

Thyroid Testing In Dogs

A Reference for Dog Breeders & Owners

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Hypothyroidism is a relatively common endocrine disease of dogs which results from progressive loss of functioning thyroid tissue. While epidemiological studies are few and somewhat conflicting, it appears that hypothyroidism occurs more frequently in certain breeds and lines, especially in large breed dogs. It is usually caused by lymphocytic thyroiditis (most likely an immune-mediated disease) or idiopathic thyroid atrophy (shrinking of thyroid tissue from unknown causes). The disease can occur in either sex and usually becomes apparent in young adults (1-3 years of age) or middle-aged dogs (4-9 years of age). Clinical signs may include hair loss, seborrhea, bacterial skin infections, excessive skin pigmentation, lethargy, obesity, muscle or nerve weakness, slow heart rate and reproductive disorders. Abnormal blood tests may reveal mild anemia and an increase in lipids, especially cholesterol.

Making a definitive diagnosis of hypothyroidism can be difficult because signs of hypothyroidism can be confused with other diseases. In addition, interpretation of diagnostic tests for hypothyroidism can be confusing and not specific for thyroid dysfunction. Also, there is variation within normal thyroid hormone concentrations which must be considered when evaluating any test results. Younger dogs will have higher thyroid hormone concentrations than older dogs. Certain breeds, such as the greyhound, normally have lower thyroid hormone concentrations. At this time, there is no ideal test, which is readily available and affordable, that is specific for diagnosing hypothyroidism.

The most commonly used thyroid tests are those that evaluate thyroid gland function by measuring thyroid hormone concentrations in the blood. The Total T4, Total T3, Free T4 and TSH concentration tests measure various hormone concentrations at rest. The TSH and TRH stimulation tests measure hormone concentrations after stimulation of the thyroid gland. Other available tests evaluate thyroid gland pathology and include the measurement of antibodies to thyroid hormones (T3 or T4) or thyroglobulin, as well as thyroid biopsy.

A much less common diagnostic procedure is the Radioactive Iodine Uptake (RAIU) study. RAIU studies do not accurately reflect thyroid gland function but simply determine the turnover of iodine in the thyroid gland. Results can be affected by the amount of iodide in the diet and non-thyroid illness. Because of these factors, as well as the expense, radiation exposure and the necessity for sedation, RAIU studies and radioisotope scans are not routinely used to diagnose hypothyroidism in dogs.

Hormone concentrations are usually measured by radioimmunoassay (RIA), although chemiluminescence, fluorescence polarization (TDX) and enzyme-linked immunosorbent (ELISA) assays are also being used. Regardless of the assay, it is important that the laboratory uses an assay that has been validated for use in dogs and that each laboratory establishes its own normal canine reference ranges. While most veterinary laboratories have done this, it is

less common in human reference laboratories. It is essential to verify this when ordering tests from a human laboratory.

TOTAL T3 CONCENTRATION

Total T3 concentration (TT3) is the amount of total (free plus protein-bound) tri-iodothyronine (liothyronine) measured in the blood. It is the most active form of thyroid hormone at the cellular level. However, it is not the major hormone made by the thyroid gland and is formed by conversion from T4 (or thyroxine) in the peripheral tissues of the body. Hypothyroid dogs will often have normal concentrations of TT3 in the blood. Some veterinarians propose that there can be a defect in the conversion of T4 to T3, thus causing hypothyroidism. This defect is rare in people and not well documented in dogs. Occasionally, hypothyroid dogs form antibodies to circulating T3 in the blood, due to immune-mediated disease. These dogs can show elevated concentrations of TT3 in the blood. Because of these facts, measurement of TT3 in the blood is not an accurate reflection of thyroid gland function.

TOTAL T4 CONCENTRATION

Total T4 concentration (TT4) is the amount of total (free plus protein-bound) thyroxine measured in the blood. It is the predominant hormone made by the thyroid gland. While it has some biological activity itself, its major purpose is to be converted to T3, the more active thyroid hormone, in the peripheral tissues of the body. The free portion of T4 is the active available form of the hormone, while the protein-bound portion serves as a reservoir for hormone in the circulating blood. The protein-bound portion of TT4 can be lowered by non-thyroid illnesses, other hormones (cortisol) and certain drugs. Drugs that can lower protein-bound T4 (and therefore TT4) include steroids (cortisone, prednisolone, prednisone, etc.), non-steroidal anti-inflammatory drugs (phenylbutazone and others), anticonvulsant or seizure drugs (Phenobarbital), certain potentiated sulfa drug antibiotics and potentially other drugs. Therefore, a dog with normal thyroid gland function could have a low TT4 due to lowering the protein-bound portion of TT4. It is very important that the veterinarian evaluating a dog for thyroid function know about all medications and any other medical conditions that the dog might be suffering from. Results of the measurement of TT4 can be misleading if one of the above mentioned conditions exist.

FREE T4 CONCENTRATION

Free T4 concentration is the amount of free or active thyroxine not bound to protein in the blood. As previously explained, protein-bound T4 can be lowered by non-thyroid illness and drugs. The free T4 portion is less likely to be affected by non-thyroid illness and drugs; therefore, it more accurately reflects true thyroid gland function. However, steroids (or a condition of excess internal cortisol called Cushing's Syndrome) can lower free T4 concentrations in some cases.

When measuring free T4 concentrations, it is important that the laboratory utilize a technique called equilibrium dialysis in performing the free T4 assay. Other "analog" techniques are not as useful in obtaining accurate results. Due to this equilibrium dialysis step which properly separates the free T4, this test is more expensive than other hormone assays. A formula incorporating the concentration of free T4 measured by equilibrium dialysis and cholesterol may be utilized to improve the diagnostic accuracy of a single blood sample.

TSH STIMULATION TEST

The current "gold standard" test for measuring thyroid gland function is called the TSH stimulation test. In this test, T4 concentrations in the blood are measured before and 6 hours after the administration of TSH (thyroid stimulating hormone or thyrotropin). TSH is the hormone normally produced by the pituitary gland which stimulates the thyroid

gland to produce hormone. If the T4 concentration in the blood sample taken 6 hours after giving the TSH is not significantly greater than that in the blood sample taken before the TSH is given, then this means that the thyroid gland is not able to produce adequate thyroid hormone (primary hypothyroidism). Unfortunately, the utility of this test is limited by the availability and cost of TSH. The veterinary TSH product is currently not available and the human TSH product is difficult to find. Costs of TSH range from \$160.00 to \$220.00 for 10 units of TSH (1-5 units needed per test).

TRH STIMULATION TEST

The TRH (thyrotropin releasing hormone stimulation test) is another test for measuring T4 before and after thyroid gland stimulation. TRH is produced by the hypothalamus to stimulate the pituitary gland to produce TSH which in turn stimulates the thyroid gland to produce T4. The TRH stimulation test is not as good a test as the TSH stimulation test because the amount of increase in T4 after administration is not as great in normal dogs as with TSH, so it makes it harder to distinguish normal from subnormal response. Also side effects (salivation, vomiting, diarrhea, panting and rapid heart rate) are frequent when TRH is given. The cost of TRH is not as great as TSH but it is also difficult to find.

TSH CONCENTRATION

The direct measurement of TSH (thyroid stimulating hormone or thyrotropin) concentration in the blood is the test of choice to evaluate thyroid gland function in people. With primary hypothyroidism (the thyroid gland is the sight of dysfunction), the pituitary gland produces extra TSH to try to additionally stimulate the low functioning thyroid gland. Therefore, the measurement of high concentrations of TSH in a patient with low T4 and signs of hypothyroidism, would indicate that the patient has primary hypothyroidism. (Dogs with secondary hypothyroidism, where the problem is a lack of secretion of TSH by the pituitary gland results in lack of stimulation of the thyroid gland are rare.) Unfortunately, the assay used to measure TSH in people can not be used to measure canine TSH, due to a species difference in the structure of the molecule. However, recently a species specific TSH assay for the dog has become commercially available. This assay can distinguish known hypothyroid dogs by measuring elevated TSH concentrations in the hypothyroid dogs when compared to TSH concentrations measured in clinically normal dogs. After limited use in practice (over 200 samples during 6 months), the authors have found the TSH concentration to be a valuable tool, when used with the measurement of free T4 concentration, in assessing thyroid function in dogs.

CIRCULATING ANTIBODIES AGAINST THYROGLOBULIN

When hypothyroidism is due to lymphocytic thyroiditis, the dog's body may produce antibodies to a protein portion of the thyroid hormone molecule called thyroglobulin. As many as 60% of hypothyroid dogs tested for thyroglobulin antibodies were positive; however, normal dogs may also have thyroglobulin antibodies but at a much lower incidence. The measurement of thyroglobulin antibodies has the potential value of early recognition of lymphocytic thyroiditis. This test is only available at the College of Veterinary Medicine at the University of Florida.

CIRCULATING ANTIBODIES AGAINST THYROID HORMONES

In addition to forming antibodies against thyroglobulin, dogs with hypothyroidism due to lymphocytic thyroiditis can also produce antibodies against thyroid hormones (T3 & T4). Thyroid hormone antibodies can also occur in normal dogs. Besides indicating that the process of lymphocytic thyroiditis may be occurring, the presence of thyroid hormone antibodies may result in elevations in the measurement of TT3 and TT4. Measurement of free T4 by equilibrium dialysis will show normal concentrations early in the immune-mediated process before clinical hypothyroidism is apparent. As thyroid gland tissue is destroyed by the lymphocytic thyroiditis, thyroid gland function

decreases, free T4 concentrations decrease and the dog shows clinical signs of hypothyroidism. The measurement of thyroid hormone antibodies is available at certain veterinary laboratories across the country.

THYROID BIOPSY

A surgical biopsy of the thyroid gland can be taken to evaluate the pathological process occurring in the thyroid gland. Cellular changes such as lymphocyte infiltration or follicle atrophy can be defined by microscopic examination of the thyroid biopsy. The advantage to biopsy is that it is specific for thyroid disease and can detect early changes even before significant decreases in circulating blood hormone concentrations. However, the degree of thyroid pathology observed microscopically may not necessarily parallel thyroid production of hormones or clinical hypothyroidism. Thyroid biopsy can be helpful in distinguishing primary hypothyroidism (defect is the thyroid gland) from secondary hypothyroidism (lack of stimulation of the thyroid gland by lack of TSH from the pituitary gland). Thyroid biopsy requires surgery and anesthesia and is moderately expensive (\$75.00-\$150.00)

In summary, definitive diagnosis of hypothyroidism can best be made at this time by a combination of abnormal specific thyroid test results coupled with successful long term response to thyroxine supplementation. The ideal tests currently available seem to be either a TSH stimulation test or when not available or affordable, a free T4 concentration by equilibrium dialysis, with or without a cholesterol measurement. Measurement of TSH concentration, along with measurement of free T4 concentration, are currently the diagnostic tests of choice in evaluating thyroid function in the dog. Caution should be made in over-interpretation of any thyroid function tests in clinically ill dogs.

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TABLE I
 THYROID TESTS FOR DOGS

| TEST | ADVANTAGES | LIMITATIONS |
|----------------------------|--|---|
| TT3 | Simple, Inexpensive | Does not reflect thyroid function |
| TT4 | Simple, Inexpensive | Can be falsely low due to drugs or non-thyroid illness |
| FREE T4 | Better reflects true thyroid function | Relatively expensive |
| TSH STIMULATION | Currently the best indicator of thyroid function because it measures thyroid gland response to stimulation | Cost & Availability |
| TRH STIMULATION | Also measures thyroid gland response to stimulation but harder to interpret | Cost & Availability Side Effects |
| TSH CONCENTRATION | Potentially may be best indicator of thyroid gland function when used with Free T4 | Limited use so far; moderately expensive |
| THYROGLOBULIN ANTIBODIES | Early detection; Specific for immune-mediated thyroid involvement | Can be positive in normal dogs; Currently available at Southwest Veterinary Diagnostics, Phoenix, Arizona |
| THYROID HORMONE ANTIBODIES | Early detection; Specific for immune-mediated thyroid involvement | Can be positive in normal dogs; Available at Michigan State University |
| THYROID BIOPSY | Demonstrates specific thyroid involvement; distinguishes primary from secondary hypothyroidism | Cost, surgery, anesthesia; May not reflect degree of thyroid dysfunction |