

## Submission Guidelines and Test Protocols

### General Information

Proper sample collection and handling are essential to accurate test results.

- A. Do not send primate samples.**
- B. Do not send whole blood for any test procedure.
- C. Do not send samples in serum-separator tubes (polymer-gel-tubes). This causes hemolysis of samples and the gel is known to precipitate drugs.
- D. Label sample containers adequately
  - a. Owner's name
  - b. Animal's name
  - c. Test desired
  - d. Time of collection (baseline, post-stim, etc.)
- E. For serum samples:
  - a. Clot at room temperature, centrifuge within 1 hour, remove serum and freeze.
  - b. Alternatively, store blood samples in the refrigerator for 2 to 4 hours to clot, centrifuge, remove serum and freeze.
- F. Packaging samples for shipment
  - a. Ship samples in insulated container with adequate cold packs and insulating materials (peanuts, paper), being careful to pack to avoid leakage or breakage (plastic tubes are recommended).
  - b. Seal lid juncture with tape to preserve cold.
  - c. Ship samples Monday-Thursday by overnight delivery service (UPS, FedEx, DHL).
  - d. Do not send immediately prior to a holiday!
  - e. Be sure to include a brief history of the case, and especially include the sex status and age of the animal.

### Thyroid Function

(All samples should be *serum*; do not send in polymer gel tubes)

\* Thyroid function tests (T3, T4, TSH) are run on Monday, Wednesday, and Friday each week

- A. T3 and/or T4 – DOG, CAT**
  - a. Prepare 0.5 ml of serum (sufficient for both tests).
  - b. Consider requesting the combined T4/TSH assay for dogs.
- B. Post Pill T4**
  - a. Collect serum (0.5 ml) post-pill at 4-6 hrs.
- C. T3 Suppression**
  - a. Collect basal serum sample for T4. Administer liothyronine (T3 or Cytobin™) at an oral dose of 25 µg 3x/day for 7 doses.
  - b. Collect sample for T4 on morning of 3rd day, 4 hrs after 7th dose of liothyronine.
  - c. **NOTE:** Request T3 determination also, if there is concern for owner compliance, or drug intake by the cat.
- D. Canine TSH**
  - a. Prepare 0.5 ml of serum.
  - b. Freeze sample and ship next-day delivery with proper packaging and cool packs.

## Adrenal Function

(All samples should be *serum*; do not send in polymer gel tubes)

\* Cortisol assays are run on Monday, Wednesday, and Friday each week

### A. Cortisol – ACTH Stim Test \* – DOG, CAT

- a. For dogs, collect baseline serum sample (1.0 ml) then give synthetic ACTH (Cortrosyn™) at 5 µg/kg IV (preferred) or IM and collect post-ACTH serum sample (1.0 ml) 1 hr later.
- b. For cats, collect baseline serum sample (1.0 ml) then use Cortrosyn™ as for dogs, but give 25 µg/kg IM (maximum dose of 125 µg/cat). Collect post-ACTH serum sample at 60 minutes. If using ACTH gel, give 2.0 IU/kg IM. Collect post-ACTH serum sample 1 hr and 2 hrs later.
- c. \* Cortrosyn™ (versus compounded ACTH) is the preferred ACTH for use in testing since quality is consistent.

### B. Cortisol-Dex Supp/ACTH Stim Test – DOG

- a. Collect baseline serum sample (1.0 ml). Administer dexamethasone at 0.1 mg/kg (IV).
- b. Take a post-Dex serum sample 4 hrs later.
- c. Following the Post-Dex collection, perform the Cortisol-ACTH Stim Test as above.

### C. Cortisol-Low Dose Dex Supp Test – DOG

- a. Collect baseline serum sample (1.0 ml).
- b. Administer dexamethasone at 0.01 mg/kg (IV).
- c. Collect Post-Dex serum samples at 4, 6 and 8 hrs (1.0 ml).

### D. Cortisol-High Dose Dex Supp Test – DOG

- a. Collect baseline serum sample (1.0 ml).
- b. Administer dexamethasone at 0.1 mg/kg (IV).
- c. Collect Post-Dex serum samples at 4, 6 and 8 hrs (1.0 ml).

### E. Adrenal Panel for Dogs and Cats (ACTH Stim) -Test for congenital adrenohyperplasia-like syndrome (dogs, cats), or Alopecia-X, Atypical Cushings Syndrome

**Note:** All Adrenal function tests use serum samples.

\* Adrenal panels are batch-run weekly (Aldosterone is no longer available)

- a. Collect baseline serum sample (2.0 ml).
- b. Centrifuge sample as soon as possible, separate and freeze.
- c. Administer the ACTH Stim test as above (3A). Collect Post-ACTH serum sample (2.0 ml) at 1 hr (2 hrs if gel-ACTH is used).
- d. Centrifuge sample as soon as possible, separate and freeze. (NOTE: If samples are grossly hemolyzed, repeat test in one week).
  - i. The following hormones will be assayed: Cortisol, Estradiol, Androstenedione, 17-hydroxyprogesterone, Progesterone and Testosterone.

### F. Adrenal Panel with Combined Dex Supp/ACTH Stim Test for Dogs

- a. Collect baseline serum sample (2.0 ml). Administer dexamethasone at 0.1 mg/kg (IV).
- b. Take a post-Dex serum sample 4 hrs later (an 8 hr post-Dex sample is optional).
- c. Following the 4 hour post-Dex collection (or 8 hour post-Dex collection, if performed), administer synthetic ACTH (Cortrosyn™) at 5 µg/kg (IV), and collect post-ACTH serum sample (2.0 ml) 1 hr later. Use of synthetic ACTH (IV) is the preferred method however if using ACTH gel administer 2.0 IU/kg (IM) and collect post-ACTH serum sample (2.0 ml) 2 hrs later.
- d. This test will provide the same information as the adrenal panel with ACTH stimulation only however it will also provide information as to whether the hormones are suppressed by

dexamethasone. With this approach, results from two separate testing procedures (dexamethasone suppression test and ACTH stimulation test) are achieved with one test.

**G. Adrenal Panel for Ferrets -Test for adrenocortical disease in ferrets**

- a. Collect baseline serum sample (0.5 ml).
- b. Centrifuge samples as soon as possible, separate and freeze.
- c. Ship frozen samples as for the Adrenal Panel for Dogs method above (3G).
- d. The following hormones will be assayed: Estradiol, Androstenedione and 17-hydroxyprogesterone. The assay is run each week.

**H. Adrenal Panel for Rabbits (neutered rabbits only)**

- a. Collect baseline serum sample (1 ml).
- b. Centrifuge sample as soon as possible then separate and freeze. Hormones analyzed are progesterone, 17-hydroxyprogesterone, androstenedione, testosterone, and cortisol.

Reproductive Hormones

(All samples should be *serum*; do not send in polymer gel tubes)

\* Reproductive hormone assays are run weekly. Breeding progesterone is run same day as submitted; please notify laboratory personnel in advance of sample submission

**A. Estradiol\*, Progesterone\*, or Testosterone\* - DOG, CAT**

- a. Collect 0.3 ml of serum for each.
- b. Centrifuge samples as soon as possible and separate serum.
- c. \* If analyzing sample for all 3 hormones send 0.5 ml serum.

**B. Testosterone – hCG Stim – For cryptorchidism detection in the horse.**

- a. Collect baseline serum sample (1.0 ml) in the morning hours (a.m.).
- b. Administer hCG at 10,000 units total dose (IV). Collect Post-hCG serum sample (2.0 ml) 72 hours later (a.m.).
- c. Centrifuge samples as soon as possible and separate serum.

**C. Testosterone - hCG stimulation - For completeness of castration in dogs and cats.**

- a. Collect baseline serum sample (1.0 ml).
- b. Administer hCG at 50 IU/kg IM.
- c. Collect Post-hCG serum sample at 2 hours.
- d. Centrifuge samples as soon as possible and separate serum.

**D. Progesterone – hCG Stim -For detecting ovarian remnant in dogs\* or cats\***

- a. Collect baseline serum sample (0.5 ml).
- b. Administer 50 IU of hCG/kg I.M.
- c. Collect post hCG samples at 7 and 21 days.
- d. Process samples per general information guidelines.
- e. \* Perform test when in estrus.

**E. Progesterone – hCG Stim – For detecting ovarian remnant in rabbits**

- a. Collect baseline serum sample (0.5 ml).
- b. Then administer 50 IU hCG IV.
- c. Collect serum sample (0.5 ml) 4 days later.

**F. Testosterone – hCG Stim – For detecting retained testicular tissue in rabbits**

- a. Collect baseline serum sample (0.5).
- b. Then administer 1500 IU hCG IM.
- c. Collect serum sample (0.5 ml) 3 hours later.