CALIFORNIA STATE SCIENCE FAIR
2013 PROJECT SUMMARY

Name(s)

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Project Number

S0503

Project Title

A New Genetic Transformation Method: Agrobacterium and CowpeaChloric Mosaic Virus in a Replication Independent Matter

Abstract

Objectives/Goals

The purpose of this project was to develop a new cheaper, efficient, and effective way of introducing recombinant DNA into viral cells. In this project, Cowpea Chloric Mottle Virus (CCMV) was used to create a new transformation method with Agrobacterium pCass4- Rz vector. It was initially hypothesized that the transformation will be successful and the infected Black Eyed Pea plant will then successfully express via basal replication/translation the CCMV virus.

Methods/Materials

To initiate elucidation upon the gene delivery system between Agrobacterium and the CCMV virus, amplification of 3 CCMV inserts and the pCass4-Rz vector through the Polymerase Chain Reaction (PCR) technique was done as well as purification of the products using the Qiagen Purification Kit. Digestion of both the insert and the pCass4-Rz Agrobacterium was simultaneously done and important conditions were also applied considering the different NaCI concentrations. Ligation between 3 pieces of DNA insert from the CCMV virus which was correspondent to the genome of the pCass4-Rz vector derived from Agrobacterium as well as transformation of the cells was also done following the infection of Black Eyed Pea plants via agroinfiltration.

Results

Transformation of the 3 CCMV inserts and pCass4-Rz vector was successful and expressed via basal replication/translation of the CCMV virus in a controlled environment using Black Eyed Pea Plants. Further studies were done using results from Gel Electrophoresis technique and DNA concentrations were reviewed using The Thermo Scientific NanoDrop# 1000 Spectrophotometer.

Conclusions/Discussion

Further experiments site- directed mutagenesis on the CCMV virus for amino acid point mutations. Through this, further understanding of how viruses work and how they can be genetically mutated efficiently and effectively will be studied. Developing this method in Agrobacterium may prove to be the breakthrough needed to successfully insert foreign DNA into genomes of other organisms in order to try to apply such favorable traits such as disease resistant genes.

Summary Statement

A new genetic transformation method by using a vector from a bacterial cell (pCass4-Rz vector from Agrobacterium tumefaciens) to control a viral cell (Cowpea Chloric Mosaic Virus) to express specific genes.

Help Received

Devin Brandt from the Department of Chemistry and Biochemistry of UCLA helped me practice many of the laboratory techniques as well as helped me design the protocol correctly using the correct measurements.