Comparison of the Binding Strengths of known Mortaparibs with mortalin, p53 and PARP1

Project Report

Submitted as a part of **BBD451** B. Tech Major Project

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2020BB10010

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DECLARATION

I certify that.

a. The work contained in this report is original and has been done by me under the guidance of my supervisor(s).
b. I have followed the guidelines provided by the Department in preparing the report.
c. I have conformed to the norms and guidelines given in the Honor Code of Conduct of the Institute.
d. Whenever I have used materials (data, theoretical analysis, figures, and text) from other sources, I have given due credit to them by citing them in the text of the report and giving their details in the references. Further, I have taken permission from the copyright owners of the sources whenever necessary.

Signed by:
Aryan Godara

CERTIFICATE

It is certified that the work contained in this report titled "**Comparison of the binding strengths of known mortaparibs with mortalin, p53 and PARP1**" is the original work done by Aryan Godara and has been carried out under my supervision.

Prof. D. Sundar

Date:19^{th} sept. 2023

ABSTRACT

Mortaparib is a novel inhibitor of mortalin, p53 and PARP1. Where, Mortalin and PARP1 are two proteins that are essential for cancer cell survival and proliferation. Mortaparibs work by binding to and inhibiting both mortalin and PARP1. Mortaparibs are used as they interact with and inhibit these proteins, resulting in growth arrest and apoptosis signaling in cancer cells, in vitro and in vivo. It's advantageous as it is a small molecule, making it easier to administer. Plus, it's interaction with the proteins are reversible in nature, which means that it is less likely to cause any long-term side effects. The side effects range from nausea and vomiting to fatigue. And it can interact with other drugs, so it's important to keep up to date with the other medicines in use by the patients who are prescribed mortaparibs. And lastly, it's not effective against all types of cancers. In this study we focus on studying and comparing the binding strengths of known mortaparibs with mortalin, p53 and PARP1. It is important to study the binding strengths to evaluate the ease and extent of inhibition, since different Mortaparibs have varying levels of affinity for different mutations of p53 and/or PARP1. We will use computation techniques like the ones previously used in identifying novel Mortaparib drugs. And we'll create a dataset comparing the binding strength of known Mortaparibs against p53 and PARP1. And then use the observations to compare the mortaparibs and find the most effective ones. The mortaparibs used in this study are Mortaparib^{Mild}, and Mortaparib^{Plus}.

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List of abbreviations

Abbreviation	Description
PARP	Poly (ADP-ribose) polymerase
HSP	Heat Shock Protein
ADP	Adenosine diphosphate
ACE2	Angiotensin-converting enzyme 2
His296	Histidine 296
Lys31	Lysine 31
Glu35	Glutamic acid 35
Glu75	Glutamic acid 75
Ser441	Serine 441
Ser436	Serine 436

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Chapter 1

Introduction

Mortaparibs are a class of small-molecules inhibitors that target the DNA repair proteins p53, PARP1 and Mortalin. It works by binding to and inhibiting to each of mortalin, p53, and PAPR1 in different ways. Mortaparibs and currently being evaluated in clinical trials for the treatment of a variety of cancers, including breast cancer, ovarian cancer and prostate cancer. It's still in early-phase clinical trials, so it's a novel drug. And in these trials it has shown to be safe and well-tolerated, and to have promising anti-tumour activity. Albeit, having minor side effects like nausea, vomiting and fatigue. And it's not really effective against all types of cancer.

However, more data is needed on the binding strengths of mortaparibs with the three proteins in order to optimize their clinical use.

Mortalin is an HSP, that helps to repair DNA damage. And PARP1 is an enzyme that helps to repair single-stranded DNA breaks. When Mortaparib inhibits both mortalin and PARP1, it creates a double-stranded DNA break that cell can't repair. This results in apoptosis.

Mortalin is Hsp70 chaperone protein that is found in the mitochondria, endoplasmic reticulum, plasma membrane, cytoplasmic vesicles, and cytosol. It's a highly conserved protein that is found in call eukaryotic cells. It's also involved in the development and progression of Cancer. Overexpression of mortalin has been linked to increase in cancel cell proliferation, invasion and metastasis. Mortalin has also been shown to inhibit the activity of tumour suppressor proteins like p53. It plays a roles in a variety of cellular process (including, but not limited to) Protein folding and assembly, DNA repair, apoptosis (programmed cell death), stress response, immune response, viral infection.

What is PARP1?

PARP1 is a nuclear protein that is involved in a variety of cellular processes, including DNA repair, transcription, and cell death. PARP1 is activated by DNA change, and it binds to DNA break to repair them.
PARP1 activity is essential for the survival of cancer cells. They (cancer cells) often have defects in their DNA repair machinery, and they rely on PARP1 to repair their DNA damage. Therefore, inhibition of PARP1 can be an effective way to kill cancer cells.

What is p53?

P53 is a tumour suppressor protein that plays a critical in cell growth and division. p53 is activated in response to DNA damage and it can induce cell cycle arrest or apoptosis (programmed cell death) to prevent the proliferation of damaged cells.
Mutations in p53 are common in cancers, and these mutations can lead to the development of tumours. Mortaparibs have been shown to bind to p53 and activate its function. This can lead to apoptosis in cancer cells, even in cells with p53 mutations.

What is Mortalin?

Mortalin, also known as Heat Shock 70kDa Protein 9 (HSPA9), is a multifunctional protein that plays a crucial role in various cellular processes. It is a member of the heat shock protein 70 family, known for their response to stress conditions in cells. Mortalin is involved in the regulation of cell proliferation and apoptosis, which are essential for maintaining cellular homeostasis. It operates in the mitochondria, where it assists in protein folding and the protection of cells from stress-induced damage. Additionally, Mortalin has been implicated in the aging process and in certain diseases, including cancer and neurodegenerative disorders like Parkinson's disease. Its dual role in both normal cellular functions and disease states makes it a significant focus of research in the fields of cell biology and medicine.

Binding Strengths of Mortaparibs with PARP1, Mortalin and p53

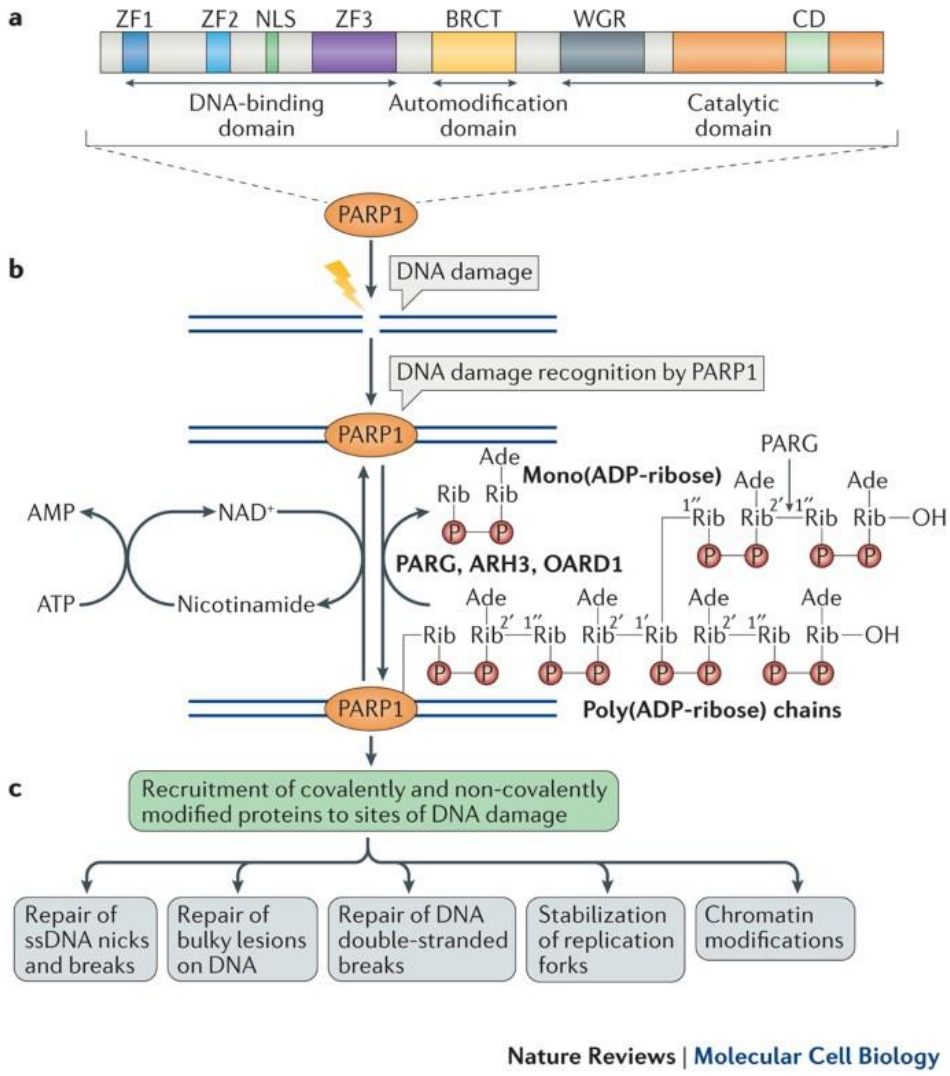
The binding strength of a Mortaparib with PARP1 or mortalin or p53 is a key determinant of its biological activity. A higher binding strength results in more effective inhibition of PARP1 activity or activation or p53 function, and greater sensitization of cancer cells to radiation therapy and chemotherapy.
The binding of mortaparibs to p53 is a secondary effect. Mortaparibs were not designed to target p53, and their binding affinity for p53 is generally weaker than their binding affinity for PARP1.

Clinical Implications of the Binding Strengths of Mortaparibs

The binding strengths of mortaparibs with PARP1 and p53 have important clinical implications. Mortaparibs have a higher binding strength with PARP1 are likely to be more effective in inhibiting PARP1 activity and sensitizing cancer cells to radiation therapy and chemotherapy. Mortaparibs with a higher binding strength with p53 may also have additional clinical benefits. E.g.: they may be more effective in inducing apoptosis of cancer cells and in preventing the development of drug resistance.
More research is needed to determine the optimal binding strengths of mortaparibs with PARP1 and p53 for the treatment of different types of cancer.

This chapter of the report focuses on reviewing the current research available on the topic, and to discuss which approaches are best for comparing the binding strengths of known Mortaparibs with p53 and PARP1.

PARP1



PARP1 stands for Poly(ADP-ribose) polymerase 1. It is an enzyme that is involved in DNA repair. PARP1 is activated when DNA is damaged. It then attached ADP-ribose molecules to other proteins, which helps to repair the DNA damage.

PARP1 is also involved in cell death. When DNA is damaged and can't be repaired, PARP1 can become overactivated. This can lead to cell death by apoptosis.

PARP1 is overexpressed in many types of cancer, including breast cancer, ovarian cancer, and pancreatic cancer. This is because cancer cells are often more prone to DNA damage. The overexpression of PARP1 can make cancer cells more sensitive to PARP inhibitors.

Some of the ways in which PARP1 is involved in cancer:

- **DNA Repair:** PARP1 is involved in the repair of DNA damage. When DNA is damaged, PARP1 attaches ADP-ribose molecules to other proteins, which helps to repair the damage.
- **Cell Death:** PARP1 is also involved in cell death. When DNA is damaged and can't be repaired, PARP1 can become overactivated. This can lead to apoptosis.
- **Oncogene:** PARP1 can act as an oncogene is come cases. This means that it can promote the growth of cancer cells.
- **Tumour Suppressor:** PARP1 can also act as a tumour suppressor in most cases. This means that it can inhibit the growth of cancer cells.

Interaction of Mortaparib with PARP1

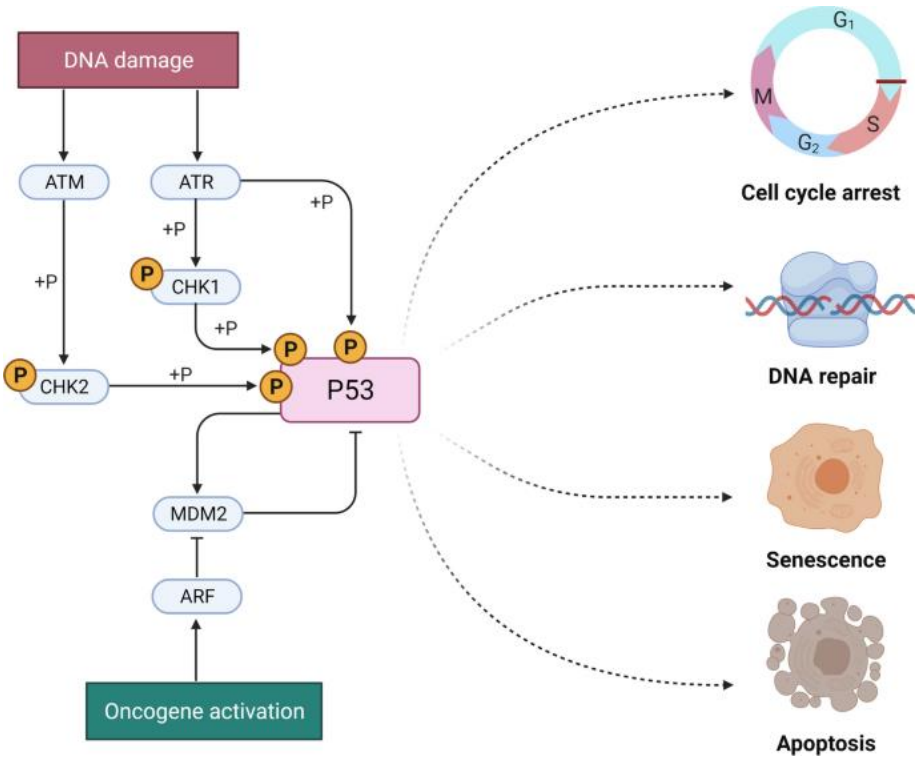
Mortaparib is a PARP inhibitor that binds to the active site of PARP1. This prevents PARP1 from attaching ADP-ribose molecules to other proteins, which blocks PARP1 from repairing DNA damage.

The active site is a pocket on the surface of PARP1 that is essential for its function. Mortaparib fits into this pocket and blocks PARP1 from binding to its substrates.

The binding of Mortaparib to PARP1 is reversible. This means that PARP1 can still be activated if Mortaparib is removed. However, the binding of Mortaparib to PARP1 is very strong, so it is unlikely that PARP1 will be able to be activated in the presence of Mortaparib.

The inhibition of PARP1 by Mortaparib can lead to cell death in cancer cells that are dependent of PARP1 for DNA repair. This is because cancer cells aren't able to repair DNA damage are more likely to die.

p53



p53 is tumour suppressor gene. This means that it helps to prevent cells from becoming cancerous. p53 becomes does this regulating the cell cycles, DNA repair, and apoptosis (programmed cell death).

When DNA is damaged, p53 can activate a number of genes that help to repair the damage. If the damage can't be repaired, p53 can also activate genes that lead to apoptosis. This helps to prevent cells with damaged DNA from dividing and becoming cancerous.

p53 is often referred to as the "**guardian of the genome"** because it plys such an important role in preventing cancer. Mutation in the p53 gene are found in about half of all human cancers. These mutations can lead to p53 becoming inactive, which can allow cells with damaged DNA to divide and become cancerous.

There are a number of ways to target p53 in cancer therapy. One approach is to use drugs that activate p53. These drugs can help to repair DNA damage and prevent cancer cells from dividing. Another approach is to use drugs that inhibit the activity of proteins that can inactivate p53. These drugs can help to restore p53 functions and prevent cancer cells from becoming cancerous.

Some of the ways p53 is involved in cancer:

- **Cell cycle arrest:** p53 can trigger cell cycle arrest, which means that the cell stops dividing. This gives the cell time to repair DNA damage.
- **DNA repair:** p53 can activate genes that are involved in DNA repair. This helps to repair DNA damage that could lead to cancer.
- **Apoptosis:** p53 can trigger apoptosis, which are programmed cell death. This helps to remove cells with damaged DNA that could become cancerous.

Interactions of Mortaparibs with p53

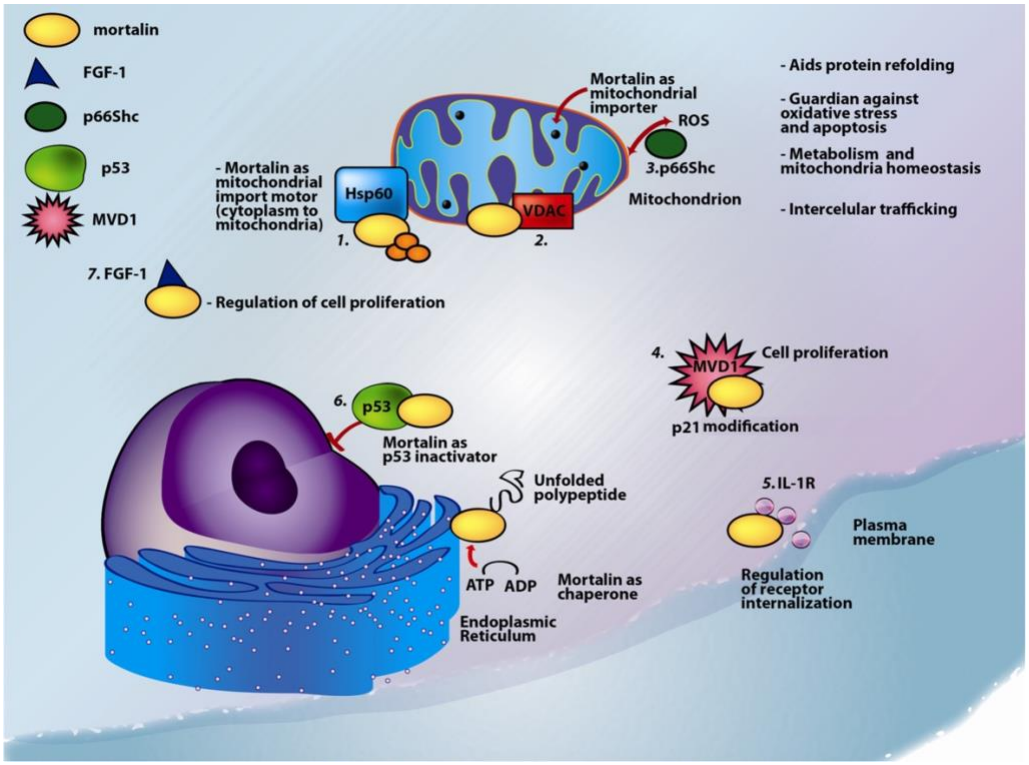
Mortaparib doesn't directly interact with p53. However, it can indirectly affect p53 by inhibiting PARP1.

PARP1 is an enzyme that is involved in DNA repair. When DNA is damaged, PARP1 helps to repair the damage. However, if PARP1 is inhibited, it can lead to the accumulation of DNA damage. This can activate p53, which can then trigger cell cycle arrest, DNA repair or apoptosis.

Some of the ways in which Mortaparib can indirectly affect p53

- **Inhibiting PARP1:** Mortaparib can inhibit PARP1, which can lead to the accumulation of DNA damage. This can activate p53, which can then trigger cell cycle arrest, DNA repair, or apoptosis.
- **Restoring p53 function:** Mortaparib can restore p53 function in cells that have mutated p53 genes. This can help to prevent cells from dividing and becoming cancerous.
- **Inhibiting proteins that inactivate p53:** Mortaparib can inhibit proteins that can inactivate p53. This can help to restore p53 function and prevent cancer cells from becoming cancerous.

Mortalin



Mortalin as a Regulatory Protein: Mortalin, also known as HSPA9, is a chaperone protein that is crucial for mitochondrial function and cellular stress response. It plays a key role in the maintenance of mitochondrial integrity, import of proteins into the mitochondria, and the prevention of mitochondrial-associated cell death.

Mortalin's Role in Cellular Protection and Aging: Mortalin interacts with various cellular components to assist in protein folding, protection from oxidative stress, and regulation of the cellular life span. It's implicated in the aging process, and its dysfunction can be associated with age-related diseases.

Mortalin in Cancer: Overexpression of Mortalin has been observed in several types of cancers. It helps cancer cells evade apoptosis, contributing to the immortalization of the cancer cells. Mortalin's inhibition is being studied as a potential therapeutic approach to trigger the death of cancer cells.

Interactions of Mortalin with the p53 Pathway: Mortalin can bind to the tumor suppressor protein p53, regulating its function. By sequestering p53 in the cytoplasm, Mortalin prevents it from executing its tumor suppressive functions such as DNA repair and apoptosis.

Mortalin as a Therapeutic Target: Strategies targeting Mortalin are focused on disrupting its interaction with p53, which could restore the tumor-suppressive activities of p53. Additionally, inhibiting Mortalin's chaperone activity could impair the survival of cancer cells, making it a potential target for anti-cancer therapies.

Some of the ways Mortalin is involved in cellular regulation:

- **Mitochondrial Function**: Mortalin ensures proper mitochondrial function by aiding in protein transport and folding within the organelle.
- **Stress Response**: It helps cells respond to stress by preventing the aggregation of misfolded proteins.
- **Cellular Senescence and Apoptosis**: Mortalin has a role in inhibiting apoptosis, which can contribute to cellular senescence and tumorigenesis when dysregulated.

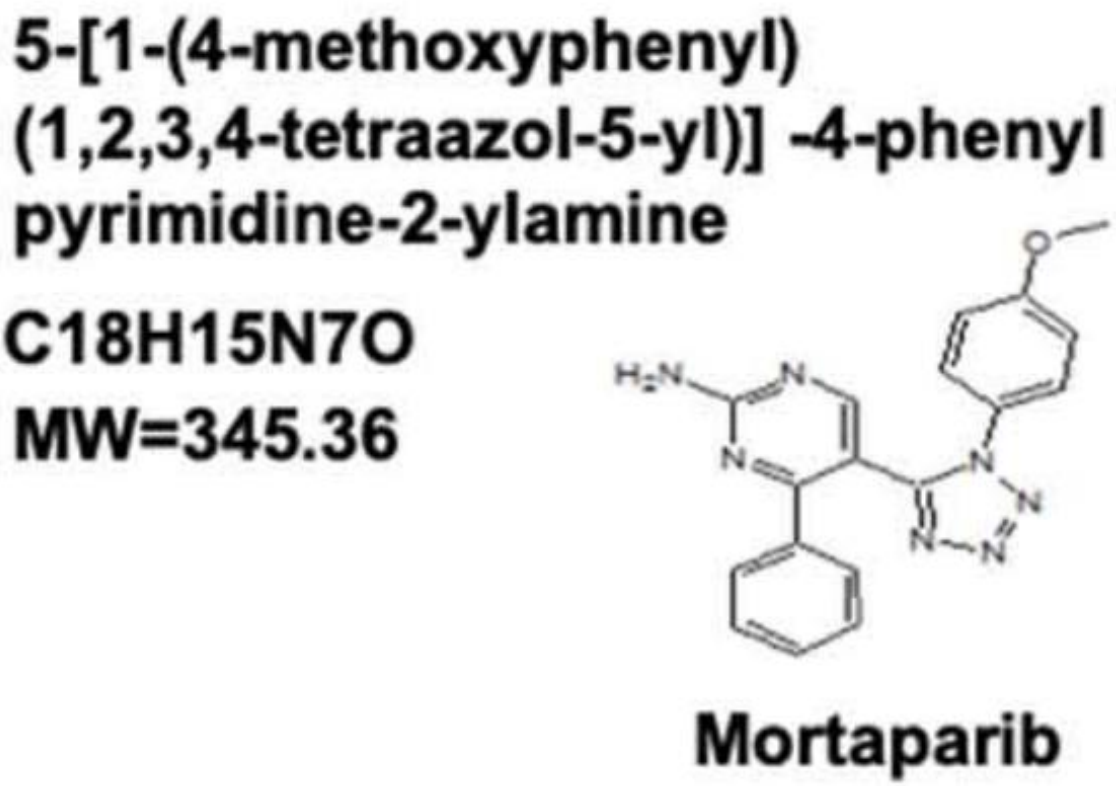
Interactions of Therapeutics with Mortalin:

- **Inhibition of Mortalin**: Small molecule inhibitors that can disrupt Mortalin's function could potentially reactivate apoptotic pathways in cancer cells.
- **Restoration of p53 Activity**: By inhibiting Mortalin, it may be possible to release p53 from its cytoplasmic sequestration, allowing it to resume its role in DNA repair and apoptosis.

Some of the ways in which therapeutics can indirectly affect Mortalin:

- **Disrupting Mortalin-p53 Interaction**: Agents that can disrupt the Mortalin-p53 complex could restore p53 function, leading to the induction of apoptosis in cancer cells.
- **Chaperone Inhibition**: Drugs that inhibit Mortalin's chaperone activity could increase the susceptibility of cancer cells to stress, leading to their death.

Structure of Mortaparib:



Why use Mortaparib?

5-[1-(4-methoxyphenyl)(1,2,3,4-tetraazol-5-yl)]-4-phenylpyrimidine-2-ylamine

https://pubchem.ncbi.nlm.nih.gov/compound/2825874

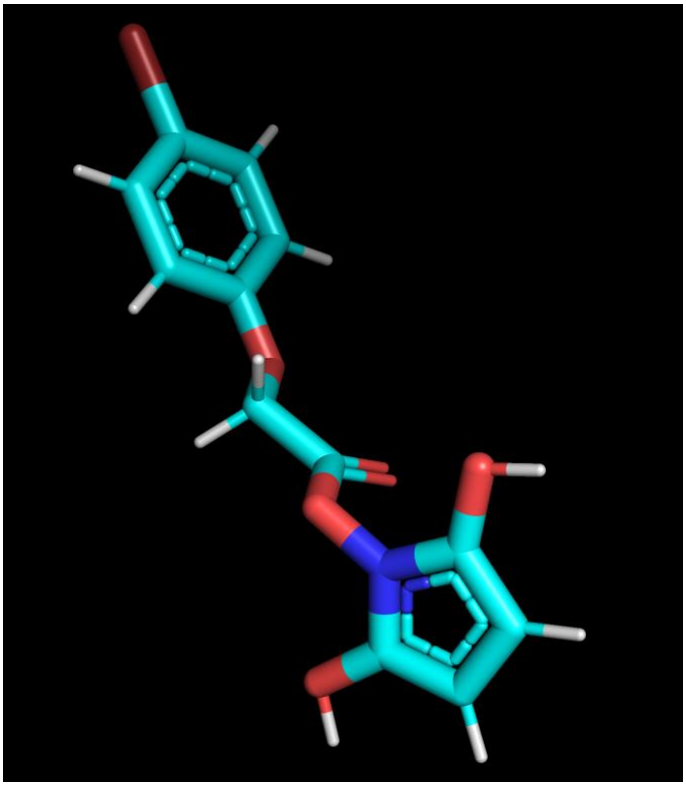
Mortaparib structure PubChem link

In the research paper titled **"Mortaparib, a novel dual inhibitor of mortalin an PARP1, is a potential drug candidate for ovarian and cervical cancers."** , it was concluded that Mortaparib inhibits both Mortalin and PARP1.

This results in activation of growth arrest and apoptosis signalling in cancer cells (both in vivo and in vitro).

Known Mortaparibs :-

Mortaparib^{Mild}



Mortaparib-mild is a novel triazole derivative that targets mortalin and PARP1 proteins, essential for cancer cell survival and proliferation. It is a small molecule that is designed to specifically bind to the active site of PARP1.

The active site is a pocket on the surface of PARP1 that is essential for its function. Mortaparib-mild fits into this pocket and blocks PARP1 from binding to its substrates.

The binding of Mortaparib-mild to PARP1 is reversible. This means that PARP1 can still be activated if Mortaparib-mild is removed. However, the binding of Mortaparib-mild to PARP1 is very strong, so it is unlikely that PARP1 will be able to be activated in the presence of Mortaparib-mild.

The inhibition of PARP1 by Mortaparib-mild can lead to cell death in cancer cells that are dependent on PARP1 for DNA repair. This is because cancer cells that are unable to repair DNA damage are more likely to die.

Mortaparib^{Plus}

Mortaparib-Plus is an innovative chemical entity emerging in the field of oncology for its dual-action mechanism that targets critical proteins in cancer cell physiology. Its primary mechanism of action involves the inhibition of mortalin, a mitochondrial chaperone protein, which usually binds to and inactivates the tumor suppressor protein p53. By inhibiting mortalin, Mortaparib-Plus facilitates the release and activation of p53, thereby promoting the execution of its tumor suppressive functions including DNA repair, cell cycle arrest, and apoptosis. This reactivation is crucial, especially in cancer cells where p53 functionality is often compromised due to mutations, playing a pivotal role in tumor growth suppression.

In addition to targeting mortalin, Mortaparib-Plus acts on PARP1, an enzyme that is essential for repairing single-strand breaks in DNA. By impeding PARP1 activity, Mortaparib-Plus causes an accumulation of DNA damage in cancer cells, leading to their death. This is particularly effective in cancer cells that are deficient in other DNA repair pathways, a condition often found in tumor cells, making them uniquely vulnerable to PARP1 inhibition.

Molecular docking of Mortaparib^{Mild} with p53, mortalin, and PARP1

Firstly, I procured the PDB structures of p53(10LG), Mortalin(4KB0), and PARP1(4ZZZ).
The PDB IDs were publicly available; so the structures had to be download in the .pdb (protein data bank) format.
Because the licence for all the tools in Schrodinger wasn't available, and only the academic-free version was working; a workaround had to be developed for it. And after some trials and errors. A new workflow was created.

Building chemical structures for the Mortaparibs.

The structure for Mortaparib^{Mild} was available publicly on PubChem, so it was download from that repository. (CID = 91464252) .
For Mortaparib and Mortaparib^{Plus}, the structures were made manually from ChemSketch with the version freely available for college students

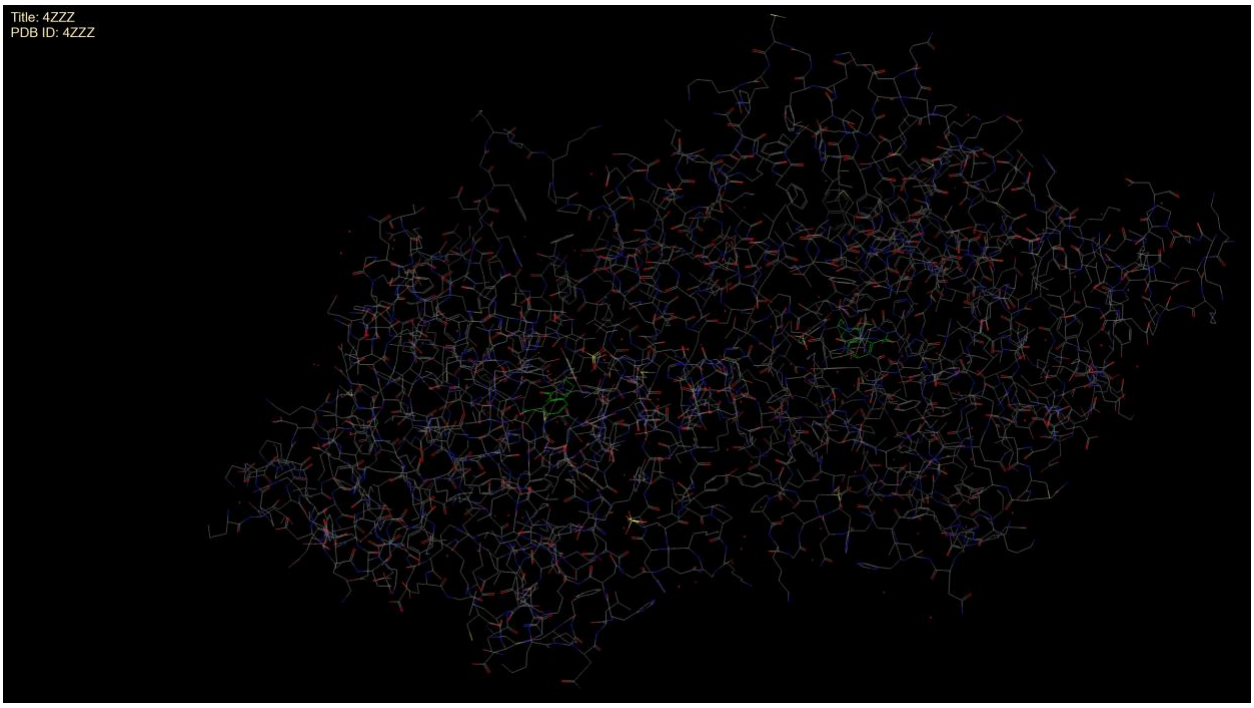
This resulted in errors down the line, after the docking on PyRX. After which Mortaparib had to be dropped in the end, as the ***mortaparib.sdf*** file was getting corrupted after starting over multiple times.

Protein Preparation on Schrodinger Maestro:

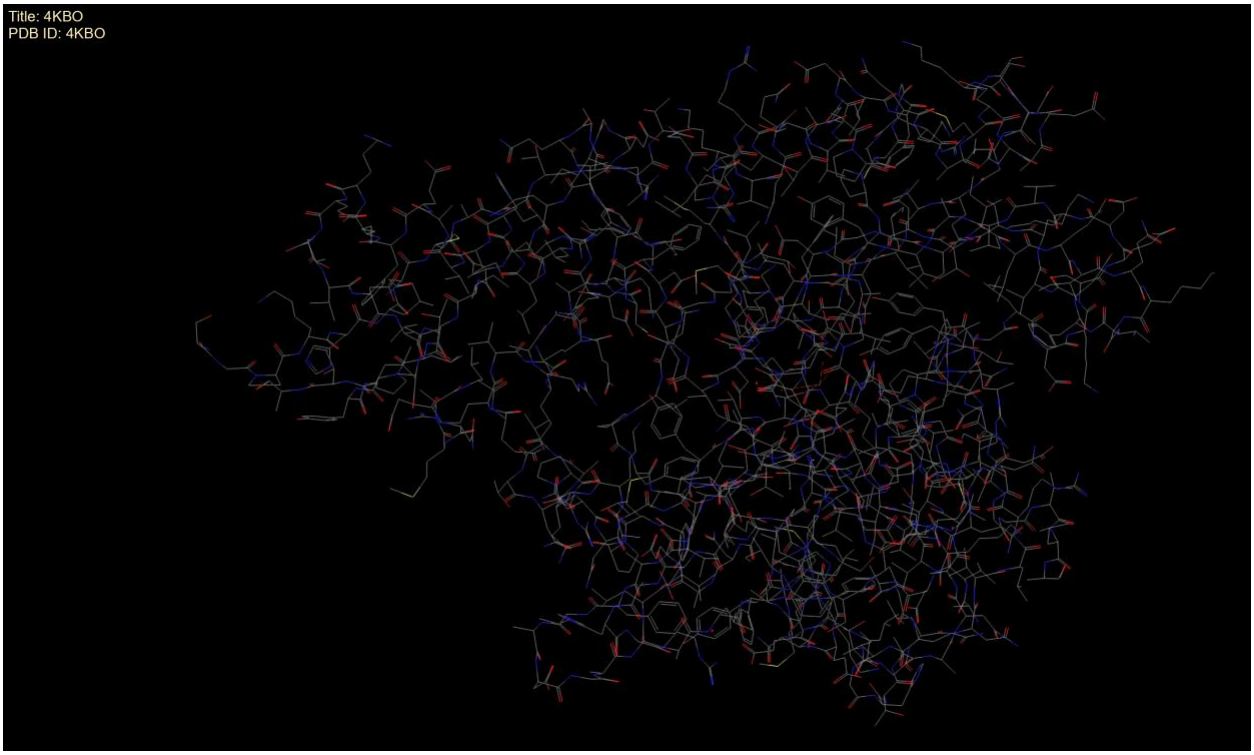
- **Project Setup**: A dedicated project workspace was established in Schrödinger Maestro, an integrated suite for molecular simulation, providing a platform for visualizing, manipulating, and analysing biomolecular data. This advanced software facilitates rigorous computational tasks, often essential for structural biology research.

- **Structure Acquisition**: Atomic coordinates for the protein structures of Mortalin, p53, and PARP1 were procured in the form of Protein Data Bank (PDB) files. These files were individually loaded into the project, each serving as a template for subsequent computational refinement and simulation.
- **Employment of Protein Preparation Wizard**: The Protein Preparation Wizard, a feature within Schrödinger Maestro, was utilized to refine the protein structures. This automated tool is crafted to execute several pre-processing tasks including the assignment of correct bond orders, addition of hydrogen atoms, and identification of disulphide bonds.
- **Initial Pre-processing**: The 'Pre-process' function within the Wizard was engaged with default settings, which included the removal of water molecules beyond 5 Å from the hetero groups, creation of zero-order bonds to metals, and filling in of missing side chains and loops utilizing Prime, Schrödinger's structure prediction technology.
- **Hydrogen Bond Network Optimization**: After pre-processing, the 'H-bond Optimization' was initiated under standard parameters. This function adjusted hydroxyl, thiol, and amine group orientations to form optimal hydrogen bonds, and performed a water orientation optimization using a sampling of water orientations in conjunction with the Epik state penalties at pH 7 ± 2. The default setting also included a restrained minimization step to relax the structure, applying a root-mean-square deviation (RMSD) cut-off of 0.30 Å for heavy atoms.

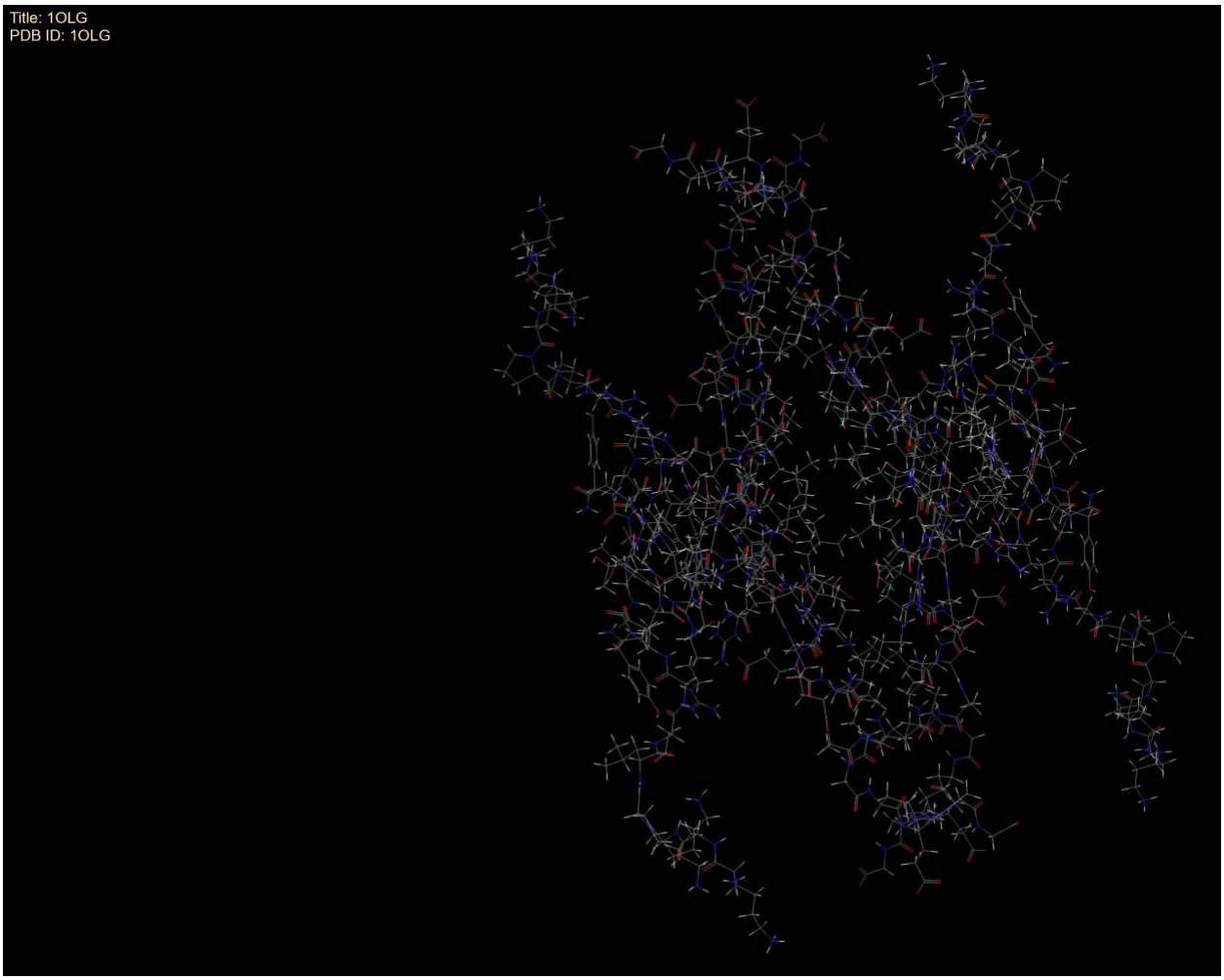
The integration of these steps, executed with Schrödinger Maestro's default parameters, ensured that the protein structures of Mortalin, p53, and PARP1 were systematically and uniformly prepared. This standardization is crucial for the reliability and reproducibility of molecular simulations, which in turn informs the understanding of protein behaviour in biological systems.



4ZZZ (PARP1 in Schrodinger)



4KB0 (Mortalin in Schrodinger)



10LG (p53 in Schrodinger)

Molecular Docking in PyRX

Docking grids were generated around the mortalin binding region of p53 (312–352), the p53 binding region of mortalin (253–282), and the Olaparib-binding site in the catalytic domain of PARP1 (862–880)

OpenBabel is integrated into PyRx, which is a virtual screening software, to facilitate the conversion and management of ligand files. For Ligands, OpenBabel serves the critical function of interconverting chemical formats, ensuring that the ligand structures are compatible with the different molecular simulation tools used within PyRx. It can translate various chemical file formats to the ones required by PyRx for docking simulations, thereby streamlining the process of setting up and running virtual screenings for potential drug compounds. This conversion is crucial because it allows researchers to utilize a wide array of ligand databases and formats, optimizing the process of identifying promising therapeutic molecules.

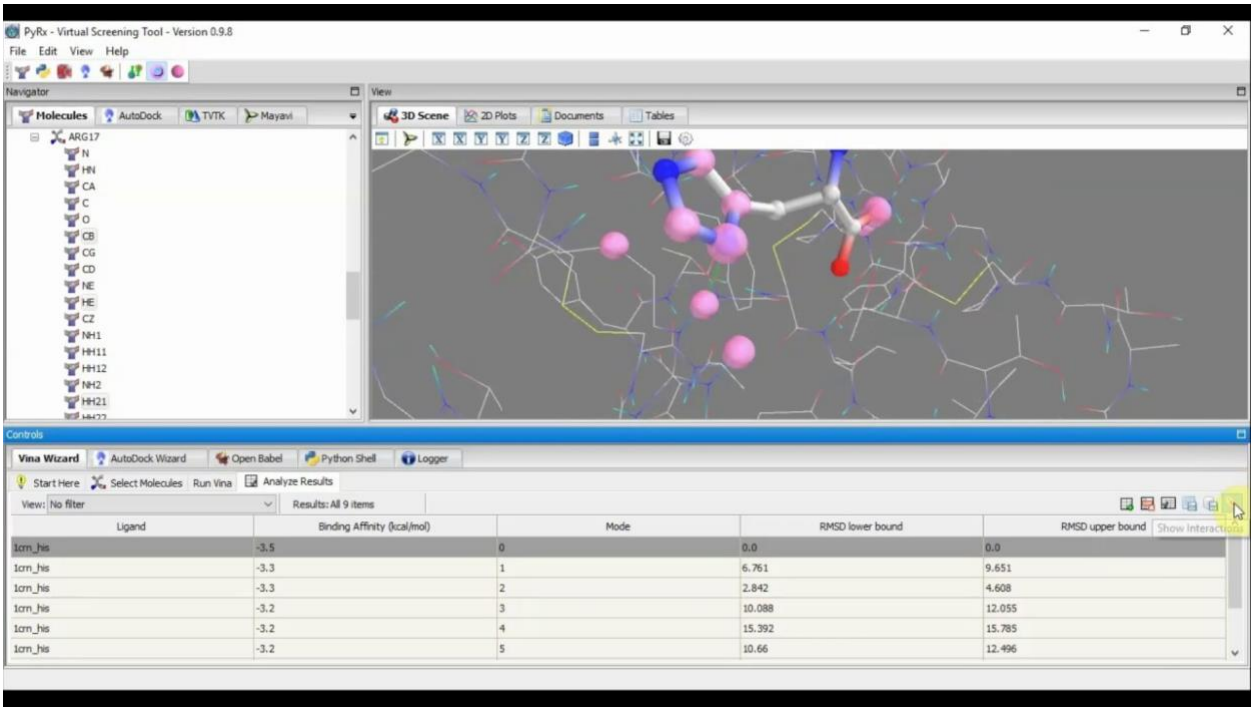
After this, Molecular Docking was perform in **Vina Wizard** of PyRX. The steps for this are:-

So, I ran the OpenBabel module on the three Mortaparibs (Mortaparib, Mortaparib$^{Plus, and}$ Mortaparib^{Mild}), and exported the resulting structures in .pdb and .sdf format.

Vina Wizard simplifies the process of setting up and running molecular docking simulations, which predict how small molecules, such as drugs, bind to a receptor of known 3D structure. The wizard provides a user-friendly approach to configure the docking parameters, allowing for the meticulous exploration of ligand-receptor interactions. This is crucial for the identification of potential binding affinities and the subsequent development of pharmaceutical agents.

- **Loading Molecules**: Upon initiating the PyRx interface, two molecules were selected for the docking study. The ligand, chosen from the Mortaparib series (Mortaparib, Mortaparib-plus, or Mortaparib-mild), and the target protein (either Mortalin, p53, or PARP1) were loaded into the workspace. Each molecule was individually imported into the Vina Wizard, ensuring that the ligand and protein files were in the correct format for compatibility with the docking software.

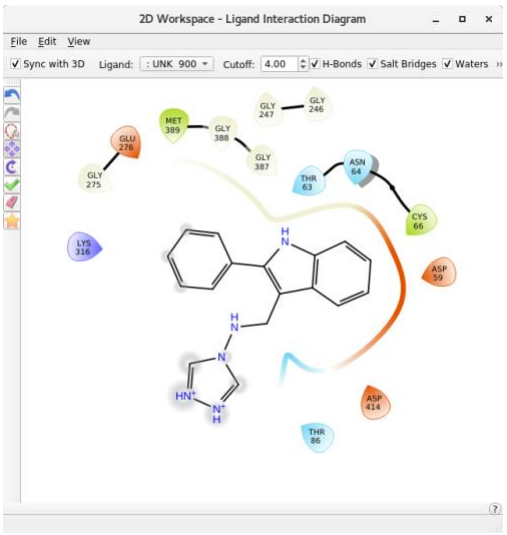
- **Binding Region Identification and Atom Tagging**: The next step involved the identification of the binding regions on the protein surface. These regions were carefully selected based on known active sites or predicted interaction domains. Within the selected binding region, specific atoms were tagged to guide the docking process, ensuring that the ligand would interact with functionally relevant areas of the protein.
- **Grid Setting and Docking**: An orthogonal grid box was created around the active site of the protein to define the search space for the docking simulations. The dimensions of the grid box were adjusted to encapsulate the entire active site, allowing for sufficient space for the ligand to orient itself in various conformations. Subsequently, the Vina Wizard was executed to perform the docking operation, leveraging the computational algorithms to predict the optimal binding conformations of the ligands with the protein.
- **Results Analysis and Saving**: After the docking simulation was completed, the results were evaluated to determine the binding affinities and the most favourable ligand-protein conformations. The docking poses were saved in Structure-data file (sdf) format for each ligand-protein pair. This format was selected for its compatibility with various molecular visualization tools, facilitating further analysis and presentation of the docking study.



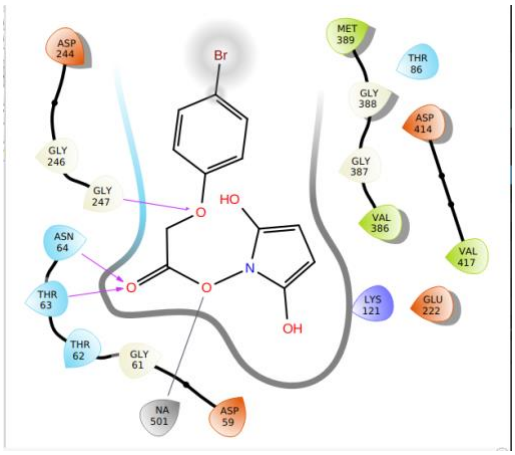
System Builder in Maestro

After saving the results, I was back in Maestro to Merge the protein and mortaparib into 6 respective groups:-

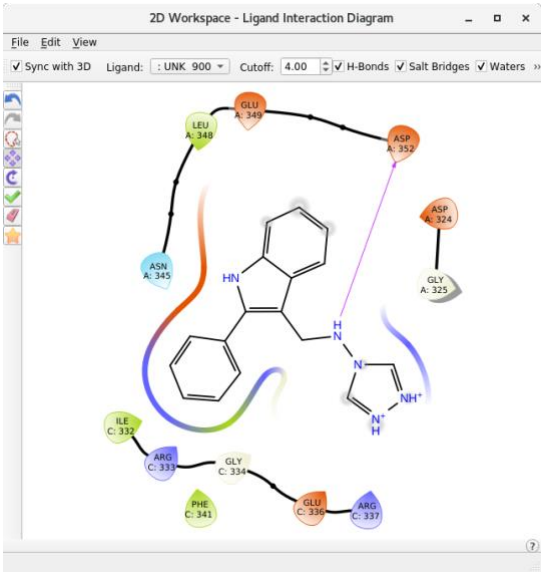
1. 4KB0 and Mortaparib^{Plus}



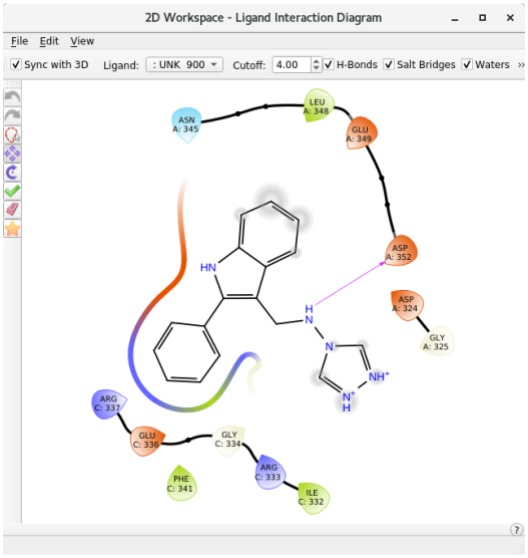
2. 4KB0 and Mortaparib^{Mild}



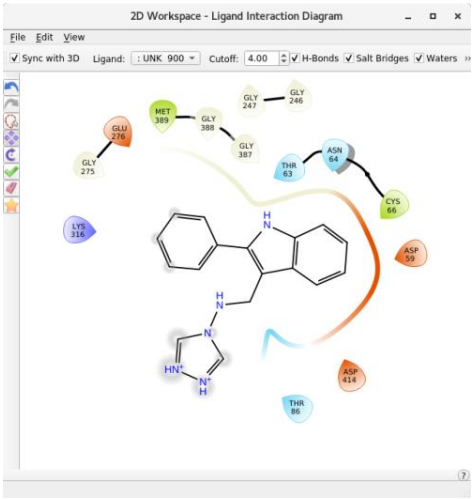
3. 10LG and Mortaparib^{Plus}



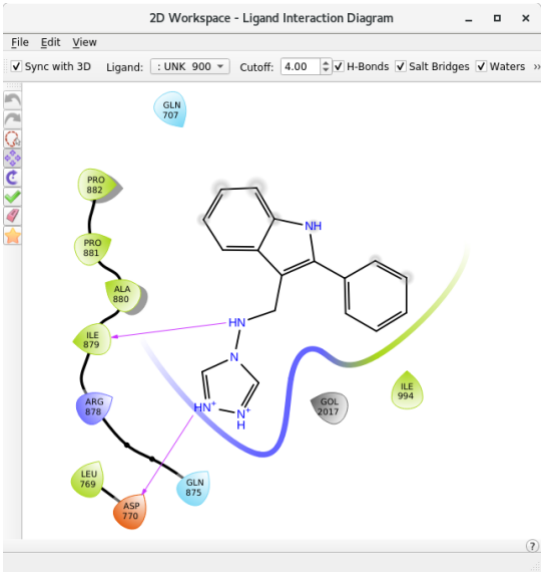
4. 10LG and Mortaparib^{Mild}



5. 4ZZZ and Mortaparib^{Plus}



6. 4ZZZ and Mortaparib[Mild]



Merge Ligand + protein

The first step was to merge the two structures (docked structure of Ligand from Vina Wizard). Then, use the preprocessed structure of protein.

After this, the two were merged in Schrodinger, and the Protein preparation tooling was run again.

System Builder

Utilizing the System Builder, scientists can construct and refine the molecular systems they wish to study. This includes the ability to add solvents, create membranes, and adjust ion concentrations to mimic physiological conditions, thereby enhancing the biological relevance of the simulations. The System Builder's intuitive interface streamlines the process of setting up the system prior to simulation, ensuring that the protein and ligand are optimally prepared.

So, I ran the System Builder wizard to solvate the ligand + protein structure.

Minimization

After this, I ran Minimization with default settings for 100 ps.

Molecular Dynamics

Finally, MD simulations were performed for 50seconds for each pair. And on average it took 30 hrs to perform one simulation.

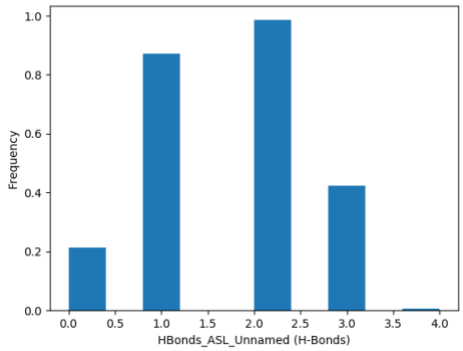
For our simulation tasks, we utilized the Desmond module from the Schrödinger suite (referenced from the 2020 edition). The initial structures of our protein-ligand complexes, post-docking, were immersed in a simulation box using Desmond's "system builder" functionality, employing the standard TIP3P model for water molecules. We defined the simulation box's boundaries with an orthorhombic shape, ensuring a 10 Å buffer space around the complexes. Charge neutrality was achieved by the judicious addition of sodium and chloride ions tailored to each specific complex.

Once we had our solvated complexes ready, we proceeded with an energy minimization step. This was conducted using a Brownian dynamics approach for a duration of 20 picoseconds at a reduced temperature setting of 10 Kelvin within the NVT ensemble, which helped in alleviating any potential spatial conflicts between atoms.

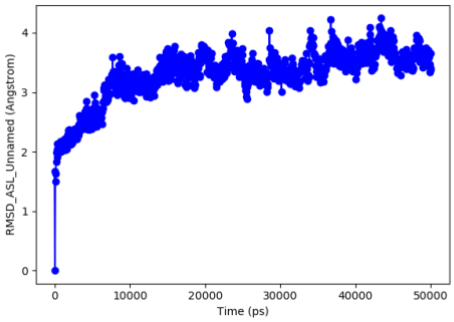
Subsequent to energy minimization, our systems underwent an equilibration phase. This consisted of a seven-stage process alternating between NVT and NPT ensembles, adhering to a predefined relaxation protocol available in the Desmond module of the Schrödinger suite.

The final stage of our simulation was the production run, extending over a 100 nanosecond timescale within the NPT ensemble and incorporating a 2 femtosecond timestep. We maintained our system's temperature at 300 Kelvin with a 1 picosecond relaxation time and the pressure at 1 atm with a 2 picosecond relaxation time. For short-range electrostatic interactions, we established a cutoff radius of 9 Å. Throughout these simulations, we opted not to impose any restraints on the molecules, and all remaining parameters were kept at their default settings.

4ZZZ and Mortaparib^{Mild}

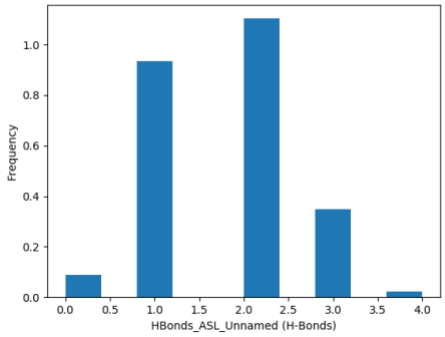


H-bond Frequency Histogram

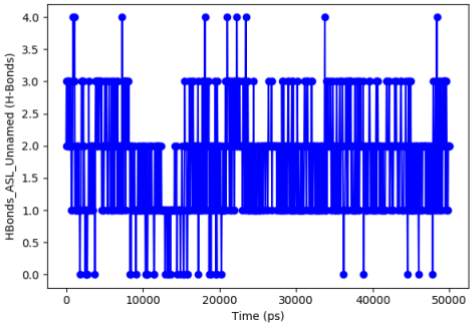


RMSD Time-Series

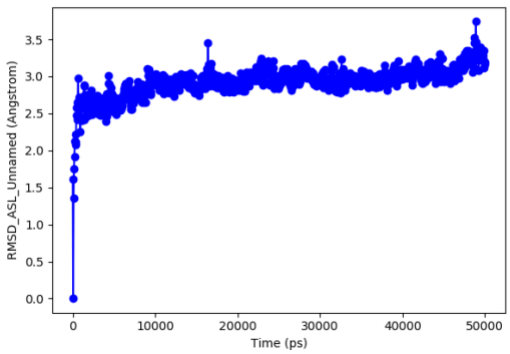
4ZZZ and Mortaparib^{Plus}



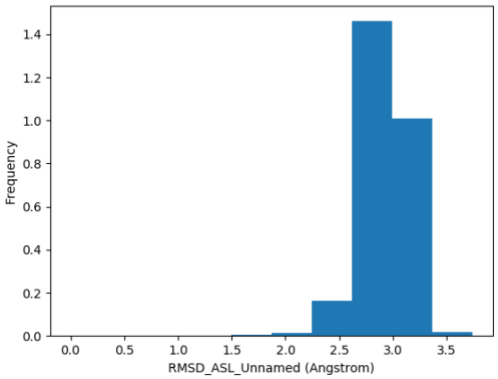
H-bond Histogram



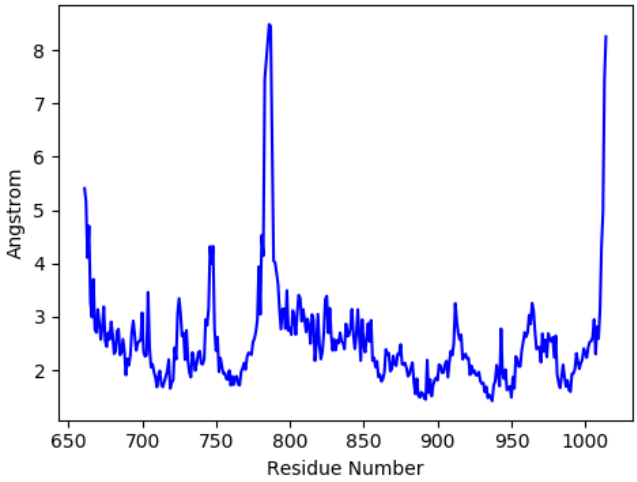
H-bond time series



RMSD Time-Series

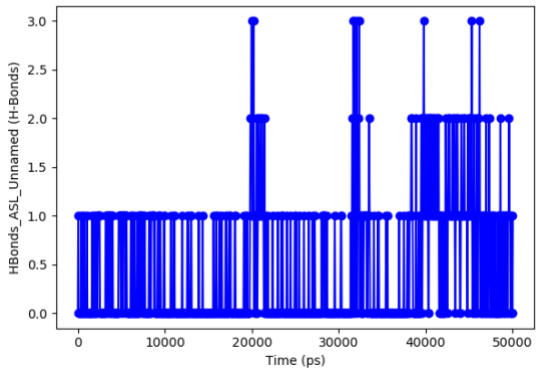


RMSD Histogram

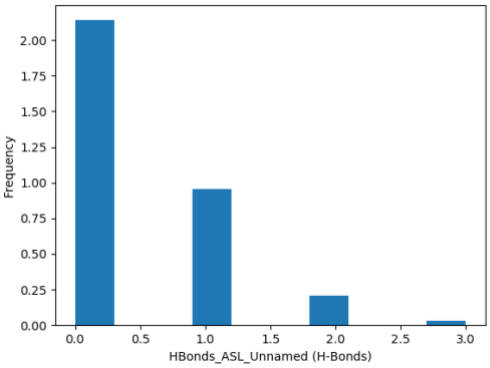


RMSF Curve

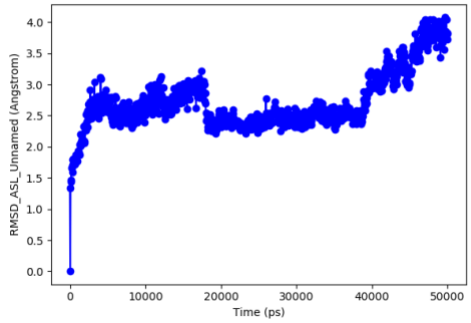
4KB0 and Mortaparib[Plus]



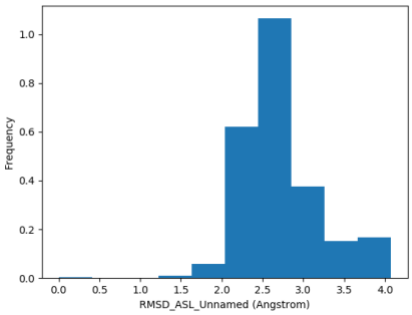
H-bond Time-series



H-bond histogram

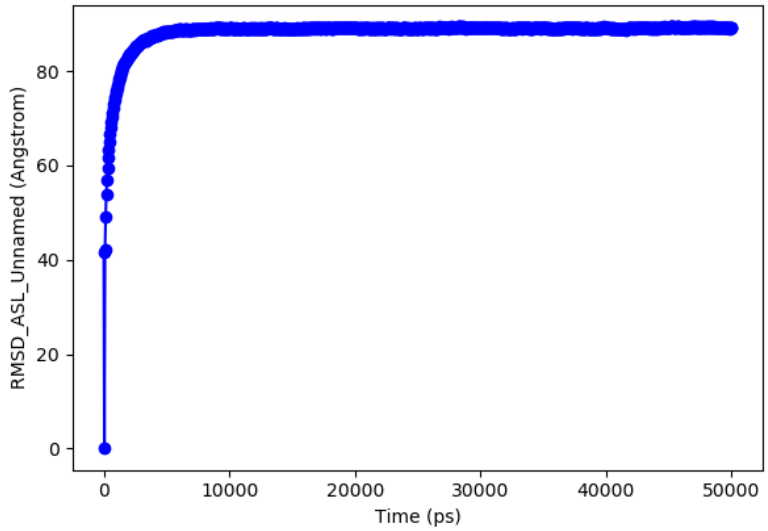


RMSD Time-Series



RMSD Histogram

4KB0 and Mortaparib[Mild]



RMSD Time-series Curve

10LG results are still pending. I will append this in the presentation.

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