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Astrotactin-2: Docking with Flavoinoids, Functional, Pathway and Disease Associations

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Abstract

Astrotactin protein that is ASTN-1 and ASTN-2 are important perforin proteins that are involved in neurodevelopment. Mutations in this protein is associated with neurodevelopmental disorders and has been found functional in Alzheimer's, schizophrenia, Parkinson's diseases, Autism Spectrum Disorder, cancer etc. Genes for ASTN protein expressed in brain and functional in brain development any mutation in this gene leads to abnormal growth and functioning of brain. ASTN-2 protein id functional in protein trafficking and regulation of neuronal growth and development increased expression of this protein can also result in neurodevelopmental disorders. In this study detailed analysis has been done on ASTROTACTIN-2 protein to identify potential ligands that show strong interaction with ASTN-2 protein. 7 ligands were selected from PubChem database and Docking was performed to analyze the protein-Ligand interaction map. Result shows that Morin and Quercetin shows the best interaction with the Glide Score of -8.26 and -8.15 and at positions GLN790, LEU833, THR878 and EU833, GLN962 respectively. Protein -Protein interaction study was also done to identify interacting proteins that are functional in neurodevelopment and can be associated with neuronal disorders. This study detailed about the ligand interaction, protein function and protein interaction Astrotactin-2 protein.

Keywords: ASTN1, ASTN2, Neurodevelopment, Neuronal Disorder, Docking, Schrodinger

Introduction

Astrotactin-2 (ASTN-2) a novel member of the astrotactin gene family, regulates the trafficking of ASTN1 during glial-guided neuronal migration (1). ASTN-2 protein belongs to the family of Zinc Finger proteins that binds to DNA. ASTN-2 protein is important protein having function in neurodevelopment (2). ASTN-2 protein functions as signaling protein that transfers information for synaptic responses. Mutation in ASTN-2 protein is related to neurodevelopmental disorder, Alzheimer's,schizophrenia, Parkinson's diseases, Autism Spectrum Disorder, cancer etc. (3). There are various symptoms that are associated with ASTN-2 related diseases the premonitory phase can start from severalhours up to several days before the headache appears. In this phase, affected individuals canexperience extreme tiredness (fatigue), concentration problems, and muscle stiffness in the neck (4). Awide variety of additional signs and symptoms can occur including excessive yawning, food cravings, irritability, depression sensitivity to light, and nausea (5). Auras commonly include temporary visual changes such as blind spots (scotomas), flashing lights, and zig-zagging lines of color. Additional features of aura can include numbness, difficulty with speech and language, episodes of extreme dizziness (vertigo), and double vision (6). ASTN-2 protein is associated with different neurodevelopmental protein of these other important protein associated with neuro developmental disorder is DNAAF4 (7).

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Disease susceptibility is associated with variations affecting the gene represented in this entry. A chromosomal aberration involving DNAAF4 has been found in a family affected by dyslexia (8). Axonemal dynein assembly factor required for ciliary motility. Involved in neuronal migration during development of the cerebral neocortex. May regulate the stability and proteasomal degradation of the estrogen receptors that play an important role in neuronal differentiation, survival and plasticity (9). Disease susceptibility is associated with variations affecting the gene represented in this entry. A chromosomal aberration involving DNAAF4 has been found in a family affected by dyslexia. A relatively common, complex cognitive disorder characterized by an impairment of reading performance despite adequate motivational, educational and intellectual opportunities (10). It is a multifactorial trait, with evidence for familial clustering and heritability. A disorder characterized by abnormalities of motile cilia. Respiratory infections leading to chronic inflammation and bronchiectasis are recurrent, due to defects in the respiratory cilia (11). Patients may exhibit randomization of left-right body asymmetry and situs inversus, due to dysfunction of monocilia at the embryonic node. Primary ciliary dyskinesia associated with situs inversus is referred to as Kartagener syndrome (12).

In current work ASTN-2 protein was focused to study its structural properties, binding efficiency, protein -protein interactions, functional and disease associations.

Materials and Methods

Homology modeling of ASTROTACTIN2 protein was done using Prime tool of Schrodinger software (13). 7 ligands viz. Morin, Quercetin, Anisodamine, Silymarin, Trolox, Zilascorb Bisphenol A, these compounds belong to natural chemical compound having different biological property were used for docking. The list of ligands along with their molecular weight, molecular formulae has been mentioned in table 1. Ligand files was downloaded, and its properties were studied from PubChem Database.

To perform Docking, Glide Dock tool of Schrodinger software (14) was used, methodology adopted for this work along with databases and software's used has been shown in figure 1.

Glide dock score was studied to identify the best interacting ligand. Ligand-Protein interaction map was analyzed using LigPlot tool. Interaction map provides detail of bonds, bonds type, amino acids, position, and strength of bonds. Interaction map for all ligands were studied and shown in table 2.

Protein- Protein interaction study of ASTN-2 protein was done using STRING database (https://string-db.org), clustering, functional enrichment was further done using Cytoscape software (15) . Functional and pathways analysis of interacted protein was done using KEGG pathway (https://www.genome.jp/kegg/ pathway.html) , Gene Cards (https://www. genecards.

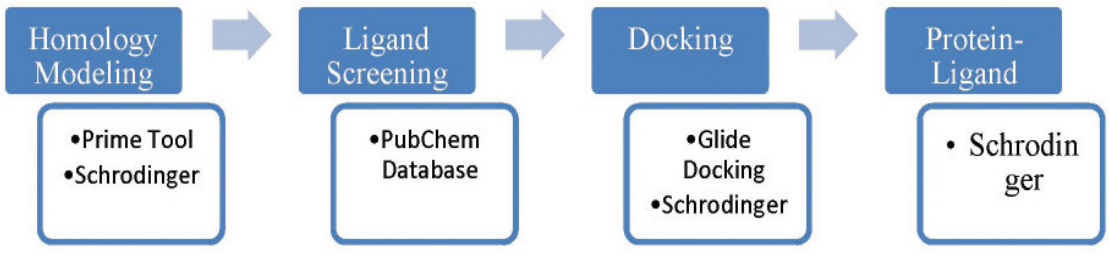

Fig. 1. Methodology and softwares used for docking study of ASTROTACTIN2 protein

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Fig. 2. Template selection for the Homology Modeling of Astrotactin-2 protein using Prime tool of Schrodinger Software

org/), Reactome and literature survey. All results were compared to identify the significance of ASTN-2 protein and its association with neuronal disorders.

Results and Discussion

Homology modeling of astrotactin-2 protein: Astrotactin-2 Protein involved in Neuronal Migrationhaving main function in Protein transport and localization (16). The structure of proteinis important parameter to study the function of protein and its interaction with ligands. Astrotactin-2 complete proteinstructure is important to screen potential ligand that shows interaction. Astrotactin-2 protein sequence was retrieved from Uniprot database with UniProt KB - O75129 (ASTN2_HUMAN). Homology Modeling Astrotactin-2 protein was done using Schrodinger software Prime tool as shown in figure 2. Homology model was built on template 3V65_B, with the Identitiesof 31%, positives 44%and Gap 20% modeled structure of Astrotactin-2 protein was shown in figure 3.

Docking of astrotactin-2 with antioxidants : Molecular interaction study of Astrotactin-2 protein was done with group of natural compounds mostly flavonoids that have antioxidant property and anti-cancer, anti- bacterial and of high importance in

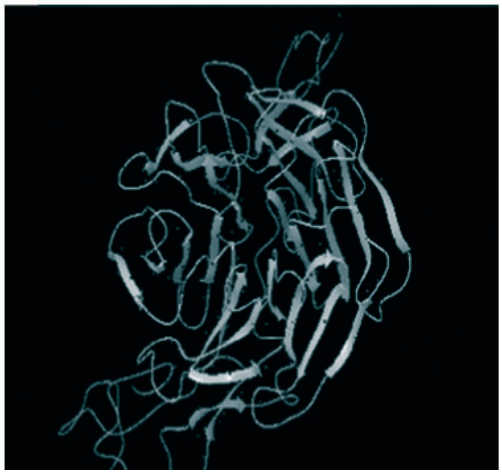

Fig. 3 Homology modeled structure of Astrotactin-2

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Table 1. Docking resultof ModeledAstrotactin-2 protein with ligands

S. No Name (g/mol)	PubChem Formulae	Molecular Weight	Molecular	Glide Score	Protein-Ligand Interaction Bonds	Figure
1.	Morin	C15H10O7	302.23	-8.26	GLN790, LEU833 THR878	Figure-4
2.	Quercetin	C15H10O7	302.23	-8.15	LEU833, GLN962	Figure-5
3.	Anisodamine	C17H23NO4	305.4	-7.709	LEU833, GLN792	Figure-6
4.	Silymarin	C25H22O10	482.4	-7.371	LEU833, GLN792	Figure-7
5.	Trolox	C14H18O4	250.29	-7.273	GLY876, VAL1011, GLN790	Figure-8
6.	Zilascorb	C13H12O6	265.24	-7.042	THR878	Figure-9
7.	Bisphenol A	C15H16O2	228.29	-7.099	GLY 20 LYS 38 THR 25	Figure-10

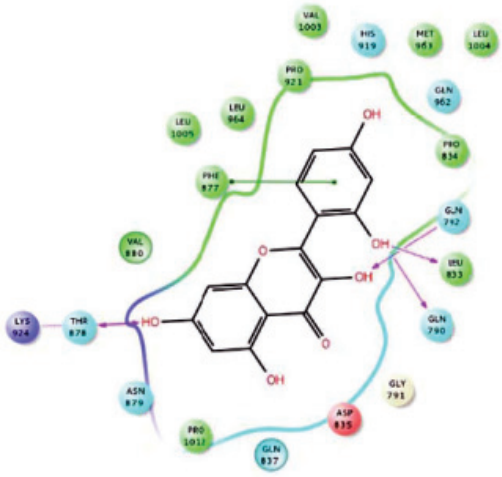

Fig. 4: Interaction map of Morin ligand shows Hydrogen bond at THR 878, GLN 790 and LEU 833 positions.

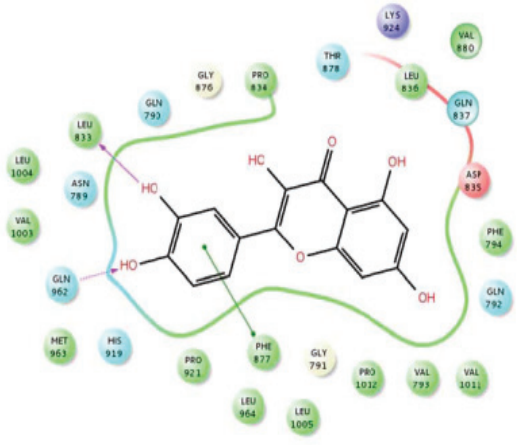

Fig. 5: Hydrogen bond with Quercetinat positions LEU 833 and GLN 962

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pharmacological implications and medications (17). The list of selected ligands and their properties has been shown in table 1.

Morinligand shows interaction with ASTN-2 protein with theGlide score-8.26 and make bonds at positions THR878, GLN790 and LEU833 as shown in figure 4. Quercetin have Glide score-8.15 and interact with different amino acids at different position LEU833 and GLN962 (Figure 5). Anisodamineligand binds with target protein with the Glide score -7.709 at positions GLN792 and LEU833 (Figure 6)

Docking of Silymarinligand with ASTN-2 protein shows interaction with the glide score -7.371at positions LEU833 and GLN792(Figure 7). Trolox ligand binds with ASTN-2 protein with glide score -7.273and at different position GLN790, VAL1O11 and GLY876 (Figure 8)

Zilascorbshows the glide score of -7.042and interact at position THR878(Figure 9). Bisphenol A shows the binding with protein with the glidescore -40.808 at positions GLY(A:22), LYS(A:37) and THR(A:25) at the given position (Figure 10)

Protein-protein interaction (PPI): Protein-protein interaction analysis is important study to understand the protein functional and molecular mechanism. To study the ASTN-2 protein interaction STRING database was used and also visualized in Cytoscape tool as shown in figure 12. Protein interaction network shows that ASTN-2 protein interacts with AIMP2, AP4E1, ARHGAP21, ASTN2, CDKL5, GAR1, KIAA1462, MAP6, MYO10, NDST4 and NR3C2 as shown in figure 11.

Protein interaction network was clustered by using K-means method with the value of k=3. Figure 11 shows the clustering result of PPI using STRING database, different colors correspond to different cluster (Pink, green and blue). Result shows that ASTN-2 protein strongly interacts with GAR1, AIMP2, NDST4 and NR3C2.

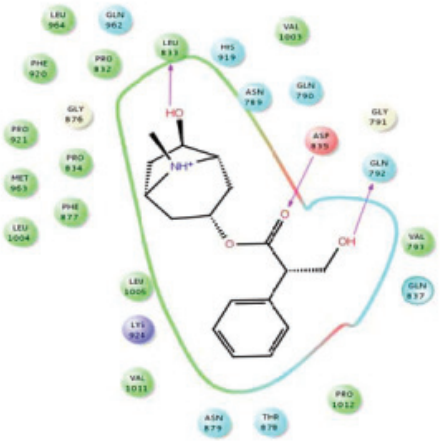

Fig. 6: Interaction map with Anisodamineat different bond position like- H bond with GLN792 and LEU833.

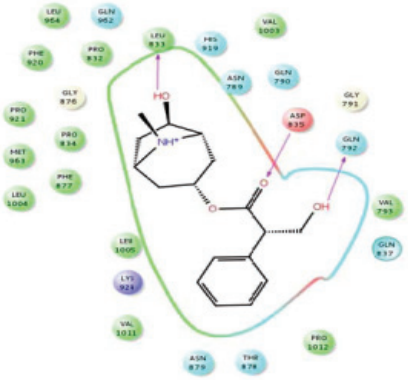

Fig. 7: Interaction map ofSilymarin at positions LEU 833and GLN792.

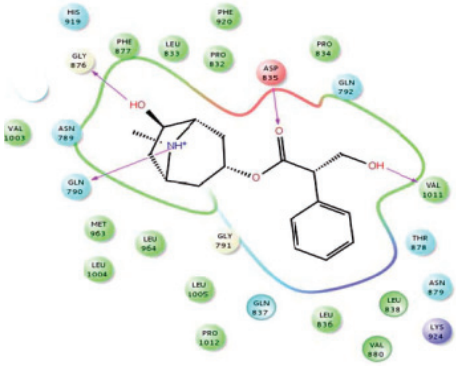

Fig. 8: It shows interaction Troloxwith amino acids at different positions Hydrogen show with GLN 790, VAL 1011, GLY 876 position.

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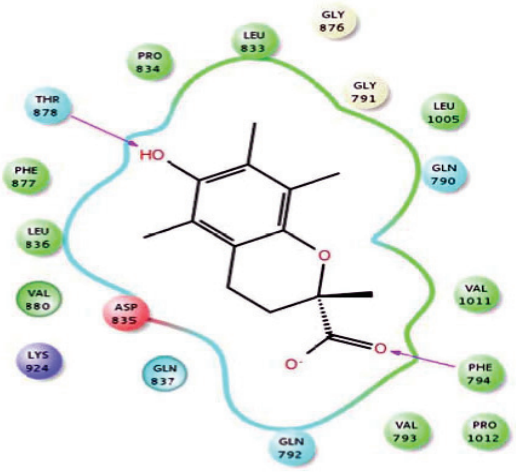

Fig. 9: Ligand Zilascorbshow interaction at H bond THR878.

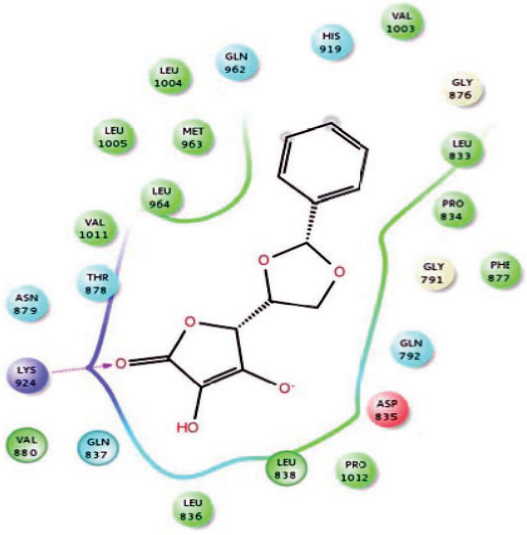

Fig. 10: It shows interaction of Bisphenol Awith amino acidsat different positions

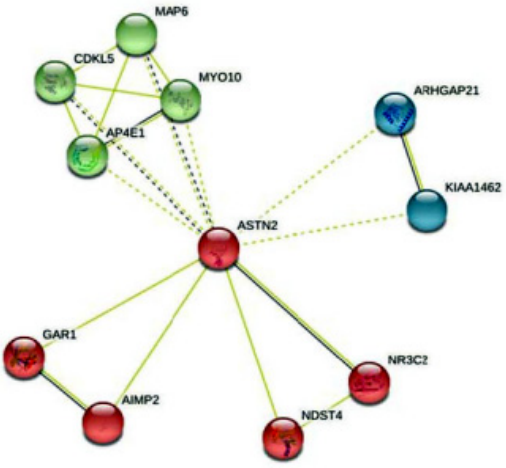

Fig. 11. Protein-Protein Interaction map of ASTN-2 protein from STRING database

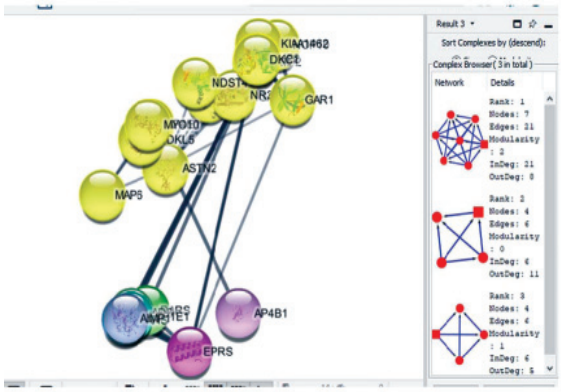

Fig. 12. Visualization of Protein network in Cytoscape for pathway analysis and identification of closest neighbors

Figure 12 shows the analysis of PPI of ASTN-2 protein yellow nodes are clustered together and shows strong interaction with ASTN-2 protein. Further functional annotation and pathway analysis was done to study the function of interacting proteins and their importance in neuronal disorders. Result of function enrichment was shown in table 2

From pathway analysis it was identified that CDKL5 is a important protein associated with Brain-Derived Neurotrophic Factor (BDNF) signalling pathway and mutation in this gene is reported in neurodevelopmental disorders (18)]. Other proteins like ASTN2, GAR1, KIAA1462, MAP6, MYO10, NDST4 and NR3C2 are associated with different types of neuronal

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Table 2 Functional annotation of interacting proteins with ASTN-2

S. No	Protein Symbol	Protein Name	Pathways	Diseases
1	AIMP2	Aminoacyl tRNA synthase complex- interacting multifunctional protein 2	Viral mRNA Translation and tRNA Aminoacylation	Leukodystrophy, Hypomyelinating
2	AP4E1	AP-4 complex subunit epsilon-1	Lysosome	Hereditary spastic paraplegia
3	ARHGAP21	Rho GTPase-activating protein 21	Signalling by GPCR and p75 NTR receptor-mediated signalling	Ciliary Dyskinesia
4	ASTN2	Astrotactin-2		Bardet-Biedl Syndrome 11 and Muscular Dystrophy, Limb-Girdle, Autosomal Recessive 8, schizophrenia
5	CDKL5	Cyclin-dependent kinase-like 5	Brain-Derived Neuro-trophic Factor (BDNF) signalling pathway.	neurological diseases, Rett syndrome
6	GAR1	H/ACA ribonucleoprotein complex subunit 1	Ribosome biogenesis in eukaryotes, Pentose and glucuronate interconversions	Pentosuria, Familial tumoral calcinosis, Spinocerebellar ataxia (SCA), Cartilage-hair hypoplasia
7	KIAA1462	Junctional protein associated with coronary artery disease	NA	Alzheimer Disease
8	MAP6	Microtubule-associated protein 6	NA	Schizophrenia and Disease of Mental Health.
9	MYO10	Unconventional myosin X	Fc gamma R-mediated phagocytosis, Pathogenic Escherichia coli infection	Chronic granulomatous disease, Nemaline myopathy, Immune thrombocy topenia, Activated PI3K-delta syndrome
10	NDST4	Bifunctional heparan sulfate N-deacetylase /N-sulfotransferase 4	Glycosaminoglycan biosynthesis-heparan sulfate / heparin	Multiple exostoses, Desbuquois syndrome, Spondyloocular syndrome, Multiple joint dislocations, short stature, craniofacial dysmorphism, and congenital heart defects, Immunoskeletal dysplasia with neurodevelopmental abnormalities
11	NR3C2	Mineralocorticoid receptor	Aldosterone-regulated sodium reabsorption	Bartter syndrome, Liddle syndrome, Alternating hemiplegia of childhood, Hypertension exacerbated in pregnancy

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development disorder like Bartter syndrome, Liddle syndrome, Bardet-Biedl Syndrome 11, Muscular Dystrophy, Limb-Girdle, Autosomal Recessive 8, schizophrenia, Alzheimer Disease, Rett syndrome etc (19-20). as shown in table 2. This study shows that ASTN-2 protein interacts with other protein that have function in neuro development disorders.

Conclusion

ASNT-2 protein is neurodevelopmental protein any defect in genes expressing this protein can lead to many neuronal development disorders. Copy number variation of this protein also associated with different types of cancer. Abnormality in this protein function is responsible for synaptic disorders, abnormal neuronal migration, disorder in brain development etc. This protein has significant role in neurodevelopmental disorders and can be of significance in drug discovery and development process. ASNT-2 protein and ligand interaction were done to study the possible binding efficiency of this protein. Result show that ASNT-2 protein interacts with Morin and Quercetin. These are natural compounds and have antioxidant property, anticancer, and antibacterial property. These properties of Morin and Quercetin compound make them suitable for pharmacological application. Protein-Protein interaction analysis of ASTN2 protein shows that ASTN2 protein interacts with proteins that are functional in neurodevelopmental disorder. Functional and Pathway analysis of interacting protein verifies that ASTN2 protein can be possible protein target for drug designing and development.

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