

## **Abstract**

Pancreatic Cancer is the fourth leading cause of cancer death with the lowest survivability. Usually by the time of diagnosis, the cancer has already spread throughout the body and there has not been a better treatment option in 40 years. Farnesol is an Isoprenoid, which is an experimental drug class that has been shown to inhibit cell development. By using it with the chemotherapeutic agent 5-Fluorouracil, these two drugs have the potential to be good combination therapy. By measuring cell growth with different concentrations, my objective is to find evidence that these two drugs will be better synergistically than either of them alone.

## **Significance of Project**

Cancer is a disease where cells of the body grow without being inhibited. They are not normal cells because they do not die and can rapidly grow. Some types of cancer can form a tumor, like breast and lung, but does not necessarily have to, as seen in leukemia. When it has not invaded other tissue or migrated to other parts of the body it is called a benign tumor. If it has invaded into the basement membrane of a tissue then it defined as malignant. Once malignant tumors begin to spread to other tissues and organs it is called metastasis.

The pancreas is an organ in the abdomen cavity that is about 6 inches long and 2 inches wide. It contains both exocrine and endocrine properties, meaning it produces enzymes and proteins to send to other parts of the body and for itself. The main function of the endocrine gland is to make insulin so the body can take up glucose. Beta cells on the islet of Langerhans produce insulin, while alpha cells make glucagon and delta cells produce somatostatin. Exocrine gland acinar cells make digestive enzymes. The exocrine gland epithelial cells which are

responsible for making sodium bicarbonate and digestive enzymes are the cells which cause most people with pancreatic cancer to die.

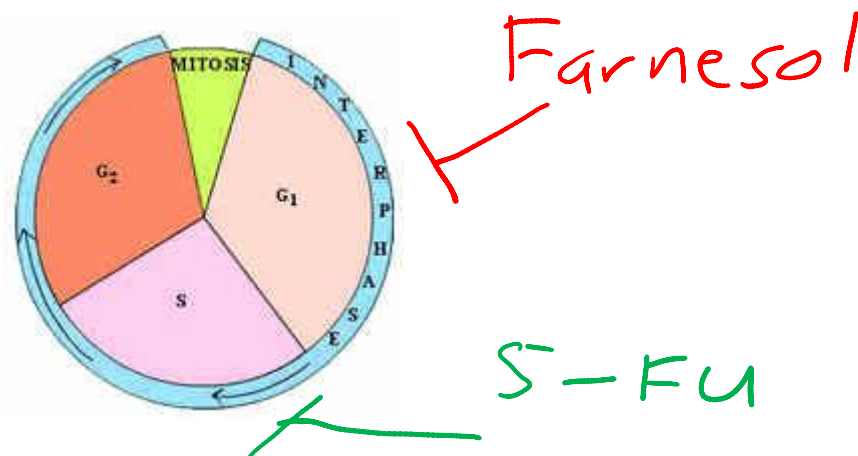
Tumors can form on either the exocrine or endocrine glands of the pancreas, but usually it is on the exocrine. Pancreatic cancer is the fourth leading cause of cancer mortality in the United States. There were 44,000 new cases of pancreatic cancer diagnosed in 2011, along with 37,600 deaths. The 5-year survival rate is 14% if caught in the earliest phase of the disease.<sup>3</sup> That number decreases as the cancer progresses to different phases.

Pancreatic cancer has one of the lowest survivability ratings of all human cancers, and the current ways to treat it are not enough. Currently there are 3 ways to treat this cancer: chemotherapy, radiation therapy, and surgery. Surgery is only a viable option if you discover the disease early on. Normally when a person is diagnosed, the cancer has already metastasized and doing the surgery would not cure it. Radiation therapy can be used as an adjunct therapy to chemotherapy. It helps shrink the cancer cells. Currently, chemotherapy is the best option at treating pancreatic cancer.

Chemotherapy is used to kill or inhibit the cells, so it will stop spreading to different parts of the body. It can be given in intravenous or pill form, so it is not a local therapy. A new experimental drug class for pancreatic cancer is called Isoprenoids. Although the exact mechanism of action is not known, isoprenoids exhibit anti-growth effects by interrupting the G<sub>0</sub>/G<sub>1</sub> part of the cell growth cycle. They increase expression of p21<sup>Clp1</sup> and p27<sup>Kip1</sup>, which are cyclin kinase inhibitor proteins that allow for cell inhibition.<sup>5</sup> The Isoprenoid Farnesol, which will be used in this research, is plant based from chamomile and lemongrass.

Combining an Isoprenoid with a chemotherapeutic agent already proven effective should allow for more cell inhibition. This project will be using 5-Fluorouracil (5-FU), which is an antimetabolite that has been used to treat cancer for 40 years. It is a pyrimidine analogue that incorporates itself into RNA and DNA. Once 5-FU is incorporated, it stops the cell from replicating and eventually causes cell death. It works to stop cell growth in the S-phase.<sup>1</sup>

These two drugs stop cell growth in two different phases of cell replication. Hopefully combining them will produce synergistic effects. By doing this research, we hope to find a better treatment for pancreatic cancer and increase survivability. More research needs to occur with this cancer because the pathogenesis of this disease is quick and deadly.



### Statement of Central Objective

My objective in doing this research is to determine if the Isoprenoid Farnesol and 5-Fluorouracil are more effective at inhibiting cell growth together than either one alone.

### Methods

I will be using two different cell cultures, BX-PC3 and MIA PaCa2, to determine cell inhibition. Both of these cultures are from human pancreatic adenocarcinoma cell lines, and are available to purchase from the American Type Culture Collection (ATCC).

For each cell line there will be a control group, where no drug will be added. This will allow me to compare how much growth was inhibited by each of the medications. The first drug I will administer is Farnesol, which will show me what the cells look like when they are interrupted at the G<sub>0</sub>/G<sub>1</sub> stage of cell growth. I am going to use doses from 50-500 μM to determine the best concentration to inhibit cell growth. The next medication I will use is 5-Fluorouracil with a concentration range of 5-100 μM. This will allow me to determine what the cells look like when interrupted during the S phase of cell development. After measuring both the Farnesol and 5-FU, I will determine which dose of each was the most effective at inhibiting cell growth. With choosing which concentrations to use in combination, it is a good starting point to choose the doses with 40% inhibition, and then work my way up to see if I can get a maximum effect.

These cell lines are coming in very concentrated. I will have to detach the cells from their shipping container and plate them out, allowing them to reattach overnight in a less concentrated plate and allowing them to grow. Next, I will administer the drugs for 1-3 days and measure the cell growth, first by counting the cell numbers with a coulter counter and then with an MTT assay.

A coulter counter is a device that has microchannels between two electrolyte solutions with the cells in one of the chambers. When the cells go through the microchannels they change how much electrical resistance is being measured by the counter and the numbers begin to add up.<sup>4</sup> The other cell counting device will be the MTT Assay. This works by measuring how many living cells there are in each separate section by a varying purple color. The darker

the purple means that more cells are living because they are producing more enzymes to reduce the MTT and release the purple color.<sup>2</sup>

### **Progression of Project**

Spring 2012

This spring I am doing independent study for the Research Program for the COPHS. I will be learning proper techniques on cell culturing and administering medications.

#### BSI

Week 1: Begin cell cultures

Week 2: Administer Farnesol

Week 3: Administer 5-Fluorouracil and measure cell growth

Week 4: Administer Farnesol and 5 Fluorouracil and measure cell growth

Week 5: Measure Cell Growth

Week 6: Additional Experiments with more doses and time points

Week 7: Additional Experiments with more doses and time points

Week 8: Data Analysis/Conclusion

Week 9: Prepare presentation

### **Feasibility/Working on campus**

All equipment needed is in the Pharmacy research lab. I will be on campus over the summer.

### **Personal**

When I decided I wanted to become a Pharmacist, I knew I was interested in the research aspect that Butler could provide. I came into pharmacy school thinking eventually I would get a PhD and research. The thought of being able discover a new drug or do research to discover that next big milestone has always been present in my head. When I found out that Dr. Pamela Crowell does pancreatic cancer research, I knew I wanted to get involved. All of my grandparents have died of some cancer related disease and there are just so many possibilities to increase our knowledge on how to prevent and treat these diseases. I have just completed my business minor and the possibility of working with a drug company like Eli Lilly has always interested me. I think doing BSI and this research could better set me up in the future to get

into a graduate school or work for a drug company. I have also always been interested in working in a lab. I have taken microbiology with a lab, so I know how to work with cultures and basic techniques. Through courses such as Biotechnology I have learned of different lab equipment and methods of analyzing data to reach a conclusion. The only other person that will be working with me is my mentor Dr. Crowell, and she is going to teach me the techniques and how to use the equipment.

### **Presentation**

Upon completion of BSI I will submit to have my work presented in the Pharmacology Journal, Cancer Chemotherapy and Pharmacology Journal, or the Journal of Pharmacology and Experimental Therapeutics. Also, I plan to present at the Butler Undergraduate Research Conference the following year. Another goal would be to present at the annual American Association for Cancer Research (AACR) meeting.

### **Research Approvals**

N/A

### **References**

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