



**CALIFORNIA STATE SCIENCE FAIR  
2016 PROJECT SUMMARY**

<b>Name(s)</b> <p align="center"><b>Geneva D. Trovato</b></p>	<b>Project Number</b>  <p align="right">36459</p>
<b>Project Title</b> <p align="center"><b>Effects of CaCl<sub>2</sub> Temperature in the Creation of Competent Host Cells: A Study in Genetic Engineering of E. coli</b></p>	
<p align="center"><b>Abstract</b></p> <p><b>Objectives/Goals</b>  I wanted to find out if protein production could be maximized by changing part of the transformation process. Transformation is the process of inserting a foreign piece of DNA that makes it produce a protein of interest. This experiment looked at the individual steps of transformation and considered the easiest way to maximize the result: a host cell that is producing the greatest volume of protein. Temperature of CaCl<sub>2</sub> was the focus of this experiment. The results revealed that more transformed colonies grew from the 42°C CaCl<sub>2</sub> than any other CaCl<sub>2</sub> temperature.</p> <p><b>Methods/Materials</b>  I used BactoBeads (attenuated E. coli), plasmids (with +amy gene inserted for screening), CaCl<sub>2</sub> (at three temperatures), agar plates (poured myself), pipettes and tips for transfer of cells. Standard Transformation protocols were used with one exception: CaCl<sub>2</sub> temperature was a variable.</p> <p><b>Results</b>  After transforming the E. coli cells, at the various CaCl<sub>2</sub> temperatures, the number of colonies formed (CFUs) were counted and recorded in a table. The two control plates were as expected and were an indication that the conditions were held constant and in working order. The three plates exposed to the E. coli cells, made competent with the 42°C CaCl<sub>2</sub>, had 30, 84 and 25 total CFUs. The 4°C CaCl<sub>2</sub>, had 22, 17 and 25 total CFUs. And, the 27°C CaCl<sub>2</sub>, had 15, 31 and 18 total CFUs.</p> <p><b>Conclusions/Discussion</b>  Transformation is of critical importance in genetic engineering. It includes the selection, propagation, expression and purification of a particular gene, such as INS-the gene that codes for Insulin. By examining the number of colonies formed, one can calculate transformation efficiency. In this investigation, the temperature of CaCl<sub>2</sub> at cold (4°C), room temperature (27°C) and hot (42°C), affected the competence of E. coli cells when transformed with plasmids. It was determined that the CaCl<sub>2</sub> temperature most likely to produce the highest transformation efficiency, was 42°C.</p>	
<b>Summary Statement</b> Effects of CaCl <sub>2</sub> Temperature in the Creation of Competent Host Cells: A Study in Genetic Engineering of E. coli showing that Hot CaCl <sub>2</sub> produces the most competent cells.	
<b>Help Received</b> 1. Dr. Nick Webster, UCSD, Assistance with brainstorming, ideas, and procedure. He also provided many of the tools necessary. 2. Kathy Blakemore, EMS Science Teacher, scientific inspiration. 3. Irene Trovato, ThermoFisher Scientific (& Mom), guidance and scientific discussions 4. Ed Trovato, Scientist,	