

## **Brood Bitch Fertility**

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While the stud dog has the advantage of a longer age-related fertility period and the opportunity to have many multiples of litters within each year, the true quality of a breeding program depends upon the exceptional brood bitches. With a maximum breeding potential of only twice a year, and age-related issues more insidious than with the stud dog, it is often imperative that the owner of the bitch determine her peak fertility period within the estrous cycle. One of the most frequently misunderstood physiological processes, the estrous cycle of the bitch is as complex as that of people. Veterinarians, owners, even experienced breeders may not realize the opportunities they have missed in obtaining a pregnancy if they do not understand the hormonal fluctuations which signify changes in fertility as well as other biological phenomena which can influence the chances of pregnancy and reproduction.

There are several different hormones involved in estrous, pregnancy and parturition of a brood bitch. These include estrogen, progesterone, follicle stimulating hormone (FSH), luteinizing hormone (LH), prostaglandins (PGF2, etc.), and oxytocin. The estrous cycle will be explained in detail, then the maintenance of pregnancy, and lastly the chemical signals which lead to whelping. Other physiological and biological problems which can lead to infertility will also be discussed.

The main point of the estrous cycle of the bitch is that while she has signs of being "in heat" for up to three or four weeks, she is only actually fertile for a 4 or 5 day window within the heat cycle. For bitches that do not stand or flag, or for stud dogs that are inexperienced or with low libido, the actual window may be missed by the breeder if he/she is depending on behavioral changes or vaginal discharge or cytology to signal "readiness." There are four stages to the estrous cycle: proestrus, estrus, diestrus, and anestrus.

Proestrus begins with estrogen levels rising slowly over a 10-14 day period, resulting in vulvar swelling, vaginal discharge (usually dark red and copious) and an increased production and sloughing of vaginal epithelial cells. During this time, progesterone is low, and the LH surge has not occurred. Estrus begins with the LH surge and rising progesterone levels. Concurrently, estrogen begins to decline, and the bitch may show behavioral signs such as flagging and winking. This is the fertile period of the bitch, and lasts only 4-5 days. Diestrus begins about 7-9 days post LH surge, with the characteristics of progesterone levels which remain elevated, low estrogen levels, and return of non-cornified epithelial cells. Anestrus is the period between diestrus and proestrus, during which the uterine lining is rebuilding, and the ovaries are quiescent.

A summary of the hormonal fluctuations during the estrous cycle and pregnancy begins with estrogen slowly rising, reaching a peak, and then rapidly declining. During this time follicle stimulating hormone is inducing the eggs to begin maturation processes within the ovaries. Meanwhile, progesterone maintains a steady state at low

concentration, begins to slowly rise, then maintains high levels throughout diestrus or pregnancy, if bred. LH is initially low in the cycle, spikes over a 24-48 hour period and then remains low. LH is the hormone responsible for signaling the ovaries to release eggs, resulting in ovulation. Progesterone must remain high post-ovulation in order to maintain implantation of the eggs and pregnancy. Finally, at the end of pregnancy, waning progesterone levels result from the destruction of the corpus luteum by prostaglandin release. Concurrently, rising oxytocin levels signal the beginning of parturition, with elevated oxytocin levels resulting in uterine contractions leading to the whelping of puppies.

There are several methods of “timing” the fertile period of the bitch. In the past, breeders have used the change in color of vaginal discharge as well as behavioral changes such as standing, flagging, and winking, to determine when a bitch is ready to be bred. These methods can be variable in success, as some bitches have heavy or light discharge throughout their cycle, and may never stand or appear receptive behaviorally. Alternatively, the use of measuring vaginal epithelial cell cornification has also been used to estimate ovulation. The problem with this method is that the epithelial cells are changing appearance (becoming more “cornified”) in response to rising estrogen levels, which can be extremely variable among bitches. Some bitches show 100% cornification very early, while still in proestrus, whereas other may never show absolutely 100% cornification. Estrogen is a very unreliable hormone upon which to base an estimate of ovulation timing. More recently, testing of LH and progesterone has become available, which are much more reliable methods for determining the fertile period within the estrous cycle. Most importantly, one must have a veterinarian who is familiar with the reproductive system of the dog and bitch and is experienced in dealing with fertility issues.

Infertility more often results from improper ovulation timing than from any other cause. If one is using fresh chilled or frozen semen, which further reduces the fertility of the sperm, then proper ovulation timing is crucial. Ovulation occurs approximately two days after the LH surge and the eggs then undergo several divisions over a 2 to 3 day period to mature into an egg which can be fertilized. The eggs will only live for a short period of time if not fertilized and then implanted into the uterine lining. Therefore, the most fertile period of the bitch is approximately 4-6 days after the LH peak. Depending on the fertility of the stud dog, and viability of his sperm, correct timing is often imperative to achieving pregnancy.

While improper ovulation timing is the most common cause of female infertility, there are other less common conditions which can influence the chance of a successful pregnancy. These include chromosomal abnormalities such as XO syndrome and XX sex reversal, female pseudohermaphroditism, excessively long or short intervals between estrous cycles, luteal insufficiency, cystic ovaries and/or uterus, infectious disease such as the herpes virus and brucellosis, low thyroid hormone, and improper nutrition or body condition.

The two most common chromosomal abnormalities which result in the outward appearance of a female but with resulting infertility are XO syndrome and XX reversal. XO syndrome occurs when one of the sex chromosomes is missing. Because there is only one X chromosome, and no Y chromosome, the puppy will appear female, but will not develop a normal uterus or ovaries, and will never have an estrous cycle. In XX sex reversal, the puppy will appear female or possibly as an abnormal male, but will have a mixture of ovarian and testicular tissue or have undescended testicles. Depending on the combination of chromosomes and reproductive organs, these dogs may also be termed true hermaphrodites, in which they can appear as one sex or the other, but contain internal reproductive organs representative of both sexes. There is evidence that XX sex reversal is a familial disorder in some breeds of dogs.

Pseudohermaphroditism, on the other hand, occurs when the puppy has an XX chromosome composition, normal ovaries, but ambiguous external genitalia. External appearance can range from mild masculinization such as an enlarged clitoris, to a nearly complete male appearance. The appearance of pseudohermaphrodites within a litter is usually due to the presence of androgens during a sensitive period of fetal development, either from internal or external sources. Most commonly, the administration of certain steroid hormones during pregnancy can be attributed to the development of pseudohermaphrodites. Whatever the cause, pseudohermaphrodites are unable to reproduce.

While pseudohermaphrodites can have abnormal or absent "heats", even normal bitches can have excessively long or short periods between estrous cycles. When the period between estrous cycles is excessively long, it is important to look for underlying disease or other cause to determine if ovulation is not occurring due to inadequate body reserves. For instance, a bitch who is not receiving the proper nutrition will often not cycle at all, as her body is in a "starvation" mode and any unnecessary body functions are shut down, including ovulation, lactation, etc. Other possibilities are low thyroid hormone levels, which has been correlated with abnormally long estrous cycles and inability to maintain pregnancy. Very short intervals between estrous cycles, on the other hand, may lead to insufficient time for adequate regeneration of the lining of the uterus, which is necessary for implantation and nutrition for the fertilized eggs.

Other influences on implantation of eggs and maintenance of pregnancy include luteal progesterone production as well as the health of the ovaries and uterus. High progesterone levels are maintained throughout pregnancy by the presence of the corpus luteum. If there is luteal insufficiency, and progesterone levels fall below a certain level, spontaneous abortion or resorption can occur. Physical abnormalities such as cystic ovaries and/or uterus can also result in abortion or resorption. If the bitch has cystic ovaries, or small, fluid-filled pockets within the ovarian tissue, ovulation may be disrupted or prevented, despite proper hormonal levels and signals. Alternatively, a cystic uterus may result in prevention of implantation within the uterine lining, or successful implantation but inhibition of growth of the fetuses once they enlarge enough to encroach on the cystic areas. The administration of megestrol acetate, also known as ovaban or "check drops" has been correlated as a possible cause of cystic ovaries and/or

uterus. While these are relatively uncommon causes of infertility, they should be considered in a bitch that is unable to conceive despite ruling out more common underlying causes.

Infectious disease can also cause spontaneous abortion or resorption during a pregnancy. The two most common causes are the canine herpes virus, and brucella canis. A high bacterial population in the cervical area has also been theorized to reduce fertility, but is considered a less common cause. It is also generally difficult to determine if a positive bacterial culture is the result of normal flora which lives within the vaginal vault, or a more pathogenic problem. There is testing available for the herpes virus as well as brucella canis, simply requiring a blood serum sample. The brucella canis test is relatively inexpensive and common for bitch and stud dog owners to request, while the herpes virus test is more expensive and less commonly understood. It should be noted that the herpes virus is an extremely ubiquitous organism and its presence as a factor in infertility needs to be considered under specific circumstances.

When breeding a bitch it is recommended to have a complete blood count, serum chemistry profile, urinalysis, negative brucella test, thyroid panel, and appropriate genetic testing for the breed to be completed. One should consider breeding an inexperienced bitch to an experienced stud dog with a known fertility history. Keep in mind that as a bitch ages, her fertility will decrease. It is generally accepted that a bitch is most fertile between the ages of 2-6 years in small breed dogs, 2-5 years in medium to large breed dogs, and somewhat less than that in giant breeds. It is not recommended to breed a bitch with a questionable fertility history, either due to inexperience, age, or other causes, with either fresh chilled or frozen semen. If a bitch appears to be infertile, and proper ovulation timing and sperm fertility has been determined, then other testing including ultrasound, karyotyping, serological testing, etc. can be used as tools to rule in or out other possibilities for infertility problems mentioned in this article. Overall, if a brood bitch owner is absolutely determined to breed an apparently infertile bitch, working with a veterinarian who is experienced in canine reproduction is imperative.

Once a bitch has been bred at the proper time and pregnancy has been determined and appears to be progressing normally, there are several methods to monitor the fetuses. Pregnancy can be detected through a variety of methods, including palpation of the ~1cm uterine swellings at about 3 weeks, ultrasound detection of the amniotic cavities also at about 3 weeks, relaxin hormone detection at about 4-5 weeks, and lastly x-ray of skeletal components at about 7 weeks or later. Pregnancy lasts about 64 to 66 days post-LH surge, approximately 63 days from appropriate breeding. There are blood tests available to monitor progesterone levels, serial ultrasounds which can be performed to monitor development and fetal monitors which can be attached to the bitch to detect changes in fetal heart rate, uterine contractions, etc. Overall, veterinary medicine has become much more advanced concerning canine reproduction, and many infertility and pregnancy issues can be overcome with proper advice and medical knowledge.