Negative feedback control of pituitary thyroid-stimulating hormone synthesis and secretion by thyroid hormones during metamorphosis in *Xenopus laevis*

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A basic understanding of the endocrinology of the hypothalamic-pituitary-thyroid (HPT) axis of anuran larvae is necessary for predicting the consequences of HPT perturbation by thyroid-disrupting chemicals (TDCs) on the whole organism. This project examined negative feedback control of pituitary thyroid-stimulating hormone (TSH) synthesis and secretion by thyroid hormones in *Xenopus laevis* tadpoles. Plasma thyroid hormone (TH) concentrations in anuran larvae rise rapidly during metamorphosis. Such a rise in an adult anuran would inevitably trigger a negative feedback response resulting in decreased synthesis and secretion of TSH by the pituitary. However, pituitary TSH mRNA expression also increases during metamorphosis in concert with plasma TH concentrations. If negative feedback by THs on the pituitary is operative during metamorphosis, how TSH production and circulating TH concentrations rise concomitantly during this transformative process is still unclear. We used *ex vivo* and *in vivo* experiments to examine the hypothesis that the set-point for negative feedback on pituitary TSH synthesis and secretion by THs changes during metamorphosis to allow for this seemingly paradoxical, concurrent increase in TH concentrations and TSH synthesis. Pituitaries from *X. laevis* tadpoles were cultured in media containing increasing, physiologically-relevant concentrations of thyroxine (T4) or triiodothyronine (T3). Media and pituitaries were collected and analyzed for TSH mRNA expression, intracellular TSH protein, and TSH protein secretion. Results indicated that pituitaries from pre- and prometamorphic tadpoles were more sensitive to TH-induced inhibition of TSH synthesis and release than pituitaries from climatic tadpoles. The observed decrease in sensitivity of pituitary TSH mRNA expression to negative feedback by THs from premetamophosis to metamorphic climax was confirmed by *in vivo* experiments in which tadpoles were reared in water containing T4 or T3. Collectively, this *ex vivo* and *in vivo* data provide support for the hypothesis that the set-point for negative feedback control of TSH synthesis and secretion by THs increases as metamorphosis progresses allowing for the concomitant rise in TSH synthesis and plasma concentrations of THs. This abstract does not necessarily reflect EPA policy.

Impact statement:

The Endocrine Disruptor Screening Program (EDSP) includes an amphibian metamorphosis assay as part of the proposed Tier I screening battery. This research examines compensatory mechanisms of the HPT axis of anuran larvae during the stages of TH-dependent metamorphosis by establishing the details of the regulation of TSH release from the pituitary as a function of circulating TH levels. The results of this effort increase our basic understanding of the endocrinology of the HPT axis of anuran larvae. In addition, the results could be incorporated into an HPT systems model to serve as the framework for predicting the consequences of HPT perturbation by endocrine-disrupting chemicals on the whole organism.