CALIFORNIA STATE SCIENCE FAIR
2013 PROJECT SUMMARY

Name(s)

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Project Number

33669

Project Title

Iron Based Catalytic Biosensor for Reactive Oxygen Species: Detection to Fighting Chemical Terrorism

Abstract

Objectives/Goals

My research is to study the effects of Reactive Oxygen Species(ROS) and Oxidative Stress that affect our environment and everyday life. I harnessed the power of the natural occuring enzyme- horseradish peroxidase(HRP) by extraction and chemical modifications and developed a novel biosensor to detect peroxide type ROS in aquatic as well as from chemical explosives.

Methods/Materials

I extracted HRP-Enzyme from=0Athe roots of the horseradish and modified it by chemical conjugation to=0Achromogenic substrate (Fluorescein) by NHS ester to promote the cell permeation in zebra fish cells (ZFL). I cultured ZFL cells for 3-4 days in media and treated with various amounts of copper and phenol (water contaminants) to investigate if they produce intracellular ROS, which can be measured by known H2DCF-DA substrate. I used HRP-Fluorescein as a=0Adetection reagent to detect intracellular ROS spectrometrically. I planned to detect peroxide type ROS that are byproducts of the decomposition of chemical explosives such as TATP (Triacetone Triperoxide). I adsorbed HRP and ABTS dye onto silica beads (H-A-S) and determined the structural morphology by Scanning Electron Microscope (SEM). Using solid acids such as Amberlyst 15, Nafion, and Citric acid to decompose TATP into peroxide, I developed a colorimetric sensitive test method to detect explosives. Then, I developed a prototype pen=0Adevice using my silica- HRP as biosensor to detect harmful peroxide-based explosives analogous to TATP.

Results

I discovered that copper (20mg/ml) and phenol(5mg/ml) produce concentration dependent intracellular ROS. My HRP-Fluorescein conjugate permeates ZFL cells and acts as an intracellular ROS detection reagent, while native HRP did not.

Using my Silica adsorbed HRP (H-A-S), I optimized conditions between solid acids and TATP and designed several prototype pen models to deliver a most effective one step device for chemical explosive detection.

Conclusions/Discussion

I have shown that a natural enzyme can perform multiple functions by chemical modifications which is shown by my HRP-Fluorescein and H-A-S.

I conclude that my HRP-Fluorescein is very efficient in detecting intracellular ROS using native HRP as control. My modified HRP had useful and unique properties that enabled to act as a biosensor for intracellular ROS.

My novel prototype pen device using HRP-ABTS-Silica that detects these peroxide based explosives has

Summary Statement

The purpose of this project is to devise a sensitive biosensor to detect Reactive Oxygen Species (ROS) from chemical explosives like TATP and by industrial wastes in our aquatic ecosystems by chemically modifying the natural enzyme HRP

Help Received

I worked in the department of chemistry at Amylin Pharmaceuticals under the supervision of Dr. Lawrence D#Souza-Sr. Scientific Investigator.