Recent studies have shown that targeting VEGF is a promising anti-cancer treatment since it is known to be responsible for angiogenesis. Blood supply plays a dual role in tumor growth: it supplies oxygen and nutrients, but also carries chemotherapeutic and other drugs to the tumor. Tumor vascularization is often abnormally fast and has poor blood flow. VEGF antagonism actually improves blood flow and drug access, but probably also improves oxygenation of the tumor. Therefore, inhibiting VEGF binding is likely important in cancer treatment. Studying VEGF inhibition was done mainly with mammalian cell culture models. The active site was characterized with modeling software and VEGF inhibitor targets were designed based on these modeling studies. The designed ligands were optimized using quantum mechanics based computational methods and several homology studies were done to assess the viability of targeting the VEGFR2 protein.

### INTRODUCTION

Angiogenesis plays an important role in tumor growth and metastasis. In the early 1970s, Folkman and colleagues identified a tumor angiogenesis factor that is mitogenic to capillary endothelial cells in human and animal solid tumors and suggested that blocking this factor might allow control of tumors with a tiny diameter (few millimeters). This later was called vascular endothelial growth factor (VEGF).

VEGF is recognized as an essential regulator of normal and abnormal blood vessel growth. It regulates both vascular proliferation and permeability, and it functions as an antiapoptotic factor for newly formed blood vessels. It is expressed in response to hypoxia, oncogenes, or cytokines, and its expression is associated with poor prognosis in several types of cancer.

In mammals, the VEGFs are encoded by a family of genes including VEGF-A, -B, -C, -D, and placental growth factor (PIGF). VEGF-A, -B, and PIGF are predominantly required for blood vessel formation, while VEGF-C and -D are essential for the formation of lymphatic vessels. The biological functions of VEGF require binding to receptor tyrosine kinases; VEGFR-1, VEGFR-2, and VEGFR-3. VEGF-A binds to VEGFR-1 and -2, while VEGF-C and -D bind to VEGFR-2 and -3 (Fig 1).

Several signaling pathways are activated by VEGFR2, including the PI-3 kinase/Akt pathway and the classical Ras-dependent signaling cascade impinging on MAP kinases such as ERK1 and ERK2 (Fig 2).

Recent studies have shown that targeting VEGF is a promising anti-cancer treatment since it is known to be responsible for angiogenesis and inhibition of apoptosis. Blood supply in tumors has a dual role that includes transporting not only oxygen and nutrients, but also chemotherapeutic and other drugs. VEGF inhibition has been shown to decrease tumor size and improve the survival of both animal and human tumors, by improving what is normally a leaky tumor vasculature. This has a dual effect of increasing both drug access and oxygen supply to the tumor. Thus, inhibiting VEGF should increase blood flow and improve the clinical outcome of chemotherapy and radiotherapy treatment.

VEGF has also been linked to dormancy induction of tumor cells because it has the ability of signaling to “damp down” the PI3K-AKT pathway. This should not only potentiate the pro-apoptotic effects of concurrent cytotoxic therapy, but also increase the probability of reversing the disease to dormant state.

### METHODOLGY

Cheminformatics:

1. Spartan v7.0.2.0, all the designed ligands were geometrically optimized at Hartree-Fock level of theory using 3-21G basis set.
2. Chemaxon2006 (Chem3D & Chemdraw) to draw chemical structure.

Biotechnologies:

1. Homology studies were done using Blast server provided through NCBI http://blast.ncbi.nlm.nih.gov
2. Prediction of secondary protein structure was done via SOPMA software installed on ESPasy Proteins tools web site: www.espasy.org/tools

### RESULTS

**Fig 3 & 4:** 2-D representation of the most important amino acids involved in VEGF-C/VEGFR2 interaction with the distances in Å.

**Fig 5 & 6:** 2-D representation of six designed ligands.

**Fig 7:** 3-D rendering of ligand 5 in the VEGFR2 active site.

### SOPMA analysis

**Fig 8:** Diagram representing the active site of VEGFR2.

**Fig 9:** Non-human homology study

**Fig 10:** Human homology study

### FUTURE WORK

Dr. Wilkinson’s lab has recently been working on whether a second gene influences how well VEGF can activate its receptor (KDR). His lab measures receptor activation by tyrosine phosphorylation of specific residues, which are needed for downstream signals. In their tests, they found that "tied" protein can directly associate with KDR to enhance activation and that this protein, called ESCR, is controlled via sRNA to reduce or increase its expression. They can then measure sensitivity to VEGF. The Wilkinson group has recently become interested in KDR as being presented on the cell surface as a complex, which makes drugs to both extracellular domain hold promise of altering its sensitivity or signaling output. Future research in anti-VEGF therapies will most likely combine anti-tumor therapy with anti-VEGF treatments (combination therapy and dosage time).

### SUMMARY

The project allowed us to use what we have learned in biochemistry and apply it to a current research project involving an important protein (VEGF). We were able to learn about the metabolic pathways that include VEGF and how it plays a significant role in angiogenesis and inhibits the tumor growth. The project was expected to enhance the knowledge of the protein, and to have a better sense of the nature of the binding. Lastly, work on this project required us to learn some very useful software and techniques that can be applied in a wide array of fields.

### REFERENCES