

CALIFORNIA STATE SCIENCE FAIR 2003 PROJECT SUMMARY

Name(s)

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Project Number

S1321

Project Title

The Feasibility of Transforming E. coli Utilizing High Voltage Electroporation

Objectives/Goals

Abstract

DNA can be inserted into cells to achieve desired characteristics previously unexpressed. It is challenging to insert a DNA sequence through the cell membrane. Furthermore, a researcher typically needs to transform (change the genetic makeup of) many cells at a time to have observable results. In this experiment, the two techniques of heat-shock (0°C-42°C) and a novel method of electroporation are compared to determine the feasibility of high voltage in transforming Escherichia coli. Electroporation disrupts the insulating matrix, forming aqueous pores by which DNA can enter. To test the global hypothesis that high voltage electroporation would be more efficient, both techniques are performed, using a Tesla coil to supply the needed high voltage.

Methods/Materials

The trait applied to the E. coli is the gene for making green flourecent protein (GFP), which glows under ultraviolet radiation. The Tesla method involves a heat-sink, which allows electrocution for longer periods of time without an increase in temperature. Time periods from 1-4 seconds as well as longer controls are observed. The E. coli cells are plated on Petri dishes (agar) with arabinose (a simple sugar) and ampicillin antibiotic. Arabinose switches on the operon that makes GFP. Under ultraviolet light, transformed cells supplied with arabinose will glow.

Regults

Outcomes supported the practicality of the novel take on electroporation. The hypothesis was supported by the data in that there was a greater amount of electrically-shocked cells than heat-shocked ones. A benefit of this new method is that Tesla Coil pulses were observed to negligibly increase the temperature of the bacterial suspension due to the water bath heat sink. In addition, a consistent, exponentially greater amount of satellite colonies emerged from the electrically shocked bacteria.

Conclusions/Discussion

Each bacterial suspension contained millions of cells and could not be counted, however the mass of the pGLO was known in advance. Hence, the efficiency could be related back to how much pGLO originally was in each suspension. The results indicated that electroporation is more effective in opening pores in the cell membrane than heat-shock. If more cells transform initially, more colonies can be grown and more of the desired trait harvested before the cells lose potency from lack of telomeres. Efficiency in transformation is crucial to biotechnology.

Summary Statement

A new method of high voltage electroporation is compared to heat shock in transforming (changing the genetic makeup of) E. coli.

Help Received

Thanks to Nancy Cannon for her advice and our parents for their money.