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Estrogen in the Water: Impacts of Sewage Wastewater on Feminization and Vitellogenin Expression in Male Fathead Minnows *(Pimephales promelas)*

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Estrogenic compounds (primarily from substances like birth control drugs) are commonly found in domestic wastewater effluent. These compounds can feminize male fish (e.g., decrease male secondary sex characteristics), reducing competitive advantage during spawning. Exposure to estrogenic chemicals can also lead to the production of female-specific proteins such as vitellogenin (VTG). VTG is a yolk-precursor protein synthesized by the liver of egg-laying females after the stimuli of estrogen. We hypothesize that upon exposure to estrogen-containing wastewater, adult male fathead minnows (*Pimephales promelas*) will express this female-specific protein. Adult males were caged at two different sites in the West Fork of the White River. The downstream group was placed directly below the outflow from the Muncie Water Pollution Control Facility (MWPCF), while another group (upstream) was placed 0.25 km upstream from MWPCF. Following a two-week exposure, secondary sex characteristics were examined, and livers were processed through quantitative polymerasechain reaction (qPCR) to determine expression of VTG. While no significant differences resulted from comparison of secondary sex characteristics between the study groups, downstream males showed a VTG up-regulation of ~ 14-fold (SD = 2.4) when compared to the control group. These results are in agreement with a previous study in this same area that found "feminization" of native populations of bluntnose minnows (*Pimephales notatus*). The "estrogenic" compounds that elicited this response remain unknown.

Research advisor Maria Sepulveda says, "Pharmaceuticals and personal care products are an emerging class of environmental pollutants, having been detected in streams and rivers downstream from sewage wastewater treatment plants. We know very little about their effects on fish."

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