IN-SILICO CHARACTERIZATION OF MERISTEM DEFECTIVE (MDF) PROTEIN ASSOCIATED WITH REGULATION OF ROOT MERISTEMS IN *ARABIDOPSIS THALIANA*

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ABSTRACT: Meristem defective protein (MDF) plays its role in proper development of meristem patterns in *Arabidopsis* through regulation of auxins homeostasis. It also enables plants to survive in stress. Present study was designed to characterize MDF protein in *A. thaliana* to get an insight into its molecular aspects. Sequence of protein was retrieved from uniprot database and subjected to CELLO, SOPMA, SWISSMODEL, MEME server and SRING tools. The protein was also docked with four proteins i. e. SR4, RSZ33, PLT1, 2 and 3. The protein was found to localize in nucleus with secondary structure comprising of alpha helix (47.20%), extended strand (6.34%), beta turn (5.37%) and random coil (41.10%). MDF was analyzed as a conserved protein with monomeric structure. Proteins observed as interacting partners of MDF included STA1, T13D8.9, LSM5, LSM2, LSM4 and LSM8. Docking analysis revealed highest and lowest affinities of MDF binding with RS4, RZS33, PLT1, PLT2 and PLT3 in case of FGRTLTPKEAFRLLSHKFHG and IQGQTTHTFEDLNSSAKVSSDYFSQ conserved motifs, respectively. Features of MDF protein explored in this investigation can be exploited for engineering and production of plants with efficient root meristems development.

Keywords: meristem defection protein, *Arabidopsis thaliana*, STRING, alpha helix, configuration.

(Received 11.12.2023 Accepted 13.03.2024)

INTRODUCTION

Meristem defective protein (MDF) also known as Defectively Organized Tributaries 2 (DOT2) is encoded by a splicing factor gene. MDF is a serine-arginine (SR) related arginine-serine (RS) protein comprising of a polypeptide of 820 amino acids (Kakkar, 2019). Sequence of this protein is homologous to squamous cell carcinoma antigen recognized by T-cells 1 (SART1) human protein (de Luxán-Hernández et al., 2023). MDF has two motifs i. e. nuclear localization C-terminus and RNA-binding N-terminus. This protein regulates the development of root meristems and cell division (Kakkar, 2019).

Arabidopsis thaliana commonly called as thale cress and mouse-ear cress belongs to family *Brassicaceae*. Complete genome annotation, shot term reproductive cycle, easy transformations, simple structure of roots, and susceptibility to artificial methods of mutation inductions, has made this plant an excellent model organism for research purpose (Fonseka et al., 2013; Fouracre and Poethig, 2019; Montazeaud et al., 2023; Pasternak et al., 2023). Several genes involving in arsenic resistance, bud and flower development, elongation of primary and lateral roots, photosynthesis and phytohormones synthesis have been deeply investigated in *A. thaliana* (Alvarez et al., 2023; de Luxán Hernández, 2022; Lin et al., 2023; Ranjan et al., 2023; Smyth, 2023; Zeng et al., 2023).

MDF protein, via alternative splicing, regulates the concentration as well as the distribution of auxins in roots in *A. thaliana*. It also plays role in inhibiting the cell death as well as differentiation in root meristems under the conditions of stress (Calixto et al., 2018). Alternative splicing induces diversity at the level of transcriptome and proteome through encoding a large number of transcripts from smaller number of genes. During this process, MDF plays crucial role in spliceosome assembly. A study involved the generation of MDF mutants in *A. thaliana* revealed defective meristems contributed by abnormal transcriptome variations (Casson et al., 2009; de Luxán-Hernández et al., 2023).

During spliceosome complex formation, ribonucleoproteins U1 and U2AF attach to 5' and 3' splice sites, respectively. The later ribonucleoprotein is also bound by polypyrimidine tract i. e. YYY-rich. This step follows the binding of MDF protein with exon enhancing splicer sequence (ESE) of pre-mRNA via its N-terminus end. Afterwards, the U2 and U2AF binding induce conformational change in RNA molecules which promotes binding of tri-snRNP complex of three ribonucleoproteins i. e. U1, U5 and U6 (Wang et al., 2008). Association of MDF with this tri-snRNP complex has been reported to regulate proper splicing of 2037

transcripts (de Luxán-Hernández et al., 2023). In addition to this, MDF controls splicing via interacting with other proteins like SR34, RSZ33, ACC1, PIN family of proteins and PLETHORA gene family i. e. PLT1, 2 and 3 (Figure 1) (Kakkar, 2019; Thompson et al., 2023; Wisniewska et al., 2006).

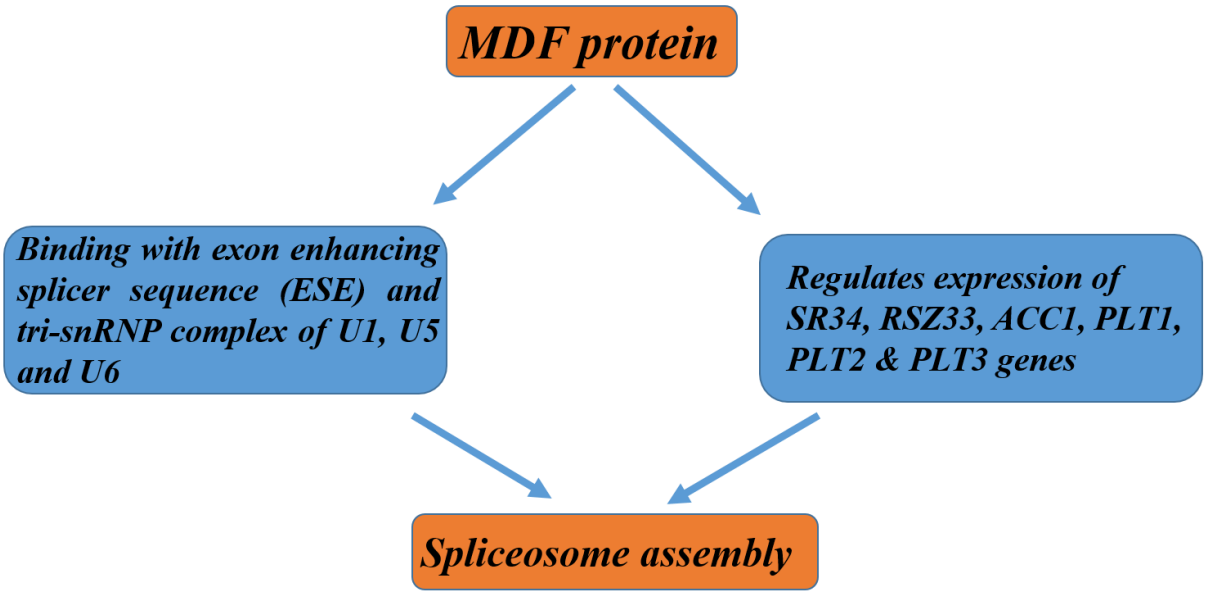

Figure 1: Role of MDF protein in spliceosome assembly and auxins homeostasis

Keeping in view the significance of MDF protein in plant roots development and the wide adaptability of *A. thaliana* as model system, we initiated present study to target the characterization of MDF protein in *A. thaliana*. This characterization included the prediction of sub-cellular localization, two dimensional (2D), three dimensional (3D) configuration, conserved domains and interactions with other proteins. This analysis can be helpful in designing strategies for optimizing activity of this protein in plants root growth under stress conditions not only in *A. thaliana* but also in economically important crops.

METHODOLOGY

Sequence retrieval from Uniprot database: Sequence of MDF protein with accession no. Q9LFE0 in *A. thaliana* was retrieved from Uniprot database (https://www.uniprot.org, accessed in Sep 2023) (Consortium, 2019). The sequence is shown as follows;

MEVEKSKSRHEIREERADYEGSPVREHRD
GRRKEKDHRSKDKEKDYDREKIRDKDHRDKEKER
DRKRSRDEDTEKEISRGRDKEREKDKSRDRVKEKD
KEKERNRHKDRENERDNEKEKDKDRARVKERAS
KKSHEDDDETHKAAERYEHSDNRGLNEGGDNVD
AASSGKEASALDLQNRILKMREERKKKAEDASDA
LSWVARSRKIEEKRNAEKQRAQQLSRIFEEQDNLN
QGENEDGEDGEHLSGVKVLHGLEKVVEGGAVILT
LKDQSVLTDGDVNNEIDMLENVEIGEQKRRNEAY
EAAKKKKGIYDDKFNDDPGAEKKMLPQYDEAAT
DEGIFLDAKGRFTGEAEKKLEELRKRIQGQTTHTFE
DLNSSAKVSSDYFSQEEMLKFKKPKKKKQLRKKD
KLDLSMLEAEAVASGLGAEDLGSRKDGRRQAMK
EEKERIEYEKRSNAYQEAIAKADEASRLLRREQVQ
PFKRDEDESMVLADDAEDLYKSLEKARRLALIKKE
EAGSGPQAVAHLVASSTNQTTDDNTTTGDETQEN
TVVFTEMGDFVWGLQRENDVRKPESEDVFMEEDV
APKAPVEVKEEHPDGLTEVNDTDMDAAEDSSDTK
EITPDENIHEVAVGKGLSGALKLLKDRGTLKEKVE
WGGRNMDKKKSKLVGIVDDDGGKESKDKESKDR
FKDIRIERTDEFGRTLTPKEAFRLLSHKFHGKGPGK
MKEEKRMKQYQEELKLKQMKNSDTPSQSVQRMR
EAQAQLKTPYLVLSGHVKPGQTSDPQSGFATVEK
DVPGSLTPMLGDRKVEHFLGIKRKSEPGNSDTPPK
RPKP

CELLO: prediction of sub-cellular localization: To predict the localization of MDF protein at sub cellular level, CELLO tool (cello.life.nctu.edu.tw, accessed in Sep 2023) was employed (Xiong et al., 2016).

SOPMA tool: prediction of 2D configuration: For assessment of the 2D configuration, secondary structure prediction tool, SOPMA (https://npsa-prabi.ibcp.fr/cgi-bin/ npsa_automat.pl?page=/NPSA/npsa_sopma.html, accessed in Sep 2023) was accessed. It predicted four attributes of 2D structure i. e. alpha helix, extended strand, beta turn and random coil (Geourjon and Deleage, 1995).

SWISSMODEL: prediction of 3D configuration: For interpretation of 3D configuration, homology modelling

server SWISSMODEL (https://swissmodel.expasy.org, accessed in Sep 2023) was used. Modelling was performed via searching for the template and target-template alignment (Waterhouse et al., 2018).

MEME server: prediction of conserved domains: MEME suite version 5.5.5 (Multiple Em for Motif Elicitation) was employed (https://meme-suite.org/meme/tools/meme, accessed in Sep 2023) (Bailey et al., 2015). For this analysis, for motif site distribution the option of any number of repetitions (anr) and 10 number of motifs was selected.

STRING tool: prediction of protein interactions: Protein-protein interactions network functional enrichment analysis was performed using STRING tool (https://string-db.org, accessed in Sep 2023) developed by Swiss Institute of Bioinformatics (SIB) (Bajpai et al., 2020).

HDOCK server: Docking of MDF conserved motifs with ligands: Docking was performed to analyze the binding affinity of three conserved domains of MDF protein predicted in present study i. e. AQAQLKTPYLVLSGHVKPGQTSDPQ, IQGQTTHTFEDLNSSAKVSSDYFSQ and FGRTLTPKEAFRLLSHKFHG, with five protein ligands i. e. SR4, RSZ33, PLT1, PLT2 and PLT3. For analysis, HDOCK server (hdock.phys.hust.edu.cn, accessed in Sep 2023) was employed and docking score, confidence score and RMSD values were determined (Yan et al., 2020).

RESULT

Sub-cellular localization: The highest score was recorded in nucleus i. e. 4.172 followed by cytoplasmic (0.410), endoplasmic reticulum (0.110), mitochondrial (0.076), plasma membrane (0.066), cytoskeletal (0.060), extracellular (0.050), chloroplast (0.019), vacuole (0.015), golgi complex (0.010), peroxisomal (0.009) and lysosomal (0.002) (Figure 2a).

Secondary (2D) structure: Total 387 amino acids were found to make up alpha helix constituting 47.20% of protein. Extended strand, beta turn and random coil were found to be comprised of 52, 44 and 337 amino acids, respectively. Hence, 41.10% portion of protein was made up of random coil. Very few part of protein was composed of beta turn (5.37%) and extended strand (6.34%) (Figure 2b).

Three dimensional (3D) structure: The template identified was SART-1 family protein DOT2 with 100% sequence identity. The GMQE value was observed to be 0.66 (Figure 3). MDF protein was monomeric with highly complex folding (Figure 3).

Conserved domains: The conserved domains predicted at twenty different locations of protein through MEME suite. The p-values ranged between 1.05e-8 and 8.18e-12. The E-values were recorded to be 1.0e+002 to 7.0e+001 (Table 1 and Figure 4).

Interaction with other proteins: Stabilized 1 (STA1) is a pre-mRNA splicing factor, T13D8.9 is an RNA recognition motif containing protein (RRM), Sm like proteins (LSM5, LSM2, LSM4 and LSM8) which are the components of LSM protein complex that is associated with RNA processing, U5 small nuclear ribonucleoprotein component (CLO) which is a 110kDa splicing factor involved in pre-mRNA splicing, small nuclear ribonucleoprotein SmD2 (F17A22.3), U5 small nuclear ribonucleoprotein component (GFL) which is a 109kDa 110kDa splicing factor involved in pre-mRNA splicing and thioredoxin like U5 (Q6NMD4-ARATH) which is mRNA splicing factor. The scores recorded were above 0.9 in all the cases (Figure 5).

Binding affinities of conserved domains of MDF with targeted proteins: Docking analysis revealed the binding tendencies of MDF conserved domains with different ligands like SR4, RSZ33, PLT1, PLT2 and PLT3. (Table 2 and Figure 6).

(a) 1^{st} domain with SR4 (b) 2^{nd} domain with SR4 (c) 3^{rd} domain with SR4 (d) 1^{st} domain with RSZ33 e) 2^{nd} domain with RSZ33 (f) 3^{rd} domain with RSZ33 (g) 1^{st} domain with PLT1 (h) 2^{nd} domain with PLT1 (i) 3^{rd} domain with PLT1 (j) 1^{st} domain with PLT2 (k) 2^{nd} domain with PLT2 (l) 3^{rd} domain with PLT2 (m) 1^{st} domain with PLT3 (n) 2^{nd} domain with PLT3 (o) 3^{rd} domain with PLT3

Maximum binding affinity (-252.13) was observed in case of third conserved motif with PLT2. Lowest affinity (-160.83) was observed for second conserved motif with RSZ33. In all the cases, highest docking scores were obtained with third conserved motif. i. e. -217.66 (SR4), -190.11 (RSZ33), -216.03 (PLT1), -252.13 (PLT2) and -229.84 (PLT3). Lowest binding affinity was showed by second conserved motif i. e. -184.28 (SR4), -160.83 (RSZ33), -198.66 (PLT1), -192.62 (PLT2) and -162.96 (PLT3).

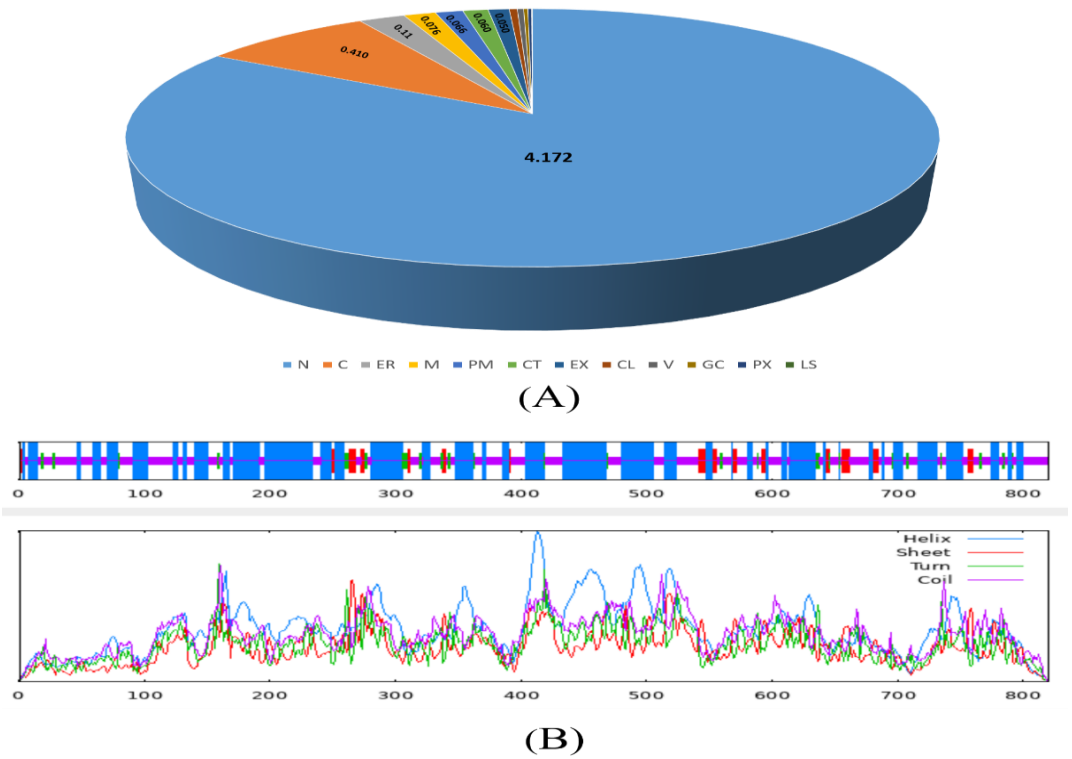

Figure 2: In-silico prediction of sub-cellular localization and 2D configuration of MDF protein
(a) CELLO tool (b) SOPMA tool

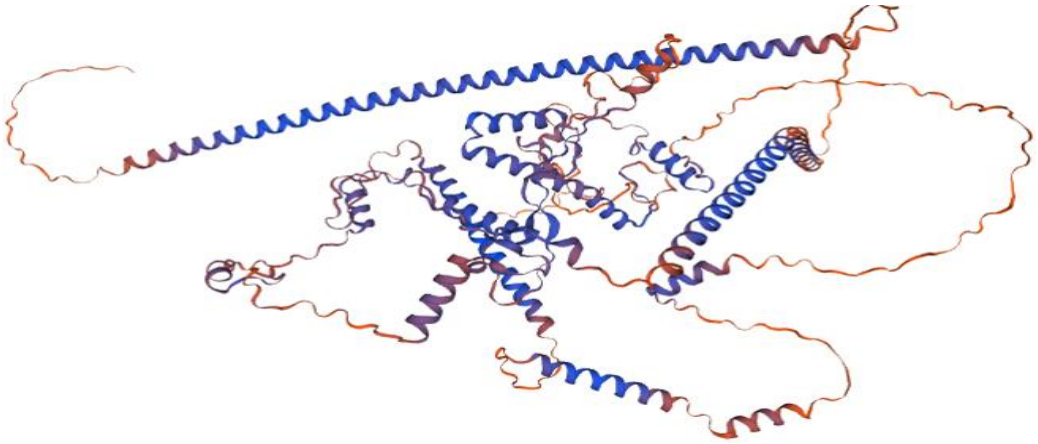

Figure 3: Three dimensional (3D) configuration of MDF protein predicted using SWISSMODEL modelling workspace.

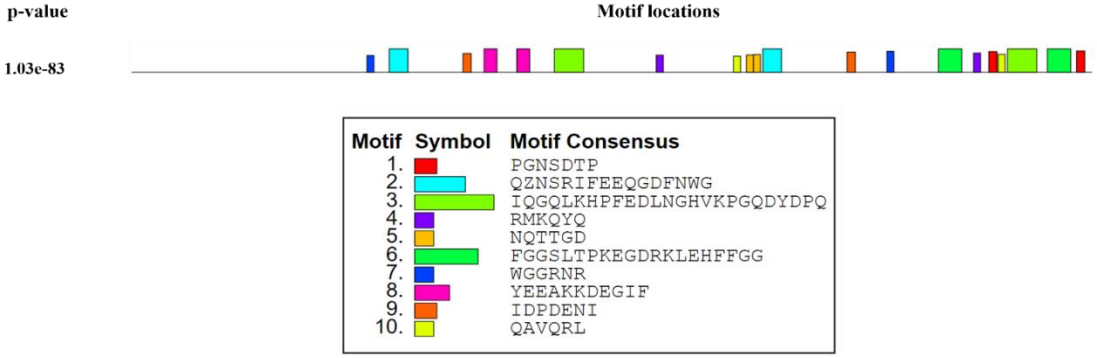

Figure 4: Prediction of conserved domains of MDF protein based on MEME suite.

Table 1: Conserved domains of MDF protein, their location, p-value and E-value, predictd using MEME suite.

#	Motif sequence	Location	p-value	E-value
1	HFLGIKRKSE **PGNSDTP** PKRPKP	808	7.87e-10	3.0e+001
2	QYQEELKLKQ **MKNSDTP** SQSVQRMREA	733	1.49e-9	
3	DDNTTTGDET **QENTVVFTEMGDFVWG** LQRENDVRKP	540	1.64e-19	7.0e+001
4	EKRNAEKQRA **QQLSRIFEEQDNLNQG** ENEDGEDGEH	221	5.88e-17	
5	PSQSVQRMRE **AQAQLKTPYLVLSGHVKPGQTSDPQ** SGFATVEKDV	749	3.97e-26	5.5e+001
6	EKKLEELRKR **IQGQTTHTFEDLNSSAKVSSDYFSQ** EEMLKFKKPK	362	2.15e-25	
7	KGPGKMKEEK **RMKQYQ** EELKLKQMKN	720	6.52e-9	1.0e+002
8	EEKERIEYEK **RSNAYQ** EAIAKADEAS	449	5.06e-8	
9	ASSTNQTTDD **NTTTGD** ETQENTVVFT	532	2.56e-8	3.0e+002
10	AVAHLVASST **NQTTDD** NTTTGDETQE	526	4.47e-8	
11	QSGFATVEKD **VPGSLTPMLGDRKVEHFLGI** KRKSEPGNSD	783	7.88e-22	3.4e+002
12	KDIRIERTDE **FGRTLTPKEAFRLLSHKFHG** KGPGKMKEEK	690	5.85e-21	
13	DRGTLKEKVE **WGGRNM** DKKKSKLVGI	646	1.10e-9	2.2e+002
14	KAEDASDALS **WVARSR** KIEEKRNAEA	202	7.36e-8	
15	PGAEKKMLPQ **YDEAATDEGIF** LDAKGRFTGE	330	5.26e-13	3.8e+002
16	IGEQKRRNEA **YEAAKKKKGIY** DDKFNDDPGA	302	8.18e-12	
17	AAEDSSDTKE **ITPDENI** HEVAVGKGLS	612	2.16e-9	4.8e+002
18	VLTDGDVNNE **IDMLENV** EIGEQKRRNE	284	1.05e-8	
19	KQMKNSDTPS **QSVQRM** REAQAQLKTP	741	2.17e-8	2.6e+003
20	IKKEEAGSGP **QAVAHL** VASSTNQTTD	515	1.43e-7	

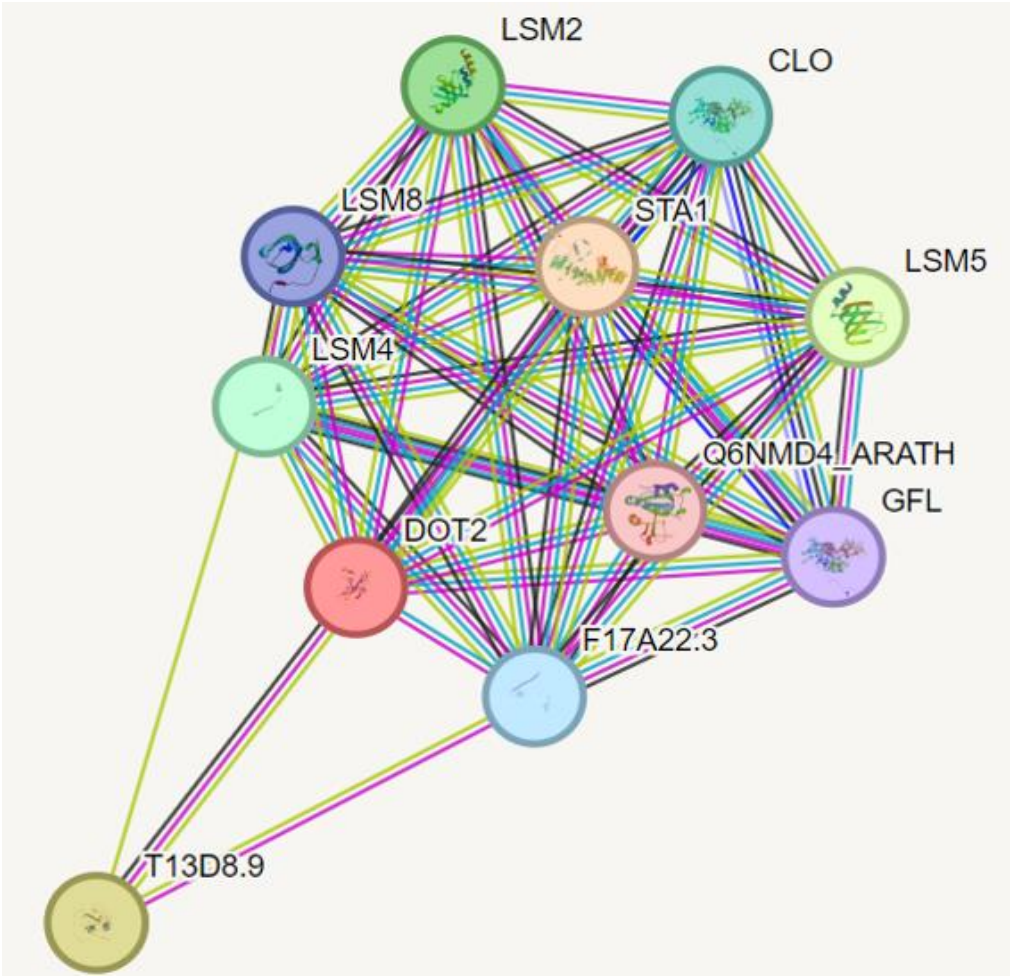

Figure 5: Prediction of MDF interaction with other proteins using STRING tool

Table 2: Docking scores, confidence scores and ligand rmsd predicted through docking of MDF conserved domains with SR4, RSZ33, PLT1, PLT2 and PLT3.

#	Conserved Domain	Docking score	Confidence score	Ligand rmsd Å
		SR4		
1	AQAQLKTPYLVLSGHVKPGQTSDPQ	-187.81	0.6805	50.21
2	IQGQTTHTFEDLNSSAKVSSDYFSQ	-184.28	0.6650	26.14
3	FGRTLTPKEAFRLLSHKFHG	-217.66	0.7947	406.66
		RSZ33		
1	AQAQLKTPYLVLSGHVKPGQTSDPQ	-172.76	0.6119	42.65
2	IQGQTTHTFEDLNSSAKVSSDYFSQ	-160.83	0.5539	39.43
3	FGRTLTPKEAFRLLSHKFHG	-190.11	0.6904	422.88
		PLT1		
1	AQAQLKTPYLVLSGHVKPGQTSDPQ	-211.89	0.7752	60.47
2	IQGQTTHTFEDLNSSAKVSSDYFSQ	-198.66	0.7258	60.90
3	FGRTLTPKEAFRLLSHKFHG	-216.03	0.7893	404.57
		PLT2		
1	AQAQLKTPYLVLSGHVKPGQTSDPQ	-195.06	0.7112	76.81
2	IQGQTTHTFEDLNSSAKVSSDYFSQ	-192.62	0.7011	64.29
3	FGRTLTPKEAFRLLSHKFHG	-252.13	0.8852	416.83
		PLT3		
1	AQAQLKTPYLVLSGHVKPGQTSDPQ	-188.93	0.6854	35.89
2	IQGQTTHTFEDLNSSAKVSSDYFSQ	-162.96	0.5644	38.70
3	FGRTLTPKEAFRLLSHKFHG	-229.84	0.8316	396.06

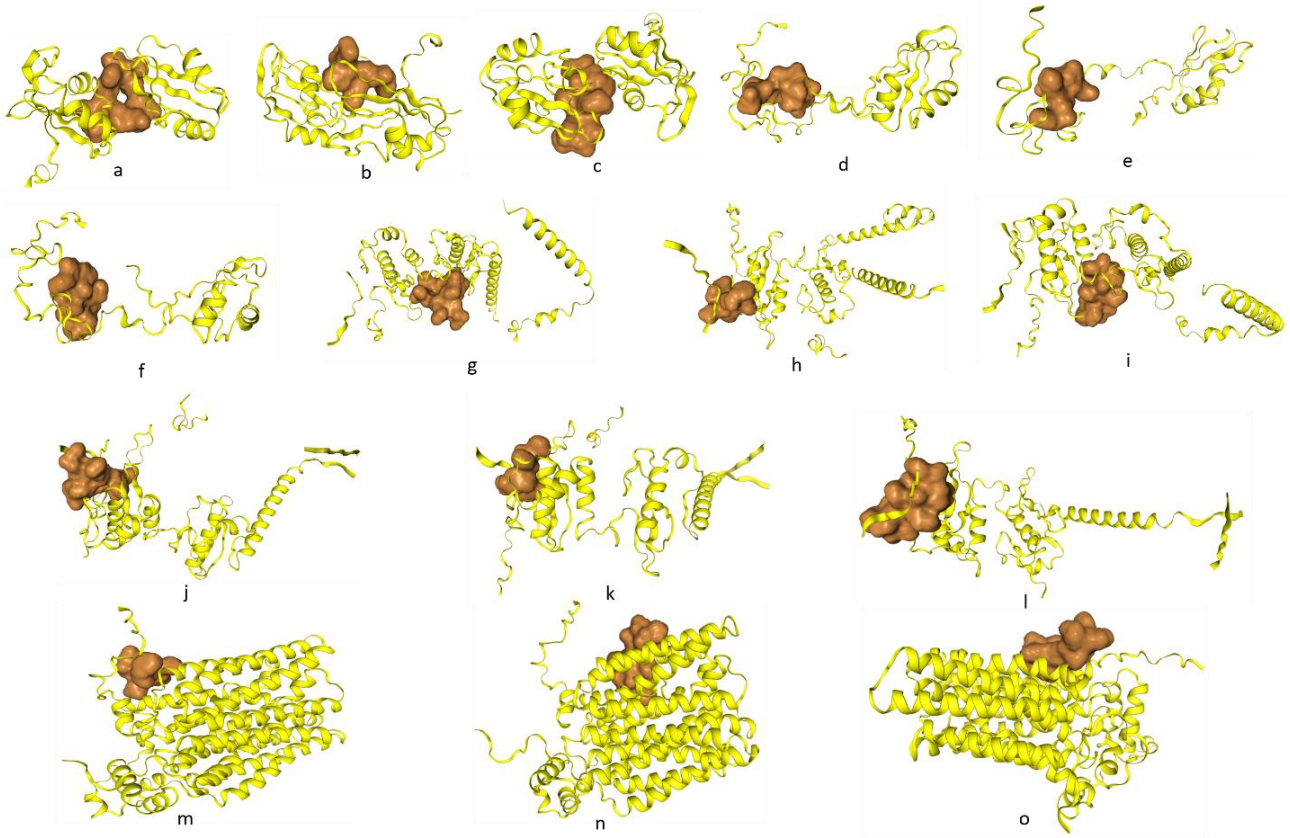

Figure 6: Docking analysis of MDF conserved domains with SR4, RSZ33, PLT1, PLT2 and PLT3 predicted using HDOCK server

DISCUSSION

Auxins are associated with patterning of root meristems at embryonic stages in *A. thaliana*. MDF protein has central role in auxins homeostasis. It regulates the spliceosome assembly and expression of various genes which are associated with auxins transport and meristems development (Shen, 2019). So far, no one has targeted the comprehensive characterization of MDF protein using in-silico tools. Therefore, present work is the first ever project reporting it's in silico analysis.

Comparison of present study findings with literature revealed that sub-cellular localization of MDF in nucleus is consistent with previously reported work. The study analyzed location of protein using a different in silico tool i. e. Plant P-Loc protein localization software (Chou and Shen, 2007). Assessment of 3D configuration via SWISSMODEL using template search mode, revealed the similarity of MDF with human hSART-1 protein which is also consistent with previous findings (Blencowe et al., 1999). Multiple conserved domains were identified in present study which is inconsistent with literature reporting only RNA binding and nuclear localization motifs and no conserved domains in MDF (Terribilini et al., 2007). An initial study has reported the interaction of MDF with STA1 protein, hence our finding is consistent with literature. However, in the same study, no interaction was observed between MDF and LSM (Thompson et al., 2023).

To maintain the auxins homeostasis and distribution, MDF regulates the expression of several other proteins. A study conducting using MDF mutants revealed the reduced level of PLT transcripts leading to disruption of root meristem patterning (Casson et al., 2009). Another gene RSZ33 encodes alternatively spliced mRNA which is abundantly present in root meristems. In a project, mis-spliced RSZ33 transcripts were reported in MDF mutants causing retarded root meristems (Thompson et al., 2023). Keeping in view these interacting genes of MDF, three long conserved domains predicted in the analysis of MDF protein, were selected for docking against SR4, RSZ33, PLT1, PLT2 and PLT3 proteins. This helped to check out the binding tendency of MDF with these protein ligands. It was found that the motifs in SR4, RSZ33, PLT1, PLT2 and PLT3 proteins, respectively have higher binding tendencies with MDF. These domains can be targeted for modifying MDF regulatory activity of spliceosome, thus improving its function of meristems development. Literature does not report any docking analysis of MDF protein of *A. thaliana*.

Conclusion: Analysis at the molecular level of MDF and its interactions predicted in present study might be helpful to explore the molecular mechanisms underlying its activity. Through modifications in MDF protein the plants seeds responses to stressors should be evaluated in different important plants. The protein imparting highest stress tolerance can be useful in development of good quality crops.

Statements and Declarations

Funding: N/A

Acknowledgements: We acknowledge the School of Biochemistry and Biotechnology, University of the Punjab, Lahore.

Author contributions: Fatima Muccee wrote the manuscript, Aleena Ahmed perceived the idea, Afreen Fatima Mirza retrieved data from database, Sara Rehman designed methodology, Bisma Nadeem Siddiqui, Aqsa Arshad and Noor Ul Huda performed analysis.

Informed consent: N/A

Competing interests: The authors have no competing interests.

Data availability statement: The sequences of protein documented in present study are available at Uniprot database (www.uniprot.org).

Submission declaration and verification: The work is not published previously and it is not under consideration for publication elsewhere.

REFERENCES

Alvarez JM, Rodríguez SM, Fuente-Maqueda F, Feito I, Ordás RJ., Cuesta C. 2023. Identification of candidate genes involved in bud growth in pinus pinaster through knowledge transfer from *Arabidopsis thaliana* models. Forests 14: 1765.

Bailey TL, Johnson J, Grant CE., Noble WS. 2015. The MEME suite. Nucleic acids research 43: W39-W49.

Bajpai AK, Davuluri S, Tiwary K, Narayanan S, Oguru S, Basavaraju K, Dayalan D, Thirumurugan K., Acharya KK. 2020. Systematic comparison of the protein-protein interaction databases from a user's perspective. Journal of Biomedical Informatics 103: 103380.

Blencowe BJ, Bowman JAL, McCracken S., Rosonina E. 1999. SR-related proteins and the processing of messenger RNA precursors. Biochemistry and cell biology 77: 277-291.

Calixto CPG, Guo W, James AB, Tzioutziou NA, Entizne JC, Panter PE, Knight H, Nimmo HG, Zhang R., Brown JWS. 2018. Rapid and dynamic alternative splicing impacts the *Arabidopsis* cold response transcriptome. The Plant Cell 30: 1424-1444.

Casson SA, Topping JF., Lindsey K. 2009. Meristem-defective, an RS domain protein, is required for the correct meristem patterning and function in Arabidopsis. The Plant Journal 57: 857-869.

Chou KC., Shen HB. 2007. Large-scale plant protein subcellular location prediction. Journal of cellular biochemistry 100: 665-678.

Consortium U. 2019. UniProt: a worldwide hub of protein knowledge. Nucleic acids research 47: D506-D515.

de Luxán-Hernández C, Lohmann J, Tranque E, Chumova J, Binarova P, Salinas J., Weingartner M. 2023. MDF is a conserved splicing factor and modulates cell division and stress response in *Arabidopsis*. Life Science Alliance 6.

de Luxán Hernández C. 2022. Characterization of meristem defective as a splicing factor involved in the DNA damage response in plantsStaats-und Universitätsbibliothek Hamburg Carl von Ossietzky.

Fonseka DLCK, Yang X, Mudge A, Topping JF., Lindsey K. 2013. Embryogenesis in *Arabidopsis*: Signaling, genes, and the control of identity. Seed Genomics: 21-42.

Fouracre JP., Poethig RS. 2019. Role for the shoot apical meristem in the specification of juvenile leaf identity in *Arabidopsis*. Proceedings of the National Academy of Sciences 116: 10168-10177.

Geourjon C., Deleage G. 1995. SOPMA: significant improvements in protein secondary structure prediction by consensus prediction from multiple alignments. Bioinformatics 11: 681-684.

Kakkar M. 2019. Investigation of the Role of MERISTEM-DEFECTIVE (MDF) in *Arabidopsis thaliana* Root DevelopmentDurham University.

Lin Q, Gong J, Zhang Z, Meng Z, Wang J, Wang S, Sun J, Gu X, Jin Y., Wu T. 2023. The *Arabidopsis thaliana* trehalose-6-phosphate phosphatase gene AtTPPI regulates primary root growth and lateral root elongation. Frontiers in Plant Science 13: 1088278.

Montazeaud G, Helleu Q, Wuest SE., Keller L. 2023. Indirect genetic effects are shaped by demographic history and ecology in *Arabidopsis thaliana*. Nature Ecology & Evolution: 1-14.

Pasternak T, Pérez-Pérez JM, Ruperti B, Aleksandrova T., Palme K. 2023. A new in vitro growth system for phenotypic characterization and seed propagation of *Arabidopsis thaliana*. Journal of Plant Growth Regulation: 1-7.

Ranjan A, Gautam S, Michael R, Shukla T., Trivedi PK. 2023. Arsenic-induced galactinol synthase1 gene, AtGolS1, provides arsenic stress tolerance in *Arabidopsis thaliana*. Environmental and Experimental Botany 207: 105217.

Shen W. 2019. Genetic control of root development in *Arabidopsis*Durham University.

Smyth DR. 2023. How flower development genes were identified using forward genetic screens in *Arabidopsis thaliana*. Genetics: iyad102.

Terribilini M, Sander JD, Lee JH, Zaback P, Jernigan RL, Honavar V., Dobbs D. 2007. RNABindR: a server for analyzing and predicting RNA-binding sites in proteins. Nucleic acids research 35: W578-W584.

Thompson HL, Shen W, Matus R, Kakkar M, Jones C, Dolan D, Grellscheid S, Yang X, Zhang N., Mozaffari-Jovin S. 2023. Meristem-defective regulates the balance between stemness and differentiation in the root meristem through RNA splicing control. Development 150: dev201476.

Wang ET, Sandberg R, Luo S, Khrebtukova I, Zhang L, Mayr C, Kingsmore SF, Schroth GP., Burge CB. 2008. Alternative isoform regulation in human tissue transcriptomes. Nature 456: 470-476.

Waterhouse A, Bertoni M, Bienert S, Studer G, Tauriello G, Gumienny R, Heer FT, de Beer TAP, Rempfer C., Bordoli L. 2018. SWISS-MODEL: homology modelling of protein structures and complexes. Nucleic acids research 46: W296-W303.

Wisniewska J, Xu J, Seifertová D, Brewer PB, Ruzicka K, Blilou I, Rouquié D, Benková E, Scheres B., Friml J. 2006. Polar PIN localization directs auxin flow in plants. Science 312: 883-883.

Xiong E, Zheng C, Wu X., Wang W. 2016. Protein subcellular location: the gap between prediction and experimentation. Plant molecular biology reporter 34: 52-61.

Yan Y, Tao H, He J., Huang SY. 2020. The HDOCK server for integrated protein–protein docking. Nature protocols 15: 1829-1852.

Zeng J, Yang L, Tian M, Xie X, Liu C., Ruan Y. 2023. SDG26 is involved in trichome control in *Arabidopsis thaliana*: Affecting phytohormones and adjusting accumulation of H3K27me3 on genes related to trichome growth and development. Plants 12: 1651.