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Integrated Multi-Omics And Biopython Approaches For Target Identification Of A-Synuclein Polymorph In Parkinson's Disease

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Abstract:
Parkinson's disease (PD) is a progressive neurodegenerative disorder characterised by the degeneration of dopaminergic neurons and the abnormal aggregation of α-synuclein protein within neuronal cells. The misfolding of α-synuclein leads to the formation of amyloid fibrils and Lewy bodies, which are considered major pathological hallmarks of the disease. Structural polymorphs of α-synuclein fibrils have been identified, among which the cryo-EM resolved polymorph represented by a proteomic sample of Parkinson disease provides important insights into fibril architecture and aggregation behavior. In this study, an integrated multi-omics and structural bioinformatics approach was employed to investigate the molecular features of the α-synuclein polymorph 8PK4 associated with Parkinson's disease. Computational analyses were carried out using Bio python-based workflows, enabling sequence retrieval, structural evaluation, residue interaction mapping, and contact map generation to identify critical structural regions involved in fibril stabilization. Structural characterisation was further performed to analyse residue interactions, secondary structure organisation, and potential functional hotspots that contribute to amyloid formation and protein aggregation. The results highlight key stabilising residues and interaction networks that maintain the integrity of the amyloid fibril structure. Identification of these structural determinants provides insights into potential therapeutic targeting sites for inhibiting α-synuclein aggregation. Overall, the integration of multi-omics information with computational structural analysis provides a systematic framework for understanding α-synuclein polymorphs and supports the development of structure-based therapeutic strategies for Parkinson's disease.

Keywords: *α-Synuclein, Parkinson's disease, Amyloid polymorph, Bio Python, Contact map, Protein aggregation, Therapeutic Targeting*

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I. Introduction:

Currently, PD affects over 10 million people worldwide. By 2025, the disease will be present in over 25 million people worldwide, due to ageing populations and environmental factors. Motor impairments associated with PD are comprised of four cardinal signs: bradykinesia, rigidity, resting tremor, and postural instability. These features are due to the degeneration of dopaminergic neurons in the substantia nigra pars compacta. Many patients will experience non-motor symptoms, including autonomic dysfunction, sleep issues, and dementia. These symptoms greatly hinder the quality of life for those living with PD.

The molecular basis of PD is alpha-synuclein (α-syn), a presynaptic protein comprised of 140 amino acids and encoded by SNCA. Misfolding of α-syn and aggregation of the protein into amyloid fibrils lead to the development of Lewy bodies within neurons, which are the neuropathological hallmark of PD. Various amyloid fibril polymorphs, including a variant known as 5A, as determined by cryo-EM structures, contain distinct arrangements of protofilament architecture. These unique arrangements result in polymorphs characterising differences in the efficiency of fibril seeding and neurotoxicity, which contribute to different presentations of PD.[1,2]

Among those described as fibrillar, the α-synuclein polymorph with PDB ID 8PK4 represents a disease-relevant amyloid fibril (with cross-β architecture) of the highest order. The structural resolution of this polymorph can provide high-quality data, but the systematic investigation of stabilising interactions, flexible regions, and the identification of druggable hotspots at the residue level has not been extensively studied; to-date, no computational models can generate mechanistically meaningful insights from static structural data into the identification of biologically relevant amyloid fibril targets for future therapies.To overcome this gap

between biochemical (such as residue organisation, interaction networks and conformational stability) and structural data (e.g. high-resolution coordinate data) that are often used to describe the properties of amyloid fibrils, computational approaches based on the structural characterisation of proteins offer a quantitative basis for deriving both types of information. In-silico drug discovery approaches have emerged as efficient strategies for identifying therapeutic candidates targeting aggregation-prone proteins involved in neurodegenerative disorders.[3] Bioinformatics frameworks such as Bio Python enable efficient processing of sequence and structural datasets derived from next-generation sequencing studies in neurodegenerative diseases.[4] Bio Python is an accessible and widely used open-access bioinformatics suite that provides an extensive range of protein structure analysis, residue-level analysis, and geometric characterisation tools available in one package for the study of proteins. Nevertheless, the use of these computational tools to comprehensively interrogate amyloid fibrils (particularly those associated with neurodegenerative diseases) has received limited attention.To complement this work, multi-omics research has begun to describe the complex regulatory processes governing α-synuclein expression, aggregation propensity and cellular toxicity. The verification of structural information (using a multi-omics approach) with systems biology context is becoming increasingly recognised as a completely essential step for identifying biologically relevant and therapeutic drug targets. Integrating multi-omics datasets with structural analysis of α-synuclein polymorphs provides a powerful framework for identifying therapeutic targets in Parkinson's disease.[5] Simultaneously, new multi-omics studies point to the complex regulation of α-synuclein expression, aggregation propensity, and cellular toxicity. It has become clear that combining structural insights with biological systems-level context will be an important step to support the identification of biologically therapeutically valid targets for drug development. Therefore, structure-based identification of regions that have a high propensity to form aggregates and have high levels of interaction with other proteins will provide complementary information for omics-based disease signatures. Recent advances in next-generation sequencing have enabled detailed analysis of protein domains associated with neurodegenerative disorders, revealing structural determinants that influence aggregation and disease progression.[6]

We report here our Bio Python-based structural analysis of α-synuclein polymorph 8PK4 at an atomic and residue level. Specifically, using chain-wise residue mapping, atomic composition profiling, B-factor-based flexibility assessment, residue mass distribution analysis, and residue-residue contact map analysis, we provide the structural features that regulate the stability and organisation of α-synuclein fibrils. Through identification of rigid core regions, flexible residues that occupy surface-exposed regions, and the mapping of contact-dense interaction hotspots, we provide structure-guided insights that will support potential drug development strategies aimed at disrupting fibril stability and propagation of α-synuclein in patients with Parkinson's disease.

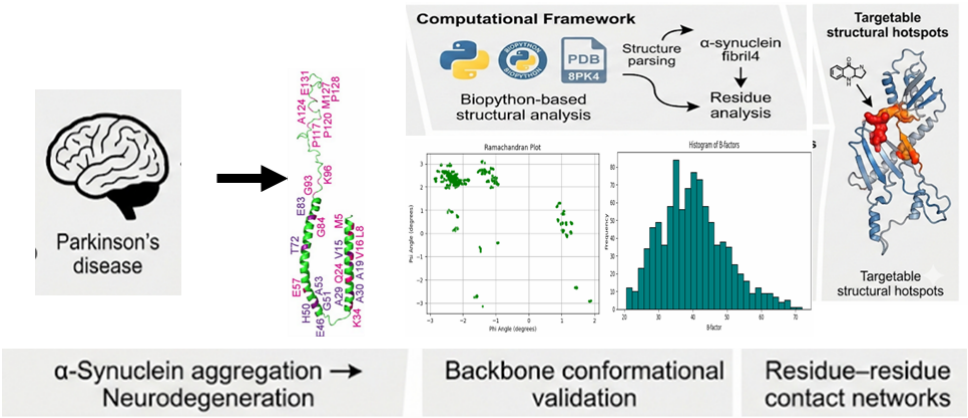

Figure1: Graphical Abstract representation

II. Materials And Methods

Structural data retrieval

The 3-dimensional atomic structure for the alpha synuclein protein 8PK4 was retrieved via the PDB database. The protein retrieved is in a Type 5A polymorph form, which plays a crucial role in the pathogenesis of Parkinson's disease.[7,8]

Protein visualisation and structural examination using RasMol

Raswin (v2.7), which is a Windows version of RasMol, was used for the visualisation of protein structure 8PK4. RasMol is a molecular visualization tools which helped visualize the structure of 8PK4 protein

in different formats, as well helped in automation of process by using the RasMol command line, so analysis of the structure becomes much more efficient and simpler.

Structural validation

To maintain structural integrity and computational efficiency, structural validation for the 8PK4 protein was carried out using SAVES V.6.1. It is a structure validation server that has tools for analysis, such as ERRAT, PROCHECK, VERIFY 3D, etc. together this helps in understanding and examining the stereochemical quality of proteins as well as evaluating geometrical parameters.[9]

Identification and Characterisation of Functional Domains and Motifs

InterProScan was used to determine if 8PK4 belongs to the synuclein family, which participates in and is a hallmark of neurodegenerative disorders like Parkinson's disease.

Interproscan is a bioinformatics tool that helps in examining protein sequences to identify domains, families and motifs within it by integrating recognition methods from databases such as Pfam, SMART and PROSITE to provide detailed functional annotations.[10]

B factor analysis

B-factor analysis was carried out for the 8PK4 protein using PyMol, which helps in the representation of atomic displacements within the protein as well as gives information of stability. PyMol is a molecular visualisation tool that helps in visualization of 3D structures of proteins, nucleic acids, and ligands using PDB files. [10]

Sequence alignment visualisation

For homology modelling, Jalview was used, which is a Java-based tool for multiple sequence alignment, which was used to compare the amino acid sequences within the same protein structure. Alignment was carried out to check conserved regions, similarity within protein as well as variations.

The atomic coordinates of the alpha-synuclein type 5A fibrillar polymorph (PDB ID: 8PK4), which is a cryo-electron microscopy-derived, Parkinson's disease-associated synuclein Pathy, were obtained from the Protein Data Bank (PDB). This fibril structure contains an amyloid structure of pH-dependent polymorphic form relevant for use during in vitro aggregation studies. The PDB file was downloaded and analysed using BioPython (version 1.81) within a Python 3.9 environment. [8,11–13]

Heteroatoms, solvent and non-protein entities were removed to allow for consistency in the analysis of residues, while retaining the protofilament chains for independent and combined evaluation. The integrity of the structural assembly, continuity of individual residues and completeness of the chains were evaluated using BioPython's PDBParser.[13]

Mapping of Residues by Chain

From the structures belonging to each protofilament, the indices of the residues were extracted via Bio.PDB and then mapped against the corresponding chain ID to assess chain length consistency, alignment and organisation of the residues. This analysis assessed the symmetry and periodicity of the fibrils.[11]

Analysis of the Composition of Atoms

The numbers of C, N, O, and S atoms were counted by iteratively traversing the Biopython atom iterators within the processed structure, thereby revealing the bonding environments and stability characteristics of the structures.[11,13]

Analysis of the Flexibility of B-Factors

B-factors were obtained using Bio python's get_b factor() on all of the atoms and the resulting frequency distributions of the calculated B-factors were plotted in histograms to represent the flexible and rigid residues.[11]

Residue Mass Distribution:

Using standard amino acid masses, the molecular weights of each of the residues were computed, and the residues were mapped into sequence to highlight the bulky/lightweight regions that impact the steric packing and stabilisation of the residues.[13]

Residue-Residue Contact Map

The pairwise Euclidean distances of the Cα atoms of the residues were calculated based on their coordinates obtained from BioPython. This produces a heat map of distance matrices that displays the cross-β interactions both within each chain (intra-chain) and across different chains (inter-chain).[11]

Backbone Dihedral Angle Analysis

The PPBuilder module of Bio Python was used to calculate the backbone dihedral angles: φ (phi) and ψ (psi). Using the α-synuclein 8PK4 structure, the peptide fragments from each residue were extracted to calculate the φ–ψ dihedral angles for only those residues with defined backbone geometry. The resulting angle distributions were represented in Ramachandran plots created with Matplotlib.

Data Visualisation and Reproducibility

All outputs have been generated using Python scripts written based on BioPython with constant parameters; scripts for B-factor analysis, contact maps, and parsing residual data are available in the Supporting Information. All analyses were run on a Linux operating system, with Python version 3.9 and BioPython version 1.81 installed, thereby ensuring reproducibility of the analyses.[11]

III. Results And Discussions:

Structural visualization and analysis

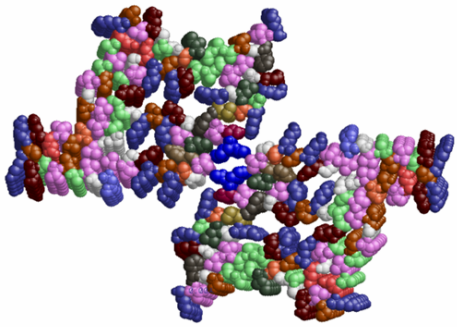

Figure 2: Three-dimensional shape representation of protein

The shapely representation is used for highlighting the amino acid composition of a protein using a predestined colour scheme. Structural observation showcases all the 140 residues of the 8PK4 protein, which has a greater number of valines, lysine, alanine and glutamate residues. These hydrophobic amino acids are generally high in alpha synuclein protein as they are responsible for misfolding and aggregation into different conformations, forming structures rich in beta sheets, which is a characteristic feature of Parkinson's disease.[7,8]

Structure validation prediction

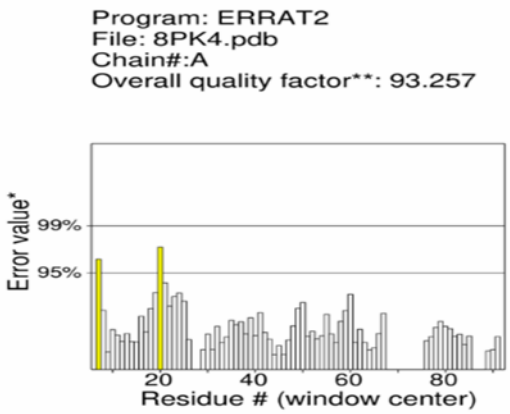

Figure 3: Quality validation and for chain A from protein 8PK4

The structure validation graph shows per-residue error value mapped along a protein sequence. ERRAT plot provides statistics of error in the three-dimensional protein structure and based on that provides quality. For chain A of the 8PK4 protein, the quality was found to be 93.257, which indicates it is an excellent quality model that can be further used for analysis. [14]

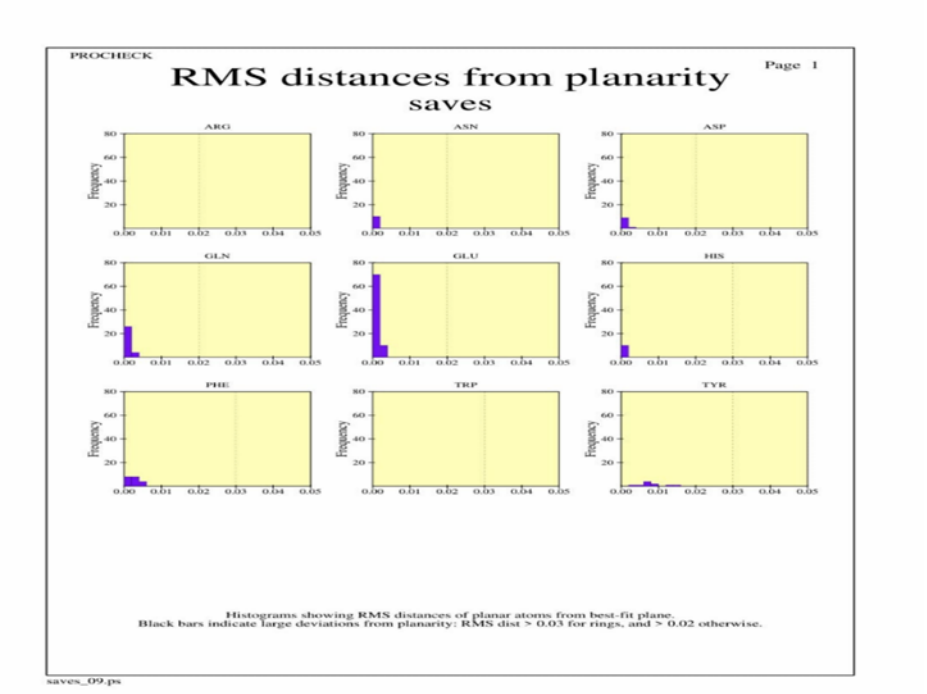

Figure 4: RMS Distance from Planarity Validation (PROCHECK Analysis)

Across all residues (Arg, Asn, Asp, Gln, Glu, His, Phe, Trp, Tyr), the histogram bars are clustered tightly near 0.00–0.01 Å. No black bars are observed in any panel, indicating no significant deviations from planarity. The frequency distributions are narrow and sharply peaked, meaning the planarity of all aromatic and conjugated systems (like benzene rings in Phe, Tyr, Trp or imidazole in His) is well preserved. Side-chain amides (Asn, Gln) and carboxylates (Asp, Glu) also maintain ideal planarity, indicating accurate modelling of double-bond character.[14]

Functional domains analysis:

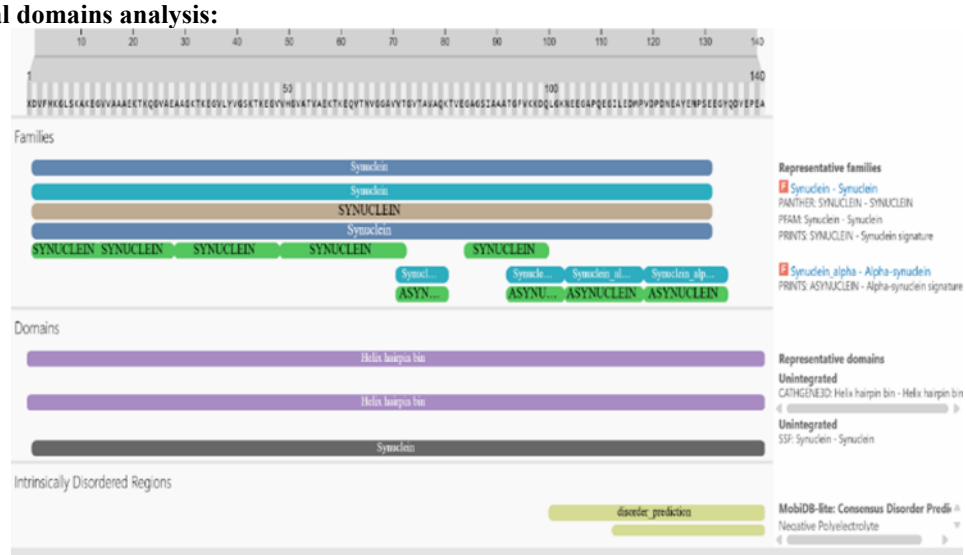

Figure 5: Protein domain organisation and family classification of the 8PK4 protein using

Inter ProScan

The grey bar represents the complete amino acid sequence below, which is represented by one-letter amino acid code. InterProScan identified 8PK4 as a synuclein protein by multiple databases. The motif

predicted is helix hairpin bin architecture with no distinct multiple catalytic domain which is consistent with the synuclein protein being a small enzymatic protein. Disorder prediction via Mobi DB-lite shows it has IDR, which are intrinsically disordered regions. The presence of IDR is a characteristic feature of synuclein as it gives the protein plasticity to interact with multiple binding proteins as well as attain different conformations within different cell environments. But the presence of IDR also contributes to misfolding and aggregation, which is the central link to Parkinson's disease, in which it promotes the protein to form beta sheet fibrils and Lewy bodies under stress such as oxidation or extreme pH change.[10]

B factor analysis

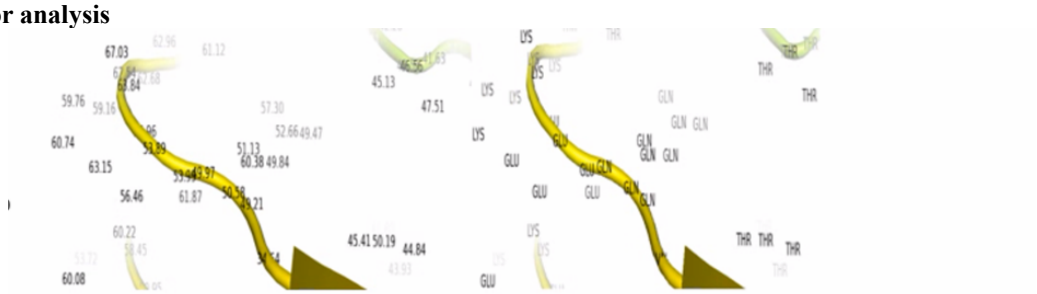

Figure 6(a): B factor representation showing score, Figure 6(b): B factor analysis showing residues

The B factor, which is based on temperature analysis, showed that a few segments have an elevated level of B factor. Usually, B factor values > than 50-60 represent a flexible region, while lower B values indicate that the structure is rigid, having secondary structure containing beta sheets and alpha helices. The PyMol visualisation of B factor is illustrated by a colour gradient in which usually colour tones like blue colour represent low B value, which is due to the stability of the structure, while warmer tones like red colour indicate flexible regions. The coloured model with yellow represents an area with flexibility with a maximum score of 67.03, which is consistent with the intrinsically disordered nature of alpha synuclein.[7,15]

Sequence alignment visualisation analysis

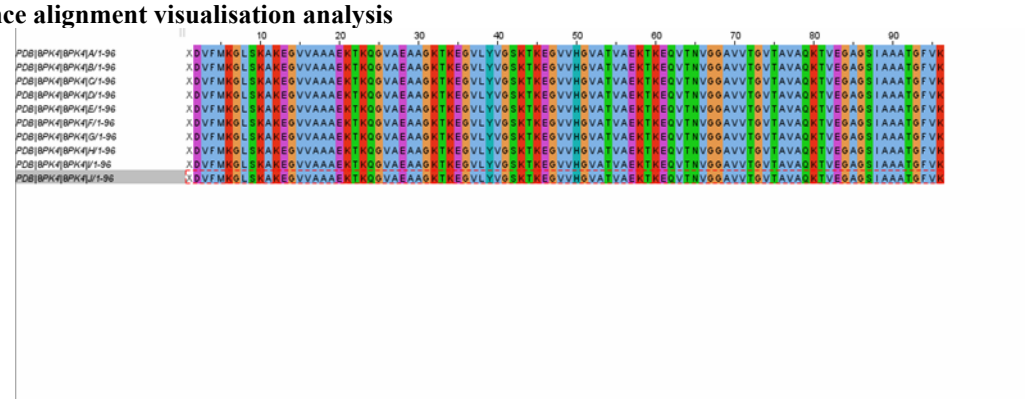

Figure 7: Jal view analysis (Multiple Sequence alignment visualisation using Jalview)

The sequence alignment using Jalview is based on the Clustal X color scheme where each colour represents different amino acid properties like red (hydrophobic residues), blue (positively charged residues), green (polar residues), Orange /yellow (small or special amino acid residues) white (less conserved residues). These Coloured pattern helps to quickly identify different regions like mutation region while similar colour represent conserved region In the sequence alignment of all the chains of 8PK4 protein each Row is representing protein sequence and each column is representing specific amino acid position in the alignment, the number 10,20,30,40 etc indicate alignment position in the protein sequence Jalview multiple sequence alignment demonstrate significant conservation across several region of the protein sequence.[16]

Structural Uniformity Among Protofilament Chains

α-Synuclein polymorph 8PK4 residue mapping by chain revealed a high degree of uniformity among protofilament chains; the lengths and positions of the residues are identical (Figure 3(h)). This high degree of uniformity reflects the repeated cross-β structure that is associated with amyloid fibrils and allows for equal contributions of each protofilament to provide overall fibril stability, which is useful for the design of targeted inhibitors.[12,17,18]

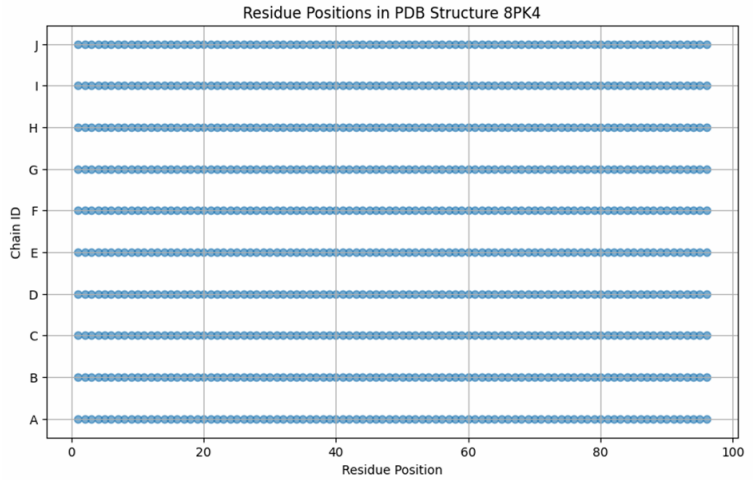

Figure 8 (h): Residue position in PDB structure 8PK4 analysis (Distribution of amino acid)

Atomic Composition Indicates Stabilisation of Hydrophobic Core

The atomic composition of the 8PK4 structure was found to be comprised of carbon atoms predominantly, then nitrogen and oxygen atoms, followed by low sulphur content (Figure 3(i)). This atomic composition illustrates that the stabilisation of the hydrophobic core is due to non-covalent interactions and extensive hydrogen bonding within the β-sheet structure, which is consistent with the mechanisms by which α-synuclein aggregates through hydrophobic collapse.[17,18]

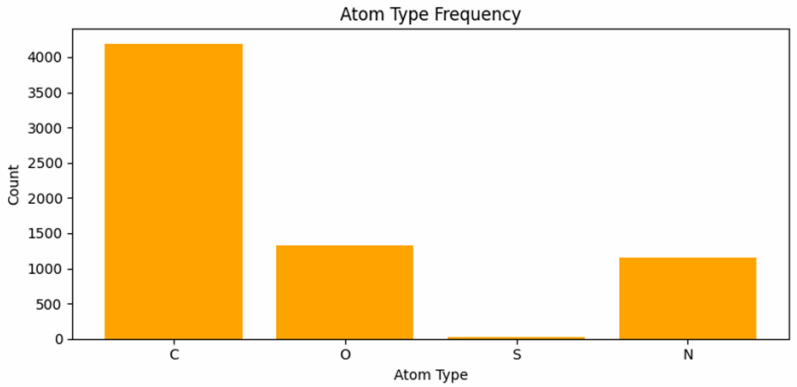

Figure 9: Bar chart shows the frequency distribution of atom types in protein structure.

B-Factor Analysis Indicates Rigid Core with Flexibility

The distribution of the b-factors revealed that the core of the fibril was rigid due to tightly packed residues (Figure 3(j)). However, some atoms had elevated b-factors and represent flexible regions at the fibril surface that can interact with other molecules in an intermolecular manner. These dynamic regions provide additional opportunities for therapy within the fibril beyond the densely packed core region.[18]

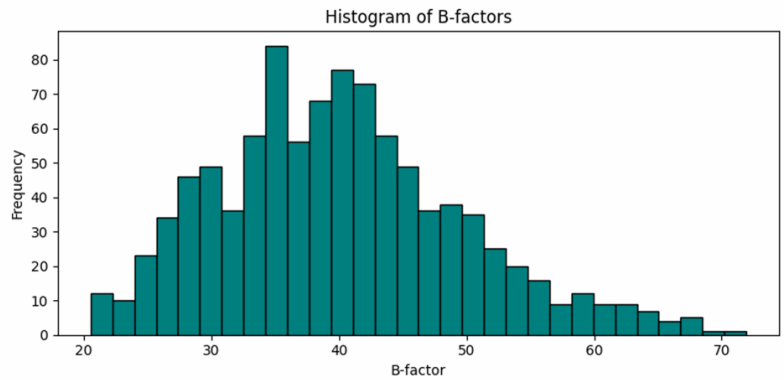

Figure 10: This histogram illustrates the distribution of the B-factor value across all the atoms

Residue Mass Distribution Indicates Heterogeneous Packing

Mapping the residues showed an alternating pattern of light and bulky residues along the polypeptide chain, indicating that there are steric "hotspots" along the polymer for stabilising inter-sheet interactions (Figure 3(k)), where the bulky side chains form a "steric zipper", and the lighter side chains allow for packing of the backbone, providing further stability of the polymorph. The residue mass distribution demonstrates a balanced representation of amino acids across the sequence, indicating a structurally consistent protein architecture. These observations collectively suggest that the analysed protein structure maintains overall stability while retaining flexible regions that may be important for biological function or ligand interactions.[12,18]

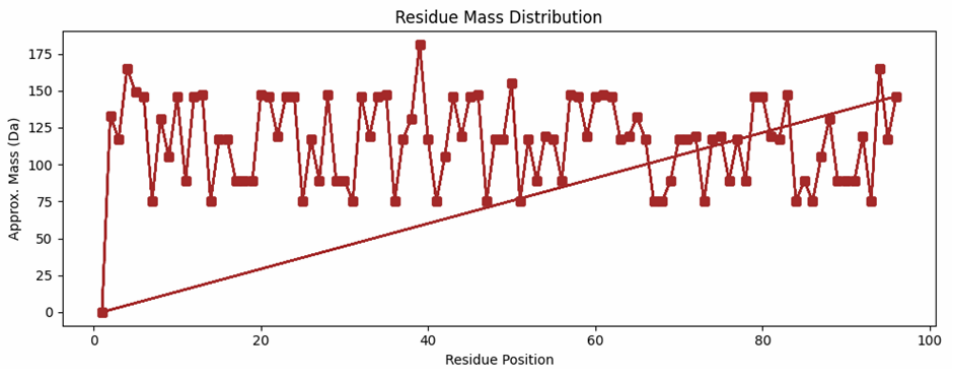

Figure 11: The graph shows the approximate molecular mass of residues along the protein sequence.

IV. Contact Map Analysis Identifies Interaction-Dense Hotspots

Contact maps have been utilised to identify contact-dense hot spots present in the structures of α-synuclein fibril assemblies. An extensive contact map for the Cα-Cα residues for the 8PK4 species reveals not only a large number of short- and long-range contacts between residues throughout the fibril assembly, but distinct clusters of contacts off the main diagonal in the contact map, which correspond to intra-chain β-strand interactions between β-sheets of the same chain and inter-chain and inter-protofilament interactions that define the helical assembly. There are also several contiguous clusters of regions with very high densities of contacts, which likely represent important interaction-dense hot spots for contributing to the overall integrity of the fibrils and are thought to be like nucleation and propagation centres recently proposed in studies of structure-function relationships by other researchers for α-synuclein fibrillar polymorphs. There is considerable evidence that disrupting these hot spot hubs would result in the destabilisation of fibrils and subsequently lead to reduced pathogenic potential for the formation of new fibrils.[18]

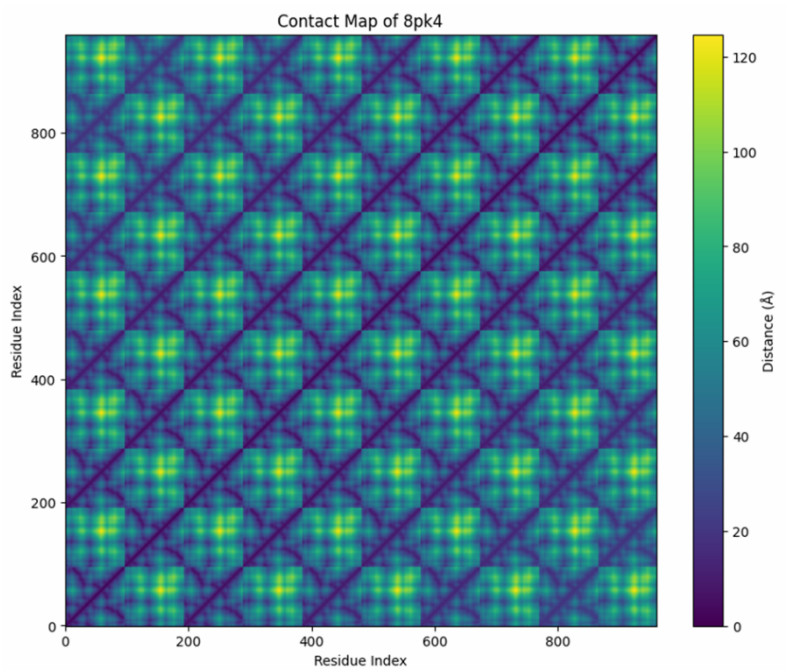

Figure 12: Structural implications of contact-rich regions for therapeutic targeting

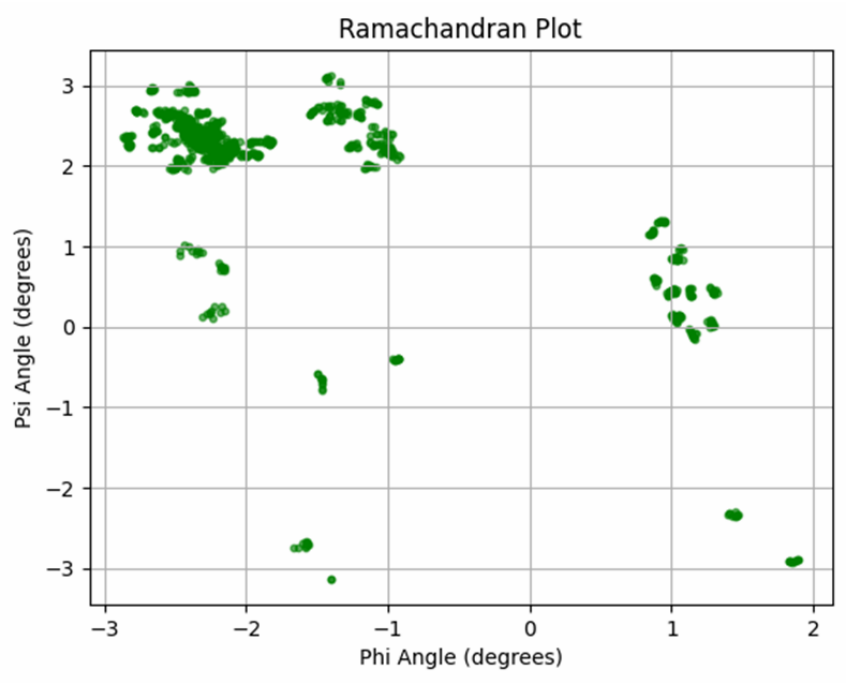

Figure 13: Ramachandran plot of α-synuclein polymorph 8PK4

Ramachandran plot by using Bio Python:

Bio Python was used in the Ramachandran Plot analysis for evaluating the conformational integrity of the backbone of the α-synuclein polymorph 8PK4. The φ–ψ angle distributions show that the majority of residues were within the sterically allowed regions and determine that α-synuclein adopts the ordered, fibrillar conformational state. The clustering seen supports the structural validity of the PDB model, which confirms that the identified regions of contact were not due to distortion of the backbone. This level of conformational consistency increases confidence in the residue-level interaction and hotspot analyses to follow.[18,19]

Implications for Structure-Guided Targeting of α-Synuclein Aggregates

Collectively, the integration of Bio Python into these analyses illustrates a strong structural heterogeneity of the eight 8PK4 polymorphs: a compact, hydrophobic, and hydrogen bond-stabilised core, and much more flexible, surface-exposed regions with a significantly greater atom mobility and density of interactions. Such a structure is consistent with current fibrillar "strain" models, wherein the conserved core of the polypeptide defines each fibrillar "strain"; the total amount of unstructured region of the polypeptide (i.e., surface-exposed region) modifies the interactome, cell responses, disease phenotype, and cellular morphology of the fibrillar assembly. In the context of Parkinson's disease, the flexible and interaction-rich nature of the surface-exposed regions and the density of contact made in the hotspots characterised here are good candidates for structure-guided compound design (e.g., small molecule, peptide, or nucleic acids) to disrupt key contacts without directing the complete disassembly of the fibril.[20]

V. Limitations And Future Directions

The results presented in this study are limited to a computational analysis of the static structure derived from cryo-EM studies and do not take conformational dynamics or cellular context into consideration. Future experimental validation through a combination of biochemical assays, structural approaches, and in vitro/animal models is required to determine the biological relevance of the identified contact hotspots and flexible regions, as shown by multi-omics and structure-function approaches of α-synuclein polymorphs.

VI. Conclusion

In this study, a comprehensive structural bioinformatics investigation of the α-synuclein polymorph 8PK4 associated with Parkinson’s disease was performed using BioPython-based computational approaches integrated with multi-omics perspectives. Detailed residue-level analyses, including structural validation, residue interaction mapping, atomic composition profiling, B-factor flexibility evaluation, and contact map analysis, provided deeper insight into the structural organisation and stability of the amyloid fibril.The findings revealed the presence of a compact and interaction-rich fibrillar core stabilised by dense residue contacts and hydrogen-bond interactions, while several surface-exposed regions exhibited comparatively higher flexibility

and atomic mobility. These flexible regions may play an important role in intermolecular interactions, fibril propagation, and cellular responses associated with Parkinson's disease pathology. Ramachandran plot assessment further confirmed that the majority of residues fall within energetically favourable conformational regions, supporting the reliability of the analysed structural model.The identification of contact-dense hotspots and structurally stable core residues highlights potential molecular regions that could serve as targets for therapeutic intervention. Structure-guided strategies focusing on these interaction sites may assist in designing small molecules, peptides, or other modulators capable of disrupting pathological aggregation of α-synuclein without destabilising the entire protein framework. This work demonstrates that the integration of BioPython driven structural analysis with multi-omics insights provides a powerful computational framework for studying aggregation-prone proteins involved in neurodegenerative disorders. Such approaches can contribute to a better understanding of the molecular mechanisms underlying Parkinson's disease and support the development of structure-based therapeutic strategies aimed at preventing or reducing α-synuclein aggregation in future research.

References:

[1]. Association Of Synuclein Alpha (SNCA) Gene Polymorphisms With Spontaneous Brain Activity In Patients With Parkinson's Disease - Pubmed. Https://Pubmed.Ncbi.Nlm.Nih.Gov/39281177/ (Accessed 2026-02-12).

[2]. Bank, R. P. D. RCSB PDB - 8H04: Major Polymorph In Alpha-Synuclein Fibril Seeded By Cerebrospinal Fluid From A Postmortal Parkinson's Disease Patient. Https://Www.Rcsb.Org/Structure/8H04 (Accessed 2026-02-12).

[3]. KUMARI, U.; Pacholee, K. In-Silico Drug Discovery-Based Approach To Treat Impairments In Patients Of Alzheimer's Disease. In JETIR; 2023; Vol. 10.

[4]. Kumari, U.; Pradhan, M.; Mukherjee, S.; Chakrabarti, S. NGS ANALYSIS APPROACH FOR NEURODEGENERATIVE DISEASE WITH BIOPYTHON. In JETIR; 2023; Vol. 10.

[5]. Singh, N.; Kumari, U. Mult-Omics-Based Drug Discovery Pipelines Incorporating Machine Learning, AI, And Biopython In Parkinson's Disease: A Case Study Of 5A Polymorph Of Alpha-Synuclein. 2026, 13 (2).

[6]. Handel, A. E.; Disanto, G.; Ramagopalan, S. V. Next Generation Sequencing In Understanding Complex Neurological Disease. Expert Rev. Neurother. 2013, 13 (2), 10.1586/Ern.12.165. Https://Doi.Org/10.1586/Ern.12.165.

[7]. Meade, R. M.; Fairlie, D. P.; Mason, J. M. Alpha-Synuclein Structure And Parkinson's Disease – Lessons And Emerging Principles. Mol. Neurodegener. 2019, 14, 29. Https://Doi.Org/10.1186/S13024-019-0329-1.

[8]. Annotations: 8PK4. Https://Www.Rcsb.Org/Annotations/8PK4 (Accessed 2026-03-02).

[9]. Protein Structure Validation. Https://Www.Biostars.Org/P/9469348/ (Accessed 2026-03-10).

[10]. Shen, N.; Song, G.; Yang, H.; Lin, X.; Brown, B.; Hong, Y.; Cai, J.; Cao, C. Identifying The Pathological Domain Of Alpha-Synuclein As A Therapeutic For Parkinson's Disease. Int. J. Mol. Sci. 2019, 20 (9), 2338. Https://Doi.Org/10.3390/Ijms20092338.

[11]. Cock, P. [Biopython-Announce] Biopython 1.81 Wheels For Python 3.12 Now On Pypi, 2023. Https://Lists.Open-Bio.Org/Pipermail/Biopython-Announce/2023-October/000185.Html (Accessed 2026-03-02).

[12]. Frey, L.; Ghosh, D.; Qureshi, B. M.; Rhyner, D.; Guerrero-Ferreira, R.; Pokharna, A.; Kwiatkowski, W.; Serdiuk, T.; Picotti, P.; Riek, R.; Greenwald, J. On The Ph-Dependence Of A-Synuclein Amyloid Polymorphism And The Role Of Secondary Nucleation In Seed-Based Amyloid Propagation. Elife 2024, 12. Https://Doi.Org/10.7554/Elife.93562.3.

[13]. Bio.PDB.Residue Module — Biopython 1.81 Documentation. Https://Biopython.Org/Docs/1.81/Api/Bio.PDB.Residue.Html (Accessed 2026-03-02).

[14]. Protein Structure Validation. Https://Www.Biostars.Org/P/9469348/ (Accessed 2026-03-10).

[15]. Subunit Fusion Unlocks Rapid In Vitro Maturation For Slowly Activating Heterodimeric [Fefe]-Hydrogenases. Pubmed Central (PMC). Https://Pmc.Ncbi.Nlm.Nih.Gov/Articles/PMC12933642/ (Accessed 2026-03-10).

[16]. Procter, J. B.; Carstairs, G. M.; Soares, B.; Mourão, K.; Ofoegbu, T. C.; Barton, D.; Lui, L.; Menard, A.; Sherstnev, N.; Roldan-Martinez, D.; Duce, S.; Martin, D. M. A.; Barton, G. J. Alignment Of Biological Sequences With Jalview. Methods Mol. Biol. Clifton NJ 2021, 2231, 203–224. Https://Doi.Org/10.1007/978-1-0716-1036-7_13.

[17]. Bank, R. P. D. RCSB PDB - 8PK4: Cryo EM Structure Of The Type 5A Polymorph Of Alpha-Synuclein. Https://Www.Rcsb.Org/Structure/8PK4 (Accessed 2026-03-02).

[18]. Kahle, P. J.; Haass, C.; Kretzschmar, H. A.; Neumann, M. Structure/Function Of Alpha-Synuclein In Health And Disease: Rational Development Of Animal Models For Parkinson's And Related Diseases. J. Neurochem. 2002, 82 (3), 449–457. Https://Doi.Org/10.1046/J.1471-4159.2002.01020.X.

[19]. Li, Y.; Zhao, C.; Luo, F.; Liu, Z.; Gui, X.; Luo, Z.; Zhang, X.; Li, D.; Liu, C.; Li, X. Amyloid Fibril Structure Of A-Synuclein Determined By Cryo-Electron Microscopy. Cell Res. 2018, 28 (9), 897–903. Https://Doi.Org/10.1038/S41422-018-0075-X.

[20]. Dilliott, A. A.; Abdelhady, A.; Sunderland, K. M.; Farhan, S. M. K.; Abrahao, A.; Binns, M. A.; Black, S. E.; Borrie, M.; Casaubon, L. K.; Dowlatshahi, D.; Finger, E.; Fischer, C. E.; Frank, A.; Freedman, M.; Grimes, D.; Hassan, A.; Jog, M.; Kumar, S.; Kwan, D.; Lang, A. E.; Mandzia, J.; Masellis, M.; Mcintyre, A. D.; Pasternak, S. H.; Pollock, B. G.; Rajji, T. K.; Rogaeva, E.; Sahlas, D. J.; Saposnik, G.; Sato, C.; Seitz, D.; Shoesmith, C.; Steeves, T. D. L.; Swartz, R. H.; Tan, B.; Tang-Wai, D. F.; Tartaglia, M. C.; Turnbull, J.; Zinman, L.; Hegele, R. A. Contribution Of Rare Variant Associations To Neurodegenerative Disease Presentation. Npj Genomic Med. 2021, 6 (1), 80. Https://Doi.Org/10.1038/S41525-021-00243-3.