The Milk Progesterone Test and Its Applications in Dairy Cattle Reproduction

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Progesterone concentrations in serum and milk are closely related to the dynamics of the reproductive cycle and status of cows. Consequently, this hormone and its methods of detection have been studied for many years in bovine reproduction. Prior to recent advances in immunodiagnostics, measurements of serum or milk progesterone were laboriously performed using radioimmunoassay (RIA).

This method is accurate but a number of factors including cost, need for specially trained personnel, and radioactive waste disposal problems make this test unacceptable for widespread commercial use. However, today's technology has overcome these problems with tests such as Open Alert/Bovi-Pro 21 and now progesterone assay can be performed rapidly and easily on-farm at minimal expense.

CYCLIC PATTERN OF PROGESTERONE

Progesterone is a hormone produced by the corpus luteum (CL) of the ovary and its levels in serum and milk increase or decrease in response to the maturation or regression of this ovarian structure. The developmental sequence of the CL goes roughly like this: several follicles proceed to develop on the ovary; however, as the cow nears estrus one follicle begins to predominate and progressively grows and matures. Following a surge of luteinizing hormone (LH) from the pituitary gland the follicle ruptures releasing its egg (ovulation). Following ovulation blood fills the space voided by the egg. Over a period of 4 to 8 days luteal cells present in this blood filled space organize and develop giving rise to a CL. The CL secretes progesterone which enters the bloodstream and subsequently the milk. If the cow becomes pregnant the CL is retained on the ovary and continues to secrete progesterone which serves to maintain pregnancy. If, on the other hand, the cow fails to become pregnant the CL gradually regresses (shrinks) and ceases to produce progesterone, thereby allowing the cow to return to estrus approximately 21 days after the previous estrus cycle.

It is important to understand that the level of progesterone in milk and serum increases slowly after ovulation. Under most circumstances the level of progesterone will be low and slowly increasing for the first 4 to 6 days following ovulation. Maximum concentrations of progesterone will not be reached until sometime between days 10 to 17 of the cow's reproductive cycle. In the non-pregnant cow, levels
of progesterone will begin to diminish fairly sharply about day 18 or 19. At about this same time estrogen levels are beginning to increase as the cow begins to cycle through another estrus period.

**INDICATIONS AND APPLICATIONS FOR MILK PROGESTERONE**

There are several potential uses for milk progesterone by dairymen and veterinarians. Some of the more important uses are discussed below.

**Diagnosis of Non-Pregnancy**

Numerous studies indicate that the accuracy of early pregnancy diagnosis by milk progesterone is only about 80%. Reasons for this include: variation in estrus cycle length between cows, estrus detection errors, uterine disease (pyometra), ovarian dysfunction (luteal or follicular cysts), and early embryonic mortality, to name a few. In short, the reliability of milk progesterone for the diagnosis of pregnancy is not satisfactory in itself and should be confirmed by palpation of the uterine tract at 40 or so days following breeding. However, with a series of samples taken at day 0 (the date of insemination) and days 21 and 24 the accuracy of making an early diagnosis of non-pregnancy approaches 95 to 100%. Therefore, milk progesterone as a tool in the consideration of early pregnancy status, should be used for purpose of determining the non-pregnant state as opposed to confirmation of the pregnant state. The advantage of this early confirmation of non-pregnancy prevents the further loss of early breeding opportunities.

**Confirmation of Estrus**

On frequent occasions cows may exhibit questionable signs of estrus such that a decision to breed is not clear. In fact, approximately 5% of pregnant cows will show signs of estrus even though pregnant. Milk progesterone can be used as a means to establish whether the behavior observed is consistent with a cow in or near estrus. Milk samples which show a high level of progesterone are not likely in heat and should be watched closely and retested at a later point.

Numerous studies have demonstrated that on average 15 to 20% of dairy cows presented for insemination are not in heat at the time of breeding. Estrus detection error rates can be as high as 50% or more on some farms. Consequently, herds experiencing infertility as evidenced by low conception rates and excessive repeat breeder cow problems may want to consider evaluating their estrus detection accuracy. This can be performed by simply saving a milk sample from cows presented for insemination over some designated period (say 2 weeks to 1 month). If more than 10% of the cows inseminated during that period of time had high levels of progesterone it may be indicative of an estrus detection error problems.

**Reproductive Problem Cows**

Reproductive problem cows are those that fail to cycle or conceive within the normally expected time post calving. In some herds a cow becomes classified as a reproductive problem as soon as she goes beyond 100 days postpartum without a breeding. The causes are several and in some herds it is simply missed heats that allow some cows to become classified as reproductive problems. However, for cows suffering uterine and/or cystic ovarian disease prompt diagnosis and treatment can save precious time and expense in re-breeding cows postpartum.

Milk progesterone assay can be a useful diagnostic aid in these problem cows for discernment of uterine disease (pyometra) and in the process of distinguishing a luteal from a follicular cyst. Studies show that accuracy of diagnosis of ovarian cysts by palpation is only about 65 to 75%. Therefore, by confirming the diagnosis one can better choose a course of therapy. After therapy is instituted the condition can be monitored to determine if the desired response is being achieved.

**Embryo Transfer Programs**

Embryo transfer programs often call for frequent examination and testing of donors as well as recipients. Proper synchronization of embryo recipients is obviously a vital step in the overall success of this procedure. Interestingly, studies indicate that the accuracy of determining mature corpora lutea by palpation alone is only 75 to 80%.
Consequently, the selective use of progesterone assay on cows with palpably questionable CLs could potentially improve the overall success of this procedure by insuring the reproductive status of recipients prior to the transfer of embryos.

**High Cost Semen**

Because of the potential for estrus detection errors, milk progesterone assay, in addition to observation of the appropriate estrus behavioral signs, is a useful estrus confirmation procedure. Testing prior to insemination, therefore, would not only prevent the waste of expensive semen but may prevent an abortion as well.

**High Producing Dairy Cows**

The stress of high milk production and associated lag in dry matter intake in early lactation is known to adversely affect cyclic activity in dairy cows. Further, negative energy balance and the hormonal dynamics of the early postpartum cow have been shown to reduce the intensity of estrus behavior. Consequently, the high producing cow is a challenge to catch in heat and successfully breed. Since the outward signs of estrus may be unclear or confusing in these cows, milk progesterone may be helpful in discerning estrus activity from non-estrus behavioral activity.

**Assistance in Breeding Programs During Times of Climatic Stress**

Environmental stress has been shown to have a dramatic effect on reproductive efficiency. Heat stress, in particular, is responsible for severely reduced conception, increased rates of early embryonic death, decreased length and intensity of estrus behavioral activity, and lowered calf birth weight. Until better strategies for the use of milk progesterone under conditions of environmental stress are defined, its role will be primarily associated with those described above.

**SUMMARY**

Through advances in biotechnology and immunodiagnostics, systems for the detection of progesterone in milk and serum are widely available commercially. Their value depends upon the operators understanding of the normal cyclic patterns of progesterone in the dairy cow. They do not pinpoint the time at which breeding should take place nor do they diagnose pregnancy reliably. However, when used sensibly in recognition of the limitations of the procedure, they are a valuable asset in reproductive management.