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3D Structure modeling & analysis of transmembrane protein EVI2A from Homo sapiens

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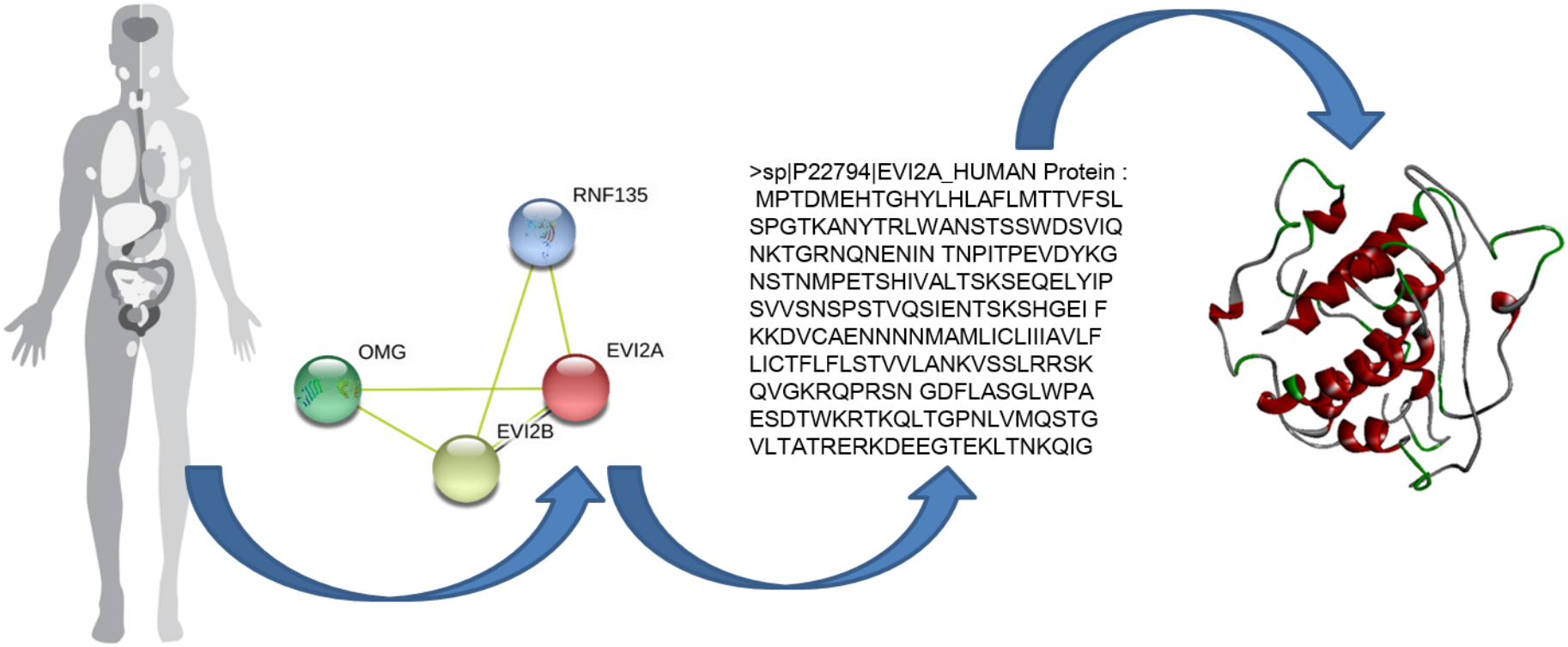
3D Structure modeling & analysis of transmembrane protein EVI2A from Homo sapiens



Abstract:
Protein EVI2A (Ecotropic viral integration site 2A) is a type 1 single pass membrane protein containing 236 amino acid residues. EVI2A is associated with several human diseases such as schizophrenia and numerous malignancies including breast and ovarian cancers. Protein 3D structure helps in understanding the molecular function of the proteins and their important role in the biological scenario if any. Till date no 3D structure of protein EVI2A has been reported in public or private databases. To fill that gap, we evaluated some computational models including comparative methods, de novo approach, ab initio and threading based methods. The multiple models, including 3D model from I-Tasser, afforded a good agreement of output and structural features. A complete model of protein EVI2A was validated by ProSa and Ramachandran analyses. Molecular dynamics (MD) simulations were performed and analyzed using the GROMACS package and active site prediction was carried out using CASTp. The predicted model could be a starting point for structural biologists, drug discovery groups, and scientific community to further enhance their studies.
Keywords: EVI2A; Protein modeling; Gromacs, Molecular dynamics, 3D structure.

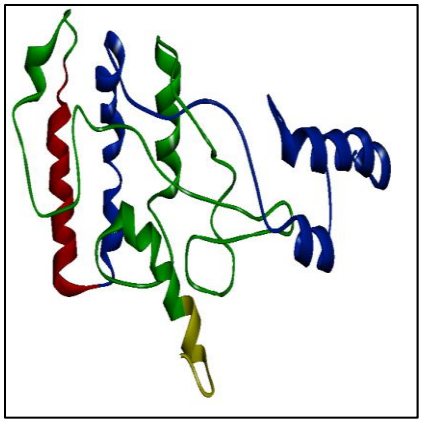
Introduction

- EVI2A stands for ecotropic viral integration site 2A
- It is a single pass membrane protein belonging to the EVI2A family.
- Sequence length : 236 Amino acids
- Function : Transmembrane signaling receptor activity
- Location : Embedded within an intron of NF1 (Neurofibromatosis 1) gene on chromosome 17q 11.2
- EVI2A shows plausible evidence in human disease such as Schizophrenia, numerous malignancy including breast and ovarian cancer.
- Membrane proteins are important to number of biological processes.
- More than 20 % and less than 40 % of proteins found in eukaryotic cells are known to have membrane proteins.
- No 3D structure of EVI2A is reported in Public or private databases.
- Our prime objective of the work is, to predict the 3D structure of EVI2A and study its structural features.
- With a poor homology with known protein structure dataset, here we adopted multiple structure prediction methods to predict the 3D structure of EVI2A (Server we used: Phyre 2; I-Tasser; Robetta; Scratch.
- The modeled structures were tested for their conformation stability using MD simulations and active sites were predicted.

Results and discussion: 2D Structure prediction

Sr no	Tool	Number of Helices	Transmembrane Helix	Inside to outside helices	Outside to inside helices
1	TM Pred (2D)	02	12-30, 134-154	12-30, 134-154	13-30, 133-154
2	DAS TMFilter (2D)	02	8-28, 129-168	--	--
3	DAS –TMPRED (2D)	04	12-24, 13-23 133-161, 134-160	--	--
4	PHOBIUS (2D)	02	5-38 128-168	143-151	144-152
5	Predict Protein (2D)	02	19-25 134-162	1-135	159-236
6	TMHMM (2D)	02	10-27 136-159	1-9, 159-236	28-135
7	HMMTOP (2D)	01	136-160	1-135	170-236
8	PolyPhobius (2D)	1	133-160	--	--
9	SPLIT 4.0 (2D)	1	175-202	--	--
10	SCAMPI (2D)	1	139-159	--	--
11	PHYRE2 (3D) (2D)	1	7-23 38-52 97-99 123-127, 133-160 161-175 214-233	--	--
12	PSI PRED (2D)	5	11-24, 133-159 161-168, 215-220 227-230	--	--

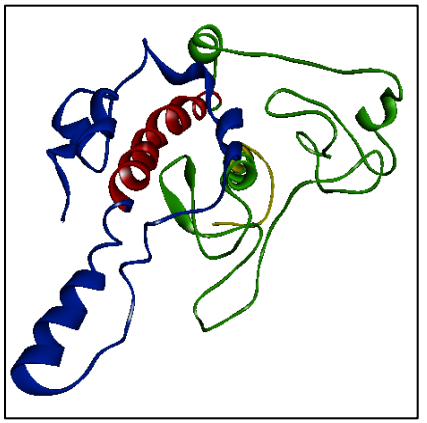
Results and discussion – 3D structure prediction



PHYRE 2

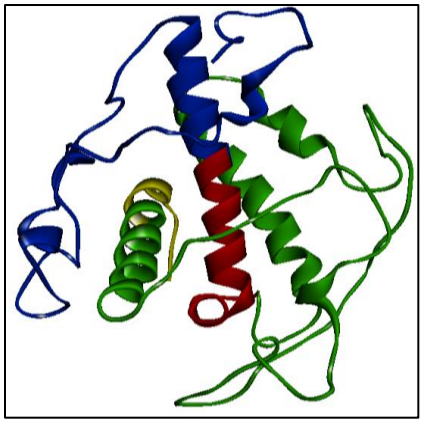
Phyre 2 model has the maximum Co-relation with 2D prédictions

High loop region and poor Co-relation with 2D prédictions



ROBETTA

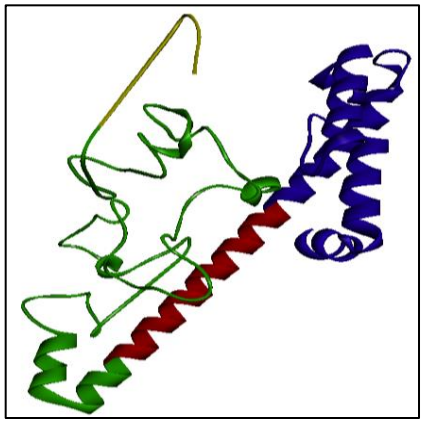
Feature key	Position(s)	Description
Topological domain	31 – 133	Extracellular (Green)
Transmembrane	134 – 154	Helical (Red)
Topological domain	155 – 236	Cytoplasmic (Blue)



I-TASSER

I-Tasser model has the maximum Co-relation with 2D prédictions

High loop region and poor Co-relation with 2D prédictions



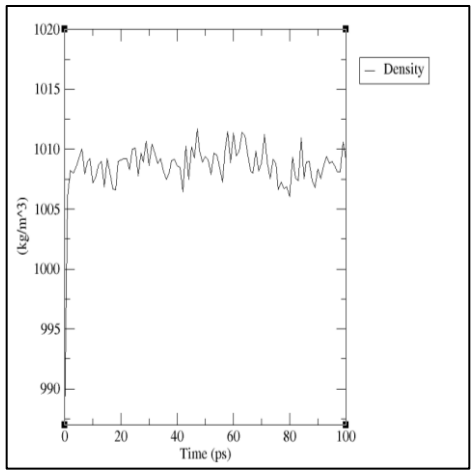
SCRATCH

Results and discussion: Structure assesment

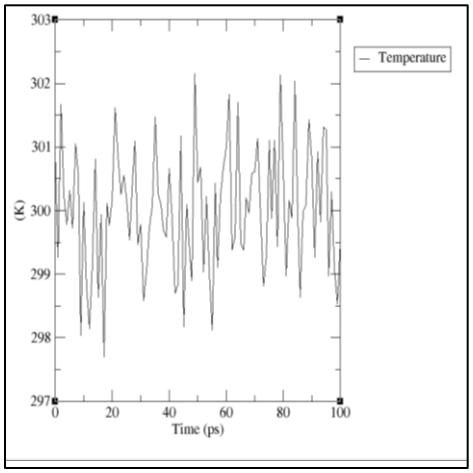
Sr no	Server / Tool	ProSA analysis Z-score	Ramachandran Plot Analysis		
			No of residues in favoured regions	No of residues in allowed regions	No of residues in outlier regions
1	Phyre 2	-3.83	75.6%	12.8%	11.5%
2	I-Tasser	-4.55	67.1%	23.9%	9.0%
3	Robetta	-6.26	90.1%	8.6%	1.3%
4	Scratch	-4.42	88.9%	7.3%	3.8%

- **3D structure predicted using Phyre2 and I-Tasser has exhbited a high degree of co-relation with 2D predictions.**
- **Whereas comparing the Z-score from ProSA and Ramachandran plot analysis 3D structure from I-Tasser is ranked higher than the Phyre 2.**
- **Here, we considered all the 04 predicted structure for further optimization using MD simulation**

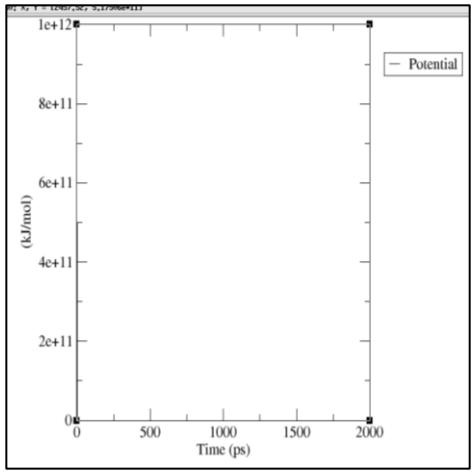
Results and discussion: MD Simulation using GROMACS
Phyre 2 model



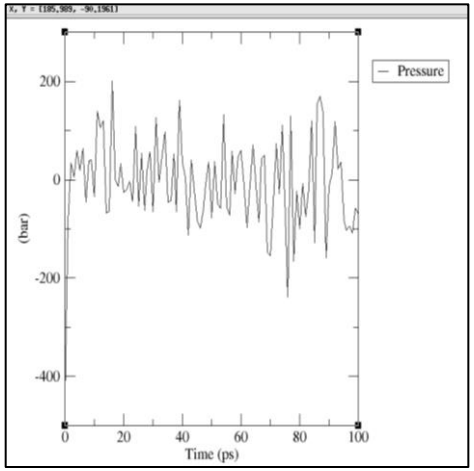
Density Vs Time



Temperature Vs Time

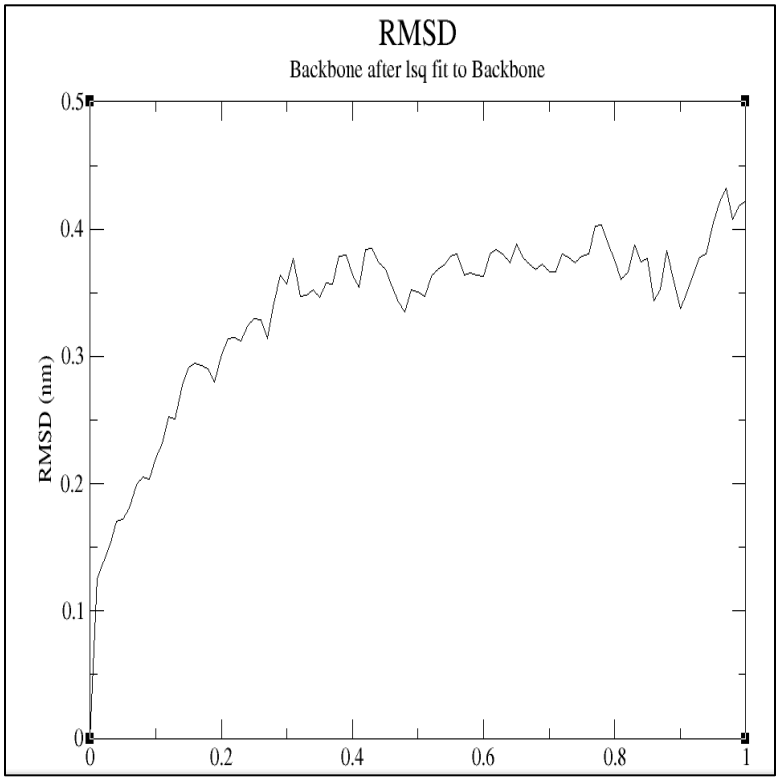


Potential Vs Time

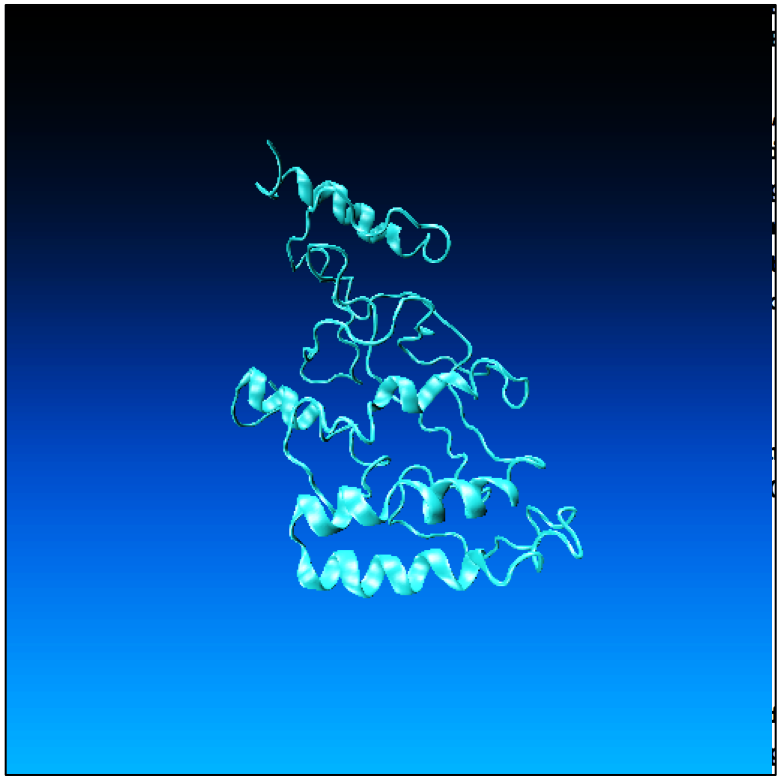


Pressure Vs Time

Results and discussion: MD Simulation using GROMACS
Phyre 2 model

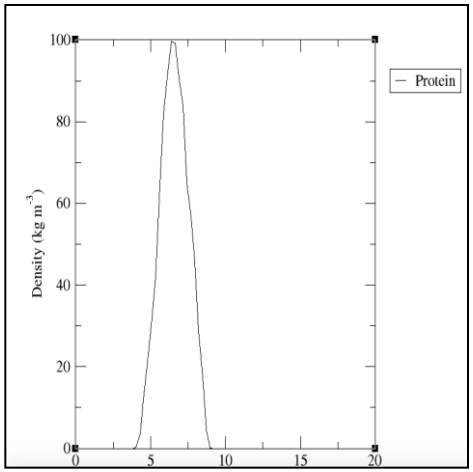


RMSD Vs Time

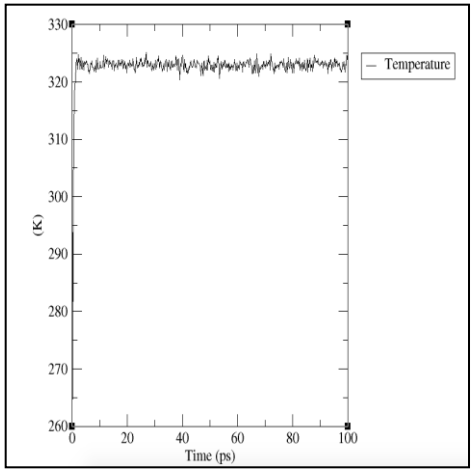


Final model after MD simulation

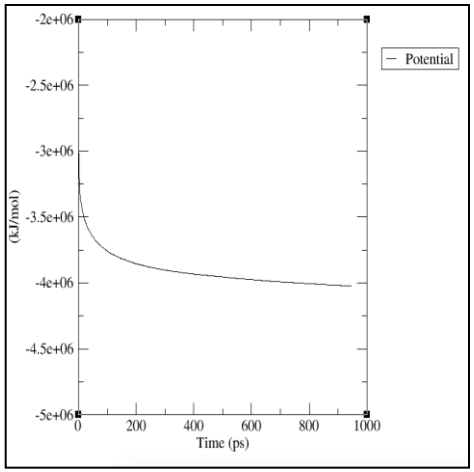
Results and discussion: MD Simulation using GROMACS I-Tasser model



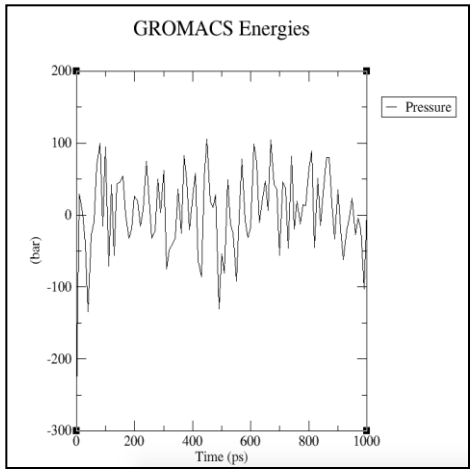
Density Vs Time



Temperature Vs Time

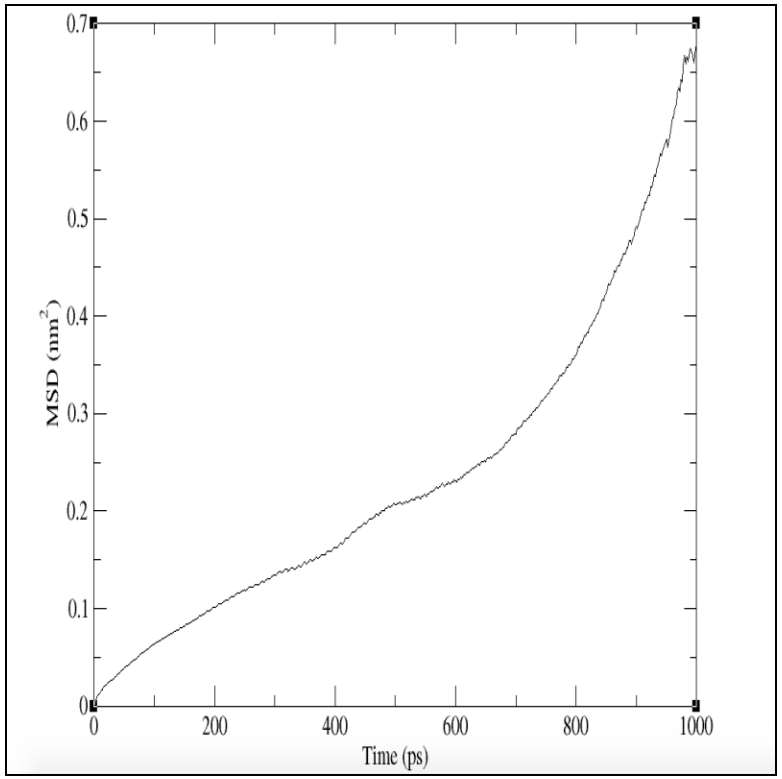


Potential Vs Time

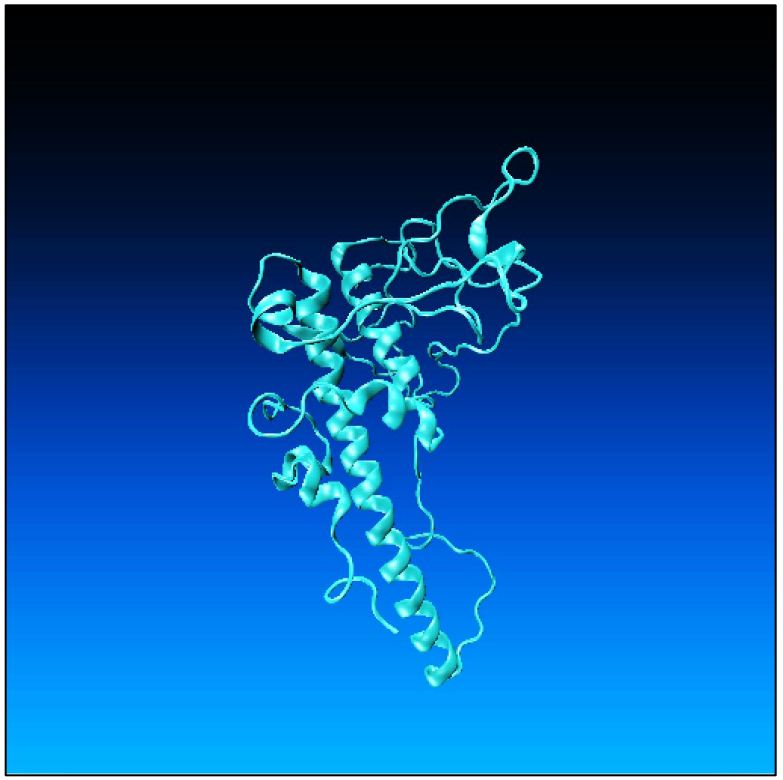


Pressure Vs Time

Results and discussion: MD Simulation using GROMACS I-Tasser model

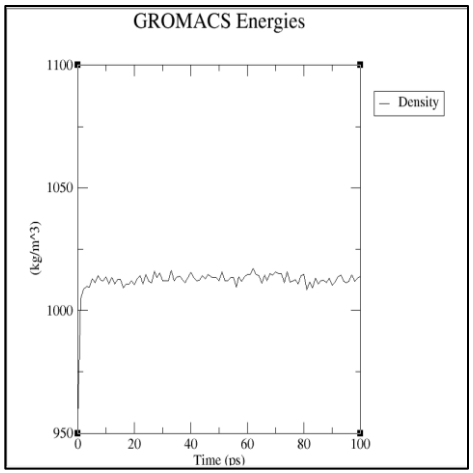


RMSD Vs Time

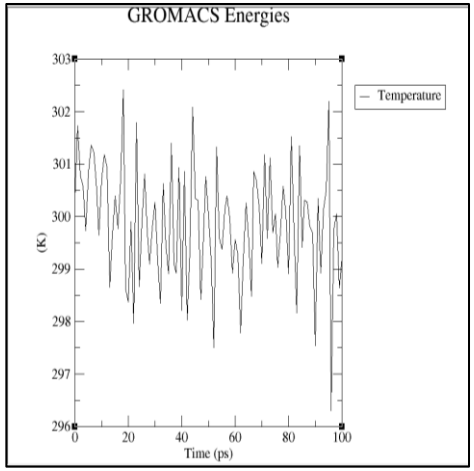


Final model after MD simulation

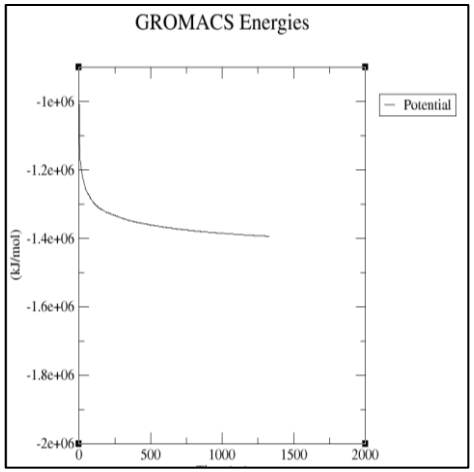
Results and discussion: MD Simulation using GROMACS Robetta model



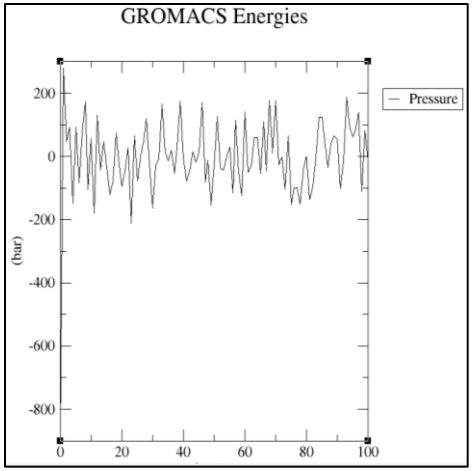
Density Vs Time



Temperature Vs Time

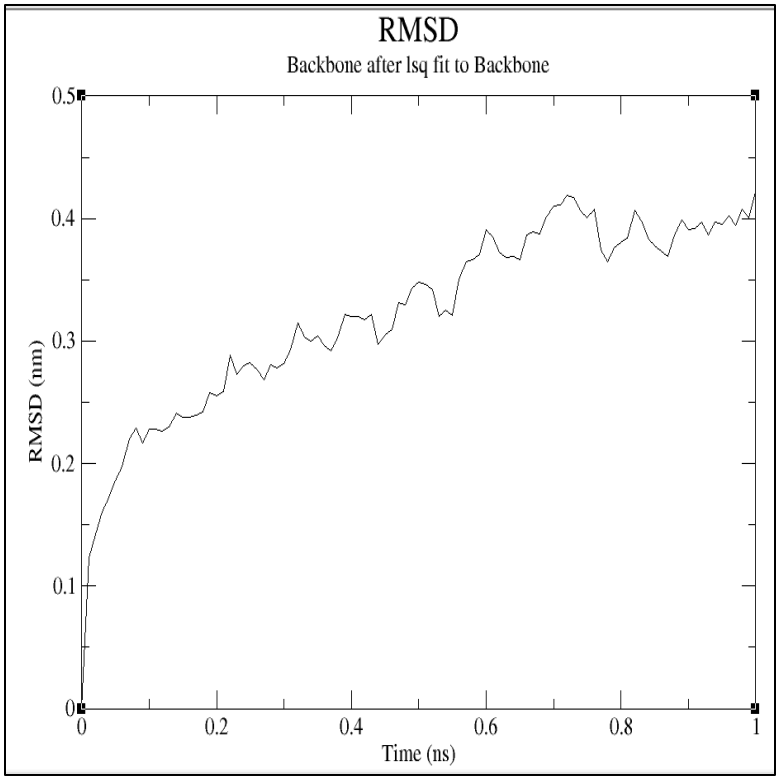


Potential Vs Time

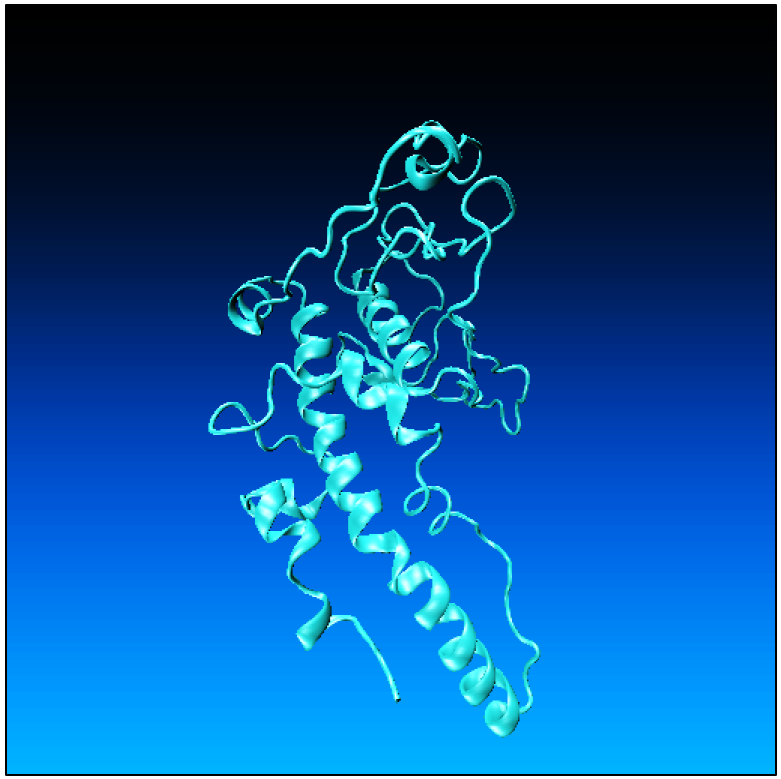


Pressure Vs Time

Results and discussion: MD Simulation using GROMACS Robetta model

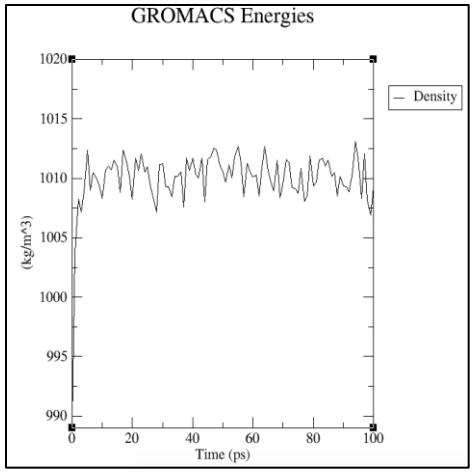


RMSD Vs Time

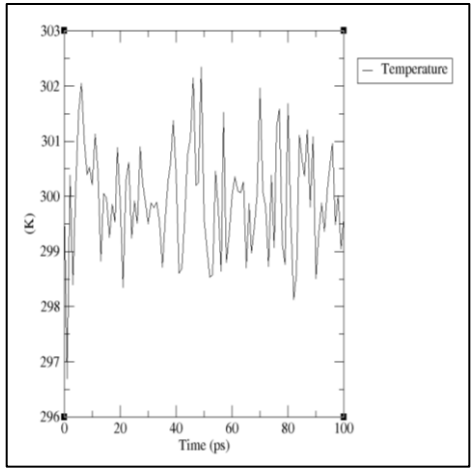


Final model after MD simulation

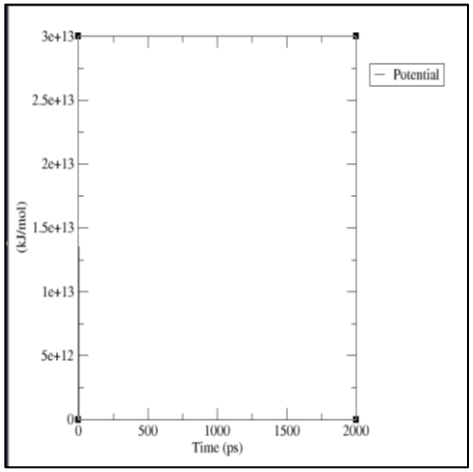
Results and discussion: MD Simulation using GROMACS Scratch model



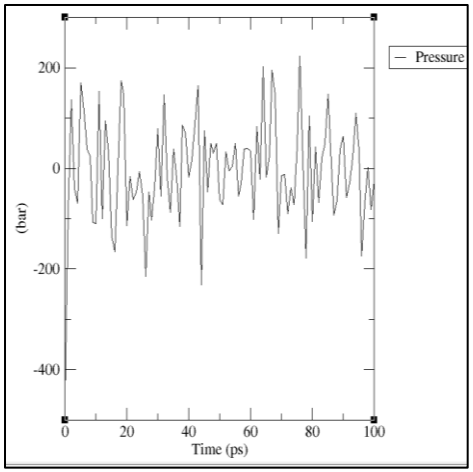
Density Vs Time



Temperature Vs Time

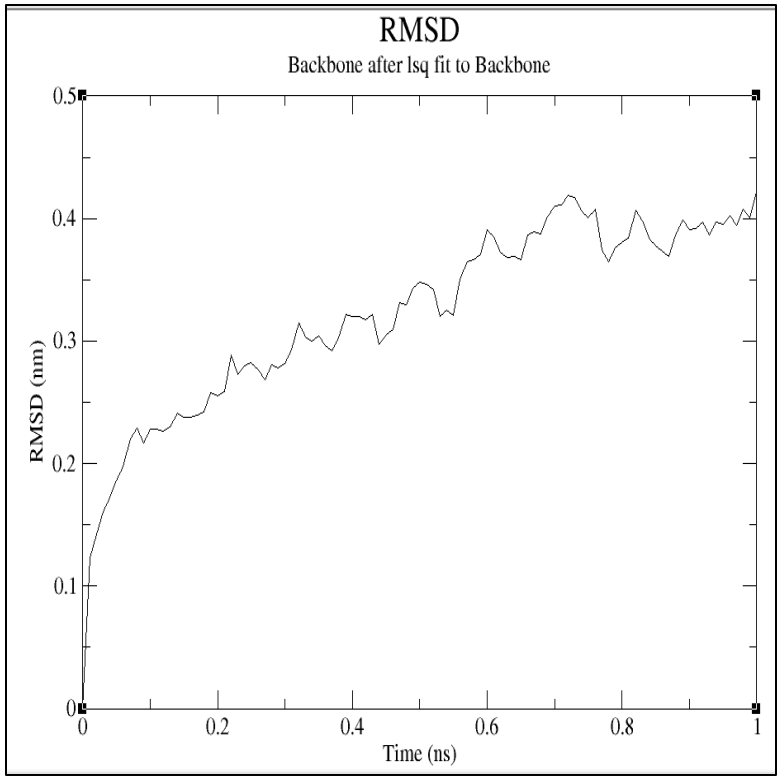


Potential Vs Time

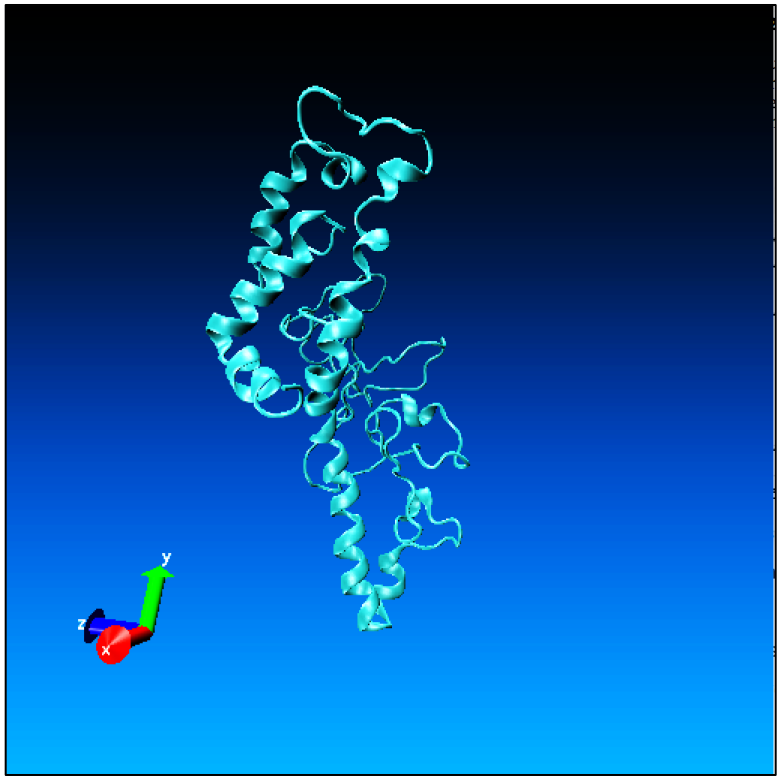


Pressure Vs Time

Results and discussion: MD Simulation using GROMACS Scratch model

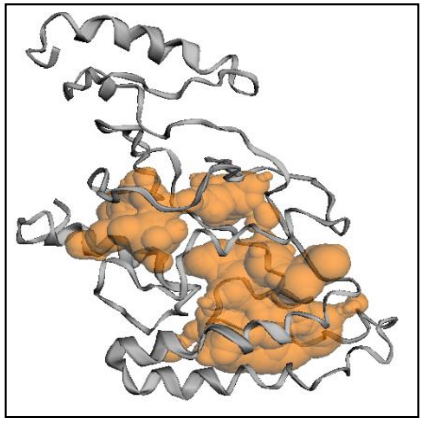


RMSD Vs Time



Final model after MD simulation

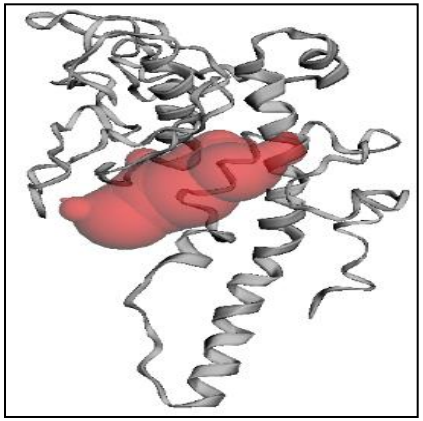
Results and discussion – Active site prediction



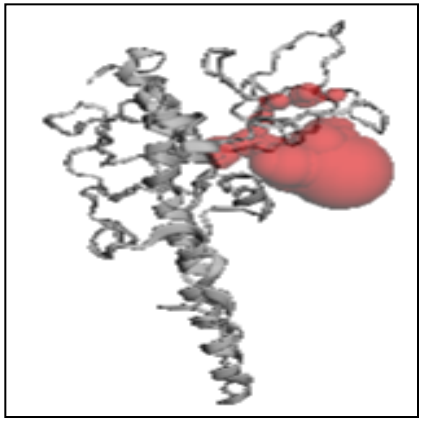
PHYRE 2

- More than 20 hydrophobic sites were predicted for each structure using CASTp online server.
- Here we have reported the cavity with the highest and voulme

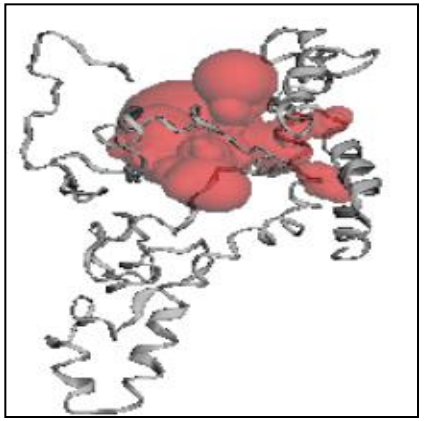
Model	Area (SA)	Volume (SA)
Phyre2	2018.183	1291.995
I-Tasser	488.558	534.480
Robetta	514.480	652.734
Scratch	1120.288	1476.620



ROBETTA



I TASSER



SCRATCH

Conclusion

- In this present study we have predicted and modelled 2D and 3D structure of EVI2A protein that has a plausible role in numerous diseases including Schizophrenia and numerous types of cancer.
- Detailed analysis of the data obtained from structure prediction methods and molecular dynamics calculation confirms the structural conformation of the protein, which may have further more conformational changes and can be detected only with experimental solved ones.
- We can further use the concepts of structure base methods and model the protein – protein interaction to identify the plausible role in numerous disease etiology.
- The current models could be a initial point to identify and model lead inhibitors also.

Acknowledgments

Self funded