



**CALIFORNIA STATE SCIENCE FAIR
2003 PROJECT SUMMARY**

Name(s) Max R. Biessmann	Project Number S0403
Project Title Expression of Mosquito Odorant Binding Protein-8	
Abstract Objectives/Goals My primary goal in this project was to successfully synthesize mosquito odorant binding protein-8 in E.coli bacteria. In order to accomplish this I had to first create a working vector which had a copy of the gene in it and to transform it into E.coli bacteria. Methods/Materials I amplified the coding region from the OBP-8 gene by polymerase chain reaction (PCR) and sequenced it to make sure it was correct. After purification, the PCR fragment was inserted into the pGEM-T-easy vector and grown in E.coli. From this plasmid, the insert was cut with EcoRI and XbaI and ligated into the ThioHis plasmid. This construct was transformed into E.coli cells. Two transformed lines were grown in LB/amp, and transcription from the plasmid was induced by turning on the lac promoter with IPTG. After every hour I took a 1 ml sample and separated the proteins on a polyacrylamide gel. The materials I used were: cDNA for the A. gambiae OBP-8, ThioHis A plasmid, E.coli bacteria, primers, restriction enzymes, Taq polymerase. Results A PCR fragment from OBP-8 was cloned into the expression vector pThioHis. I isolated several clones, checked the plasmids for size on agarose gels and verified that they had the insert by cutting with EcoRI and XbaI. Using clone #7E in SURE cells and clone #33E in XL1-blue cells, I was able to produce the mosquito OBP-8 in E.coli. This was shown by polyacrylamide gel electrophoresis, where a protein the same size as OBP-8 appeared as a band on the gel. Conclusions/Discussion I used various molecular biology techniques to engineer a plasmid, from which the mosquito odorant binding protein-8 could be produced in bacteria. This is a very important step for further study of the function of odorant binding proteins in insect antennae. These proteins are believed to play an important role in odor recognition in insects. Being able to produce a mosquito protein in E.coli will enable future researchers to determine which odor molecule they may bind. Mosquitoes are vectors of many diseases, for example malaria. By studying the odorant binding proteins we may be able to produce better insect repellents thus reducing the number of mosquito bites and helping prevent mosquito borne diseases.	
Summary Statement Production of odorant binding protein-8 in E.coli bacteria.	
Help Received Used lab equipment at UCI under the supervision of Dr. Daniel Woods and Dr. Harald Biessmann	