

## EVALUATION OF VITAMIN A STATUS AND DIAGNOSIS OF HYPOVITAMINOSIS A IN AMPHIBIANS

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### Abstract

Without supplementation, insect-based diets for amphibians and reptiles are known to be deficient in nutrients such as calcium and vitamin A (Livingston et al., 2014). In the last decade, hypovitaminosis A has been recognized as a limiting factor for amphibian conservation programs that must successfully maintain and breed endangered species for ex situ rescue, survival assurance, and reintroduction programs (Pessier, 2014; Rodriguez and Pessier, 2014). Empirical treatments, new approaches to dietary supplementation of insects (especially carotenoids and techniques for more effective gut-loading), and experimental studies have been useful for improving our understanding of this condition. (Brenes-Solo and Dierenfeld, 2014; Dugas et al., 2013; Livingston et al., 2014; Pessier, 2014). However, research and clinical management have been hampered by practical considerations (e.g., it is difficult to get sufficient samples from very small frogs), limited basic information on vitamin A metabolism in amphibians, and a lack of standardization in diagnostic methods (Glugston and Blaner, 2014; Rodriguez and Pessier, 2014). A presumptive diagnosis of hypovitaminosis A in amphibians will often be made by a pathologist observing squamous metaplasia (SM) in a normally mucus-producing or ciliated epithelium (Pessier, 2014; Rodriguez and Pessier, 2014). Although the tongue is the most common anatomic site for SM (i.e., short tongue syndrome), it is also recognized in the oropharynx, esophagus, ureter, reproductive tract, and cloaca. It is important to note that SM is not observed in every vitamin A-deficient animal, nor is it consistently observed in every anatomic site (Rodriguez and Pessier, 2014). Collection and histologic examination of a range of different tissues is suggested for every amphibian necropsy. Measurement of vitamin A levels in serum or liver is encouraged to confirm a diagnosis of hypovitaminosis A, but there are important pitfalls including awareness of what is measured and reported by different laboratories (retinol+retinyl esters or simply retinol) and the need for proper collection of samples (autolysis and light exposure influences results; Glugston and Blaner, 2014; Rodriguez and Pessier, 2014). Interpretation of vitamin A levels can be frustrating because of a lack of validated reference ranges in amphibians, wide variation in “normal” upper levels between individuals and species, and physiologic maintenance of serum retinol levels until deficiencies are advanced (serum retinol is not linear in relation to deficiency; Berkvens et al., 2014; Glugston and Blaner, 2014; Rodriguez and Pessier, 2014; Sullivan et al., 2014). However, low vitamin A levels (e.g., < 5-10 µg/g retinol in liver) should always raise suspicion of deficiency.

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