Progesterone Assays Have Revolutionized Canine Breeding Management

The reproductive features of the domestic female dog are unique when compared to those of the farm animal species. The dog is mono-estrus, that is, she has one estrus per breeding season, with most species having two breeding seasons per year. This is followed by a prolonged period of anestrus, or reproductive quiescence. During each cycle, she has prolonged follicular and luteal phases compared to those of other domestic species.

The stages of the estrual cycle are associated with hormonal and recognizable behavioral changes. Progesterone and LH are the key reproductive hormones.

Proestrus, or the follicular phase, begins when vulvar swelling and a bloody discharge is observed and lasts on the average of nine days. The bitch will not accept mating. The progesterone levels are low.

Estrus is the period of standing behavior and receptivity. The duration is variable in length (3-21 days), the average being 9 days. Standing behavior is associated with the first rise in progesterone. Ovulation occurs during this period.

Diestrus, or luteal phase, begins when behavior changes dramatically to non-acceptance of the male. The onset of diestrus is characterized as the day when exfoliated vaginal cells change from predominantly cornified to predominantly noncornified. Progesterone is produced from the corpora lutea and rises rapidly. The corpora lutea will continue to secrete progesterone for about two months.

Anestrus follows diestrus. This stage of the cycle is a period of reproductive inactivity. Anestrus lasts an average of 4 1/2 months. The duration of anestrus determines the inter-estrus interval or the time elapsed from one cycle to the next. This varies considerably between breeds, within an individual bitch or between bitches of the same breed.

The initiation of estrus behavior may be as early as five days before ovulation1 or as late as three days after ovulation and there is not always a definite correlation between the stage of the cycle and the vaginal cytology specimen.2 Because of this and because estrus is so infrequent, it is more accurate to time ovulation by detecting hormonal changes which occur during the cycle.

Accurate ovulation timing is provided when measuring the hormone progesterone in the bitch's serum. Progesterone and Luteinizing Hormone primarily determine the time of ovulation.

Ovulation is triggered by the Luteinizing Hormone or LH which is circulating in the blood in small amounts. Just prior to ovulation, it increases rapidly and peaks during a 24-hour period. Two days after the LH peak, ovulation occurs.

During the LH peak progesterone also starts to rise and continues to rise throughout the estrous cycle. By detecting the initial rise in progesterone levels, ovulation can successfully be predicted. Once the rise is detected, ovulation will occur within two days and the fertile period identified.

Although LH testing determines the peak itself, the duration of the surge is short and easily missed. LH levels are not always constant and small rises may occur early in the estrous cycle which produce false

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Breeding

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Metestrus

The transitional period between ovulation and the full development of the corpus luteum. The hormones switch from estrogen to progesterone dominance. In the bitch, this occurs in its entirety during estrus.

Ovaries

A pair of female kidney-shaped glands which are responsible for the production of the unfertilized ovum or egg.

Ovulation

The release of an ovum (or egg) from a mature ovarian follicle.

Parturition

Process of giving birth.

Proestrus

The period of the estrus cycle when rapid follicular growth occurs. Progesterone levels are low. The first day of bloody discharge is generally agreed to represent the first day of proestrus.

Progesterone

The most important hormone in the reproductive cycle. It is produced by the corpus luteum after ovulation. The rising and falling of progesterone levels directly correspond to each stage of the estrous cycle.

Glossary of Terms

Brucella Canis
A venereal transmitted infection that can cause infertility and abortion in the bitch and sterility in the stud dog.

Cytology
The study of cells under a microscope. Vaginal cytology is often used as a breeding aid.

Luteinizing Hormone (LH)
Hormone secreted by the anterior pituitary gland and released in a pulsatile fashion that stimulates the development of corpora lutea and causes ovulation to occur.

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Windys Pups

I don't know what it is that I've done to irritate "Mother Nature", but she must be pretty mad at me!

Two years ago, after years of planning, I bred my home-bred, longhaired, miniature, Dachshund, CH. Von Schaafmeister's Irish Rhapsody ("Erin"). The breeding was accomplished and we hoped for the best. But approximately one month later, trouble set in, when "Erin" began to have a bloody discharge which was diagnosed as metritis. We tried antibiotics and other things, but finally we had to spay her to save her life. I was devastated. My long term breeding plans were destroyed.

One year later, Mattie, a bitch that I co-own, was bred to a dog that I admire. However, two and a half weeks before her due date, she aborted the litter of 4-6 fetuses. Once again, it looked as if Mother Nature was getting back at me for some sin.

How to recover from the loss of "Erin" in a breeding program, is something that Paula, my friend/mentor and I discussed at length. We decided to take a chance by breeding the full sister to "Erin", "Windy", to "Rowdy". Windy is 6 years old and had never been bred and we knew it was "now or never" because of her age. Also to make matters more difficult Rowdy doesn't believe in breeding bitches. No one has ever been able to collect on "Rowdy" except Paula. We knew we were going to need Paula's expertise, and Paula and I live approximately 7 hours apart. We made plans to drive to a road-side park that is exactly half way between our houses, do the artificial insemination in my van and then return to our respective homes. Of course, we've had many laughs over the sight of two women and two dogs doing an AI in a van at a road-side park! We hope a state trooper never stops to ask what we are doing.

In the past I had relied on some of my older, experienced "boys" to tell me when a bitch was ready to be bred. Their experience, in addition to the bitch's actions were the indicators that let me know when to breed. However, in "Windy's" case, she never acted receptive. And my two best "she's ready Mom" indicator males had been neutered. Fortunately, Paula had attended a Dr. Hutchison seminar and learned about the Target tests. She suggested that my vet order some and use them to decide when we should

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Infertility in the Bitch

In my opinion, 90% of those bitches which are bred to proven sires but do not become pregnant, have no infertility problems whatsoever. Their only problem is that they are not bred at the correct time during their cycle.

Progesterone level testing done by a competent veterinarian to determine her fertile period will take care of these. The following discussion is for the other 10%. For simplicity, I will divide these bitches into two categories; those that cycle normally and those that do not. A normal heat cycle should occur at regular intervals and be of predictable duration. These may vary in length, but should be predictable for individual bitches.

A normal bitch must have a minimum of four months between cycles. This is the time that it takes for the uterus to recover (from estrus or pregnancy) and be able to accept the next pregnancy.

If your bitch falls into this normal category, I recommend the following:

- Blood test for Brucella Canis
- Exploratory Laparotomy — includes careful examination of her reproductive tract, culture and sensitivity of the uterine lumen and biopsy of the uterus and ovary (some problems may be encountered that are beyond repair and these bitches can be spayed at the same time).
- Surgical insemination of semen — the semen injected directly into the uterine body via an abdominal incision. Do not inject >12 ml. This procedure may be combined with an exploratory, but if this is done, I do not recommend taking cultures and/or biopsy at the same time. This procedure is done 48 hours after her progesterone level reaches 5ng/ml. (This is also a recommendation for increasing the chance of a fertile breeding using frozen semen.)

If you have a reason to believe that your bitch has been pregnant, but has reabsorbed the fetus, then again, an exploratory may be beneficial to rule out anatomical defects or infectious agents. Ultrasound is very helpful with this problem. Also, other causes of reabsorption are progesterone insufficiency and hypothyroidism. These causes may be identified by blood work and corrected as necessary. For hypothyroidism, use Soloxine (No Generics!). 1mg/pound(10) given two times a day.

I have experienced excellent results with thyroid supplementation even when the bitch’s T3 and T4 were normal.

Progesterone therapy is more complicated. Replacement therapy (Regumate 1cc/50 pounds, orally once a day) must be monitored closely with ultrasound and blood tests. An accurate due date must be calculated to avoid prolonged gestation and fetal death, a side effect of prolonged administration of progesterone.

Those bitches with abnormal cycles fall into four categories (an Exploratory Laparotomy may be indicated with these also):

- **Shortened estrus period** (<4 months) — You can prolong this time by using Cheque drops for 5-6 months. Therapy is started 30 days before next expected estrus. This condition tends to appear each time, so you must plan to breed her on her next heat.

- **Prolonged estrus period** — The most likely cause is hypothyroidism or Cushings Disease. Again, these may be ruled out by bloodwork. Also, this bitch may experiencing “Silent Heat,” which means she is cycling normally but detection is difficult to impossible. Try daily tissue wiping of the vulva or weekly vaginal smears. Also, sometimes the presence of an experienced male is helpful.

- **Split Heat** (sometimes called “Wolf Heat”) — These bitches appear to have a normal heat cycle but will be in heat again in 2-6 weeks. This is considered a normal variation in the canine cycle. The second heat is the fertile heat but use progesterone testing to determine ovulation.

- **Persistent estrus** (or very frequent) — The most common cause are ovarian tumors or cysts. Exploratory and/or ultrasound is diagnostic. If a tumor is found and only one ovary is involved, that ovary may be removed. The best treatment for ovarian cysts is surgical exposure and manual rupture of the cysts. Hormonal therapy (HCG) for ovarian cysts may cause pyometra.

Unfortunately, most of these are quite expensive and time consuming. Some of you may choose to spay your bitch. On the other hand, some of you may have that special girl whose contribution to our breed would be worth the extra effort.

*Nancy Rose, DVM • Newcomb Bulldogs • Elk City, OK*
Press Release

BioMetallics has just published a detailed, well illustrated wallchart entitled "Progesterone Levels and Vaginal Cytology in Relation to Canine Ovulation Timing." As well as explaining the canine estrous cycle, the chart also offers practical guidance on using progesterone ELISA assays as a tool to successful canine breeding management. If you would like a free copy, call us at 1-800-999-1961 or fax 1-800-999-1962.

Thanks for your Support!
Keep the cards and letters coming!

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Answer line

Q.

What should the color of my TARGET test results be before my bitch is bred by artificial insemination with fresh chilled semen?

A.

The test results should have just turned WHITE (C4).

It is recommended that a TARGET test be run sometime during the first five days of proestrus to determine a baseline color which is usually bright blue (C1). Starting at Day 7 or 8, the test should be run every other day until a color change to a lighter blue (C2) is seen signaling the initial rise of progesterone. Knowing when the initial rise occurs is important because it allows early planning for the breeding. Optimal breeding time will occur 4-7 days after the initial rise. Once a C2 color is obtained, test again in 2-4 days prior to breeding. The color result should have just turned white (C4) before breeding to make sure the eggs have fully ripened.
meet at the roadside park. Since it was going to be a "one shot deal", we needed to know definitively when to meet. We started testing "Windy" as soon as she came in. Paula and I kept our cars at the ready and were in almost daily contact. Days went by with no change in the reading. Every test popped up a definite, bright blue C1. This continued for 18 days. My vet assured me that while this was not the norm, it was not unheard of and to be patient. Finally, at 19 days she changed to C2 and three days later was at C3.

We were starting to panic because Paula was scheduled to fly out of state for a week on business. Dog affairs could not interfere with it. On the Sunday before Paula was scheduled to fly out, our trusty Target test said GO! I jumped in my van and headed for Louisiana; Paula jumped in her car and headed for Texas. We met at the roadside park smug and secure in the knowledge that we had the right day and that our "one shot" session would be successful. The dogs, however, had something else in mind. One and a half hours later, two frustrated women realized that there was no way that Rowdy could be bribed, convinced, coerced or made to breed Windy. Nor could we get a collection for artificial insemination.

I was at my wits end; I really wanted this breeding. When we realized that it was not going to happen, we sat stunned-looking at each other. Then we laughed at the absurdity of it all. Finally, as all resilient, but determined breeders do, we started discussing a new plan! After sorting through about a dozen options, we settled on three males including one of Paula's dogs, a dog named "Zydeco". Paula jumped in her car with "Windy" to take her back to Louisiana. I drove back to Texas very disappointed with the exhausted and uncooperative "Rowdy".

Late that night Paula successfully bred "Zydeco" to "Windy", packed her bag, and left the next morning on her trip. What friendship and dedication!

We now have three dachshund puppies that are 10 days old. Since "Windy" had an atypical season and never showed any receptiveness to a male, these puppies owe their existence to the Target tests. Additionally, when trying to co-ordinate a breeding with any distance involved, it makes it easier to know exactly when to travel. With the accuracy of the Target tests, one breeding is sufficient; although to be safe, two breedings are preferable. This makes it easier on the humans and the canines.

I guess this means Mother Nature is no longer mad at me!

Wendy Schaffmaster McQuiston, Von Schaafmeister
Longhaired Dachshunds • Red Oak, TX

Vaginal cytology is used as an aid in breeding management but should not be relied upon as the only measure. Now we have progesterone ELISA assays commercially available that can easily be done in house which detects the LH peak indirectly. These assays give us the precise time to breed with enough lead time to make arrangements for the breeding. These assays have revolutionized canine breeding management.


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When Will She Give Birth?

Gestation length has been determined to be 64-66 days after the preovulatory LH surge, independent of the day of insemination.\(^1\) Estimating the pre-ovulatory LH peak by detecting the first rise in serum progesterone is currently the most accurate way to predict the expected date of parturition.\(^2\)

\(^{1}\)Concannon PW: Reproductive Physiology of the Bitch. Proceedings of Canine Theriogenology Short Course 1993