DETERMINING A PHARMACEUTICAL HIT FOR CORONAVIRUS-19 PROTEASE USING DOCKING STUDIES AND MOLECULAR SCANS

Kerin Ingegneri
ABSTRACT

The spread of Coronavirus-19 caused a global pandemic resulting in roughly 3.14 million deaths this past year. Our efforts here are to design an antiviral drug using molecular docking studies to target the main protease enzyme used by the virus. This is a known pharmaceutical target meaning the virus can not replicate without this enzyme. There are no known human homologues of this protease thus reducing potential side effects. By employing computer software systems, we have generated a model to produce docking studies using six different criteria evaluating the virtual compounds. The virtual compounds that we employ are drug like and similar in chemical moieties to known inhibitors. The goal is to dock structures readily available to purchase and test in vitro. Then using a pivot table from excel, the duplicates of the virtual compounds with the binding criteria are revealed. These docking studies reveal how tight the virtual compounds are binding at the active site along with structural kinetic data and the end goal is to find that pharmacological hit.
PHARMACEUTICAL HIT INTRODUCTION

• Molecular docking studies with crystal structure and virtual compounds
• Generates a binding energy $\Delta G$ value looking at molecular interactions with virtual compound and active site
• Virtual compound given ten different poses/conformations evaluated via six different criteria
• Top 5% pasted into Excel using Pivot table look for duplicates
PHARMACEUTICAL HIT INTRODUCTIONS

- Generate pose at active site further evaluate, molecular interactions
- Buy compounds
- Test on enzyme
- Optimize hit using virtual compounds
- Rational Drug design Structure Activity Relationships (SAR)
Objective is to inhibit protease enzyme that generates peptides that are vital to the replication of the virus
✓ Protease - A type of enzyme that breaks down proteins into smaller proteins or smaller protein units, such as peptides or amino acids.

✓ Known pharmaceutical target

✓ The protease is essential, but has no human homologues

✓ So inhibitors of the protease have less of a chance of hitting a human protease
This enzyme processes a polyprotein chain coded by the virus’s RNA, chopping up the chain into functional proteins that the virus then uses to assemble itself and multiply.

Disrupting this key piece of the virus’s self-replication machinery could bring an infection screeching to a halt.
ENZYMES

- Bind molecule/inhibitor to stop enzymes function treat patient
- Valid pharmaceutical target

https://digitalcommons.sacredheart.edu/acadfest/2021/all/144
Ingegneri: Integrated Screening for Covid-19 Inhibitors Identification of Ph

Once the virus has fused with a host cell, the virus injects genetic material and uses the host machinery to make copies of itself. Many teams are studying viral proteins involved in replication.

**Targeting M<sub>pro</sub>**
If the virus can't build its main components, it can't replicate. Researchers are trying to find drugs that block the cutting action of M<sub>pro</sub> by fitting into its two active sites—little dimples on the surface.

**Viral RNA enters the cell**

**Host-cell ribosome**

**Drug binding site**

**Drug**

**Polyproteins**

**The virus makes and uses a protease enzyme to chop the long strands into functional proteins.**

**Main protease (M<sub>pro</sub>)**

**Non-structural viral proteins**

**Proteins involved in replication and transcription form a complex within a host vesicle.**

**RNA-dependent RNA polymerase (RdRp)**

**Targeting RdRp**
This enzyme makes copies of the full viral genome. Several novel compounds and approved drugs, such as remdesivir, bind to its active site.

**Remdesivir**

**Active site**
MOLECULAR DOCKING

✓ Sort out duplicates between the five different binding criteria
✓ Compare pose to crystal ligand pose
✓ Look for hydrogen bonding interactions
✓ Analyze fit in binding pockets look for cavities in enzyme
✓ Look for acceptable pose buy compound test on in vitro assay
✓ Generate pharmacological hit
✓ With MOE software system, the crystal structure of the enzyme’s active site and the known inhibitor produce an S-score of -10.8
✓ This represents the binding energy
✓ Known covid inhibitors are scanned through the software to make sure they are suitable for binding
Active site with known inhibitor
CONTINUED FOR KNOWN INHIBITORS

-10 or lower shows good binding energy

Compound 091
S-score = -10.5

Compound 060
S-score = -10.2
ARE THEY DRUG-LIKE IN NATURE?

- SMILES codes of these compounds were put through Swiss ADME
  - Do compounds violate Lipinski's rules?
  - Do the compounds have a MW > 500?
Criteria for drug like molecules

1. Molecular mass less than 500 Dalton
2. High lipophilicity (expressed as LogP less than 5)
3. Less than 5 hydrogen bond donors
4. Less than 10 hydrogen bond acceptors
5. Molar refractivity should be between 40-130

Examples:

• 091 = breaks rules #1 and 5
• 060 = breaks rule #1
• 033 = breaks rule #1

DO THE COMPOUNDS FOLLOW LIPINSKI RULES?
Compounds were rescored using 5 different criteria important for binding

A pivot table was produced

Compounds 72 and 91 had the most hits
Next I compiled a list of protease inhibitors

Converted this into an SDF file

Protease inhibitors are scanned through the software to make sure they are suitable for binding
WASHED PROTEASE INHIBITORS

• Receive the S-score
• Compound 35 and 1 have an s-score of -10
• Look to see how they apply to Lipinski’s rules
Both break 2 of Lipinski's rules
FUTURE WORK

• Create similarity searches of the compounds with the highest binding energy
  • In doing so, stick to finding compounds that do not violate Lipinski’s rules
• Pharmacophore elucidation
  • Create a drug that follows Lipinski's rules and has high binding energy to the main protease for drug therapy