AROMATASE ACTIVITY IN THE OVARY OF MOSQUITOFISH *GAMBUSIA HOLBROOKI*, COLLECTED FROM THE FENHOLLOWAY AND ECONFINA RIVERS, FLORIDA

Edward F. Orlando[1], William P. Davis[2], Lisa Caltabiano[1], Danielle Bass[1], and Louis J. Guillette, Jr.[1]

[1]Department of Zoology
University of Florida, Gainesville 32611
Tele: 352+392-1098
Email: efo@zoo.ufl.edu

[2]EPA – Gulf Ecology Division
1 Sabine Island Drive
Gulf Breeze, Florida 32561

Recent studies have provided a greater awareness of the adverse effects of environmental contaminants, including their ability to alter the normal development and reproduction of wildlife species by modifying the endocrine system. Female mosquitofish living downstream of a paper mill plant located on the Fenholloway River, Florida have masculinized secondary sex characteristics. This masculinization includes altered anal fin development and reproductive behavior. Prior field and laboratory studies suggested the presence of an androgen in the effluent, possibly derived from the microbial conversion of phytosterols. Induction of male secondary sex characteristics can be caused by exposure to androgens in the water or from an alteration in steroidogenic enzyme activity effecting an internal androgenic milieu in the fish. We tested the hypothesis that component(s) of the effluent are inhibiting the activity of aromatase, the steroidogenic enzyme that converts androgens (A) to estrogens (E). Inhibition of aromatase would cause a decrease in estrogens and a concomitant increase in androgens, which could alter the normal hormonal profile of a female (decreasing the E/A ratio) to that resembling a male, thereby masculinizing its secondary sex characteristics. We predict aromatase activity will be lower in the ovaries from the Fenholloway females compared to mosquitofish obtained from the reference site, the Econfina River. This hypothesis would explain the observed phenomenon, and at least one phytosterol, enterolactone, has been shown to decrease aromatase enzyme activity. We collected 50 (June 1999) and 36 (August 1999) free-ranging, adult, female mosquitofish from both the Fenholloway and Econfina Rivers. Both sites were worked on the same day. Morphometric data were taken during the June and August collections, whereas aromatase activity was measured from the August collection exclusively. Fish were transported to the lab and transferred to glass aquaria containing fresh water from their respective rivers. Over the following three days, equal numbers of fish from each site were anesthetized daily and euthanized by decapitation. Standard morphometric data were obtained, as well as anal fin length and segment number of the longest anal fin ray. Gonads were extracted, weighed, and staged prior to being cultured for aromatase activity and sex-steroid hormone production. Gambusia from the contaminated site exhibited higher aromatase activity compared to fish from the reference site. Contaminated-site fish also had greater numbers of segments on their anal fin, whereas no site difference was found in anal fin length. From the fish in the present study, ongoing collaborations with other labs will provide two additional endpoints relevant to the ovarian aromatase activity data. These include sex-steroid hormone data from the *in vitro culture* of the ovaries, and somatic tissue concentrations of sex-steroid hormones.