

### **“A Quantitative and Highly Sensitive Luciferase-based Assay for Bacterial Toxins that Inhibit Protein Synthesis”**

**Luyi Zhao, Parkway South High School, Manchester, MO – 2005-06 National Finalist**

Abstract: Bacterial toxins that inhibit protein synthesis include Escherichia coli shiga toxin, diphtheria toxin, and Pseudomonas exotoxin A. A highly sensitive and reproducible assay for susceptibility to these toxins is critical to characterizing how they interact with host cells and to developing inhibitors to toxin action. We describe here the development of a novel highly sensitive and reproducible assay for susceptibility to bacterial toxins that inhibit protein synthesis. The assay utilizes luciferase as a marker for de novo protein synthesis. Luciferase is expressed in cells through transduction with an adenoviral expression system. The assay can be adapted for use in 24, 96, and 384 well plates and was found to have significantly less variability than the conventional radioactive assay. The luciferase-assay's ability to screen for inhibitors to toxin action was validated by experiments with Vero-FS cells and a-StxB. Susceptibility of Vero, Hep2, Chang, A549, COS-1, and HeLa cells to three different toxins was also measured using the assay. This quantitative and highly sensitive luciferase-based assay will have broad applications in the study of bacterial toxins that inhibit protein synthesis.

### **“Characterizing the Properties of a Translational Regulator Expressed in Mouse Brain”**

*(Neural Science)*

**Yin Li, Stuyvesant High School, New York, NY – 2003-04 National Individual Winner**

Abstract: Neurons in the central nervous system exchange information with each other via synapses. Local synthesis of proteins at specific synapses is crucial for sustained enhancement of synaptic efficacy, the molecular correlate of learning and memory. This local protein synthesis is at least in part mediated by cytoplasmic polyadenylation element binding proteins (CPEBs), translational regulators, which are activated by kinases. Phosphorylation provides only a short lasting activation, yet ongoing protein synthesis is needed to maintain synaptic enhancement. A distinct mechanism must be involved in this maintenance of local protein synthesis. A possible clue to such a mechanism came from studies of a well-known class of yeast proteins that can undergo heritable and contagious conformational changes, which are self-sustaining—the yeast prions. Here we show, using several assays developed to characterize the classical yeast prions, that the mouse CPEB-3 has prion-like properties in yeast and in mammalian cells. Altogether, our data indicate the exciting possibility of a novel prion-based mechanism for maintaining ongoing CPEB activity and establishing long-term memory storage.

**Mentor:** Dr. Martin Theis