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Use of Supplemental Progesterone in Management of Canine Pregnancy (21-Apr-2001)

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Summary

Hypoluteoidism is a hypothesized cause of pregnancy loss in bitches. Diagnosis requires demonstration of an abnormally low serum progesterone concentration and ultrasound verification of presence of viable fetuses. Treatment is with injectable progesterone in oil or a synthetic oral progestogen; treatment must be withdrawn about 60 days post-ovulation to allow spontaneous parturition of normal puppies.

Endocrinology of Canine Pregnancy

In the bitch, estrogen concentration in serum is high during proestrus and falls just prior to the peak in luteinizing hormone (LH) that stimulates ovulation. Serum progesterone concentration begins to rise coincident with the LH peak, reaching a concentration of 4 to 10 ng/ml on the day of ovulation [1]. Serum progesterone concentration rises variably after ovulation, peaking at 15 to 90 ng/ml by 10 to 25 days after the LH peak, and then gradually declining in the latter half of pregnancy [2-5]. Serum progesterone concentration falls abruptly to less than 1 ng/ml on the day preceding or on the day of parturition [3-8] (Fig. 1).

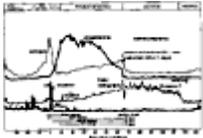


Figure 1. Diagram of typical endocrine changes during estrus and pregnancy in bitches. Adapted from Concannon et al. [5]. Note the elevation in progesterone concentration after ovulation, the peak in serum progesterone concentration at mid-gestation, the gradual decline in late gestation and the abrupt fall in serum progesterone just prior to parturition. When monitoring serum progesterone concentration during pregnancy, the normal concentration relative to the stage of gestation must be borne in mind. - To view

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Progesterone is required for pregnancy maintenance. During pregnancy, progesterone stimulates development of endometrial and mammary tissue, and prohibits uterine contractions. Dogs are dependent on luteal progesterone throughout pregnancy. Pregnant bitches ovariectomized late in gestation (days 40 to 55) abort within 2 to 2.5 days of surgery unless supported with exogenous progesterone [9,10]. The canine corpus luteum is autonomous early in pregnancy but requires support of luteotrophic hormones, such as LH and prolactin, after about day 18 of pregnancy. Pregnant bitches hypophysectomized days 4 to 10 of gestation do not show changes in serum progesterone concentrations [11,12]. Pregnant bitches hypophysectomized after day 18 of gestation, treated with serum containing antibodies against LH, or treated with prolactin inhibiting drugs, show a permanent decline in serum progesterone concentration [11-13].

Hypoluteoidism is premature failure of progesterone production by luteal tissue with a subsequent decrease in serum progesterone concentration and pregnancy loss. This is a hypothesized cause of pregnancy loss in dogs that resorb, abort or whelp prematurely with no discernible infectious or non-infectious abnormalities of the pregnancy. Supplemental progesterone can be used to counter this condition.

Case Reports

Causes of hypoluteoidism have not been defined in most cases reported. There is one report of secondary luteal insufficiency in a Great Dane with a pituitary defect [14]. One case managed by the author and two reports of apparent primary luteal insufficiency from the veterinary literature are described.

Case One - A two year old, intact, female Silky terrier presented with a history of recurrent abortions on days 49-51 of pregnancy, with birth of non-viable pups that were demonstrated to be premature at necropsy [15]. There was no evidence of viral or bacterial infection, toxoplasmosis or chlamydiosis, and serology for *Brucella canis* was negative. Serum progesterone concentrations were monitored during pregnancy with an in-house enzyme linked immunoassay (ELISA) kit, and were

demonstrated to fall to less than 5 ng/ml by day 50 of pregnancy. The bitch was treated with injectable progesterone in oil on days 50 and 53, and whelped one mummified pup and two live pups on day 57.

Case Two - A two year old, intact, female Rottweiler presented for signs of impending parturition seven days before expected parturition [16]. Whelping date had been projected as 63 days from ovulation day, assessed by measurement of serum LH (peak value two days prior to ovulation) and 57 days after onset of cytologic diestrus (six days after ovulation). The bitch had tested negative for antibodies against canine brucellosis at the time of breeding. On presentation, she had a low body temperature, was restless, and had milk leaking from engorged mammary glands. Serum progesterone concentration, measured with an in-house ELISA kit, was less than 1 ng/ml. Ultrasound revealed eight viable fetuses. The bitch was treated with a synthetic progestogen orally for 4 days. Her body temperature returned to normal and all signs of impending parturition resolved in the first 24 hours of treatment. She whelped eight live pups on the expected date.

Case Three - A five year old, intact, female Bichon Frisé presented with a history of recurrent premature labor and delivery of stillborn puppies. No specific cause had been identified for premature delivery and death of the pups. Serial serum progesterone concentrations were used to assess day of ovulation and the bitch was bred on the second and third day after ovulation. Serum progesterone concentrations were monitored during pregnancy by radioimmunoassay (RIA). Peak progesterone concentration in serum was 24.3 ng/ml, 13 days after ovulation. Ultrasound revealed pregnancy with three viable fetuses 31 days after ovulation. Serum progesterone concentration gradually declined, with concentrations of 8.1 ng/ml 35 days post-ovulation and 4.0 ng/ml 46 days post-ovulation. The bitch was treated with a synthetic progestogen orally days 46 through 60. She whelped one stillborn pup, one pup that died immediately after birth, and two live pups 62 days after ovulation.

Diagnosis of Hypoluteoidism

Hypoluteoidism should be diagnosed by demonstration of an abnormally low progesterone concentration in serum during diestrus and ultrasound verification of presence of normal viable fetuses in the uterus. Inflammatory conditions of the uterus, such as *Brucella canis* and other bacterial infections, may be able to stimulate release of prostaglandin from the endometrium with subsequent lysis of luteal tissue and decreasing serum progesterone concentration. In dogs with an abnormality of pregnancy, decrease in serum progesterone concentration alone does not imply primary luteal insufficiency. Non-infectious causes of pregnancy loss include genetic abnormalities of the puppies, systemic diseases of the bitch, and trauma [17]. Infectious causes of pregnancy loss include *Brucella canis*, *E. coli*, or *Streptococcus* sp. infections of the uterus, toxoplasmosis, and canine herpesvirus infection [17]. If the bitch has an abnormally low serum progesterone concentration and is carrying non-viable fetuses, supplementation with progesterone is not recommended as it inhibits uterine contractions and prevents the bitch from expelling the abnormal pregnancy [18]. Progesterone supplementation should not be instituted without demonstration of abnormally low serum progesterone concentrations and fetal viability. Administration of progesterone to bitches with normal concentrations of progesterone may induce masculinization of female fetuses [19]. Serum progesterone concentration can be measured either with an in-house assay or by a commercial laboratory using RIA or chemiluminescence. In-house progesterone assays are semi-quantitative, with most yielding results of low (less than 1 ng/ml), rising (1 to 5 ng/ml) or high (greater than 5 ng/ml). Although these kits are relatively inaccurate in the low and mid ranges, they are very accurate in the high range, and are useful in diagnosed bitches with serum progesterone concentrations of 5 ng/ml or less [20]. Their use in estimating the time of ovulation [23] has been described in detail separately. (see Kustritz, 2001) The RIA or chemiluminescence assays used by commercial laboratories are more accurate than the in-house assays but have a longer turn-around time.

Table 1. Schemes for the monitoring of serum progesterone concentration for diagnosis of hypoluteoidism in bitches, using either radioimmunoassay or semiquantitative in-house assay.	
Type of Progesterone Assay Used	Monitoring Scheme
Commercial assay (radioimmunoassay [RIA] or chemiluminescence) - Quantitative result	Draw blood samples weekly beginning 5 to 7 days after last breeding. If serum progesterone concentration falls to less than 10 ng/ml, draw samples every 2 to 4 days. Begin supplementation if serum progesterone concentration falls to less than 5 ng/ml before days 58 to 60 from ovulation
In-house assay (enzyme linked immunosorbent assay [ELISA]) - Semi-quantitative result	Draw blood samples twice weekly beginning 5 to 7 days after last breeding. Begin supplementation if serum progesterone concentration falls to less than 5 ng/ml (mid or low range) before days 58 to 60 from ovulation.

For diagnosis of hypoluteoidism in a bitch presented with a history of apparent premature luteolysis, the author recommends monitoring of serum progesterone concentrations using a commercial laboratory with an assay accurate at all concentrations. Samples are drawn weekly beginning 5 to 7 days after the last breeding. If serum progesterone concentration falls to less than 10 ng/ml, samples are drawn every 2 to 4 days. Supplementation with exogenous progesterone is begun if serum progesterone concentration falls to less than 5 ng/ml before day 58 to 60 from ovulation. If the less accurate in-house ELISA assay is used to monitor serum progesterone, samples should be drawn twice weekly beginning 5 to 7 days after the last breeding, and supplementation begun if the serum progesterone concentration falls into the mid or low range before day 58 to 60 from ovulation.

Treatment of Hypoluteoidism in Bitches

Hypoluteoidism can be treated with either injectable natural progesterone or orally active synthetic progestogens. Progesterone in oil is an injectable depot preparation, given intramuscularly at a dose of 2 mg/kg every 72 hours [15, 21]. This is a native form of progesterone that can be measured with either in-house or commercial laboratory assays to assess adequacy of supplementation.

Altrenogest (Regumate, Hoechst-Roussel) is a synthetic progestogen marketed for horses. It is given daily, orally, at a dose of 0.088 mg/kg (0.2 cc/10 pounds BW) [18]. This is a non-native progesterone that cannot be measured with either an in-house or commercial laboratory assay to determine adequacy of supplementation. In a study evaluating use of altrenogest at the above dose in pregnant bitches ovariectomized at mid-gestation, three of three carried the pregnancy to term, and two of three whelped live pups; one bitch lost all the pups due to dystocia [18]. Milk production was poor in the first 2 to 3 days of lactation in those bitches. Milk production was not abnormal in either the Rottweiler or Bichon Frisé described earlier. It is imperative that progesterone supplementation be withdrawn 2 to 3 days prior to expected whelping. If serum progesterone concentration is artificially maintained at a high concentration beyond the expected whelping date, the bitch will not go into spontaneous labor and the pups may be stillborn [10]. Time until whelping may be projected using breeding management information, or radiographs. Bitches whelp 62 to 64 days from ovulation. Ovulation occurs about 2 days after a progesterone concentration of 2 ng/ml; serum progesterone concentration is 4 to 10 ng/ml on ovulation day. The abrupt onset of cytologic diestrus occurs 6 days post-ovulation. Timing from accurate assessment of ovulation day is the most precise projection used in bitches. On a good quality lateral radiograph, assessment of distal extent of mineralization also may be used to predict gestational age of the puppies; this method is less accurate (Table 2).

Radiographic Finding	Days Ante Delivery *
Scapula, humerus, and femur seen	17 (15-18) d
Radius, ulna, and tibia seen	11 (9-13) d
Pelvis seen	11 (9-13) d
13 pairs of ribs countable	11 (9-13) d
Caudal vertebrae, fibula, calcaneus, paws seen	5 (2-9) d
Teeth seen	4 (3-8) d

* Numbers in parentheses indicate range

It is important to realize that the initiation of therapy after day 58 after the estimated day of ovulation can be problematic if the timing of ovulation was not done accurately, since therapy needs to be stopped by day 62. The form of progesterone or progestin supplementation is also important in this regard. Serum progesterone or progestogen concentration due to supplementation declines within a day of withdrawal of an oral supplement, and within three days of the final injection of progesterone in oil.

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