Evolution of Duplicated Isozyme Gene Expression Patterns

Adriane Berry, 2014

Enzymes play an integral role in the development and metabolic processes of all living organisms. In teleost fish, duplicate forms of enzyme genes were generated following the teleost whole genome duplication event that occurred 345 million years ago. In this study, the goal is to analyze the developmental expression patterns of four enzymatic proteins, which encompass paralogous isoforms encoded by independent loci, known as isozymes. Using the distantly related teleost fish Orzias latipes and Danio rerio, this study examines the Guanylate kinase (Guk), Triosephosphate isomerase (Tpi), Malate dehydrogenase (Mdh), and Phosphoglucose isomerase (Pgi) isozyme pairs. It has been hypothesized that the original enzymatic proteins were neutrally charged and ubiquitously expressed (Auld et al. 2012). After gene duplication, one copy became negatively charged, and it has been suggested that they are expressed primarily in the neural tissues of teleost fish (Auld et al. 2012). Using 24 and 48-hour post fertilization embryos, with RNA probes synthesized from the 3’ UTR of genomic sequences, this study qualitatively examines in situ hybridization expression patterns (Thisse et al. 2001). By analyzing tissue expression patterns, this study ultimately tests the hypothesis that the negatively-charged isoforms of these four enzymes are present strictly in neural tissues.

This summer, I was able to use genomic sequences from D. rerio’s Mdh1a/1b and Tpi1a/1b genes with cDNA to perform RT-PCR reactions that confirmed amplification of a desired gene segment. I then transformed these segments into a vector using a TOPO-cloning reaction. The products were purified via PCR Mini-Prep, and the four RNA probes were successfully synthesized. During my independent study in the fall, I will continue to explore the justification behind the development of a negative charge in these enzymes. Hopefully, the data collected will further my understanding of the evolution of genes following a duplication event.

Faculty Mentor: Bill Jackman

Funded by the IDeA Network of Biomedical Research Excellence Summer Fellowship

References:
