INTENDED USE

The Canine Proges-Check™ is a Colored Antibody Rapid Testing device (**CART)**, an enzyme immunoassay system for semi-quantitative determination of progesterone levels in serum/plasma test samples. The test is intended for professional use as an aid in the diagnosis and monitoring of conditions related to progesterone levels and reproductive state of the animal. The device is designed to help identify ovulation or pregnancy status of canine and related animals.

TEST PRINCIPLES

The progesterone semi quantitative visual Test is a solid-phase enzyme immunoassay based on a competitive binding method. A sample (serum/ plasma) containing an unknown amount of progesterone will compete with enzyme-conjugated progesterone for high affinity binding sites on a limited number of antibodies coated on the CART. After washing, the amount of labeled progesterone in the CART is inversely proportional to the concentration of the sample progesterone. A color reagent solution is then added and the enzyme is allowed to react for 2-5 minutes. The actual concentration of the sample is compared by color with three different standard concentrations provided. This kit is suitable for the direct visualization of progesterone in serum/plasma samples of canine and related animals.

MATERIALS PROVIDED

1. Progesterone antibody coated CART devices (5 units/box)
2. Enzyme Conjugate, 1.0 mL (Red Cap)
3. TMB Color Reagent, 1.0 mL.(Blue Cap)
4. Wash buffer 3 mL (White Cap)
5. Pipettes (5)
6. Instructions

STORAGE & STABILITY CONDITIONS

1. Store the kit at 2-8°C upon receipt and when it is not in use. **Do not freeze.**
2. Keep CART devices in a sealed bag with desiccants to minimize exposure to damp air.
3. Allow all the reagents to reach to room temperature before setting up the assay.
4. Do not at any time mix or use components with other manufacturer kits. Do not use the kit components after the expiration date.

SAMPLE COLLECTION AND PREPARATION

1. This kit is suitable for use with serum or heparin plasma samples. The use of hemolytic or lipemic samples will affect results and also samples with bilirubin may interfere with the assay.
2. No special preparation of the samples is required. A venous blood sample (enough to produce about 0.5 ml serum) is collected aseptically.
3. If the sample is not tested immediately refrigerate at 2-8°C. If a storage period greater than 3 days is anticipated, the specimen should be frozen and repeated thawing and freezing should be avoided.
4. If the sample is turbid or contains precipitate it may give false results. Such samples should be centrifuged before use. Serum samples with gross lipemia, hemolysis and turbidity should not be used.

ASSAY PROCEDURE

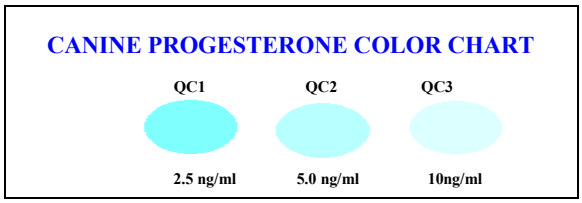
All reagents should be allowed to reach room temperature (18-25°C) before use and all CART devices should be clearly marked for further identification.

1. Add three drops (0.1mL) of wash buffer to the CART and allow it to sink completely.
2. Add three drops of test samples to CARTs (mark samples prior to addition for further identification) and allow it to sink completely. Use separate pipette provided for each sample and discard after use.
3. Add three drops of progesterone Enzyme Conjugate (Red cap) to each CART and allow it to sink completely.

4. Add three drops of wash buffer and allow it to sink completely.
6. Add three drops of TMB color solution to all CARTs and allow it to sink completely.
5. Read the results in five minutes and compare to the chart (see below) for interpretation and comparison.

RESULTS

The test results can be interpreted for ovulation depending on the levels of progesterone in the test samples. Compare the results with the color chart shown below.



1. The color intensity is close to or similar to QC1, the progesterone level in the test sample is about 2.5 ng/ml . The animal has not yet ovulated. Keep monitoring.
2. The color intensity is close to similar to QC2, the progesterone level in the test sample is about 5.0 ng/ml. The animal is close to or ready for ovulation.
3. The preparation for AI process should be underway.
4. The color intensity is close to similar to QC3, the progesterone level in the test sample is about 10 ng/ml. The animal has ovulated.

The test may also be used to determine pregnancy status near term. A minimum of 2.5 ng/ml of progesterone is required to maintain a pregnancy. A color **darker** than QC1 indicates that labor is imminent within 48 hours. This information can be useful in planning normal or surgical deliveries.

LIMITATIONS OF THE TEST

1. Endocrine's Visual progesterone test system is designed for semi-quantitative visual observation of progesterone levels in Canine and related species serum/plasma samples only. 2. The device should be adequately allowed to soak each sample to obtain reproducible results 3. The results obtained with this assay should only be used as an adjunct to other diagnostic procedures and information available to the veterinary professional. 4. Trained and skilled professional only should perform the assay. 5. For quantitative determination of progesterone in test samples, use ETI's Canine Progesterone ELISA system. Use canine LH Rapid test for accurate identification of ovulation and timing for AI process.

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