Imatinib: Challenges in Targeted Chemotherapy

Poster team: Jacklyn Ekern, Jason Wettengel, James Cruikshank
Jmol team: Patrick Pentek, Jerome Mauro
Faculty Advisor: Daniel S. Sem, Ph. D
Concordia University of Wisconsin
Mequon, WI
Preceptors: Eric Ebacher, Bob LaBello, Melissa Handcock, Harry Link, Laura Alar, Henry Eskin, Michael Bishop, Jerry Nooker, Cynthia Zernzach-Mathe, Jessica Haufschildt

Abstract

The goal of any chemotherapy treatment is to maximize efficacy and minimize toxicity. Unfortunately, most chemotherapeutic agents target both normal and cancerous cells, which increases the likelihood of side effects and toxicity. Advances in medicinal chemistry and chemotherapy have resulted in a number of targeted chemotherapeutic agents. These agents have been developed to target only cancerous cells, thereby minimizing side effects and toxicity. Imatinib is a chemotherapeutic agent that targets the BCR-ABL protein, which is expressed by cancerous cells. This type of chemotherapy is desired; however, it comes with challenges, such as resistance.

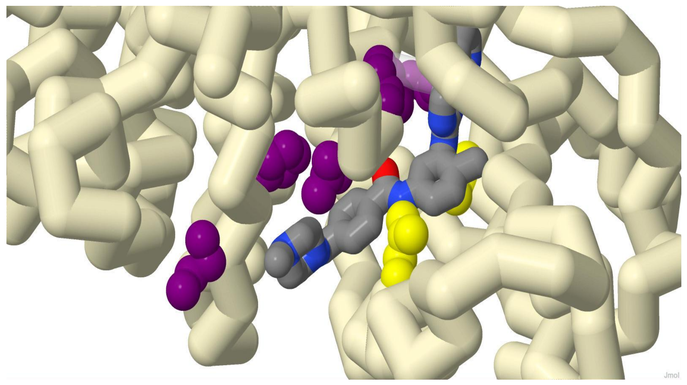

Figure 1. Imatinib in the binding pocket of BCR-ABL. The key amino acids
(Thr315 and Glu286) are highlighted in yellow.

Introduction

Prior to 2001, there was no FDA approved targeted therapy for Chronic Myelogenous Leukemia (CML). Non-specific treatment options often only delayed progression of the disease, but ultimately resulted in death.

A specific target was needed in order to attack only the CML cells. Upon decoding the human genome, a difference was discovered in the CML cells compared to normal healthy cells. This difference was a fusion of a hinged portion in the BCR-ABL protein which prevents the switching of the protein from inactive to active conformations. Imatinib binds to the kinase domain region of the affected BCR-ABL protein when it is in the inactive conformation. This binding locks the protein in the inactive conformation, preventing it from activating any second messengers within the signaling cascade that lead to cell proliferation or growth, preventing the cancer cells from spreading and continuing to grow.

Mutations to this protein however, lead to less binding affinity for the imatinib molecule and result in greater activation of the signaling cascade leading to proliferation of cancerous cells, which leads to disease progression and ultimately death.

The CREST Program is funded by grant #1022793 from NSF-CCLI.

Molecular Story

We concentrated on the molecular interactions between imatinib and only one of the ABL proteins, ABL2.

Five of the atoms within imatinib serve as hydrogen bond acceptors.

- Thr315 forms a hydrogen bond with a nitrogen.
- Met318 forms a hydrogen bond with the nitrogen in the piperidine ring.
- Glu286 forms a hydrogen bond with the amide.
- The final nitrogen located within the piperazine ring forms two hydrogen bonds with His 407 and Ile360.
- Asp 381 forms a hydrogen bond with oxygen.

Of these interactions, the hydrogen bond formed between nitrogen and Thr315 is the most important interaction to consider. It is this interaction and the interaction with the Phe382 residue that form the hydrophobic pocket.[1]

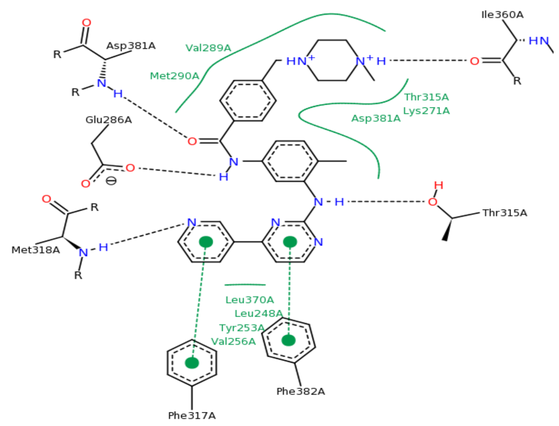

Figure 2. Key amino acids involved in drug binding in the active site. Note the location and relationship of Thr315, which is the "gatekeeper." Mutations in this amino acid confer resistance to imatinib.
http://www.pdb.org/pdb/explore/explore.do?structureId=3GVU

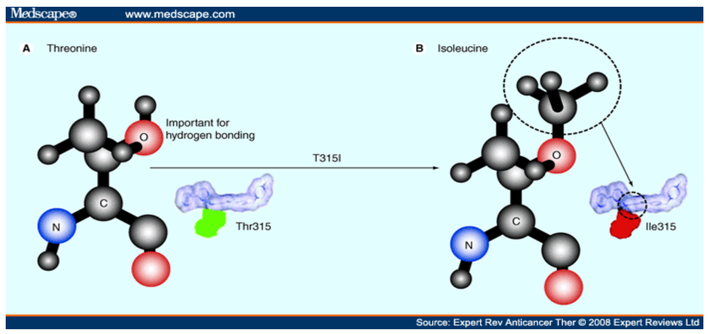

Figure 3. A mutation in Thr315 disrupts the hydrogen bonding between imatinib and the BCR-ABL protein.
http://www.medscape.com/viewarticle/580426_5

Resistance

There are a number of suspected resistance mechanisms associated with imatinib. Some leukemic cells have exhibited resistance to the drug, which has been determined to be the result of a point mutation, referred to as the "gatekeeper mutation". Imatinib is unable to bind to the ABL protein when a point mutation occurs and a hydrophobic amino acid is substituted in place of the threonine residue.[2] This indicates that the hydroxyl group on the threonine is essential in the drug binding to ABL. This single point mutation prevents effective binding of imatinib in the active site of BCR-ABL, so drug development is now geared toward investigating allosteric inhibitors.[3]

Imatinib is a substrate for P-glycoprotein. Resistance to imatinib therapy has also been attributed to over-expression of P-glycoprotein, decreasing cellular concentrations of the drug. Some individuals express greater concentrations of BCR-ABL than others, resulting in a decreased therapeutic response. It is likely a combination of mechanisms that can be attributed to decreased therapeutic outcomes.[3]

Future Research

Resistance is becoming a growing problem in cancer treatment. In the case of imatinib, point mutations in the active site render the drug unable to bind, which has resulted in resistance. Research is now being geared toward allosteric inhibitors, in an effort to circumvent point mutations in the binding site. Two new drugs have been discovered, GNF-2 and GNF-5. These drugs are competitive inhibitors of BCR-ABL, but bind to the myristate site.[4]

Resistance is bound to become a greater concern in targeted chemotherapy treatments given that many cancerous cells or tumors are exposed to radiation as a part of common treatment regimens. Radiation therapy causes alterations in DNA, which can result in mutations.

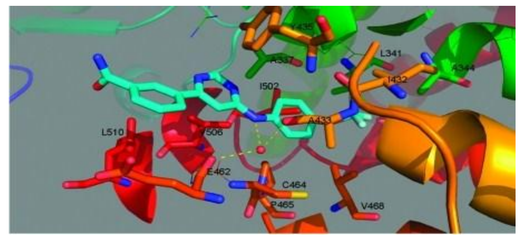

Figure 4. GNF-2 in the myristate binding site of BCR-ABL.
http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2951064/

Summary

Recent advances in medicinal chemistry have led to the development of new and exciting therapy options for many different types of cancer. These new therapies are much more targeted to cancer cells, preventing many of the unwanted side effects of classic chemotherapy options. Imatinib is a major step forward in this area. However, there are still many hurdles to overcome.

Resistance has proven to be a major player in the ineffectiveness of imatinib. However, it is through studying these resistances that researchers will one day be able to develop therapies which work similarly to imatinib, while avoiding the resistance issues. Hopefully these barriers will be overcome so we can truly aid and cure affected patients.

References

1. Wong, S. (2009). *Journal of Hematology & Oncology*, 2:10.
2. Jacob, R. E., Zhang, J., Gray, N.S., Engen, J.R. (2011).. *PLoS One*, 6(1), e15929.
3. Duckett, D. R., Cameron, M. D. (2010). *Expert Opinion on Drug Metabolism and Toxicology,* 6 (13), 1175-93.
4. Deng X, Okram B, Ding Q, Zhang J, Choi Y, Adrián FJ, Wojciechowski A, Zhang G, Che J, Bursulaya B, Cowan-Jacob SW, Rummel G, Sim T, Gray NS. J Med Chem. 2010 Oct 14;53(19):6934-46.