

**Immunohistochemical detection of vitellogenin (Vg) in formalin-fixed livers of *Lepomis macrochirus*.**

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Vitellogenin (Vg) in male fish has become a popular biomarker of estrogenic contamination. Exposure to natural or synthetic estrogens can adversely affect human health, particularly involving the reproductive system; recently, prolonged exposure to estrogenic contaminants has been linked to reproductive cancers in humans (Jobling et al 1995). Because duration of exposure to estrogens is an important aspect in assessing its effects, historical monitoring of exposure to estrogenic contaminants in formalin-fixed archived specimens may prove useful. This study develops an immunohistochemical protocol to detect Vg as an indication of estrogenic contamination in formalin-fixed livers. *Lepomis macrochirus* were exposed to estrogen in the lab, and were also captured in the field at Elevenmile Creek in Florida, where papermill effluents are discharged; Vg was detected in blood serum by western blotting. The fish were fixed in formalin, their livers embedded in paraffin, sectioned, and placed on coated slides. Immunostaining was performed using a *L. macrochirus* Vg antibody; immunoreactivity was detected by an immunoperoxidase method after antigen retrieval. Vg was detected, making this protocol an essential key in historical monitoring that may unlock the question of long-term effects of estrogenic exposure.