

Clinical Significance and Application of cProg





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a. Hormones Affecting Dog Oestrus and Pregnancy

There are a variety of hormones that help regulate the female dog's oestrus cycle and pregnancy. These include:

- Oestrogen Stimulate ovaries to lay eggs
- □ Luteinising Hormone (LH) Stimulate ovulation
- □ Progesterone: Maintain pregnancy



a. Hormones Affecting Dog Oestrus and Pregnancy

Understanding how hormone levels change can help determine the optimal reproduction time and predict production. When the level of oestrogen in the blood rises, most mammals ovulate. However, when the level of oestrogen drops and the level of progesterone rises, the dog ovulates.

Oestrogen levels can help us understand when a dog will be in heat, but it is not enough to determine when we should breed.

Vaginal cytology can also provide some basic information.

The level of progesterone (Prog) and the level of luteinising hormone (LH) are the best indicators of ovulation time and when is the best time for fertilisation. They can also be used to determine the date of pregnancy and due date.



Luteinising Hormone Levels

LH Test: It needs to be tested every day, from the appearance of oestrus to the end of the pre-oestrus.

The peak value of LH can only be maintained for 24 hours or less, so it needs to be tested every 24 hours.

If the test break time is too long, the peak value may be ignored.

The LH peak appears 48 hours before ovulation.





Figure 1 Hormone Levels During Heat and Pregnancy



Progesterone Levels

Progesterone testing should be done every 2 - 3 days, starting from 3 - 5 days in heat.

The initial progesterone level is usually less than 1.0 ng/mL until the day before the LH surge.

On the day of LH peak, the serum progesterone concentration was 2 - 3 ng/mL.

One day after the peak of LH, the serum progesterone concentration is 3 - 4 ng/mL.

Ovulation occurs at a progesterone level of 5 ng/mL.





Figure 2 Estimated Service Time and Production Time



b. Mating Time

The goal is to determine when the progesterone level reaches 2.5 ng/mL so that a mating plan can be established, or a semen sample can be prepared for collection and delivery.

Depending on the type of semen used, the best time for natural or artificial insemination is:

Mode of Reproduction	Time	Sperm Survival Time After Fertilisation	Insemination Part
Natural Mating	Prog 2.5 ng/mL after three days	5 – 7 days	
Cold Fresh Semen	Prog 2.5 ng/mL after four days or Prog 5 ng/mL after 48 hours	48 – 72 hours	Cervix
Frozen Semen	Prog 2.5 ng/mL after five days or 5 ng/mL after 72 hours	Less than 24 hours	Inside the uterus

 Table 1 Fertilisation Time of Different Sperm Types



b. Fertilisation and Implantation

It takes about 7 hours for sperm to become fertilised after ejaculation. This time is called the "capacitation time".

Egg cells also need a certain amount of time to mature, about 48 hours after ovulation.

Fertilisation is done in the fallopian tube, regardless of the method of insemination (natural reproduction, cold fresh semen, frozen semen). The fertilised egg then moves to the uterus until it is implanted into the endometrium 17 to 18 days after ovulation.

If there is a problem with the endometrium, the fertilised egg will not implant or the embryo will not grow or maintain.

A normal embryo implants into the endometrium. If implantation does not occur or the embryo grows abnormally, the foetus is absorbed.



b. Progesterone Levels During Pregnancy

After ovulation, the progesterone concentration continues to increase for 2 - 3 weeks, eventually reaching 10 - 80 ng/mL.

This concentration is necessary to maintain pregnancy. The progesterone level will remain at this level for about 60 days, regardless of whether the dog is bred and whether she is pregnant (false pregnancy).



b. Progesterone Levels During Pregnancy

	Progesterone
48 hours before delivery	2 ng/mL
24 hours before delivery	1 ng/mL

Table 2 Progesterone and Childbirth

This can help predict the correct time for childbirth, especially for dogs whose ovulation date is unknown.

The correct determination of the delivery time can prevent premature caesarean section and increase the survival rate of puppies.



Progesterone Value (ng/mL)	Estimated Ovulation Time (Days)	Estimated Mating Time (Days)	
0-1	Basic concentration, ovulation is still early	Can't match	
1 – 2	At least two days before ovulation, this concentration may last for a week or more	Expected mating time is 4 – 6 days later but it may take longer	
2 – 4	Expected ovulation time is less than 1 day	The mating time is expected to be 3 – 5 days later, but it may take longer	
4 – 6	Ovulation is coming or just happened	Expected mating time is 2 – 4 days later	
6 - 10	Ovulation does not occur more than 1 day	Expected mating time is 1 – 3 days later	
10 – 20	The egg cells are mature and have the best fertility potential	Estimated mating time 0 – 2 days	
20 – 30	The egg cells are mature but aging, and the fertility potential has declined	Estimated mating time 0 – 1 days	
>30	Too late or drastically reduced fertility potential		

Table 3 Range of Progesterone Results and Their Significance in Breeding



Sample Collection

Avoid using serum separator tubes

It is reported that the concentration of progesterone in canine serum is reduced when using serum separator tubes. It is suspected that progesterone will be absorbed by the gel material.

It is recommended to be tested after centrifugation. In addition, if the blood produces a blood clot, it should be centrifuged immediately to separate the serum from the clot.

If using a plasma sample, it is recommended to use matching lithium heparin tube.





Summary

Continuous monitoring of progesterone in female dogs has proven to be an effective diagnostic tool for managing breeding.

However, the time interval from the pre-oestrus period to the first increase in progesterone and the difference in the increase rate are very different between female dogs.

In order to determine that ovulation is about to occur or has occurred, testing should be continued until a result of 5 ng/mL or more is obtained. When the serum progesterone is between 10 - 20 ng/mL during breeding, the fertility is the best in this period.

During pregnancy, progesterone is greater than 2 ng/mL. The progesterone level is reduced to 2 ng/mL 48 hours before delivery and 1 ng/mL 24 hours before delivery.



	P4 Analyzer	8	P4 Analyzer		P4 Analyzer
	<0,2		8,0		18,0
	0,5	9	9,0		18,0
	1,0	Ì	9,0		18,0
	1,5	2	9,0		18,5
$\rightarrow \rightarrow \rightarrow \rightarrow$	1,5	$\rightarrow \rightarrow \rightarrow \rightarrow$	9,5		19,0
LH Surge	2,0	First Breeding	10,0		19,0
Looking for	2,5		10,0		19,0
significant rise in	3,0		10,5		19,5
Progesterone PG Value	3,0		11,0		20,0
Should Doble	3,0		11,0		20,0
	3,0	$\rightarrow \rightarrow \rightarrow \rightarrow$	11,6		20,5
	3,5		11,5		20,5
	3,5		12,0	$\rightarrow \rightarrow \rightarrow \rightarrow$	21,0
	4,0		12,0	Surgical or	21,5
	4,0		12,5	Frozen semen	21,5
	4,5		13,0		22,0
	4,5		13,5		22,0
$\rightarrow \rightarrow \rightarrow \rightarrow$	5,0		13,5		22,5
Ovulation	5,0		14,0		22,5
	5,5		14,5		23,0
	5,5		15,0		23,5
	6,0		15,0		23,5
	6,0		15,5		24,0
	6,5		15,5		24,0
	6,5		16,0		24,5
	7,0		16,0		24,5
	7,0		16,5		24,5
	7,5		16,5		25,0
	7,5		17,0		25,0
	8.0		17,0		25.0

Table 4 Comparison Table of Progesterone Monitoring in Europe and America



d. Do Male Dogs Contain These Hormones?

Oestrogen (E2)

Males produce small amounts of oestrogen, mainly oestradiol (E2) secreted by interstitial cells and Sertoli cells of the testis. The main victory function of E2 is to participate in spermatogenic function of testis. In human medicine, E2 is often used as one of the indicators of male infertility.

Luteinising Hormone (LH)

Luteinising hormone (LH) can stimulate the development of testicular interstitial cells and promote their secretion of testosterone (T), so it is also known as interstitial cell promoter. Insufficient secretion of LH can affect the production of T, thus affecting sperm generation and maturation.

Progesterone (P)

Increased blood progesterone levels in males are common in certain malignancies and congenital adrenal hyperplasia.





Canine Progesterone (cProg) Rapid Quantitative Test

Woodley have developed a rapid, accurate and reliable, highly sensitive detection method for Progesterone in dogs.

The InSight V-IA cProg Rapid Quantitative Test is a fluorescence immunoassay used with the InSight V-IA Veterinary Immunoassay Analyser for the quantitative determination of progesterone concentration in canine serum or plasma.

The test is used as an aid to track ovulation, determine the best time for breeding or detect early pregnancy failure.

It can be stored at room temperature.







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Thank You



