CALIFORNIA STATE SCIENCE FAIR
2013 PROJECT SUMMARY

Name(s)
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Project Number
S1512

Project Title
Mutating E. coli with the arxA gene: Creating a Novel, Practical Solution to the Global Arsenic Water Problem

Abstract

Objectives/Goals

Arsenic, a poison found in water, exists in environment in mainly two states: arsenite (carcinogenic and water-soluble) and arsenate (easily removed from water), with arsenite being most predominant. Extremophilic bacteria like MLHE-1 have a gene called arxA, which enables them to change the toxic arsenite into less-toxic arsenate.

The goal of my project is to determine if E. coli strain K-12 can be transformed to contain the gene arxA from MLHE-1. If so, will the new strain recombinantly express the protein from the arsenite oxidase gene? I hypothesized that the E. coli strain K12 can be transformed to contain arxA, and when induced, K12 will express arxA. This mutated E. coli strain is an ideal choice for practical arsenic water bioremediation.

Methods/Materials

MLHE-1 DNA was extracted, and the arxA gene was amplified using PCR. Restriction digests were conducted, and the plasmid and the insert were ligated. E. coli underwent transformation via heat shock, and was plated onto LB-lac-ampicillin plates, and surviving colonies were subcultured. Recombinant protein expression analysis and an SDS-PAGE Gel was conducted.

Results

In my ligation gel, cut plasmid moved faster than the uncut plasmid, and the ligations moved faster than the uncut plasmid. After the transformation, E. coli cells grew on LB-lac-ampicillin plates, indicating insertion of the plasmid. In the SDS-Page Gel, I saw only 1-2 bands (not the expected barcode-like protein patterns) probably because the samples were too dilute, and this part of the research is a work in progress.

Conclusions/Discussion

The first part of my hypothesis was supported. As seen by my ligation and transformation results, the plasmid with the arxA gene was inserted into E. coli. The second part of my hypothesis is currently inconclusive and a work in progress, since the sample run in the SDS Page Gel is too dilute. The next step is to concentrate the sample and conduct the gel again. If the E. coli expresses the protein, the induced sample should have one more band than the uninduced sample, representing the arxA gene.

Summary Statement

This research focused on creating a new strain of E. coli with a gene called arxA, that converts arsenite (toxic & hard to remove from water) to arsenate (easy to remove from water), and opens up a possible way to remove arsenic from water.

Help Received

Used lab equipment at Schmahl Science Workshop under the supervision of Dr. Aru Hill.