting approximately 20–30% of total SMA cases. Individuals with this type commonly encounter challenges in walking independently and frequently exhibit signs of restrictive lung disease. Many also experience scoliosis, fine tremors in their upper extremities, and muscle weakness. With appropriate supportive care, life expectancy for these patients can extend to around 25 years or more [6].

SMA type 3: The SMA type 3 represents a minority portion, accounting for 10–20% of total SMA cases. Its onset typically ranges from 18 months to adulthood. Individuals with this type may initially walk independently, but their ability to do so reduces gradually depending on the age of onset. People who lose the ability to walk often end up with scoliosis. Despite these challenges, they can generally expect a normal life expectancy [6, 8].

SMA type 4: SMA type 4 comprises a small fraction of all SMA cases and emerges after the age of 21. Affected individuals exhibit symptoms such as muscle weakness and fatigue but typically have a standard life expectancy [6, 9].

The onset of disease not only depends on the loss of the homozygous gene of SMN1 but also the presence of a number of copies of the SMN2 gene generated in the body. The SMN2 gene also produces SMN protein, which produces a high level of nonoligomerized unstable truncated proteins and a very minimum quantity of functional SMN protein which cannot perform all the functions needed to be done. This is because the SMN2 gene is 90% homologous to the SMN1 gene functionally. The low production of non-functional SMN proteins is due to the transition of the C>T base of the exon 7, leading to the deletion during the event of splicing within the SMN2 gene. The severity of the case depends upon the mutation of the SMN2 gene and the number of copies of the SMN2 gene (Table 1) [10].

The treatment of SMA is managed through several therapeutic approaches. Gene replacement therapy stands out as a highly effective treatment option, offering enduring benefits for patients. In addition, several other treatments like antisense oligonucleotide therapy and small-molecule therapies exist [11, 12, 13].

Table 1. Number of SMN2 copies and the onset of disease.

SMA type	No. of SMN2 copies	Onset
0	1	Prenatal or at-birth
1	1–2	0–6 months
2	1–2	6–18 months
3	3–4	18–3/3–30 years
4	> = 4	>30 years

Despite having these treatments, their high cost renders them inaccessible to many individuals, resulting in prolonged suffering among patients. As a response, scientists are exploring novel alternatives like other medications, therapies, and adjuvant drugs. Researchers are inclined toward the formulation of conventional medicines derived from the phytochemical constituents of medicinal plants. The plants that possess the phytochemical constituents of a long history are known for their therapeutic attributes like potential antioxidants, ability to prevent neurodegeneration, mitigate neurotoxicity, and enable the neuroprotection mechanisms in neurological disorders and *Withania somnifera* [14] (Figure 1) seems to be an ideal option for deserving further research studies.

Withania somnifera, commonly called ashwagandha, belongs to the Solanaceae family and is grown in dry and sub-tropical areas of India. This plant has been used in India and China for more than 6,000 years in medicine for its beneficial properties [15]. Its phytochemical composition includes steroidal lactones, alkaloids, phenolics, and flavonoids, which serve as antioxidants, anti-inflammatory agents, anti-cancer agents, and neuroprotective agents. Active metabolites aid in treating various diseases like cancer, diabetes, stress, anxiety, cardiovascular conditions, rheumatoid arthritis, and several others [16]. Many research studies have been conducted to explore the impact of *Withania somnifera* on neurological disorders like Parkinson's, Alzheimer's, Huntington's disease, dementia, and schizophrenia due to its perceived neuroprotective and neuroregenerative properties within the realm of neurology [17].

Researchers showed that the extracts enriched with active metabolites of *Withania somnifera* displayed potential neuroprotection to SK-N-SH, a neuroblastoma cell line, against Aβ peptide and acrolein in their in-vitro studies. These protective effects are exhibited through its antioxidant mechanism and as a cholinergic modulator [18].

In a research study, scientists demonstrated that withanoside-IV, a phytoconstituent found in *Withania somnifera*, displayed the ability to induce neurite outgrowth, promote synaptogenesis, and enhance synaptic integration as a potential therapeutic approach for Alzheimer's disease [19].

An extensive preclinical investigation carried out on albino rats revealed that *Withania somnifera* extract enhanced cognitive and motor coordination deficits. It rejuvenated and protected neurons against apoptotic cell death triggered by LPS-induced neuroinflammation in neuronal cultures treated with activated microglial-conditioned medium, as well as in microglial-neuronal co-cultures [20].

As mentioned above, recent studies demonstrated that *Withania somnifera* has ground-breaking beneficial effects in treating neurological disorders. The positive outcomes of the studies determined the study to explore the ability of phytocompounds of *Withania somnifera* through computational techniques.

Figure 1. *Withania somnifera*.

There is a notable gap in research regarding the role of *Withania somnifera* in treating SMA. While *Withania somnifera* is recognized for its neuroprotective and neuroregenerative effects, its potential to target *S. pombe* SMN YG-Dimer protein remains minimally explored. Our study aims to address this gap by investigating the neuroprotective and neuroregenerative effects of *Withania somnifera* extracts or compounds on *S. pombe* SMN YG-Dimer protein activity. This study aims to explore the promising phytochemical constituents of *Withania somnifera* targeting *S. pombe* SMN YG-Dimer protein in developing a novel therapeutic drug for treating SMA by using molecular docking and other computational techniques.

MATERIALS AND METHODS

Selection and Purification of the Target Protein

In the context of SMA, the SMN protein serves as the specific focus of attention. More precisely, the *S. pombe* SMN YG-Dimer, identified with the PDB ID 4RG5, was acquired by retrieving its three-dimensional structure in PDB format from the RCSB Protein Data Bank (PDB) (*https://www.rcsb.org/*). The acquired target protein is then visualized utilizing BIOVIA Discovery Studio 2021 v21.1.0.20298 software (*https://discover.3ds.com/discovery-studio-visualizer-download*). To procure a refined version of the target protein, all heteroatoms, including water molecules, ligand groups, other ions, and amino acid chains, were eliminated. The modified target protein was subsequently stored in PDB format for prospective docking and additional computational analyses.

Secondary Structure Prediction

Secondary structure analysis was done by using the PDB sum webserver (https://www.ebi.ac.uk/thornton-srv/databases/pdbsum/). It was studied to know the presence of structures like beta strands, alpha helices, loops, turns, and beta hairpins, which play an important role in the stability and functionality of a protein. This structure aids in a detailed understanding of specific interactions between a target protein and a ligand [21].

Moreover, PROCHECK, a software tool accessible through the PDBsum server, was employed to conduct the Ramachandran plot analysis. This method enabled the evaluation of the protein's structural integrity and conformational quality.

Selection and Preparation of the Ligand

The ligands selected for the target protein were phytocompounds sourced from the leaves and roots of *Withania somnifera.* These phytocompounds were thoroughly retrieved from a database containing medicinal plants. The Indian medicinal plants phytochemistry and therapeutics (IMPAAT) database, accessible at (*https://cb.imsc.res.in/imppat/*), was chosen as the preferred resource for phytocompounds [22]. Subsequently, the three-dimensional structures of the ligands were procured in structured data file (SDF) format from the PubChem repository (*https://pubchem.ncbi.nlm.nih.gov/*), along with their respective canonical smiles or phytocompounds. These structures will be utilized for ADME analysis in the forthcoming docking procedure.

ADME Analysis

ADME refers to the absorption, distribution, metabolism, and excretion of a drug within the human body. It plays a crucial role in assessing the optimal functioning of a drug. These parameters were utilized to examine and study the pharmacokinetics of the drug. The Swiss ADME webserver (*http://www.swissadme.ch/*) was utilized to assess the drug [23]. For evaluation, the canonical SMILES of 30 ligands were obtained from PubChem (*https://pubchem.ncbi.nlm.nih.gov/*) and inserted into Swiss ADME for ADME analysis. This study helps comprehend the biological mechanisms and effects of a drug. A drug is considered favorable when it aligns with the criteria and ranges specified for all physicochemical properties defined by Lipinski's guidelines [24] (Table 2).

Table 2. Lipinski guidelines.

Property	Optimal range
Molecular weight	<500 Daltons
MlogP	<4.15
H donors	<5
H acceptors	<10
Molecular refractivity	40–130

Molecular Docking

Molecular docking is a technique used to study the interactions between a small-molecule drug/ligand) and the target macromolecule (*enzyme/lipid/nucleic acid*). These pharmacodynamic interactions result in the binding affinity of small molecules with the target macromolecule, revealing the pharmacological effects of the drug [25]. Molecular docking can be performed by various docking tools like PyRx, AutoDock Vina, Swiss Dock, and others.

PyRx, a virtual screening tool (*https://pyrx.sourceforge.io/*) was preferred for docking purposes. For docking, five ligands identified as optimal among a pool of 30 ligands based on ADME analysis were subjected to docking with the *S. pombe* SMN YG-Dimer protein obtained from the PDB database.

Initially, the purified protein target was transformed into a macromolecule, and ligands were imported into an open-babel tool and proceeded to further processing. The grid box was generated to calculate the potential of all possible docking interactions between the ligand and the target proteins's binding site. Finally, Vina Wizard was run to obtain the binding affinities of all ligands for the interaction with protein, indicating that higher scores correspond to the highest binding affinity.

RESULTS

Extraction and Purification of the Protein

The *S. pombe* SMN YG-Dimer, identified by its PDB ID 4RG5, was acquired at a resolution of 1.7 Aº and saved in PDB format. Subsequently, the target protein was purified using the BIOVIA Discovery Studio 2021 v21.1.0.20298 Tool, which involved the removal of water molecules and other hetaatoms. The configured version of the target protein is shown in Figure 2.

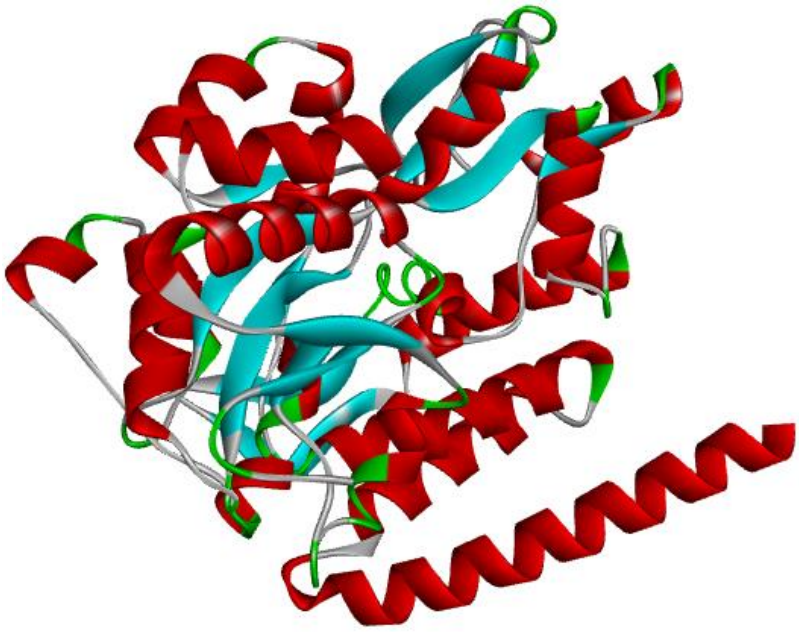

Figure 2. 3D structure of purified *S. pombe* SMN YG-Dimer.

Secondary Structure Analysis

Secondary Structure

The secondary structure of the target protein (PDB ID: 4RG5) includes 401 amino acid residues, 4 beta-sheets, 1 beta-alpha unit, 2 beta hairpins, 1 psi loop, 3 beta bulges, 15 strands, 20 helices, 27 helix-helix interact, 28 beta turns, and 3 gamma turns (Figure 3).

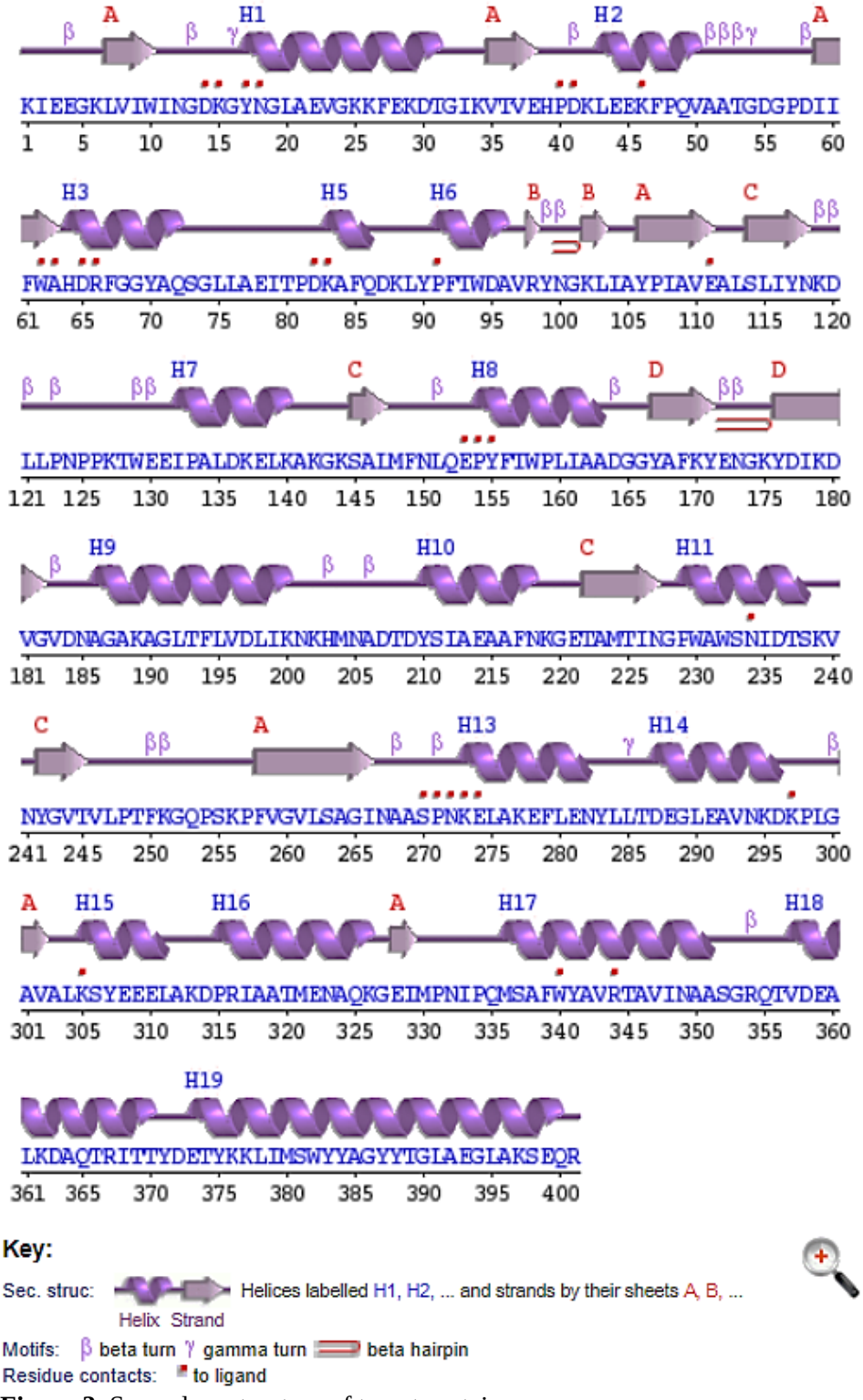

Figure 3. Secondary structure of target protein.

Ramachandran Plot

The Ramachandran plot was generated using PDBSUM. According to the plot statistics, approximately 92.6% of amino acid residues were found in the most favored regions, with 640 residues observed. Additionally, there were 48 residues (6.9%) in regions allowed with some constraints and 2 residues (0.3%) in generously allowed regions. The presence of residues in disallowed regions was minimal, accounting for only 0.1%. Furthermore, the total number of non-glycine and non-proline residues amounted to 691, constituting 100% of the amino acid residues analyzed. The number of end residues (excluding glycine and proline) was 4. In contrast, there were 64 glycine residues and 42 proline residues present, making up a total of 801 amino acid residues in the dataset (Figure 4).

Extraction of the Phytocompounds

Phytocompounds were retrieved from the IMPAAT Database. 30 phytocompounds from *Withania somnifera* were extracted (Table 3). Phytochemicals were mostly extracted from the leaves and roots of the plant. Subsequently, their canonical smiles were obtained from the PubChem database.

Table 3. Phytocompounds and part of the plant.

S.N.	Name of the phytocompound
1.	Withanolide Q
2.	Withanolide R
3.	Withanolide M
4	Withanolide D
5.	Withanolide E
6.	Withanolide G
7.	Withanolide O
8.	Withanolide P
9.	Withanolide N
10.	Withanolide S
11.	Withanolide C
12.	Withanolide L
13.	Withanolide H
14.	Withanolide I
15.	Withanolide K
16.	Withanolide J
17.	Withanolide M
18.	Cuscohygrine
19.	Anahygrine
20.	Pelleterine
21.	Nicotine
22.	Withasomnine
23.	Hygrine
24.	Withaferin A
25.	27-Deoxy-14-hydroxywithaferin A
26.	27-Deoxywithaferin A
27.	Withanone
28.	17alpha-hydroxywithanolide D
29.	D-Glucose
30.	Withasomidienone

ADME Analysis

ADME analysis was conducted using the SwissADME webserver to assess the physicochemical properties, pharmacodynamics, and drug-likeness of all 30 compounds. This evaluation aimed to assess the biological functions of the ligand or drug. The outcomes of the analysis are presented in Table 4.

Table 4. ADME Analysis of the phytocompounds.

S.N.	Phytocompound name	Formula	MW (g/mol)	H-bond acceptors	H-bond donors	MR	TPSA (Å²)
1.	Withanolide Q	$C_{28}H_{38}O_6$	470.60	6	3	129.21	104.06
2.	Withanolide R	$C_{28}H_{38}O_6$	470.60	6	2	127.49	96.36
3.	Withanolide M	$C_{28}H_{36}O_6$	468.58	6	2	127.09	96.36
4.	Hygrine	$C_8H_{15}NO$	141.21	2	0	45.47	20.31
5.	Withanolide D	$C_{28}H_{38}O_6$	470.60	6	2	127.53	96.36
6.	Withaferin A	$C_{28}H_{38}O_6$	470.60	6	2	127.49	96.36
7.	Withanolide E	$C_{28}H_{38}O_7$	486.60	7	3	128.77	116.59
8.	Withasomnine	$C_{12}H_{12}N_2$	184.24	1	0	56.58	17.82
9.	Withanolide G	$C_{28}H_{38}O_5$	454.6	5	2	128.08	83.8
10.	Withanolide A	$C_{28}H_{38}O_6$	470.60	6	2	127.53	96.36
11.	27-Deoxy-14-hydroxywithaferin A	$C_{28}H_{38}O_6$	470.60	6	2	127.53	96.36
12.	27-Deoxywithaferin A	$C_{28}H_{38}O_5$	454.6	5	1	126.33	76.1
13.	Withanone	$C_{28}H_{38}O_6$	470.60	6	2	127.53	96.4
14.	17alpha-hydroxywithanolide D	$C_{28}H_{38}O_7$	486.60	7	3	128.73	116.59
15.	Pelletierine	$C_8H_{15}NO$	141.21	2	1	45.37	29.10
16.	Withanolide S	$C_{28}H_{40}O_8$	504.61	8	5	132.12	144.52
17.	Withanolide C	$C_{28}H_{39}ClO_7$	523.06	7	4	135.75	124.29
18.	Withanolide L	$C_{28}H_{36}O_5$	452.58	5	2	127.61	83.83
19.	2R)-2-[(1S)-1-[(8R,9S,10R,13S,14R,17S)-14,17-dihydroxy-10,13-dimethyl-1-oxo-4,7,8,9,11,12,15,16-octahydrocyclopenta[a]phenanthren-17-yl]-1-hydroxyethyl]-4,5-dimethyl-2,3-dihydropyran-6-one	$C_{28}H_{38}O_6$	470.60	6	3	129.28	104.06
20.	Cuscohygrine	$C_{13}H_{24}N_2O$	224.34	3	0	74.20	23.55
21.	D-Glucose	$C_6H_{12}O_6$	180.16	6	5	35.74	110.38
22.	Somniferine	$C_{36}H_{36}N_2O_7$	608.68	9	2	171.39	100.93
23.	Hygrine	$C_8H_{15}NO$	141.21	2	0	45.47	20.31
24.	Stoindoside IX	$C_{34}H_{48}O_{11}$	632.74	11	5	159.87	175.51
25.	Withaferin A	$C_{28}H_{38}O_6$	470.60	6	2	127.49	96.36
26.	Nicotine	$C_{10}H_{14}N_2$	162.23	2	0	53.13	16.13
27.	Galactitol	$C_6H_{14}O_6$	182.17	6	6	37.93	121.38
28.	4-Hydroxy-1,26-dioxo-5,6:22,26-diepoxyergosta-2,24-dien-27-yl 6-o-hexadecanoylhexopyranoside	$C_{50}H_{78}O_{12}$	871.15	12	4	236.91	181.58
29.	Beta-Sitosterol	$C_{29}H_{50}O$	414.71	1	1	133.23	20.23
30.	Anahygrine	$C_{13}H_{24}N_2O$	224.34	3	1	74.10	32.34

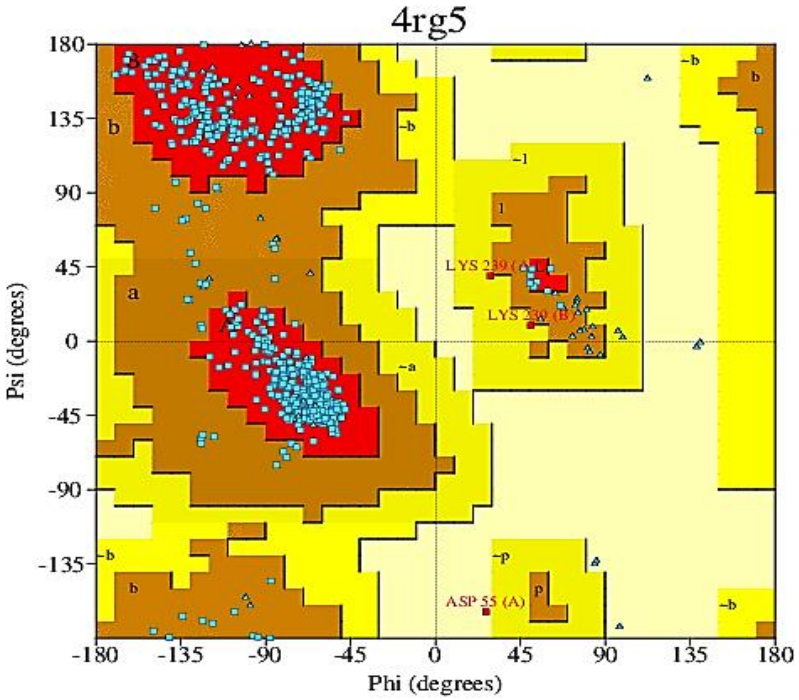

Figure 4. Ramachandran plot.

Physicochemical Properties

The factors for physicochemical properties comprise the ligand's molecular weight, number of hydrogen donors, number of hydrogen acceptors, topological surface area (TPSA), and molar refractivity. The optimal range for TPSA considered for a drug ranges <140°, while molecular refractivity falls under the expected range of 40–130. The molecular weight should not be greater than 500 daltons. Besides, the number of hydrogen donors and the number of hydrogen acceptors should be less than 5 and 10, respectively.

Pharmacokinetic Properties

Pharmacokinetics deals with gastrointestinal (GI) absorption of a drug and the permeance of BBB (Table 5). The GI absorption for a drug should be high drug besides, the permeance of the blood-brain barrier (BBB) must be positive for a drug that aids in treating neurological diseases and conditions.

Drug-likeness

Finally, for drug-likeness, the drug should adhere to the Lipinski rule with zero violations, which are listed in (Table 6). Based on the physicochemical attributes, pharmacokinetic profile, ADME analysis, and drug-likeness assessment of the entire set of 30 phytochemicals, the selected phytochemicals for docking with the target protein were identified as withasomnine, cuscohygrine, pelleterine, nicotine, and anahygrine. The SDF files corresponding to these five phytochemicals were obtained from the PubChem database. Subsequently, the ligands underwent additional energy minimization for converting from the 2D structure to the 3D structure. Final structures were then converted into AutoDock ligand format (PDBQT) for further analysis.

Molecular Docking

The molecular docking between the target protein *S. pombe* SMN YG-Dimer and the chosen 5 phytocompounds, namely anahygrine, cuscohygrine, nicotine, pelletierine, and withasomnine presents the binding affinities of their respective interactions (Table 7). The interactions were visualized by using the BIOVIA Discovery Studio 2021 v21.1.0.20298 screening tool to notify the different forms of interactions like vanderwaal interactions, hydrogen bonds, Pi-sigma bonds, alkyl bonds, and other interactions between the target protein and phytocompounds as ligands.

Table 5. Pharmacokinetics properties of the phytocompounds.

S.N.	Phytocompound name	GI absorption	BBB permeant
1.	Withanolide Q	High	No
2.	Withanolide R	High	No
3.	Withanolide M	High	No
4.	Hygrine	High	No
5.	Withanolide D	High	No
6.	Withaferin A	High	No
7.	Withanolide E	High	No
8.	Withasomnine	High	Yes
9.	Withanolide G	High	No
10.	Withanolide A	High	No
11.	27-Deoxy-14-hydroxywithaferin A	High	No
12.	27-Deoxywithaferin A	High	No
13.	Withanone	High	No
14.	17alpha-hydroxywithanolide D	High	No
15.	Pelletierine	High	Yes
16.	Withanolide S	Low	No
17.	Withanolide C	High	No
18.	Withanolide L	High	No
19.	2R)-2-[(1S)-1-[(8R,9S,10R,13S,14R,17S)-14,17-dihydroxy-10,13-dimethyl-1-oxo-4,7,8,9,11,12,15,16-octahydrocyclopenta[a]phenanthren-17-yl]-1-hydroxyethyl]-4,5-dimethyl-2,3-dihydropyran-6-one	High	No
20.	Cuscohygrine	High	Yes
21.	D-Glucose	Low	No
22.	Somniferine	High	No
23.	Hygrine	High	No
24.	Sitoindoside IX	Low	No
25.	Withaferin A	High	No
26.	Nicotine	High	Yes
27.	Galactitol	Low	No
28.	4-Hydroxy-1,26-dioxo-5,6:22,26-diepoxyergosta-2,24-dien-27-yl 6-o-hexadecanoylhexopyranoside	Low	No
29.	Beta-Sitosterol	Low	No
30.	Anahygrine	High	Yes

Table 6. Drug-likeness of the phytocompounds.

S.N.	Phytocompound name	Lipinski violations
1.	Withanolide Q	0
2.	Withanolide R	0
3.	Withanolide M	0
4.	Hygrine	0
5.	Withanolide D	0
6.	Withaferin A	0
7.	Withanolide E	0

8.	Withasomnine	0
9.	Withanolide G	0
10.	Withanolide A	0
11.	27-Deoxy-14-hydroxywithaferin A	0
12.	27-Deoxywithaferin A	0
13.	Withanone	0
14.	17alpha-hydroxywithanolide D	0
15.	Pelletierine	0
16.	Withanolide S	1
17.	Withanolide C	1
18.	Withanolide L	0
19.	2R)-2-[(1S)-1-[(8R,9S,10R,13S,14R,17S)-14,17-dihydroxy-10,13-dimethyl-1-oxo-4,7,8,9,11,12,15,16-octahydrocyclopenta[a]phenanthren-17-yl]-1-hydroxyethyl]-4,5-dimethyl-2,3-dihydropyran-6-one	0
20.	Cuscohygrine	0
21.	D-Glucose	0
22.	Somniferine	1
23.	Hygrine	0
24.	Stoindoside IX	2
25.	Withaferin A	0
26.	Nicotine	0
27.	Galactitol	1
28.	4-Hydroxy-1,26-dioxo-5,6:22,26-diepoxyergosta-2,24-dien-27-yl 6-o-hexadecanoylhexopyranoside	2
29.	Beta-Sitosterol	1
30.	Anahygrine	0

Table 7. Binding affinities of ligands and target protein.

S.N.	Ligand	Binding affinity
1.	Anahygrine	-7.2
2.	Cuscohygrine	-6.7
3.	Nicotine	-6.3
4.	Pelletierine	-5.6
5.	Withasomnine	-7.9

Finally, the results of the molecular docking process between the target protein and the ligand reveal that withasomnine exhibits the strongest affinity towards the target protein, as evidenced by its highest binding affinity. This indicates that withasomnine is the most suitable candidate for subsequent clinical evaluations.

The phytocompound withasomnine exhibited the highest binding affinity of -7.9 towards the target protein. The most robust interactions observed in the binding include conventional hydrogen bonds, Pi-sulfur, Pi-Pi stacked, Pi-Pi T-shaped, and Pi-alkyl bonds. Withasomnine engaged in conventional hydrogen bonds connected with the target protein through ASP A:65.

Pi-sulfur interactions occurred with MET A:330. Pi-Pi stacked bonds were evident with TYR A:155, TRP A:62, and TRP A:340. Pi-Pi T-shaped interactions were observed with TYR A:155. Pi-alkyl bonds were established with ALA A:63 (Figure 5).

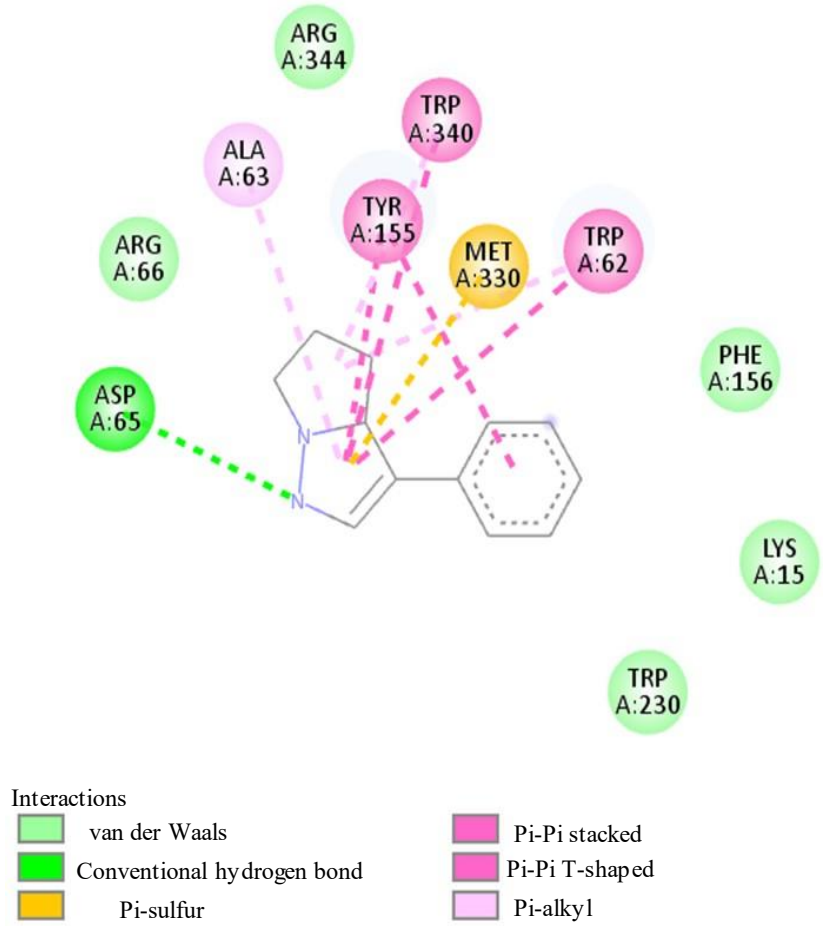

Figure 5. 2D Visualization of ligand interaction with protein.

DISCUSSION

The research study provides an in-depth analysis of the efficiency of *Withania somnifera*'s phytochemical constituents in treating SMA. As phytocompounds are widely known for their neuroprotectant and neuroregenerative activity [26, 27], this study explores their potential by targeting the *S. pombe* SMN YG-Dimer protein to develop novel therapeutic pharmaceuticals for treating SMA through computational techniques like molecular docking.

This study explains the interactions between phytocompounds of *Withania somnifera* and *S. pombe* SMN YG-Dimer through molecular docking. The phytocompounds finalized were anahygrine, cuscohygrine, pelleterine, nicotine, and withasomnine. The results indicate that withasomnine has the highest binding affinity with the target protein, signifying that it is an ideal match for developing a drug.

Molecular docking not only discusses interactions but also the binding poses and ligand efficiency. The binding pose describes the orientation and conformation of the phytocompound to the binding site [27]. The evaluation of pharmacokinetics, physicochemical properties, and drug-likeness of the phytocompound ensures the effectiveness of the phytocompound resulting in the emergence of potential drug therapies.

This research shed considerable insight on investigating the potential impacts of the phytochemical components of *Withania somnifera* in targeting the *S. pombe* SMN YG-Dimer, aiming to develop a therapeutic drug for SMA.

CONCLUSION

In conclusion, our research has demonstrated that the phytocompound withasomnine, derived from *Withania somnifera*, exhibits a high binding affinity when docked with the *S. pombe* SMN YG-Dimer target protein. This significant finding suggests withasomnine's strong potential as a prime candidate for the development of new therapeutic drugs.

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Abbreviation

SMA: Spinal Muscular Atrophy
PDB: Protein Data Bank
IMPPAT: Indian Medicinal Plants, Phytochemistry and Therapeutics
SMILES: Simplified Molecular Input Line Entry System
GI: Gastrointestinal
BBB: Blood-Brain Barrier
SDF: Structural Data File
MW: Molecular Weight
MR: Molar Refractivity
TPSA: Topological Polar Surface Area
SMN: Survival Motor Neuron

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