PROTEOMIC EXPRESSION PATTERNS IN FATHEAD MINNOWS EXPOSED TO TRENBOLONE AND FLUTAMIDE

Nancy D. Denslow[1], Christopher Martyniuk[2], Sophie Alvarez[3], Daniel L.Villeneuve[4], Gerald T. Ankley[4]

1. Department of Physiological Sciences and Center for Environmental and Human Toxicology, University of Florida, Gainesville, FL, USA
2. University of New Brunswick, St. John, NB, Canada
3. Donald Danforth Plant Science Center, St Louis, MO 63132, U.S.A.
4. U.S. EPA, ORD, NHEERL, MED, Duluth, MN, USA

Insights into androgen signaling in the liver of fathead minnow (*Pimephales promelas*) was obtained using non-gel based proteomics analysis. We exposed female fathead minnows for 48 hr through the water to a prototypical androgen (17β-trenbolone, 5 μg/L), a prototypical anti-androgen, flutamide (500 ug/L) and a mixture of the two at these concentrations. The concentrations chosen had been previously shown to reduce plasma hormones and ovulation in females therefore affecting reproductive physiology. Proteomics was performed by LC MS/MS on an Applied Biosystems QSTAR instrument using iTRAQ isobaric tags to quantify changes in protein expression. Over three hundred proteins were identified in the fathead minnow liver encompassing a wide variety of molecular functions from transcription regulation to catalytic activity and up to structural cellular function. While flutamide appeared to influence the changes more dramatically than trenbolone, there were several proteins that were altered reciprocally by trenbolone and flutamide suggesting that they may be good candidates for protein biomarkers that are regulated directly through the androgen receptor. Among the proteins directly regulated were phosphoglycerate mutase 1(PGAM1), ferritin heavy chain (FTH1), leucine amino peptidase (LAP3), betaine homocystein S-methyl transferase (BHMT), ubiquitin C (UBC), SMT3 suppressor of mif two (SMT3H1), SET nuclear oncogene (SET) and glutathione S-transferase theta 1 (GSTT1). These and others had roles in growth, cell differentiation, catabolism and secretion of proteins. Information garnered through proteomics was complementary to experiments performed previously using microarray analysis in the same tissues.