Quick Check™ Canine Progesterone Kit

Recommended For Use in Veterinary Clinics

- The main reason for unsuccessful breedings is improper timing.
- Accurate timing is important for breeding success especially when
- The breeding is done with fresh chilled or frozen semen.
- This kit will enable the user to determine the optimum time for fertilization.

DETERMINATION OF THE FERTILE PERIOD

The visible signs of heat and breeding time (such as swelling of the vulva, vaginal discharge, or flagging) are only an approximate indication of the time of ovulation. These signs can vary by more than a week.

Ovulation is triggered by the Luteinizing Hormone or LH which peaks two days before ovulation. Progesterone levels begin to rise at the same time as the LH surge. Ovulation occurs two days after the initial rise in progesterone. Predicting ovulation is done accurately by detecting the initial rise in progesterone.

After ovulation occurs, the oocytes take 2-3 days to mature, making the most fertile period 5-6 days after the initial rise in progesterone.

The relationship of the hormones, progesterone and LH, to ovulation and to the fertile period is shown in the figure.

With the Next Generation® Quick Check™ Canine Ovulation Timing kit, it is possible to detect the initial rise in progesterone. As seen in the figure, the progesterone level before ovulation is low (between 0 and 1.0 ng/ml). This corresponds to a bright blue (C1) test result. When the initial rise in progesterone occurs, the test result will be a light blue (C2).
GUIDELINE FOR WHEN TO TEST

1. In order to determine the baseline progesterone level, one serum sample can be run during the first five days of the pro-estrus cycle (characterized by the presence of blood and vulva swelling). The baseline progesterone level varies between 0-1 ng/ml in individual dogs and gives a bright blue color result similar to the C1 color on the color chart. Note the color shade.

2. To determine the initial rise in progesterone, begin testing 7-8 days after observing the first day of vaginal bleeding and vulva swelling (the beginning of pro-estrus). If the first result is the baseline color, bright blue (C1), then the progesterone level is still very low.

3. Retest every two days until the test results are a light blue that is similar to the C2 color on the color chart. This first fading of the color from C1 to C2 represents the initial rise in the progesterone level; ovulation will occur 1-2 days later.

4. Retest every day until the test results are faint blue similar to the C3 color on the color chart. This means ovulation has occurred and the egg is beginning to ripen and the fertile period begins 2-3 days later (See Figure). After obtaining a C3 color, natural mating and inseminations with fresh semen should take place in 2 days and again in 2 days. If using frozen semen, inseminate in 2 days and every other day for a total of 3 times.

5. If a white result is obtained, mating or insemination should be done immediately.

TEST PREPARATION (FOR SERUM OR PLASMA)

Allow all kit components and samples to come to room temperature. Collect the blood sample into an EDTA or heparin coated tube (purple top tube) or in a dry tube (red top tube). Immediately after sample collection, invert the tube several times to mix. Spin the blood down with a centrifuge or allow the blood to clot by staying at room temperature for 1/2 to 1 hr and pour the serum into a clean glass tube. If the sample is not run
immediately, store the sample in the refrigerator. For long term storage the sample should be frozen. Label the sample with the name, sample day and cycle day. Do not use whole blood. Do not run test with cold kits.

Other important points:
- Store the kit in the refrigerator when not in use. Do not freeze.
- Always reseal the plastic bag after removing a test cup.
- Timing of the enzyme during Step 2 is IMPORTANT: One (1) minute.
- Do not exchange test cups or reagents between different kits.
- If you want to do a control, do steps 2-7, the result should be C1.

The Next Generation® Quick Check™ Canine Progesterone kit can also be used to accurately determine the right time for a Cesarean section. About 24 hours before normal whelping, progesterone levels drop back to low baseline levels, the test result changes from white (C4) to bright blue (C1). By daily testing just before the expected whelping date, the time for the C-section can be precisely determined and emergency surgery or premature puppies can be avoided. Progesterone levels can also be measured throughout pregnancy in order to confirm that the progesterone levels remain high (C4) and the bitch is maintaining a normal pregnancy.

INTERPRETATION OF THE RESULTS

The recommendations in the following table are only a guideline and all other available information concerning the dog’s cycle should be considered.

The color result of the test corresponds to the level of progesterone present. Use the enclosed color chart as a guide for evaluating and recording the result.

- The C1 color corresponds to baseline serum progesterone levels.
- The C2 color corresponds to the initial rise in progesterone levels. This is Day 0, the day of the LH peak, which is usually 1-2 days before the ovulation.
- The C3 color corresponds to the further rise in progesterone when the egg begins to ripen after ovulation.
- The C4 color corresponds to greater than 5 ng/ml
- Progesterone when the egg is already relatively ripe.

TEST PREPARATION

Take the kit out of the refrigerator 2 hours before using it in order to allow the kit to assume room temperature. If desired, leave the kit at room temperature overnight

Intended for veterinary use only.
Not for human use.
The manufacturer warrants the kit for its intended use.
Liability is limited strictly to the value of the kit.
|   | 1. Shake blood sample.
 2. Add 8 drops to the center of the cup.
 3. **WAIT 2 MINUTES.**
  Use a new pipette for each sample. |
|---|---|
| 2 | 1. Add 4 drops of the sample
 2. Wash with *RED* Label.
 3. Wait for liquid to drain into cup.
 4. **REPEAT** this step 1 time. |
| 3 | 1. Add 3 drops enzyme from the *RED CAP BOTTLE* to the center of the cup.
 2. **WAIT ONE MINUTE**
  **Timing this step is important** |
| 4 | 1. Fill cup to the top of inner line.
 2. Use solution from the *WHITE CAP BOTTLE*.
 3. Wait for liquid to drain totally into cup. |
| 5 | 1. Prepare fresh substrate solution.
  *BLUE CAP BOTTLE*
  2. Using each dropper filled to the mark from *Substrate A* and *Substrate B*.
  3. Shake the freshly prepared substrate in the mixing bottle. |
| 6 | 1. Add 3 drops of freshly prepared substrate, to the center of the cup.**
  **The substrate solution is good for 30 minutes and should be discarded after the test is run.**
  **Save blue mixing bottle.** |
| 7 | *Record results in 9 MINUTES.*
  *Bright Blue: = C1*
  *Light Blue: = C2*
  *Faint Blue: = C3*
  *White: = C4* |