# **Environmental Sampling Research Report**

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#### **Introduction**

The environmental sampling research took place on a course of approximately 2-3 months within this timeframe students were taught and observed the geographic results of their soil. This experiment allowed students to collect solid from different geographic points and bring it back to the lab where over the weeks they would add different pesticides to observe how the bacteria degraded if it did so. As students captured their results whether the pesticide caused a reaction or not they were to collect pictures of their results to then report their final results. The final results determined if the cultures had growth meaning the bacteria was able to successfully degrade.

#### **Materials & Methods**

This experiment began with students collecting their soil from different geographical points along with recording or collecting the coordinates from where their soil was collected. For the first week students had to come in at a designated time to weigh and transfer 1mg of soil onto a test tube where the culture would then begin to be used for pesticides. Soil was then carefully added to the LB broth test tube then cap was loosely put back on. The test tube for week one is put to incubate under 30 celsius degrees at 200 rmp for one week. After the 1 week students are given 4 tubes to inoculate but due to shipment delays only 2 test tubes are able to inoculate, those are the pesticide paraxon (Px) in Carbon Selective Medium (CSM) and pesticide paraxon (Px) in Sulfur selective medium (SSM). They individually contained 4 mL of the pesticide in the test tubes, the MP pesticide is inoculated by the professor at this time for the following week. For

these test tubes 1 mL of the student soil is then added to the Paraxon in Carbon Selective medium and Paraxon Sulfur Selective medium. The cultures are to be capped and placed back on the rack for the next following week. Week 2 students are to collect their four corresponding test tubes then transfer the following pesticides add 200 microliters of CSM Px from week 1 into week 2 CSM Px, add 200 microliters of SSM Px from week 1 into week 2 SSM Px, add 200 microliters of CSM MP from week 1 into week 2 CSM MP, and add 200 microliters of SSM MP from week 1 into week 2 SSM MP test tubes. Once the student has finished transferring the pesticides into the cultures they will then place the cultures back onto the rack for the following week, and pictures were also taken of the pesticides from week 1. The same process of transfer of pesticides within the 4 cultures took place for weeks 3 and 4. All test tubes are to transfer 200 microliters of the corresponding test tube to the next following week pictures are taken for week 3 and week 4 results. For week 5 students are to add 200 ul of week 4 test tubes onto corresponding test tubes of week 5 the cultures are then to be incubated until monday. Students will meet Wednesday to interpret platelets and collect data. Week 5 cultures are incubated from wednesday to monday at 30 degrees celsius any test tubes that are bright yellow are inoculated into the LB broth. 1 mL of week 5 test tubes are taken and then added to 4 mL of LB. These test tubes are to be incubated overnight at 37 degrees celsius. Those that had growth are plated onto glycerol minimal media with pesticide. A minimal media made with glycerol had the solidifying agent agar added to it at 1.5% and is poured into a petri dish at 20-25 mL. The media was allowed to solidify overnight at room temperature. 100 microliters of a pesticide assay mix is added to the surface and spread evenly. This pesticide was allowed to sit for 2 hours at room temperature. Then any overnight LB cultures with growth were added to the surface at 100 microliters and spread evenly. The cultures grow at 37 degrees C overnight.

#### **Results**

Over the 5 weeks these cultures were transferred from one pesticide tube to another determining the reaction they developed after each week. Within these 5 weeks the cultures would have yellow tints and cloudiness formations. Pictures are taken to obtain the results from each week and compare with the final result. The colony growth is assessed the following week with the test tubes resulting in no MP cultures had the indicator p-nitrophenol turn yellow. This indicates that there are no MP degrading microorganisms. There are 20 Px bright yellow test tubes, Of those 20 only 17 had growth in LB. Students are then able to take pictures of their test tubes this will help them know if their test tube had growth or if their cultures did not result in microorganism degrading.

## **Discussion**

This environmental sampling research is an experiment to help students find different ways of degrading microorganisms from several geographical points using logical thinking, STEM activities, and communication skills. Students are able to collect soil from their own house or historic parks while recording their coordinates; this helps future studies know all the areas that contain degrading microorganisms and which ones are yet to be worked on. Within the 5 weeks students obtain data of their cultures resulting in LB growth and their result leading up to their final week before assessing their colonies. These results will be obtained and recorded on a geographical map along with experiments done before or future wise where students will learn the areas that pesticides were able to be used successfully.