

Thyroxine Testing (T4) at Phoenix Laboratory

Phoenix Lab recently discontinued T4 testing by radioimmunoassay (RIA) in favor of enzyme immunoassay (EIA). The T4 RIA has long been the gold standard but in recent years, testing by chemiluminescence and enzyme immunoassay has been validated and used in place of T4 RIA. There has been a move away from RIA testing due to regulatory licensing requirements, reduction in the availability of reliable RIA reagents, and the management of radioactive waste. Phoenix, until recently, ran Total T4 by enzyme immunoassay, repeating the assay by RIA if T4 values ran very low or very high.

But assays change over time. Manufacturers change, and manufacturing processes change. In early 2017, it was determined through a joint effort by Phoenix Lab and the Feline Hyperthyroid Center, that T4 values by RIA seemed to be running high. Phoenix Lab performed several side by side assays, running samples by RIA, enzyme immunoassay (chemistry) and chemiluminescence, and determined that the RIA was indeed running high...by 0.2 to 0.8 ug/dl. Based on these findings, Phoenix Lab elected to discontinue running T4 by RIA and instead use the enzyme immunoassay. The transition from one assay to the other was done over roughly a month, with results by both assays being reported to give veterinarians a sense of the discrepancy, before the T4 RIA was discontinued. In a publication in Spring 2017, the Feline Hyperthyroid Center discussed these findings, supported the discontinuing of T4 by RIA, and supported the use of cTSH in diagnosing occult feline hyperthyroidism (see.....).

Soon thereafter, Free T4 by equilibrium dialysis (ED), also a RIA assay, was discontinued. Free T4 (ED) had not been consistently helpful in diagnosis of either feline hyperthyroidism or canine hypothyroidism. Free T4 by chemiluminescence is analyzed in-house at Phoenix Laboratory.

Phoenix Lab is dedicated to accurate diagnostic testing *AND* a sensitivity to the environment. Moving forward to more accurate testing, and also toward environmentally sensitive testing, was important to Phoenix and to the clients we serve.

When Phoenix Lab carefully evaluated T4 testing by enzyme immunoassay, it was determined that values less than or equal to 0.6 ug/mL could not be reliably determined...the linearity of the assay (the range over which lab results are accurate in layman's terms) **does not provide consistently reproducible results. As such, concentrations of thyroxine below 0.6 µg/mL lie outside of the assay reportable range.** In order to assure our clients of the accuracy of these lower range T4s, values less than 0.6 ug/dL are verified by reanalysis. This is noted on the final report.

Thyroid Interpretation

Hypothyroidism in the Dog

Opinions vary, but many feel that hypothyroidism is over diagnosed. General blood work, including CBC and biochemistry panel, often include a total T4 determination. A low total T4 value on a general diagnostic screen could be consistent with hypothyroidism, but may also reflect nonthyroidal disease. Sometimes it does not appear that either clinical hypothyroidism or nonthyroidal disease is present in a patient. What do we do in that case?

A low total T4 on a panel could be consistent with hypothyroidism. Other findings on the panel that would suggest hypothyroidism are a mild anemia and/or hypercholesterolemia. Mild anemia, in the mid to low 30's, can be seen in up to 30% of dogs with hypothyroidism. Fasting hypercholesterolemia is seen

in roughly 75% of hypothyroid dogs. Furthermore, the patient should show clinical signs of hypothyroidism. These include poor skin and hair coat, weight gain, lethargy, recurrent otitis, etc. It is felt by many that hypothyroidism is a clinical disease and not a laboratory finding. There should be a reason to treat the patient for hypothyroidism other than a low T4.

Confirmatory laboratory testing of hypothyroidism can take several routes. Thyroid stimulating hormone (TSH) is elevated in roughly 75% of dogs with hypothyroidism, but there are false negatives and dogs with nonthyroidal disease can have an elevated TSH value. Free T4 testing can be done; a normal Free T4 and low total T4 would suggest that nonthyroidal disease and not true hypothyroidism is present. Both total and Free T4 can be decreased with nonthyroidal disease. Panels with Free T4 and TSH can be run. A low Free T4 and high TSH would be expected with hypothyroidism. Dogs on anticonvulsant therapy can have a low Total T4, Free T4 and elevated TSH: very confusing. *Again, the key: do clinical signs suggest hypothyroidism?*

A thyroid supplementation trial can be done, monitoring monthly to make sure the Total T4 value does not go high. If the dog is hypothyroid, a true clinical improvement of signs consistent with hypothyroidism should be seen within two to three months. The change should be significant not a little better; in the latter scenario, the dog is likely not hypothyroid. Keeping a dog on thyroid supplementation for a prolonged period of time could render the patient reliant on supplementation as endogenous production of Total T4 is shut off.

Hyperthyroidism in Cats

Finding a total T4 in the older cat above the normal range is fairly self-explanatory. However, in the grey area of 2.5 mg/dL and above (some folks even say 2.3 mg/dL and above) occult hyperthyroidism can be present. It is thought that these cats may have some sort of concurrent disease present, often renal disease, which brings what might be expected to be a high total T4 into the high normal range.

Free T4 (ED) was often performed as a confirmatory test to the high normal Total T4. But the assay, as noted above, has changed over the years and recent experience is not finding this test helpful, it is not high. Currently, the recommendation is to use the canine TSH assay to confirm occult hyperthyroidism. An undetectable value for cTSH in the cat (<0.03ug/dl) supports hyperthyroidism. A detectable value can occasionally be seen in cats with occult hyperthyroidism. In those cats, monitoring the Total T4 for an increase might be necessary.

Summary

Low Total T4: Dog

- Clinical signs consistent with hypothyroidism
- Mild anemia and/or hypercholesterolemia
- Confirmatory testing: high cTSH (+/- low Free T4)
- Thyroid supplementation trial (2-3 months)

High normal Total T4 in the Older Cat

- 2.5 (2.3) mg/dL to 4.4 (upper limit of normal range)
- Confirmatory testing: undetectable cTSH