CALIFORNIA STATE SCIENCE FAIR
2006 PROJECT SUMMARY

Name(s)
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Project Number
S0417

Project Title
Snake Venom and Cancer: Production of Functional Disintegrin through Metalloproteinase

Abstract

Objectives/Goals
Contortrostatin (CN) is a protein found in the Southern Copperehad snake and it is known to contain disintergins that inhibit angiogenesis, thus blocking the cancerous cells from spreading and growing due to no food or transportation. The interesting things we noticed was when we purified the venom to separate the proteins, we also found a couple of fractions that reacted to the CN antibodies, so we hypothesize that there is a pro-protein containing a metalloproteinse and an disintegrin (CN). But the disintegrin is tested and found to be inactive, thus the goal of my experiement is to try to activate the metalloproteinase, in return it will cleave itself and release a functional disintegrin, doubling the yield of our protein (CN).

Methods/Materials
First, we purified our protein using HPLC. Then all fractions were tested for CN using AN anitbodies in Western Blots and ELISAs. Then a Platelet Aggregation Assay was performed to test for platelet inhibition, a function of CN. Then many more ELISA'a and Westerns followed in order to prove that the protein does contain CN because it is responsive to the CN antibodies. Lastly, we activated the pro-protein using APMA(Amino-phenyl mercuric acetate), and did a Fibrin Plate Assay to see if the metalloproteinase released a dunctional disintegrin by measuring the amount of lysis through an spectrophotometer.

Results
Our results indicate that the metalloproteinase did not release a functional disintegrin because the lysis in the wells was inconsistent, and our negative control failed because it was not suppose to have any fibrin eaten since it didn't have any activated protein, but our spectrophotometer showed lysis, thus making our results invalid.

Conclusions/Discussion
We conclude that the disintegrin is inactive and attached to the metalloproteinase, thus not doubling the yield. Possible errors include making the fibrin plates incorrectly or improper purification of the protein. Other types of method we are thinking about are zymograms to evaluate the activity of the disintegrin.

Summary Statement
My project is about the production of a disintegrin through activation of a metalloproteinase

Help Received
I recieved tremendous help from Fritz Costa and Dr. Steve Swenson at Cancer Research Laboratories located at USC Keck School of Medicine.