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Development of an Amphibian Metamorphosis Model for Detecting Thyroid Axis Disruption

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Metamorphosis in *Xenopus laevis* represents an elaborate process of post-embryonic development which is thyroid hormone (TH) dependent. The development of a functional thyroid axis and the responses of tissues to different TH concentrations are well defined in this species, providing the rationale for the concept that amphibian metamorphosis could be used as a model system to screen chemicals for TH disruption. The most likely type of TH disruption is inhibition, since environmental thyroid agonists are apparently rare. Several chemicals have been shown to inhibit metamorphosis in anurans, but most studies have not been conducted to establish dose response data, nor has there been an effort to determine the relative sensitivity of different developmental stages to the effects of thyroid inhibitors. Based on this, we have conducted experiments comparing the sensitivity of pre-metamorphic (stage 51) and pro-metamorphic (stage 54) larvae to the model thyroid axis antagonists methimazole (control, 6.25, 12.5, 25, 50, 100 mg/L), 6-propylthiouracil (control, 1.25, 2.5, 5, 10, and 20 mg/L), and perchlorate (control, 15.6, 62.5, 250, 1000, 4000 ug/l). Larvae were exposed for a 2 wk period, and developmental stage and thyroid histology were examined at 1 and 2 wks post exposure. Methimazole, 6-propylthiouracil, and perchlorate, which are all thyroid hormone synthesis inhibitors, caused a concentration-dependent inhibition of metamorphic development. Further, these three compounds caused concentration-dependent changes in thyroid gland morphology. These changes were characterized as reduced colloid, glandular hypertrophy, and cellular hyperplasia. As determined by an analysis of these endpoints, there were only minor differences in the sensitivity between the 2 stages examined. These results indicate that larvae in early stages of metamorphosis are sensitive to thyroid axis inhibition and that development of a short term amphibian-based thyroid screening assay is feasible. Although this approach is feasible, it still does not provide diagnostic data indicating the specific mode of action of a chemical, which is needed to provide the basis for interspecies extrapolation. Based on this uncertainty, we are developing an approach to use multiple gene expression and the biochemical status of the thyroid axis as a means to indicate the specific mechanisms affected. Gene expression and biochemical data will be evaluated for diagnostic patterns related to the mechanism of each chemical and the sensitivity of these endpoints will be evaluated in the context of apical effects. This abstract does not necessarily reflect EPA policy.