

Halogens engineering-based design of agonists for boosting expression of frataxin protein in Friedreich's ataxia

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Abstract. – OBJECTIVE: Decreased expression of the mitochondrial protein frataxin is the cause of the neurodegenerative disorder Friedreich's ataxia. In patients with cardiac disorders, the death rate of this disease is very high, up to 66%. In order to combat Friedreich ataxia, which is a potentially toxic disorder, de novo drug discovery and design have been created utilizing the approach of compound engineering with halogens. This study aimed to investigate the potential for effective treatment of Friedreich ataxia.

MATERIALS AND METHODS: The screening of twenty different agonist compounds was carried out in order to find the most promising agonist compound that may be used for molecular docking prediction against the Frataxin Protein. The compound with the lowest binding energies is then optimized by halogens. The final candidate's drug-like properties are identified through Absorption, Distribution, Metabolism, Excretion, and Toxicity (ADMET) profiling. Lipinski's rule of five was checked. Molecular dynamic stimulations were evaluated.

RESULTS: The most potent agonist compound was identified out of twenty different compounds utilizing a docking approach against the Frataxin Protein. The compound with the lowest binding energies was next subjected to optimization by halogens. The optimized agonist 9-[1-[(1S, 5R)-8, 8-dimethyl-8-azoniabicyclo[3.2.1]octan-3-yl]triazol-4-yl]fluoren-9-ol has higher binding energy of -10.4 Kcal/mol with molecular weight of 705.63 g/mol. Drug-like properties are identified through ADMET profiling, having water solubility of about -7.59, skin permeation -7.08 cm/s, bioavailability score 0.17, and high GI absorption. The candidate fulfills the Lipinski rule of five and portrays efficient molecular dynamic stimulations.

CONCLUSIONS: The selected agonist is one of the most potent compounds in increasing Frataxin

in protein expression. Furthermore, optimization with halogens can be a productive approach to improve the candidate's drug efficacy. The development of effective medications for the treatment of Friedreich ataxia would be aided by the results of these computational investigations.

Key Words:

Halogens engineering, Friedreich ataxia, Agonists, Frataxin protein.

Introduction

The most prevalent form of inherited ataxia is called Friedreich ataxia (FRDA), and it is a neurological disorder that is passed down through generations in an autosomal recessive manner¹. The study of the etiology of Friedreich ataxia has advanced quickly as a result of the latest finding of the gene, *FXN*, which is mutated in this disorder. A highly conserved protein called frataxin has been linked to numerous roles in cellular iron regulation. In intron 1 of the gene, approximately 98% of all mutant variants have an expanded GAA trinucleotide repeat. As a result, the protein frataxin is present in lower amounts. This disease is caused by insufficient amounts of frataxin protein in the mitochondria². There is mounting evidence that an excess of iron in the mitochondria is responsible for the oxidative stress and cell death seen in Friedreich ataxia. There is currently no recognized medication that can change the disease's natural course. The identification and potential role of the *FXN* gene

have sparked optimism regarding the possibility of devising rational treatment strategies. Cardiac failure is widely recognized as the leading cause of death in patients with Friedreich ataxia³. Aspiration pneumonia, heart problems (60%), diabetic coma, stroke, and trauma sequelae are typical causes of death, while heart malfunction (60%), suspected cardiac dysfunction (3.3%), and non-cardiac (27.9%) are causes of death⁴.

Multiple drugs, including Idebenone, coenzyme Q10, vitamin E, Vincerinone, and others, have been tested on FRDA patients or are currently being researched for this condition. Additionally, as FRDA is characterized by oxidative stress and mitochondrial respiratory chain malfunction, patients may gain from treatments that prevent the production of free radicals⁵. But all these mentioned therapies may reduce the prevalence of this disease but do not cure it properly. Despite such therapies, the mortality rate of Friedreich ataxia is still high. Even though finding new drugs has been difficult, there are currently some innovative and interesting potential treatments for FRDA, such as pharmaceuticals and gene therapy⁶. Novel tactics for frataxin augmentation and genetic manipulation, together with compounds that boost mitochondrial activity (such as Nrf2 activators, dPUFAs, catalytic antioxidants, and agonist compounds), are being explored to treat this devastating condition. An agonist is a substance that triggers a biological reaction by binding to and activating a receptor. Depending on the agonist and the receptor, binding can occur in a variety of sites and ways. Binding is specific to the receptor-agonist interaction, although it does activate the receptor by inducing a conformational change⁷.

In this case, *in silico* methods have shown promise and have grown into powerful tools for the search for medical solutions. Computer-aided drug design (CADD) approaches offer a solution to boost drug development efficiency while decreasing both time and expense because traditional drug discovery is costly and time-consuming⁸. In this study, we suggested a drug design employing chemicals that are antagonistic to Friedreich ataxia that can boost the expression of the frataxin protein. The optimal agonist compound for Friedreich ataxia has been determined using homology modeling, screening of compounds, interaction analysis, Absorption, Distribution, Metabolism, Excretion and Toxicity (ADMET) profiling, and application of Lipinski's rule for drug safety. This study suggests the potential for effective treatment of Friedreich ataxia, which

may lead to reduced mortality rates in the future particularly among people with heart issues^{9,10}.

Materials and Methods

Prediction of Primary Structure of Frataxin Protein

Frataxin protein FASTA alignment Data retrieved and forecasted by the National Center for Biotechnology Information (NCBI) available at (<https://www.ncbi.nlm.nih.gov/>) with Accession ID: NP_000135.2. A complete record of reference sequences, maps, genes, nucleotide sequences, nomenclature, pathways, phenotypes, variants, and links to the genome phenotype can be accessed from the most reliable database, NCBI¹¹.

Prediction of Secondary Structure of Protein

PSIPRED tool, which is regarded as the most cited and reliable tool used for protein secondary structure prediction, predicted the secondary structure of the Frataxin protein (available at: <http://bioinf.cs.ucl.ac.uk/psipred/>)¹². This tool describes the strands, helices, and coils that are found in protein structure in detail. In order to forecast results, the protein sequence was entered after the job name and email ID were entered.

Prediction of Tertiary Structure by TrRosetta

TrRosetta (available at: <https://yanglab.nankai.edu.cn/trRosetta/>), which is regarded as the best tool for predicting the tertiary structure of proteins, identified the 3D structure of Frataxin protein. The trRosetta algorithm predicts protein structures quickly and precisely^{13,14}. It constructs the protein structure using constrained Rosetta and direct energy minimizations. The complex network that is employed in the restrained Rosetta, together with the restrained Rosetta, which is based on the idea of energy minimization, is used to analyze the protein Frataxin's three-dimensional structure.

Refinement of 3D Structure of Protein

The three-dimensional structure of the Frataxin protein was improved using the Galaxy Refine web server, which is accessible at (<https://galaxy.seoklab.org/cgi-bin/submit.cgi?type=REFINE>). By simulating molecular dynamics, it rebuilds and repacks the side chains, which ultimately relaxes the entire system¹⁵. It makes use of a framework that improves when adjustments are made.

Validation of 3D Structure

By using the PROCHECK (available at: <https://saves.mbi.ucla.edu/>) to generate Ramachandran plots for the Frataxin protein, the tertiary structure was validated¹⁶. PROCHECK examines the protein's stereochemical characteristics and the geometry of the residues that make up the protein structure. Based on the locations of Rama's favorite places in the plots, the Ramachandran plot was analyzed.

Active Site Prediction

The active sites of the Frataxin protein were predicted by the online server CAST p, which is available at (<http://sts.bioe.uic.edu/castp/index.html?3trg>). It is the most cited and authentic tool used for the prediction of active sites of protein¹⁷.

Protein-Protein Interaction

The interaction of the Frataxin protein with other proteins was done by STRING (search tool for the retrieval of interacting genes/proteins) which is the most authentic site for protein interaction (<https://string-db.org>)¹⁸. It is openly available and updated frequently. The information in the STRING database comes from various places, such as open text collections, computer prediction methods, and experimental data.

Selection of Compounds

Agonists compounds were used because of their ability to promote regeneration in motor neurons and have the capability to bind to synaptic receptors and enhance the effect of the neurotransmitters; furthermore, they are a novel and innovative class of therapeutic agents for the treatment of a wide range of disorders in living organisms to increase the expression of proteins and provide a practical strategy for the creation of highly selective drugs. 20 agonist compounds were retrieved from PubChem, available at (<https://pubchem.ncbi.nlm.nih.gov>). It serves as a public database for knowledge about chemical compounds and their biological effects. PubChem assists in drug development in a variety of ways, including lead identification and optimization, compound-target profiling, polypharmacology research, and explanation of unidentified chemical identities¹⁹. Later on, BIOVIA Discovery studio (<https://discover.3ds.com/discovery-studio-visualizer-download>, Dassault systems, Velizy-Villacoublay, France) was used to convert 3D structures into Protein Data Bank (PDB) from Structural Data File (SDF) format.

Screening of Compounds

The PyRx docking method was used to screen 20 different compounds. This virtual screening software explores libraries of antagonists against pharmacological targets in the context of computational drug development and discovery^{20,21}. The substance with the highest energy was chosen for additional optimization to boost its effectiveness²².

Engineering of Halogens

The selected agonist was engineered or optimized on the level of halogens as literature supports the addition of halogens to increase the efficiency of a drug by using Swiss Bioisostere²³, available at (<http://www.swissbioisostere.ch>). The Molecular Modelling group of the SIB Swiss Institute of Bioinformatics worked on its development²⁴. The ability to access information on specific molecular substitutions and create drugs for bioisosteric alterations is particularly useful.

Interaction Analysis

Interaction analysis presented the docking of frataxin protein with the halogen-engineered agonist. Molecular docking was possible using the free tool Autodock Vina because it is a docking engine²⁵. A grid box containing x, y, and z values was used to modify the binding site of the protein. The active sites were identified, and the grid was established after the protein and ligand were prepared. Following that, docking was done between the ligand and the targeted protein.

Validation of Interaction Analysis

The interaction between the Frataxin protein and halogen-engineered agonist was validated by the Protein-ligand interaction profiler (PLIP) available at (<https://plip-tool.biotec.tu-dresden.de/plip-web/plip/index>). This is an online tool that provides the type of bonds such as hydrogen bonding, Vander Waals forces, hydrophobic interactions, and bond lengths²⁶.

Pre-Clinical Testing

SwissADME performed ligand pre-clinical testing. It is a website where a medication candidate can endure pre-clinical testing (<http://www.swissadme.ch>). It predicted key characteristics like toxicity, dispersion, metabolism, and absorption²⁷. The optimized PDB structure of the compound was included in the input, and its drug's likeness qualities were identified.

Validation of Lipinski's Rule

The drug-like qualities of the improved molecule were investigated using the Molinspiration tool.

The smiles of compound were provided as input. Among the many factors that may be calculated with molinspiration are log P, polar surface area, mass, atomic number range, O or N range, OH bond range, ion channel modulator range, enzyme range, nuclear receptor range, and Lipinski's rule violation range²⁸.

MD Simulations

Simulations of molecular dynamics are possible due to the IMods server (<http://imods.chacon-lab.org>). In order to simulate the interaction in the host's body, it forecasts how the protein will behave when interacting with the molecule and gives specifics regarding the verification of the interaction analysis^{29,30}.

Results

Prediction of Primary structure of Frataxin protein

The targeted Frataxin protein with accession ID: NP_000135.2 was retrieved from NCBI (National Center for Biotechnology Information), having amino acids 210.

Prediction of Secondary Structure of Protein

PSIPRED predicted the secondary structure of Frataxin Protein, and the results were color-coded according to the different protein secondary structures that were anticipated to be present, such as yellow for strands, pink for helices, and grey for coils. The secondary structure of Frataxin protein is predicted in given Figure 1A.

Prediction of Tertiary Structure

TrRosetta was used to predict 3D structure of Protein. Five different models were generated by this tool but only one model was selected for further analysis on the basis of a high TM score. The selected model had a TM score of about 0.740. The tertiary structure of Frataxin protein is shown in Figure 1B.

Refinement of 3D Structure of Protein

As the percentage of residues in the areas with the greatest favor was about 87.2%. For better outcomes, the Protein's 3D structure was refined using Galaxy Refine (<https://galaxy.seoklab.org/cgi-bin/submit.cgi?type=REFINE>). After one modification, which produced better results than the prior one, the structure was improved. This

tool showed 5 models, but only one model was selected on the basis of Rama's favored score and clash score. One model was selected, which had a high Ramachandra Score. The refined 3d structure of Protein is predicted in Figure 2A.

Validation of 3D Structure

The 3D structure of Frataxin protein was validated by using the online server PROCHECK (<https://saves.mbi.ucla.edu/>). The projected quality score as a whole was 96.1%. Ramachandran plot showed that 173 residues are present in most favored regions i.e., 96.1%, 5 residues are present in additional allowed regions i.e., 2.8%, and only 2 residues are present in disallowed regions i.e., 1.1%. The Ramachandran plot of the 3D structure of the protein is shown in Figure 2B.

Active Site Prediction

The active sites of Frataxin protein were predicted through the online server CASTp. The three-dimensional regions were marked, delineated, and measured. As in the protein, Richard's surface and connolly's surface were fully visible and marked red. At this point, the ligands can fully interact with the protein pockets. The cartoon representation style was selected. The list of different residues present in active sites of protein is shown in given Table I.

Protein-Protein Interaction

Frataxin protein interaction with other proteins was visualized using STRING (a search engine for the retrieval of interacting genes/proteins). The *FXN* protein having 11 nodes had a direct link with other proteins like *FDX1*, *FDX1L*, *ISCU*, *HSCB*, *PMPCB*, *GLRX5*, *LYRM4*, *NFS1*, *FECH*, and *HSPA9*. The *FXN* had higher interaction with *ISCU*, *NFS1*, *FECH*, *LYRM4*, having a score of about 0.999. Each of these related proteins played a role in the development of Friedreich ataxia. The pathway of *FXN* protein is shown in Figure 3.

Selection of Compounds

20 Agonists compounds were selected from PubChem then their PDB structures were utilized for further analysis. The provided Supplementary Table I illustrates the PDB format of 20 selected ligands.

Screening of Compounds

Twenty agonists were chosen and docked against the Frataxin protein using Pyrx in order to identify the best compound. Based on their bin-

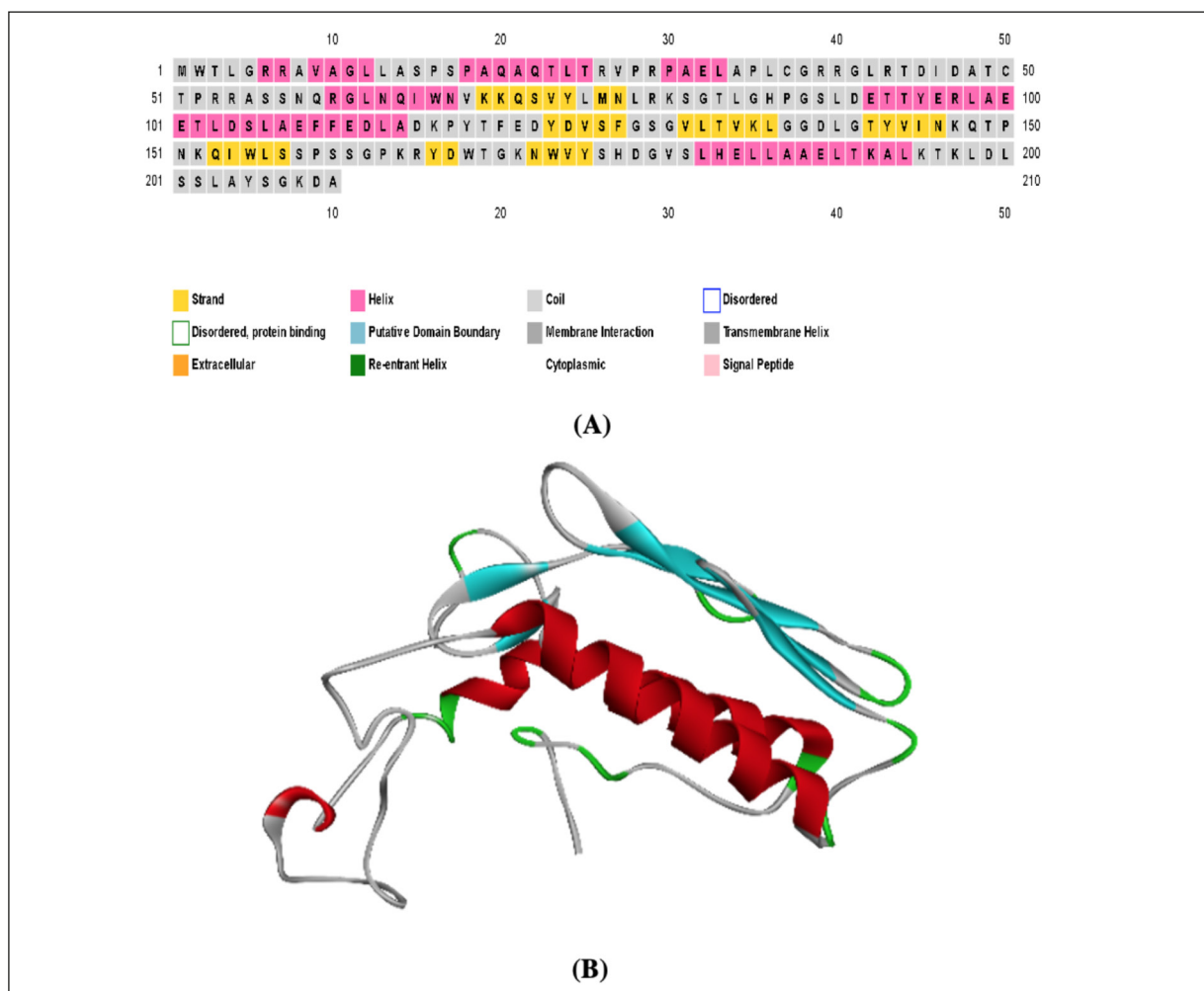


Figure 1. A, Secondary structure of Frataxin protein, yellow color (strands), pink color (Helix), grey color (coils). B, Three-dimensional (3D) structure of Frataxin Protein.

ding affinities, the 20 compounds were evaluated. Energy levels range from -7.8 to -3.9 Kcal/mol. It was determined to use the compound 9-[1-[(1S, 5R)-8, 8-dimethyl-8-azoniabicyclo [3.2.1] octan-3-yl] triazol-4-yl] fluoren-9-ol because it had the lowest binding affinity with the protein Frataxin, which was roughly -7.8. The given Supplementary Table II displays the list of binding affinities for selected compounds.

Engineering of Halogens

The selected agonist compound was functionally optimized through SwissBioisostere. As the given ligand contains two Nitrogen which are attached through a double bond, and both of them have hydrogens. During optimization, 2 bromides, 1 chlorine, and 1 Iodine was added because halogens can increase the overall efficiency of the compounds. The optimized structure of 9-[1-[(1S, 5R)-8, 8-di-

methyl-8-azoniabicyclo [3.2.1] octan-3-yl] triazol-4-yl] fluoren-9-ol is depicted in given Figure 4A.

Interaction Analysis

Using Autodock Vina, the docking was done between the Frataxin protein and the halogen-engineered agonist. The docked complex had binding energy of about -10.4 Kcal/mol. The docked complex is shown in given Figure 4B.

Validation of Interaction Analysis

The validation of interaction analysis was accessed by using the protein-ligand interaction profiler (PLIP), which allowed for further interpretation of the interaction data. As the typical length of a hydrogen bond is between 2.7-3.3 angstroms, the one hydrogen bond was detected as 2.60 angstroms. Due to the slightly greater distance of the bond length of van der Waals forces (3.3-4.0 angstroms), one hydrophobic

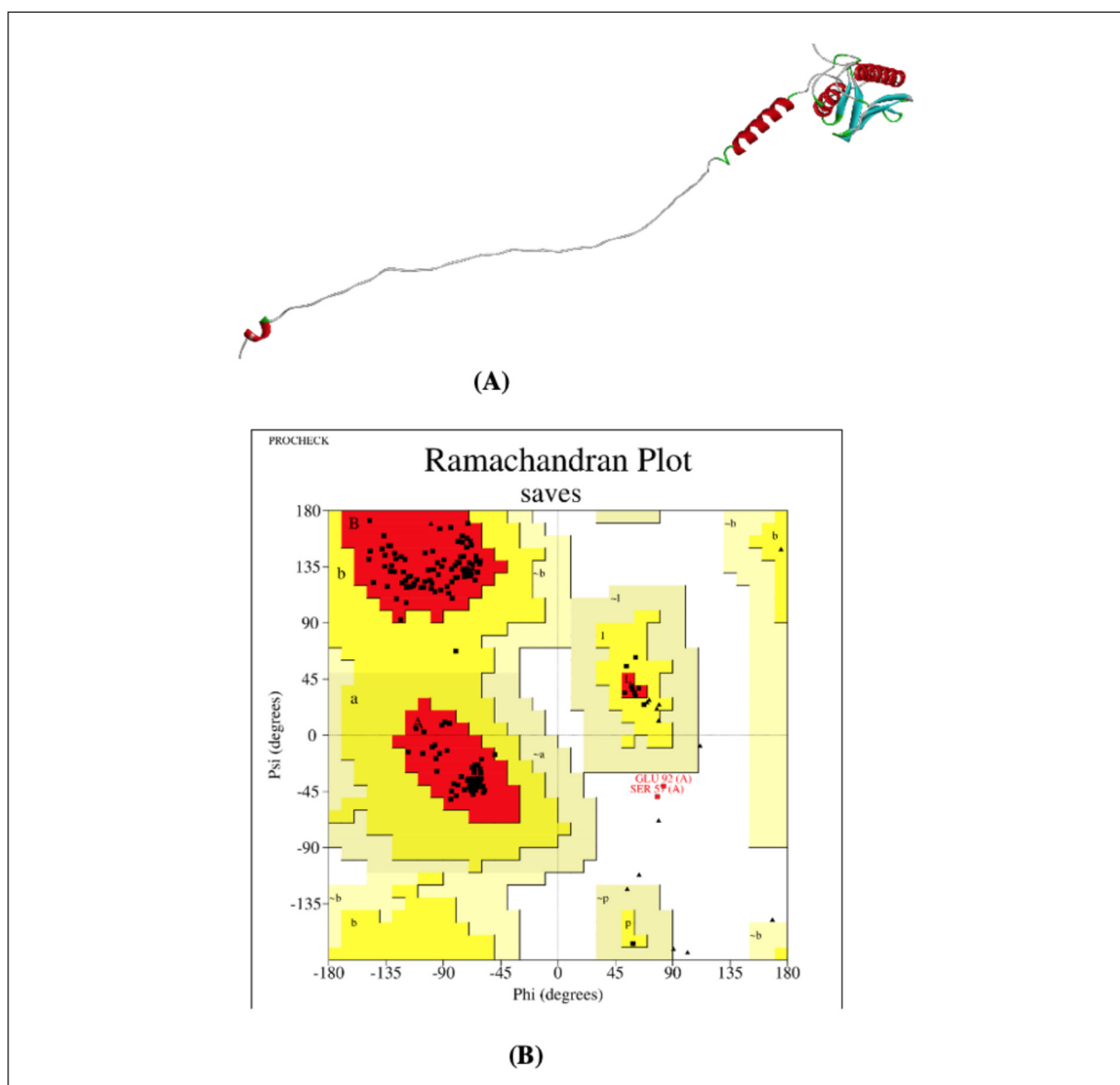


Figure 2. A, Refined 3D structure of the protein. B, Ramachandran plot of Frataxin protein.

interaction was projected as 3.47 angstroms. The interactions predicted by Protein-ligand interaction profiler (PLIP) are shown in given Figure 5A and Table II.

Pre-Clinical Testing

Swiss ADMET represented the drug's physiochemical characteristics, water solubility, gastrointestinal (GI) absorption, topological polar surface area, skin permeability, bioavailability score, and synthetic accessibility. There are only two violations indicated in accordance with the International Standard Drug Likeness Rules, i.e., the number of atoms is larger than 70. The ADMET parameters of the selected ligand are shown in Table III.

Boiled Egg Model

The SwissADME web server predicted the boiled egg model. The boiled egg model foretells a quick, intuitive, easy-to-repeat, remarkable robust technique to research the absorption of small compounds through the gastrointestinal tract and the entry of those compounds into the brain, which is very beneficial for the development and administration of medications. It is hypothesized that a molecule can penetrate the blood-brain barrier when it is present in the yellow area of the boiled egg model, however, it is anticipated that the molecule will be absorbed through the digestive system when it is present in the white area.

Table I. Active site residues of Frataxin protein.

Chain	SeqID	Residue	Atoms
A	4	LYS	C
A	5	GLU	N
A	8	GLU	C
A	9	THR	CA
A	10	CYS	SG
A	35	LEU	O
A	36	GLN	O
A	37	LEU	CA
A	38	THR	OG1
A	54	CYS	C
A	55	GLY	N
A	56	GLU	N
A	59	SER	CB
A	86	VAL	CG1
A	87	GLU	O
A	88	ASP	CA

According to the depicted Figure 5B, the molecule is present within the yellow region, indicating its ability to permeate the blood-brain barrier.

Validation of Lipinski's Rule

Table IV displays the results of Molinspiration's estimations of the parameters of Lipinski's rule, including log P, polar surface area and mass, atomic number range, O or N atomic number range, OH atomic number range, and rotatable bond atomic number range.

MD Simulations

IMods is a molecular simulation tool that adjusts the force field of the structure in connection to different time intervals to be used for structural research. The complex structure's anticipated Eigen value was $1.940727e-07$. Better interactions between the various residues are indicated by high co-related regions on the heat map and a low RMSD value. Figure 6A shows how protein structure can be used to predict MNA mobility; Figure 6B illustrates deformability, showing that minor changes occur at the

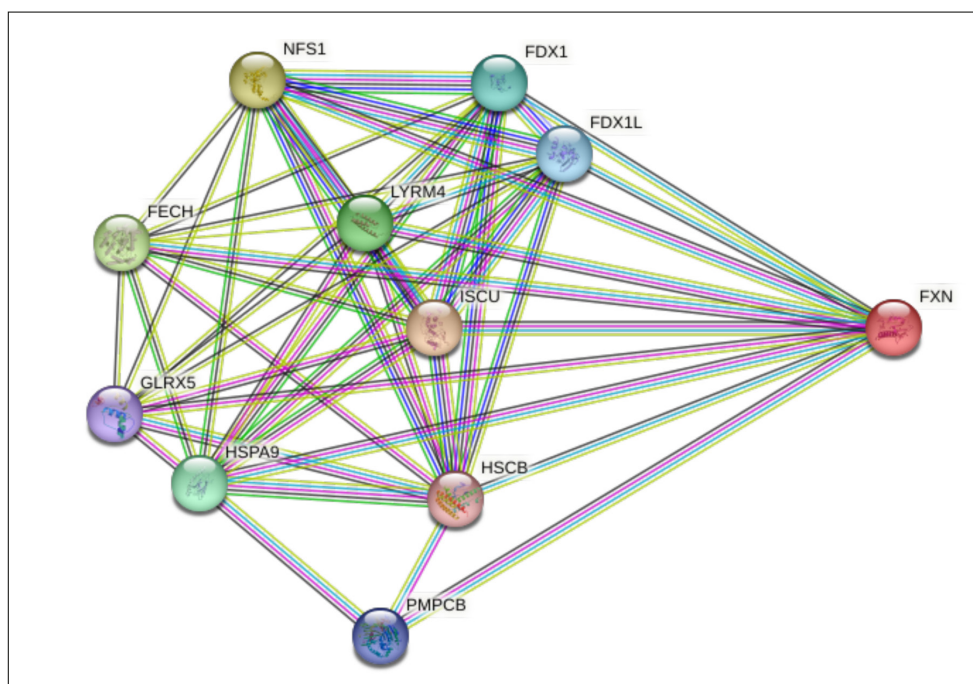
**Figure 3.** Pathway of FXN protein in Homo sapiens with other proteins.

Table II. Residues and their interacting bonds predicted by PLIP.

Residues	Amino Acids	Distance	Interaction force
71A	GLN	2.60	Hydrogen bond
74A	TYR	3.47	Hydrophobic interaction

residue level; Figure 6C indicates B-factor; Figure 6D Eigen value's prediction as $1.305321\text{e-}04$; Figure 6E exhibits variation, which is expanded upon in both green and red; Figure 6F displays covariance; Figure 6G represents the elastic network, with the stiffer regions shown by darker grey patches.

Discussion

Autosomal recessive Friedreich ataxia is the most common form of congenital ataxia, affecting the nervous system³¹. Recent discoveries of the

mutant FRDA gene have accelerated research into the disease's origin in Friedreich ataxia. FRDA is a severe multisystem disease caused by a GAA repeat expansion in the first intron of the *FXN* gene. ISC biogenesis is hindered, iron homeostasis in the mitochondria is upset, and the body's response to oxidative stress is drastically altered as a result of this frataxin deficiency. These activities give a comprehensive picture of cell toxicity and mortality across the cardiovascular, nervous, and endocrine systems. The resulting phenotype varies widely but is always characterized by a loss of mobility, heart dysfunction,

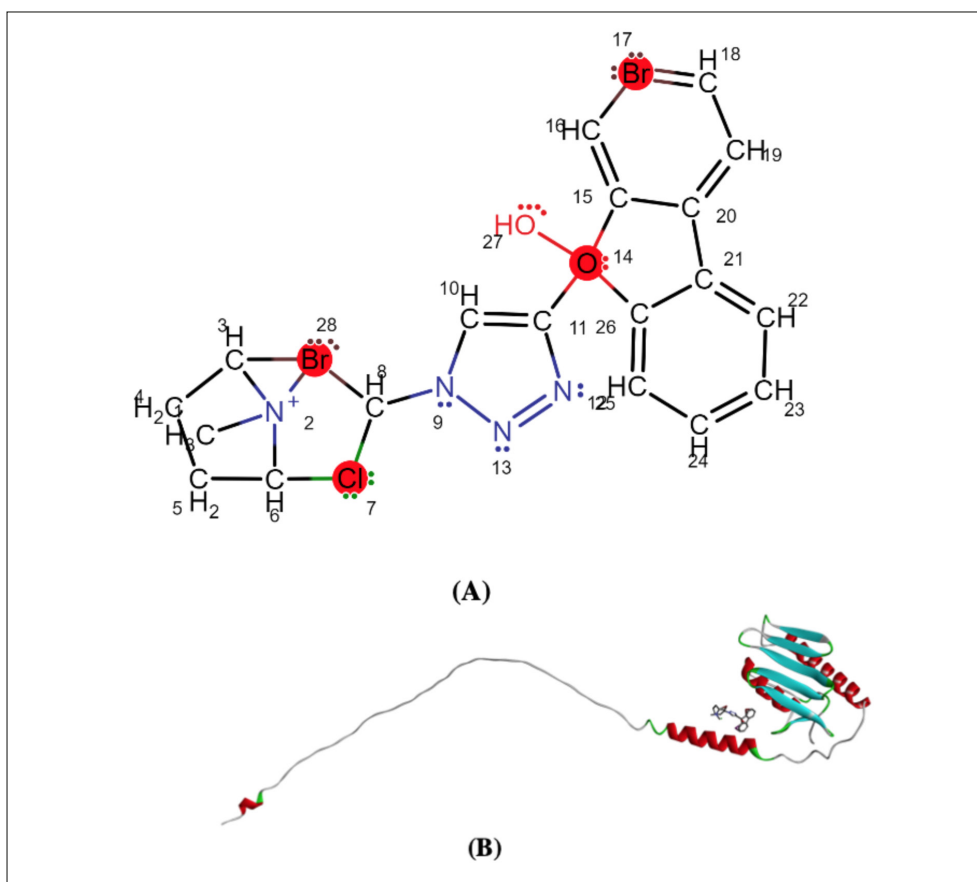


Figure 4. A, Optimized Structure of 9-[1-[(1S,5R)-8,8-dimethyl-8-azoniabicyclo[3.2.1]octan-3-yl]triazol-4-yl]fluoren-9-ol. B, Docked Complex of frataxin protein with 9-[1-[(1S,5 R)-8,8-dimethyl-8-azoniabicyclo[3.2.1]octan-3-yl]triazol-4-yl]fluoren-9-ol.

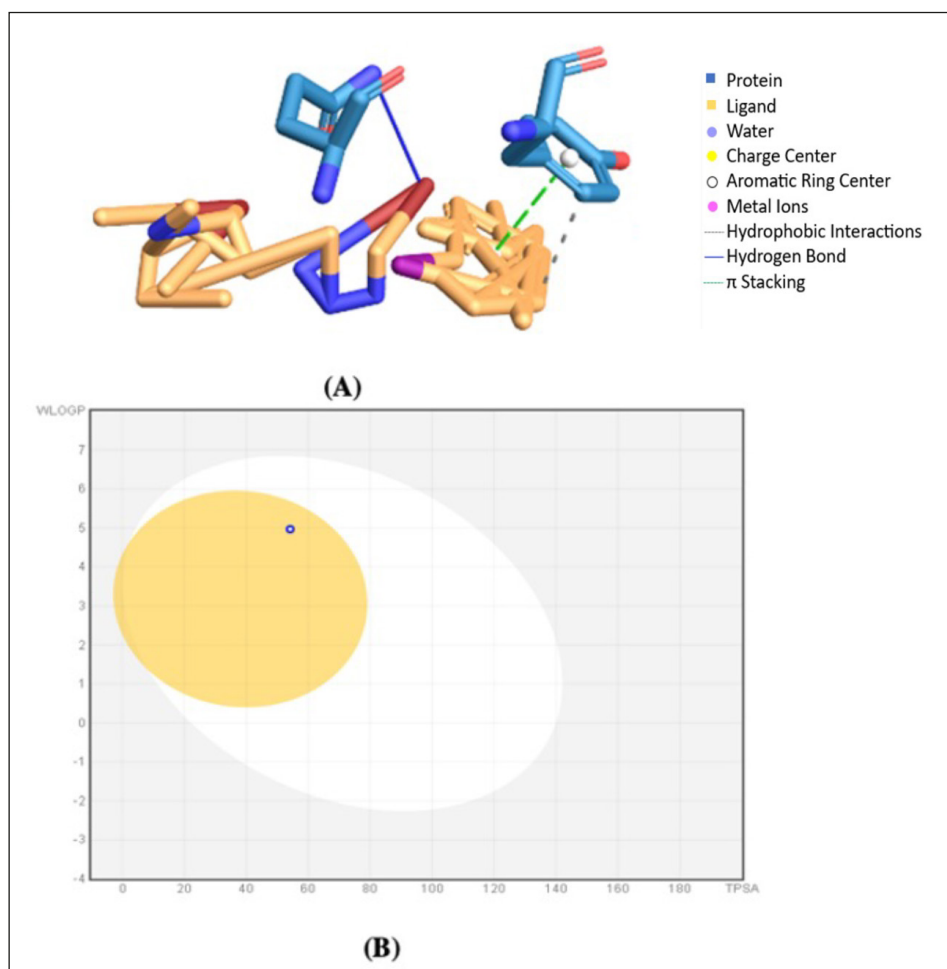


Figure 5. A, Interaction forces prediction by protein-Ligand interacting profiler (PLIP). B, Boiled egg of 9-[1-[(1S, 5R)-8, 8-dimethyl-8-azoniabicyclo [3.2.1] octan-3-yl] triazol-4-yl] fluoren-9-ol, drug (blue dot), blood brain barrier (yellow), gastrointestinal area (white).

and premature mortality³². The management of FRDA must be approached from a broad interdisciplinary perspective due to the disease's complicated and changeable clinical presentation. Muscle weakness, joint pain, difficulty walking, weariness, altered sensation, and slurred speech are all symptoms of Friedreich ataxia. Without early treatment, these symptoms are likely to worsen. Multiple methods for curing Friedreich ataxia have been recently investigated³³, however, studies^{34,35} suggest that this has not significantly reduced the disease's mortality rate.

Earlier clinical trials³⁶ explored antioxidant and iron chelation medications as monotherapy and in combination, with mixed results, focusing on the downstream effects of frataxin deficiency, particularly the roles of oxidative stress and

iron buildup in FRDA. Alternative methods³⁷ have explored the possibility of treating FRDA by increasing frataxin production. It has been demonstrated, for instance, that the glycoprotein erythropoietin raises frataxin levels in FRDA models, although clinical therapeutic advantage has not been consistently demonstrated in trials. Despite attempts at alternative approaches, they were either too costly or produced no significant outcomes. For this research, we have utilized Agonists to boost Frataxin Protein Expression. An agonist is a substance that triggers a biological response by binding to and activating a receptor. In this particular instance, we have utilized agonists employing *in silico* approaches to build a medication that increases the expression of the frataxin protein, which is associated with Friedreich ata-

Table III. ADME Parameters of pre-clinical testing of optimized agonist.

ADMET parameters	Parametric values
Formula	C ₂₄ H ₂₃ Br ₂ ClIN ₄ O
Molecular weight	705.63 g/mol
Num. heavy atoms	33
Num. arom. heavy atoms	17
Fraction Csp ³	0.42
Num. rotatable bonds	2
Num. H-bond acceptors	3
Num. H-bond donors	1
Molar Refractivity	150.44
TPSA (Topological Polar Surface Area)	54.18 Å ²
Water Solubility Log S (ESOL)	-7.59
GI absorption	High
Skin Permeation (Log K _p)	-7.08 cm/s
Bioavailability Score	0.17
Synthetic Accessibility	6.09
Drug likeness (Ghose)	2 violation i.e. #atom>70

xia. This process of treatment is less expensive as compared to conventional therapeutic solutions. CADD strategies provide a means to improve efficiency without increasing either cost or time.

Another study³⁸ suggested that the deadly disorder of Friedreich ataxia may be mitigated by the use of several medicines, such as mitochondrial cofactor, antioxidants, or iron chelating. However, these drugs were prohibitively expensive and came with undesirable side effects like itching, vomiting, a racing heart, hypotension, and cramping in the stomach or legs. In contrast, agonist compounds were utilized in this research because of their quick responsiveness toward the receptor. Agonists have a significant capacity to stimulate the expression of proteins. Docking analysis, along with computational analysis, was utilized to investigate the screening of agonists that result in potential attachments and interactions with the Frataxin protein. Because of its greater effectiveness with Frataxin protein, the 9-[1-[(1S, 5R)-8, 8-dimethyl-8-azoniabicyclo[3.2.1] octan-3-yl] triazol-4-yl] fluoren-9-ol agonist was chosen for this study. This research recommends a stronger, more focused treatment

to reverse organ damage and mortality. Gaining a thorough understanding of the molecular genetics, bioenergetics, and cellular iron dynamics involved in this treatment holds out hope for a disease-free future.

Conclusions

Friedreich ataxia is an autosomal recessive neurological disorder characterized by a variety of symptoms, including soreness in the joints, difficulty walking, exhaustion, impaired sensation, and slurred speech. Friedreich ataxia cells, according to an increasing body of data, die from oxidative stress brought on by an accumulation of iron in their mitochondria. Collectively, these activities paint a picture of cell toxicity and death in the cardiovascular, neurological, and endocrine systems. The ensuing phenotype can seem very different from person to person, but it always includes decreased mobility, heart malfunction, and early death. The previous method of treating this disease was not only costly, but it also failed to significantly reduce mortality rates. Through the use of an agonist, which promotes

Table IV. Molinspiration's estimations of the parameters of Lipinski's rule.

Optimized compound (9-[1-[(1S,5R)-8,8-dimethyl-8-azoniabicyclo[3.2.1] octan-3-yl]triazol-4-yl]fluoren-9-ol)	logP	Molecular Weight	noN	Noh, NH	Violations
	2.0070	5.63 g/mol	5	1	1

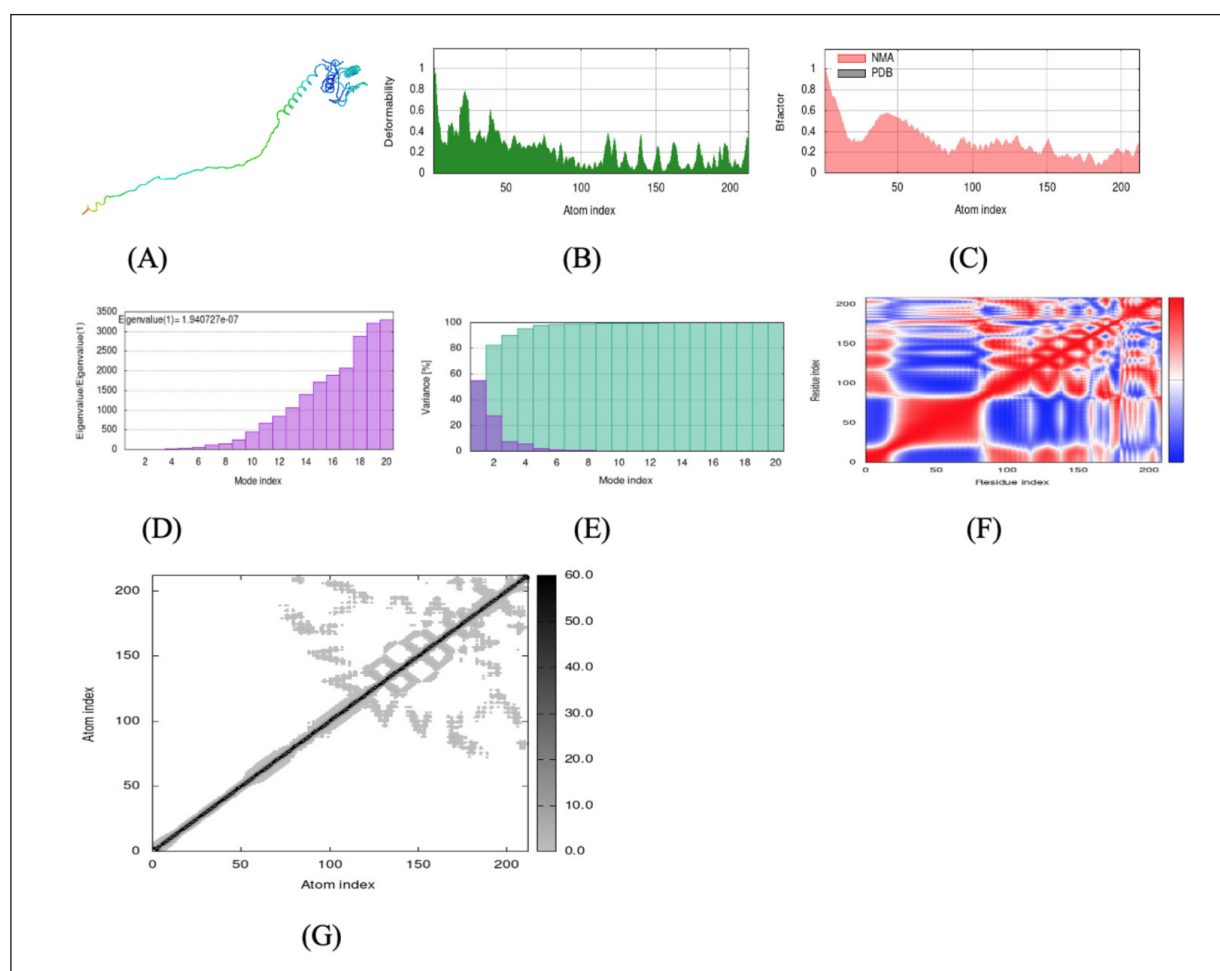


Figure 6. Results of molecular dynamic simulations through IMods, (A) Protein tertiary structure based on MNA mobility (B) Deformability (C) B-factor (D) Eigen values (E) Variance (individual variations are purple, cumulative variances are green), correlated (red) and anti-correlated (blue) motions are shown on the covariance map in (F), and (G) elastic network (darker grey regions indicate stiffer regions).

the production of extra Frataxin protein, a successful medication was developed for the treatment of Friedreich ataxia. Individuals who have a variety of complications due to Friedreich ataxia may one day find their conditions much easier to manage and treat with the properly applied drug proposed in this study. This *in silico*-based drug design would be operative, non-toxic, safe, and patient-tailored. The findings will appear to be supportive to synthesize these halogen-engineered agonists for their *in-vitro* and *in-vivo* implementations.

Authors' Contributions

Conceptualization, M.N, I.A, N.U, M.A, and T.A; methodology, M.N, N.U, I.A, M.A and T.A; software, M.A; validation, A.A.S; formal analysis, T.A; investigation, M.N, N.U,

I.A, M.A, and T.A; resources, M.A and A.A.S.; data curation, T.A.; writing—original draft preparation, T.A, and M.N.; writing—review and editing, T.A and M.N; visualization, N.U; supervision, T.A, and M.N.; project administration, A.A.S and M.A; funding acquisition, T.A.

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Ethics Approval

Not applicable.

Conflict of Interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Informed Consent

Not applicable.

Availability of Data and Materials

All the data generated during this research study has been included in the manuscript.

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