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COMPUTATIONAL ENRICHMENT ANALYSIS OF SAP PROTEIN ON LYSOSOMAL STORAGE DISORDER – METACHROMATIC LEUKODYSTROPHY

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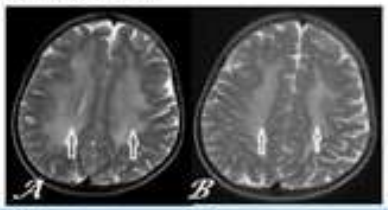
Abstract

A rare, yet fatal disease, know as Metachromatic leukodystrophy (MLD), is a neurodegenerative, hereditary malady, which falls under lysosomal storage disorder.
No other molecule can compensate for the ARSA enzyme nor a cure for MLD is available. This study reports, the molecular analysis was done, with the help of research methods, that includes quantitative and qualitative data, gained from machine learning algorithms and computational tools, to study particularly deleterious mutations that occur in the ARSA gene of MLD patients, which will foster future wet lab research and molecular diagnosis.

Introduction

MLD is an autosomal recessive, rare hereditary disease, is caused due by the mutations in the Arylsulfatase A gene (ARSA), this leads to deficiency of ARSA enzyme.
Lack of ARSA enzyme, paves way for the accumulation of 3-O-sulfogalactosylceramide, in lysosomes of various cell types including oligodendrocytes, Schwann cells, microglia, and subpopulations of neurons, thus affecting the white matter of the brain (leukodystrophy).
Symptoms such as loss of sensation, incontinence, seizures, paralysis, an inability to speak, blindness, and hearing loss. Eventually, they lose awareness of their surroundings and become unresponsive



Methodology

1. Identification of gene target and retrieval of SAP sequences from various literature and data sources.
2. The retrieved dataset is validated and screened for identifying deleterious SAP's using SNPs&GO based on Support Vector Machines (SVM).
3. The Selected Protein is Modelled and Mutated using Swiss PDB Viewer
4. Mutated compounds are validated, through an energy minimization with YASARA.
5. Comparative Analysis is performed for both Native and Mutant proteins, with the help of single model analysis.

Mutation	Prediction	Probability
T274M	Disease	0.947
R311Q	Disease	0.929
R370W	Disease	0.945
R390W	Disease	0.958
C489G	Disease	0.946

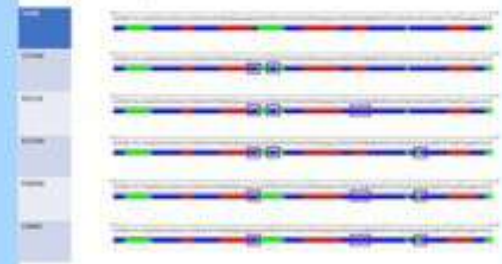
6. Single model analysis is performed through various software's and analytical tools as listed below.

- Intra Molecular Interactions
- Secondary Structure Analysis
- Contact Map Analysis
- Fluctuation Analysis
- Docking

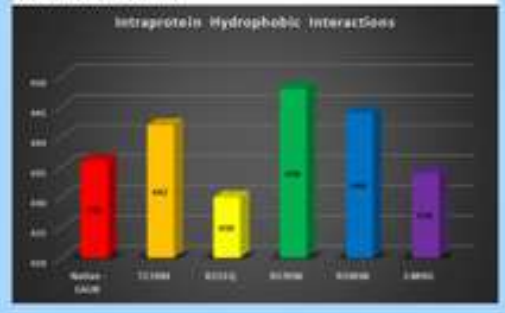
This exploration is done for both native and mutant models of the ARSA gene.

Results

Deleterious SAP's T274M, R311Q, R370W, R390W and C489G were filtered out with a SVM algorithm, called SNPs&GO, Adding on these SAPs were mutated and modelled into their respective mutants with the help of Swiss PDB view. Furthermore, energy minimization was done, and was predicted that it is energetically stable.

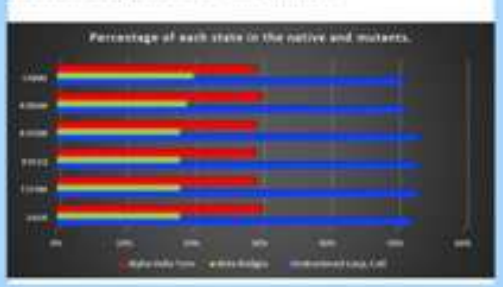


The variations produced from single model analysis, it was visible that the native was always in a different conformational analysis, when compared to the mutants. Which was further backed up by structural analysis, done in the contact map analysis and secondary structure analysis, an approx. average was calculated, to support quantitative analysis.
Intra molecular interactions, such as hydrophobic interaction, also varied drastically form each other.



Conclusion

Finally, to overcome the diagnostic challenges of MLD, integrated biochemical and genetic analyses are needed. This study helped in the molecular diagnosis, of the Arylsulfatase gene, with the help of computational tools and qualitative analysis. Docking was also done, with help of Pymol, to understand the structural changes between the native and the mutant. The variations caused, at the basic state of alpha helix, beta strands and coils were also studied and graphed for easy understanding. This research also emphasizes the importance of molecular diagnosis in all MLD patients so that MLD families can receive genetic therapy, and prenatal diagnosis for the future.



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