

# The Use of Radioimmunoassay to Monitor Reproductive Status of Dairy and Beef Cattle and The Effect of Farm Size and Management System on Reproductive Performance

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## Summary

A Study was conducted to improve breeding efficiency of dairy and beef cows on Cheju Island. Milk progesterone and plasma progesterone profiles were determined using radioimmunoassay techniques during the oestrus cycle, onset of oestrus and during the gestation period. Progesterone concentrations in milk were very low during the oestrus with a mean value of 2.4 nmol/l and 4.6 nmol/l at the onset of oestrus but gradually declined 4 to 9 hr of oestrus. In the cyclic cow, milk progesterone concentrations in the early part of the cycle, ranged from 3 to 5 nmol/l and increased from 6.0 to 11.6 nmol/l on day 13 to 21 of cycle. Milk progesterone concentrations of pregnant cows ranged from 5.0 to 40 nmol/l during the gestation and from basal line to 3.5 nmol/l for non-pregnant cows. Plasma progesterone concentrations of beef during pregnancy period ranged from 3.0 to 33 nmol/l and basal levels ranged from 0 to less than 2.5 nmol/l for non-pregnant beef cows.

## Introduction

Cheju Island includes 110,000 hectares of native grassland which comprises 51% of its total land mass. The island is an important sector for beef production in the Republic of Korea. The grassland area has long provided a tremendous potential for the grazing of native cattle. To increase the

animal productivity and to fulfill the rapid increase of domestic demand for dairy and beef products, dairy and beef projects were initiated in 1970's. Therefore, the dairy and exotic beef industries of the island are comparatively new, compared to the traditional native cattle production.

The first dairy cattle introduced were 188 Holstein heifers from Australia in 1977, to

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establish dairy farming on the grassland belt. Additional dairy herd were introduced from overseas in successive years for dairy production. A total of 1,531 dairy cattle and 13,141 exotic beef cattle were introduced to the island by 1987. However, breeding performance of the introduced beef and dairy cattle has not been optimum. By 1987, total number of beef and dairy were 41,923 and 1,531 respectively. This slow increase in the cattle population was affected by several factors: 1) slow adaptation of cattle to their new environment, 2) differences in cattle management practices and 3) low fertility.

Suboptimal fertility is a major source of loss to efficient dairy and beef production. Another component may be the extended interval between parturition and subsequent conception. Kim et al. (1986) reported that age at first conception of introduced dairy heifers ranged from 18 mo to 20 mo, and Kim et al. (1984) reported the age at first calving of exotic beef heifers ranged from 41 mo to 47 mo. In a study of the reproductive status and calving rate of Cheju native cattle, Kim et al. (1983) indicated that age at first insemination was 28.7 to 31.6 mo and that age at first calving ranged from 38.2 to 40.5 mo. Thus the breeding performance of native, exotic beef and dairy cattle in Cheju all are poor. It is necessary to improve breeding efficiency of cattle on Cheju island, and radioimmunoassay techniques are a potential tool to enhance reproduction.

The determination of progesterone concentration in milk and plasma has frequently been used to study the corpus luteum in dairy and beef cows (Gimther et al., 1974; Hoffman et al., 1976; Dobson et al., 1975; Wishart et al., 1975; Lamming and Bulman, 1976). Lukaszwska and Hensel (1980) suggested that progesterone concentration in plasma

rose more rapidly after a proper insemination than an improper one. Also, it has been suggested that the probability of conception is related to concentrations of plasma or milk progesterone during the ovarian cycle (Rosenberg et al., 1977). Concentration of progesterone in fat-free milk (Pope et al., 1976) and in whole milk (Dobson et al., 1975; Bulman et al., 1978) were closely correlated with those in plasma. Therefore the milk assay could become a useful technique for field work concerning reproductive status and pregnancy testing in cattle (Bulman et al., 1978). The objective of this study was to measure milk progesterone profiles during the estrus cycle and pregnancy of dairy cattle and plasma progesterone concentration of beef cows during pregnancy, under farm conditions, for monitoring diagnosis of pregnancy.

## Materials and Methods

Two cyclic four-year-old Holstein cows from Cheju Experiment Station were used for progesterone profile study during the oestrus period (48 hrs). Two Holstein cows (3-year-old) from the Livestock Development Center participated in the study of progesterone level during the oestrus cycle (30 days). Cows were individually fed, and the hay and concentrate diet was supplied on the basis of milk yield of cows. Milk samples were taken every hour from the onset of oestrus until the end of the oestrus period. In order to measure progesterone levels during the oestrus cycle, milk samples from two cows were taken twice daily from the day of artificial insemination (AI) for 24 d. A total of 178 milking Holstein cows in local dairy farms and 100 beef cows (Angus, Hereford and crossbred) in large scale

beef farms were used to monitor progesterone profile for pregnancy diagnosis.

Twenty milliliter milk samples were collected from individual cows and stored at  $-5^{\circ}\text{C}$  until assay. Blood samples from beef cows were taken from the jugular vein by veinpuncture into heparinized evacuated tubes. Plasma samples obtained immediately after centrifugation were stored at  $-15^{\circ}\text{C}$  until analysis. Pregnancy diagnosis by rectal palpation was conducted by veterinarians. Data records of subsequent breeding or estrus were obtained from dairy cattle owners. For laboratory assay, fat-free milk progesterone and plasma progesterone were determined using the FAO/IAEA progesterone solid-phase RIA kit.

## Results and Discussion

### 1. Progesterone profiles during oestrus

The results of analysis of milk progesterone from two oestrus cows are illustrated in Fig. 1.

Progesterone concentrations in milk were very low during the oestrus period with a mean value of  $2.4 \text{ nmol}/\ell$ . Progesterone concentration was  $4.6 \text{ nmol}/\ell$  at the onset of oestrus but gradually declined at 4 to 9 hr of oestrus. Another increase in progesterone level occurred at 10 to 12 hr after onset of heat and was then maintained at the plateaued level. These results are similar to those of Dubson et al. (1975) who reported

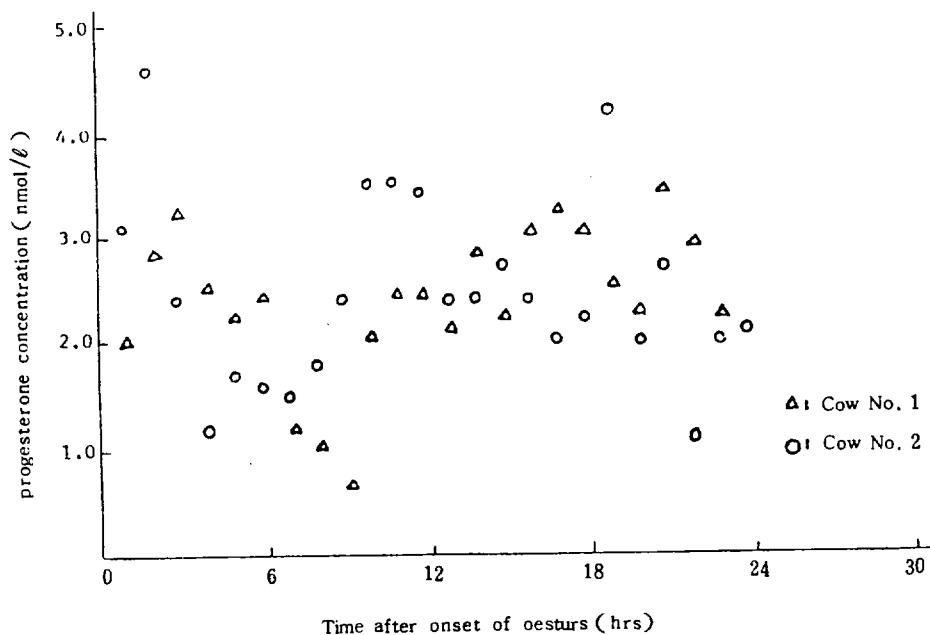


Fig. 1. Milk progesterone profile of dairy cow during oestrus.

that plasma concentration of progesterone was very low ( $0.3 \pm 0.07 \text{ ng/ml}$ ) around oestrus and that it increased with corpus luteum development. Higher progesterone concentrations in the early stage of oestrus ( $4.6 \text{ nmol/l}$ ) may be due to the high progesterone concentration before the oestrus. Milk progesterone values of the present study also agreed with the results obtained by Bulman et al. (1978).

## 2. Progesterone profile during the oestrus cycle

Progesterone profiles in morning and evening milk from two pregnant cows are shown in Fig. 2.

In the early part of the cycle, from d 5 to d 10, values ranged from 3 to 5  $\text{nmol/l}$ . From d 13 to 21, concentrations increased from 6.0 to

11.6  $\text{nmol/l}$ . Also, from d 21 to 28, values were 8.4 to 13.8  $\text{nmol/l}$ , progesterone concentrations were higher than early in the cycle. Hensel (1981) reported higher progesterone concentrations in pregnant Holstein cows at d 10 to 18 post-AI, compared to those of non-pregnant cows. Mean progesterone concentrations for pregnant and non-pregnant cows were similar until d 13. After that, the concentrations for the non-pregnant group declined, whereas those of pregnant cows continued to increase (Bulman et al., 1978). It was confirmed by rectal palpation at d 60 after insemination that the two cows examined were pregnant. Therefore it was not possible in this experiment to characterize the progesterone profile of the non-pregnant cow. Pope et al. (1976) found a three-fold increase in both fat and milk progesterone concentrations in the fore

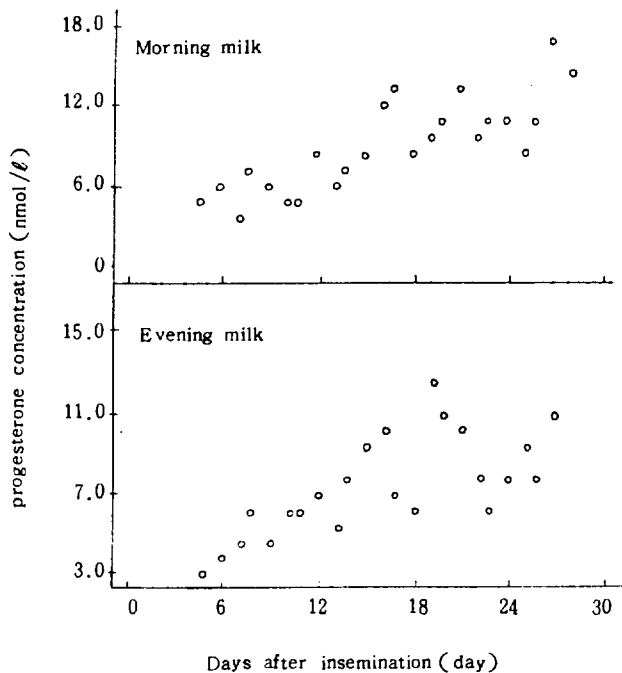


Fig. 2. Milk progesterone profile of dairy cow during early pregnancy.

milk compared to that of stripping milk. Present data indicated that higher progesterone concentrations were found in the morning milk than in evening milk. However, the concentrations of progesterone in morning and evening milk were highly correlated. Based upon present findings, it is suggested that milk progesterone concentrations above 10 nmol/ℓ at d 20 after insemination are diagnostic of pregnancy in Cheju Hostein cows.

### 3. Milk progesterone concentrations of pregnant dairy cows during the gestation period.

Distributions of milk progesterone concentrations of pregnant cows during gestation are shown in Fig. 3.

Among 178 milking cows examined in this study, 81 cows (45.5%) were pregnant and 97 cows had not conceived. Milk progesterone concen-

trations of pregnant cows ranged from 5.0 to 40 nmol/ℓ during gestation and from basal line to 3.5 nmol/ℓ for non-pregnant cows. However, there was no consistency in progesterone levels according to stage of gestation. Approximately 31% of pregnant cows showed 5 to 10 nmol progesterone per ℓ of milk, 23% showed 10 to 15 nmol/ℓ progesterone concentration in the milk tended to increase between d 50 and d 150 of gestation and gradually decrease toward the end of gestation. Considerable work has been done on progesterone level during pregnancy, but the major emphasis has been on pre and post-partum changes. Cheu et al. (1977) found that progesterone levels were sigmoid during pregnancy. Progesterone level during pregnancy in this study were slightly higher than those of Cheju native cows, as previously reported by Choong et al. (1986). Present data of milk progesterone from 178 dairy cows indicated that pregnancy could be

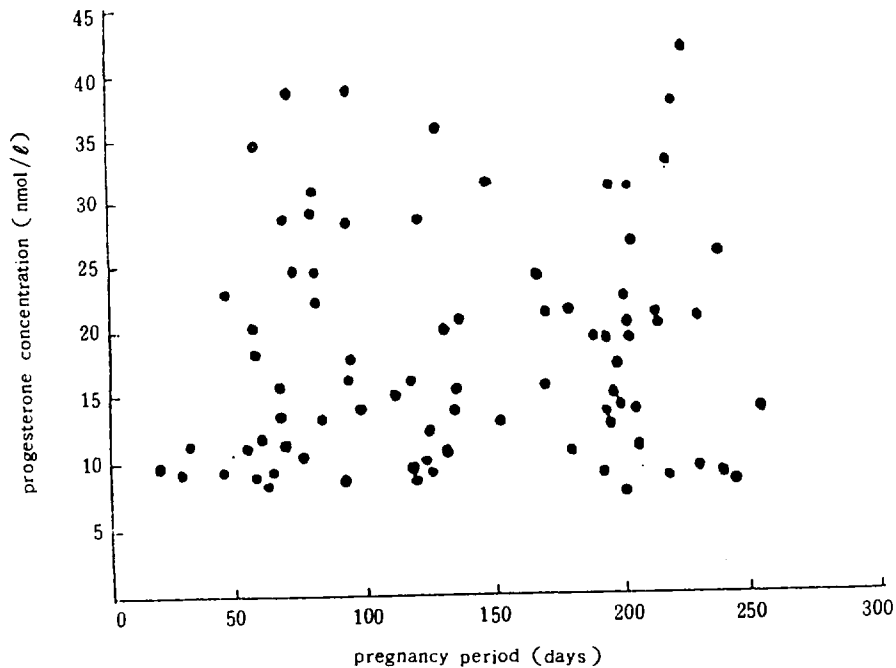


Fig. 3. Distribution of milk progesterone concentration during pregnancy period.

diagnosed using milk progesterone concentrations during gestation with above 8 nmol/l progesterone in milk.

#### 4. Plasma progesterone concentrations of beef cows during pregnancy period

Distribution of plasma progesterone concentrations of cows are presented in Fig. 4.

Of the 100 beef cows examined at the commercial beef farm, 55 cows were pregnant and 45 cows had not conceived. Pregnancy was confirmed by rectal palpation at the end of grazing season. Large variations were observed in the day of conception between pregnant cows. Plasma progesterone concentrations of beef cows were slightly lower than those of milk progesterone of pregnant dairy cows. Plasma progesterone concentrations of pregnant cows ranged from 3.0 nmol/l to 33 nmol/l, but basal levels

ranged from 0 to less than 2.5 nmol/l for non-pregnant cows. Approximately 35% of pregnant cows showed plasma progesterone concentrations of 5 to 10 nmol/l, 25% showed 10 to 15 nmol/l and 23% exhibited over 20 nmol/l during the pregnancy period.

Most progesterone measurements (Wishart et al., 1975; Pope et al., 1976; Lamming and Bulman, 1976; Bulman et al., 1978; Nebel et al., 1987) have been conducted on d 20 to 22 of the oestrus cycle, to evaluate ovarian activity together with early pregnancy diagnosis. However, few studies were conducted to measure progesterone during the entire pregnancy period. Randel and Erb (1971) reported that average progesterone level in the plasma of pregnant cows increased from 2 ng/ml on the day of breeding to 9 ng/ml on d 7. Levels decreased from d 42 (20 ng/ml) to d 125 (10 ng/ml) and then increased to 24 ng/ml by d 200 following a decrease to 15 ng/ml

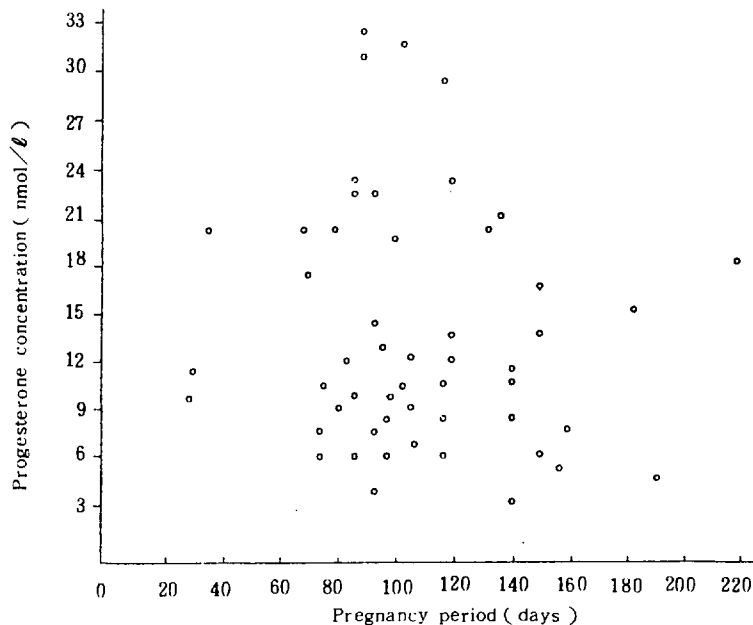


Fig. 4. Distribution of plasma progesterone concentrations of beef cows during pregnancy period.

on d 230 and 14 ng/ml on d 260. We are unable to demonstrate this with plasma progesterone due to difference in pregnancy period between cows.

Plasma progesterone profile has been widely used for the early pregnancy diagnosis of dairy and beef cows, (Dobson et al., 1975; Pope et al., 1976; Lamming et al., 1976; Bulman et al., 1978; Wishart et al., 1987). However, it is difficult to apply this technique to the beef cows under grazing conditions. For early pregnancy diagnosis using plasma progesterone measurement must be made 20 to 22 days after insemination or natural mating. It is more practical approach to diagnosis pregnancy during the gestation period, to eliminate anestrus, embryonic death and repeat breeder cows using the progesterone profile, unless synchronized oestrus is practiced in beef breeding herds.

Present data suggested that plasma progesterone levels above 5 nmol/l are indicative of pregnancy. The data indicated that plasma progesterone measurement is useful as a test of pregnancy in the beef cows under grazing conditions.

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