GENE EXPRESSION ANALYSIS USING MICROARRAYS OF PRION POSITIVE AND NEGATIVE YEAST STRAINS

Amanda Watkins[1]*, John Brothers II*[2]*, Yolande Tra*[3]*, and Irene Evans**[4]

[1]Biotechnology, Department of Biological Sciences, Rochester Institute of Technology, Rochester, NY 14623, fizzywater313@hotmail.com
[2]Bioinformatics, Department of Biological Sciences, Rochester Institute of Technology, Rochester, NY 14623, jfb4497@rit.edu
[3]Department of Mathematics and Statistics, Rochester Institute of Technology, Rochester, NY 14623, yvtsma@rit.edu
[4]Department of Biological Sciences, Rochester Institute of Technology, Rochester, NY 14623, imesbi@rit.edu

Gene expression in eukaryotes containing prion proteins (PrP) can be easily studied in the model organism, *Saccharomyces cerevisiae,* due to its relatively small and well-characterized genome. Experimentally shown to lack nucleic acids, these 'infectious' protein-only structures manage to convert the 'normal' form of a functioning protein into a different conformation. In the study of prion proteins found in yeast, two genetically identical strains of yeast that differ only in the conformation of Sup35, a prion-like yeast protein, are analyzed. In the [PSI+] strain, Sup35 assumes a prion conformation while in the [psi-] strain, the Sup35 gene product is in its non-prion conformation and functions as a translation release factor protein. The way in which conformational differences of the Sup35 gene product affect gene expression in yeast can be experimentally investigated using multiple DNA microarrays. DNA microarrays are powerful tools used to analyze gene expression in vivo because they allow simultaneous measurement of the level of transcription of all genes in the genome. Through use of the Array 50 Kit provided by Genisphere, both the [PSI+] and [psi-] cDNA isolates are tagged with a 3DNA capture reagent containing either Cy3 (green fluorescence) or Cy5 (red fluorescence) and hybridized to a glass array slide on which the entire yeast genome is printed. Statistically defined standards of data reproducibility are met by eliminating any non-biological factors through the refinement of the entire microarray procedure. Non-biological factors shown previously to interfere with data reproducibility include the oxidation and degradation of the Cy5 fluorescent label, the presence of high background and flares on array slides, light-sensitive reagents causing signal fading as well as the presence of contaminating DNA in isolated samples. The results of eight to twelve data sets will be discussed. Over and under expressed genes will be confirmed with the use of Real Time RT-PCR technology.