Use of a Microarray to Analyze Gene Expression Profiles of Acute Effects of Prochloraz on Fathead Minnows (*Pimephales promelas*)

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Microarray technology is a powerful tool to investigate the gene expression profiles for thousands of genes simultaneously. In recent years, microarrays have been used to characterize environmental pollutants and identify molecular mode(s) of action of chemicals including endocrine-disruptors. Prochloraz is a fungicide known to cause endocrine disruption through effects on the hypothalamic-pituitary-gonadal (HPG) axis. The key enzyme affected by prochloraz converts testosterone to 17β-estradiol is aromatase, CYP19. In this study, microarray analysis was used to examine gene expression profiles of the ovaries in reproductively active female fathead minnows (*Pimephales promelas*) exposed to prochloraz at concentrations of 0 or  $300 \,\mu\text{g/L}$ . The fish were exposed for a 24 hour time course study with sampling periods at 6, 12, and 24 hours. The fish were anesthetized and sampled at each time point collecting blood plasma, gonads, pituitary, and brain. A small subsample of the ovaries was used for the microarray analysis. The 15,000-gene oligonucleotide microarray designed for the fathead minnow, resulted in gene expression profiles where about 1,300 genes had either been up or down regulated. From these genes, 7 were further analyzed using quantitative real-time polymerase chain reaction (RT-PCR) to solidify the results of the microarray. The results of the quantitative RT-PCR for prostaglandin, oxysterol binding protein, fibroblast growth factor 8, cytochrome P450 4F, cdc42, Rcd1, and retinol dehydrogenase 8 were similar in trends to the microarray results; however, there were some disagreements in the capabilities of the methods to pick-up significant differences (p<0.05). In addition to the microarray analysis, the effects of prochloraz on the HPG axis was analyzed by quantitative RT-PCR, radioimmunoassay, and enzyme linked immuno-sorbent assay assays to determine steroid and gene expression levels important to the HPG axis. This study, will give insight to the early effects of inhibitory chemicals on the HPG axis as well as aid into the understanding of adverse effects on fish reproductive health to chemical stressors that can be determined by microarray analysis. This abstract does not necessarily reflect US EPA policy.