

## **Social status modulates gene expression and metabolite profiles in the fathead minnow males.**

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The fathead minnow (FHM) is a valuable small fish model for genomic research in ecotoxicology. Our recent studies have successfully used genomic and metabolomic analyses to evaluate responses to endocrine disrupting compounds (EDCs) in urine of the FHM, but these results indicated an occurrence of substantial individual variation. To improve use of FHM for genomic and metabolomic aspects of EDC research, we investigated whether one source of this variability could be male dominance status. Specifically, we hypothesized that social status may modulate endocrine-dependent processes (e.g., expression of androgen-dependent secondary sex characteristics (SSCs)), expression of genes involved in regulation of immune system, behavior and reproduction, as well as urine abundance and endogenous metabolite composition. To test this, we paired two males with two females and one spawning substrate (for which males competed) and examined their behavior, SSCs and stored urine volume prior to and after territory acquisition. We also measured plasma androgen concentrations, gene expression patterns in the brains (using a 22,000 gene microarray specific for FHM), and conducted NMR-based metabolomic analyses of urine once the territories/and social hierarchies were established. Our results show that circulating androgens, expression of SSCs, and urine abundance increase with territory acquisition and either remain unchanged or decrease in non-territorial, subordinate individuals. Metabolomic analysis of the urine samples clearly distinguished the metabolite profiles of territorial and non-territorial males. Some of the affected metabolites included taurine, creatine, trimethylamine n-oxide, lactate and choline. Differential release of taurine, a strong chemosensory attractant which modulates both aggressive behavior and osmoregulation (and possibly urine release), suggests that urine-borne taurine may serve as an “honest” signal of social status in males. Social status affected expression of multiple genes (*circa* 400) involved in immune and stress responses, steroid synthesis and signaling, reproduction and behavior. In conclusion, we demonstrated that social status significantly contributes to variability in gene expression and metabolite composition in male FHM thereby providing important baseline data for interpretation of the effects of EDCs on fish. *This work may not necessarily reflect official U.S. EPA policy.*