The Use of Radioimmunoassay to Monitor Reproductive Status of Cheju Native Cattle and the Effect of Supplementary Feeding on Reproduction.

1. Body Weight Changes, Breeding Performances and Progesterone Levels from Weaning until First Calving

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방사선 동위원소 면역기술을 이용한 濟州韓牛 營養給與의 수준이 繁殖성적에 미치는 영향

정창조 · 김중계 · 김동철

國 文 要 約

濟州韓牛의 能力向上을 위하여 營養給與 수준과 Hormone 水準의 相互關係를 규명하였다. 8頭의 濟州韓牛 未經産牛를 利用, NRC 100%와 70%給與區를 設定 增體量, 繁殖狀況 및 發情期間과 姙娠期間中의 Progesterone 의 變化를 調査하였다.

NRC 100%給與區의 增體成績은 6個月에 155kg에 比해 70%區는 137kg로서 標準區의 發育이 월등하였다. 卵巢의 크기는 標準區에서 2.1×1.6cm(左側)과 2.6×1.8 cm(右側)이였으나 70%區의 卵巢는 이보다 多少 적었다. 初發情은 標準區에서 14.6個月(體重 265kg)이였으며 初産月令은 28.9個月(體重 436kg)이였던 反面 70%區에서는 初發情 23.0個月(體重 250kg), 初産月令 38.9個月로 標準區에 比해 10個月이 지연되고 있었다. Progesterone 水準은 標準區에서 姙娠 14週와 18週에 最高에 達하여 6.4-6.5ng/ml 이였으며 24週까지 이 수준이 유지된다고 姙娠末期에는 2.1ng/ml로 減少하였다. 70%區에서는 16週까지는 標準區와 유사했으나 全 姙娠期間을 通해 Progestone 수준은 標準區보다는 떨어지고 있었다.

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Introduction

Cheju Island is situated in the Korean stratit, South of the Korean mainland. The island, dominated by Mount Halla which rises to 1950 meters, and has a subtropical climate. It possessess 110,00- hectares of undeveloped grassland which comprises 51% of the total land mass. This area of grassland provides tremendous potential for the grazing of cattle and horses.

During the 1970s there were in excess of 70, 000 head of kept on this island but by 1985 this number had been reduced to 45,000. The main reason for this reduction was the poor breeding performance of the native cattle and a dramatic increase in meat consumption.

Kim et al (1980) reported that Cheju native cattle were on average 27.8 months old at first oestrus and the average age at first calving was 38.0 months. When compared with mainland cattle and exotic beef breeds Cheju native cattle were on average 10 months later in reaching maturity. On average 174 days elapsed from calving to first oestrus when bi-annual calving cows were included. Kim et al (1983) stated that the average percentage rate of bi-annual calvers was 15.4%. This poor breeding perfomance is undoubtabley the main limiting factor to increased cattle numbers, reflected in a loss of farmers income. Poor breeding performance is always thought to be due to under-nutrition and lack of essential mineral supplmentation. Although this conjecture is probably correct it is nevertheless necessary to clarify the true reasons for poor fertility. The development of a satisfactory livestock industry depends upon solving these problems.

Much work has been done on the measurement of progesterone, estradiol, estrone and estrogen in cow blood plasma using radioimmunoassay techniques(Graverick et al., 1970; Chew et al., 1977; Humphery et al., 1983). Henricks et al., (1971) and Gonsales et al., (1975) measured the progesterone and estradiol levels in heifers prior to reaching maturity and Plotka et al. (1967), Sprague et al. (1971) and Swanson et al. (1972) measured progesterone levels during cows oestrus cycle. However in most cases (Wattmann et al., 1972; christensen et al., 1974; Ireland et al. 1984; Walterus et al., 1984) measured estradiol, together with progesterone. Chung et al. (1984) measured hormone levels from the non-pregnant period to early stage of pregnancy.

Eley et al. (1981) measured changes in estradiol and progestrone levels during pre and postpartum. Considerable work has been carried out on postpartum hormone measurements but little work has been done on prepartum hormone evaluation. Verne La Voie et al. (1981) evaluated the effect of different calf suckling methods on progesterone levels. Other research works have been done on progesterone and estradiol (Humphery et al., 1983), estradiol -17β (Kesler et al., 1979; Goodal et al., 1978) and steroid hormone of postpartum cow. However the aformentioned works only concentrated on physiological aspects and did not include nutritional levels

To improve the breeing performance of Cheju native cattle the relationship between hormone levels and nutritional levels should be investigated. This investigation was necessay to clarify the reasons for poor fertility, delay in oestrus after parturition and silent heat.

Materials and Methods.

The trial was initiated in June 1984 and 8 Cheju native calves after weaning were randomly assigned in equal numbers on two supplementary feeding levels, these were designed to provide either 100% (standard group) or 70% (restricted group) NRC. All animals in standard group were fed silage, hay and balanced ration over winter period according to NRC feeding standard. On the other hand animals in the restricted group were fed good quality hay but not balanced ration. Heifers in both groups were grazed on improved pasture from May to October. All animals were weighed twice per month at the same time during the experimental period. Rectal palpation was performed on the corpus luteum of ovaries, ovarian size and Graffian follicle size at the onset of oestrus in heifers. After parturition ovarian activity, such as the first postpartum ovulation day, corpus luteum diameter, active follicles and ovarian size were determined by rectal examination at 7 days intervals. Blood samples were collected 1400 and 1600 hrs from November 1984 until early 1986.

Blood samples for progesterone and estradiol assay were taken at the onset of oestrus cycle and thereafter every 48 hrs until the 19th day and then every 24 hrs until the end of cycle. During the heat period, blood sample were taken 3 time daily.

The collected samples were ketp at room temperature for 2-4 hrs and centrifuged at 5,000 r.p.m. after which serum was decanted and stored at -20℃, until radioimmunoassay. Progesterone and estradiol were assayed using Coat-A-Count progesterone and Coat-A-Count Estadiol, products, produced by the Diagnostic Products Corporation, U.S.A.

Results and Discussion

1. Body weights

Table 1 shows body weight changes according to different feeding levels. Mean body weight at 6 months of age was 155kg when fed 100% NRC ration but it was only 137kg when heifers received the 70% NRC ration. At 10, 15 and 20 months of age the body weight was

Table 1. Body weight change on supplementary feeding levels(kg)

Nutritional level	Cow No.	Body weight at 6	10	15	20	25 (months)
NRC 100%	1	127	173	230	317	360
	2	154	219	281	305	370
	3	172	252	230	441	552
	4	167	218	282	352	392
	Mean	155.0	215.5	255.8	353.8	411.0
NRC 70%	1	106	132	179	200	295
	2	144	144	186	193	276
	3	150	167	212	246	323
	4	148	153	165	264	287
	Mean	137.0	149.0	185.5	225.8	295.3

66, 160 and 115kg, respectively, showing that heifers fed the standard ration gained weight rapidly. There was a difference in body weight between the standard and restricted feeding groups. The body weight gain of the restricted group is similar to Sul's (1981a) data on the mainland Korean native cattle (177kg at 12 months and 229kg at 24 months). The standard group gave similar body weight gains to holstein and Korean cross breds (255kg at 12 month and 406kg at 24 months). The body weight gain at weaning of the standard group is much less than the Herford, Aberdeen Angus, Charolais, and Charolais cross Korean(Sul, 1981b). The small size of the Cheju native cattle is mainly due to the poor feeding management of the local farmer.

2. Ovarian activity

The ovarian activity judged by palpation at the first oestrus is presented in Table 2. The rectal palpation was performed when female young stock was reached at puberty.

Average size of left ovary in the standard feeding group was 2.1cm long, and 1.6cm wide and the right ovary was 2.6cm long and 1.8cm wide. However in the restricted feeding group the ovaries were found to be smaller (left 1.5 x 1.2cm, right 2.0 x 1.4cm).

Diameter of Graffian follicles followed a samilar tendency to ovarian size in the two groups. Graffian follicles in three heifers were over 10mm in diameter in the standard group, but only one heifer in the restricted group had follicles in excess of 10mm in diameter. When all heifers fed the standard ration showed normal oestrus and all conceived.

Two corpus luteum were found in the left ovary and 6 in the right, bearing a close relationship to the findings of Rajakaski(1950). The size of the corpus luteum was larger in the standard group than in the restricted group. This result was consistent with work of Apgar et al. (1975) who reported heigher ovary weights in heifers reveiving a high level of nutrition. Ovary sizes in the present trial were found to be similar to the results obtained by Chang and Kim(1982) but were much smaller than those found in Holstein (Rajakoshi, 1960 : Salisbury

Table 2. Ovarian activities judged by palpation at first oestrus

Nutritional levels	Cow No.	Corpus luteum L	Corpus luteum R	Ovarian size(cm) L	Ovarian size(cm) R	Follicle size dia. 5-10 (mm)	Follicle size (mm) 11-15	Oestrus condition
NRC 100%	1		1	1.5×1.0	1.8×1.2	1	1	+++
	2	1		2.2×1.8	3.0×2.2		1	+++
	3		1	2.3×2.1	2.8×2.0	1	1	+++
	4		1	2.6×1.5	2.8×1.8	1	1	+++
Tatal or Mean		1	3	2.2×1.6	2.6×1.8	3	3	
NRC 70%	1		1	2.0×1.5	2.6×1.7	1	1	+++
	2	1		2.0×1.3	2.5×1.5		1	++
	3		1	1.2×1.6	2.0×1.6	1	1	+++
	4		1	1.0×0.8	1.2×0.8	1	1	+++
Total or Mean		1	3	1.6×1.2	2.6×1.4	3	1	

L ; Left, R ; right, Dia ; diameter

and Van Demark et al., 1961).

The smaller ovaries found in the experimental animals were probably due to the small body size of Cheju native heifers. Swanson et al. (1972) reported 0.9 cm diameter of ovaries at first oestrus but after reccurence of three consecutive oestrus cycles, the ovaries increased their diameter to 1.2-1.4 cm. Suzuki et al. (1982) studied on the relationship between oestrus and nutritional levels and proved conclusively that low nutrition causes reduction in oestrus length and increases silent heat, leading to infertility.

3. Reproductive performance

Table 3 shows reproductive performance of heifers according to level of supplementary feeding. The first oestrus in the standard feeding group appeared at 14.6 months when the body weight was 265 kg. Age at first pregnancy was on average 19.1 months when the average body weight was 274.3 kg and the age at first calving was on average 28.9 months at a body weight of 436 kg. On the other hand when heifers were fed the restricted ration the first oestrus appeared at 23.0 months at a body weight of 250 kg. Average age at first calving was 38.9 months, which was 10 months later than the average in the standard feeding group.

With exception of one case of distocia in the standard group and a case of retaines placenta in the restricted group no other calving disorders were experienced. All calves were born healthy and in good condition.

4. Hormone levels

The progesterone concentration during pregnancy and oestrus cycle at two different feeding levels in shown in Figure 1 and 2. In the standard feeding group the progesterone level was 2ng/ml after two weeks of pregnancy and gradually increased to 4.9ng/ml at 10 weeks. The level of progesterone peaked at 6.4ng/ml at 8 weeks and this peak was maintained for 24 weeks. After 26 weeks it gradually started to fall back and fell to 2.1ng/ml at 40 weeks. The progesterone level in the restricted group up to 12 weeks was 5.0ng/ml displaying little

Table 3. Reproductive status and body weight on suppelmentary feeding levels

Nutritional levels	Cow No.	First oestrus		First pregnancy		First calving	
		Age (month)	Body weight (kg)	Age (month)	Body weight (kg)	Age (month)	Body weight (kg)
NRC 100%	1	13.3	200	13.9	207	23.3	360
	2	13.0	234	15.4	234	25.0	370
	3	12.3	291	26.0	303	36.1	555
	4	19.8	335	21.3	353	30.5	459
	Mean	14.6	265	19.1	274.3	28.9	436.0
NRC 70%	1	20.6	197	25.3	285	34.9	379
	2	26.0	257	36.0	336	45.1	360
	3	25.0	297	27.1	318	44.6	425
	4	20.2	250	21.2	256	30.8	340
	Mean	23.0	250.1	27.4	298.8	38.9	376.0

difference from the standard group but from 14 weeks the progesterone level in the restricted group dropped, and this reduction (4-4.5ng/ml) was maintained to 36 weeks when the progesterone level drops rapidly over the remaining 4 weeks until it reached 1.5ng/ml.

When the pregnancy period was divided into three equal parts there were significant difference in hormone levels in the middle scetion(12-28 weeks) but no hormone difference in part 1 and 3 between the two groups. Few studies so far have been conducted on the relationship between progesterone level and nutrition. Corah et al. (1974) reported that plasma progesterone immediately prepartum and postpartum was not affected by energy

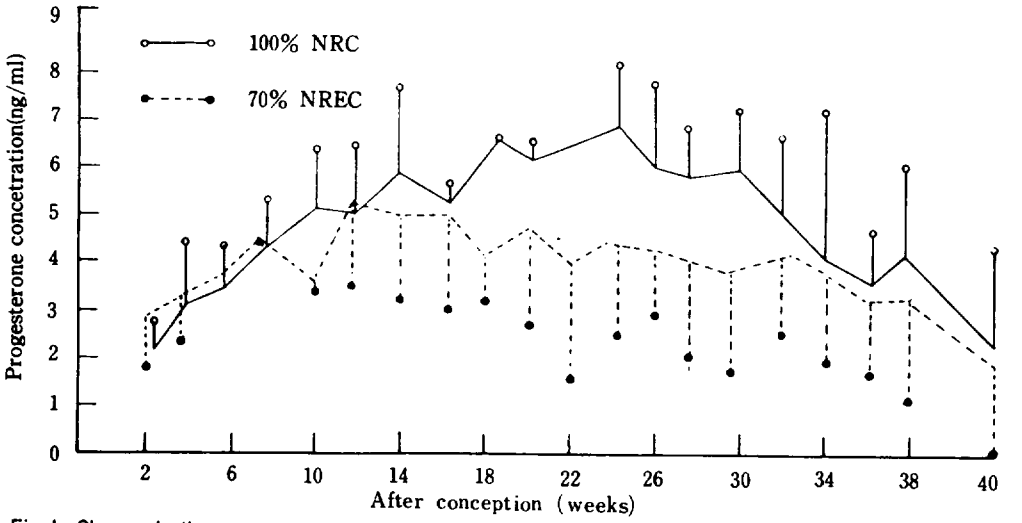

Fig 1. Change in the serum progesterone concentration in cows during pregnancy on different feeding levels

restriction 100 days prior to calving. Heifers fed high energy rations tend to have slightly higher progesterone level 3-5 days before calving.

In the prsent trial progesterone levels were measured throughout the entire pregnancy. Although Randal and Erb (1971) measured progesterone levels without regard to nutritional status their findings were nevertheless similar. Chung et al. (1978, 1984) reported that the progesterone level at 20-60 day after conception were 5.9-9.1ng/ml in Korean native cows. Henricks et al. (1971) stated that the progesterone level in heifers which returned to oestrus 18-20 days after mating was significantly lower than incalf helfers.

Considerable work has been done on progesterone levels during pregnancy but the major emphasis has been on pre and post -partum. Chew et al. (1977) found that progesterone levels were sigmoid during pregnancy. Shin et al. (1979) found higher progesterone levels (5.1-11.7ng/ml) from 30 days to 3 days before calving and these levels declined to 1.6ng/ml at 24 hours prior to calving. Progesterone levels during present trial were slightly lower than those reported by Shin et al. (1979), Randal and Erb (1971), Henricks et

al. (1971), and Ariji et al, (1974) but results obtained by Shemesh et al. (1973) and Corsh et al. (1974) were similar.

Variations in progesterone levels may be due to different hormone assay techniqued or use of Coat-A-Count for human diagnosis.

Standard and restricted groups followed similar patterns of progesterone concentration during the oestrus cycle (Fig. 2); progesterone concentration peaked 10-12 days in both groups, 2.4ng/ml in standard and 2.25ng/ml in restricted groups. These levels were maintained up to 16 days and thereafter the concentration gradually dropped towards the end of the cycle. There were no statistically significant differences in progesterone levgels between standard and restricted groups but there was variation bwtween individual animals.

Estradiol measurements were not possible due to the strong interference between bovine and human matrix in the RIA kit supplied. However as E2 levels in cows are highter than those in humans the samples should be diluted in zero matrix prior to assay.

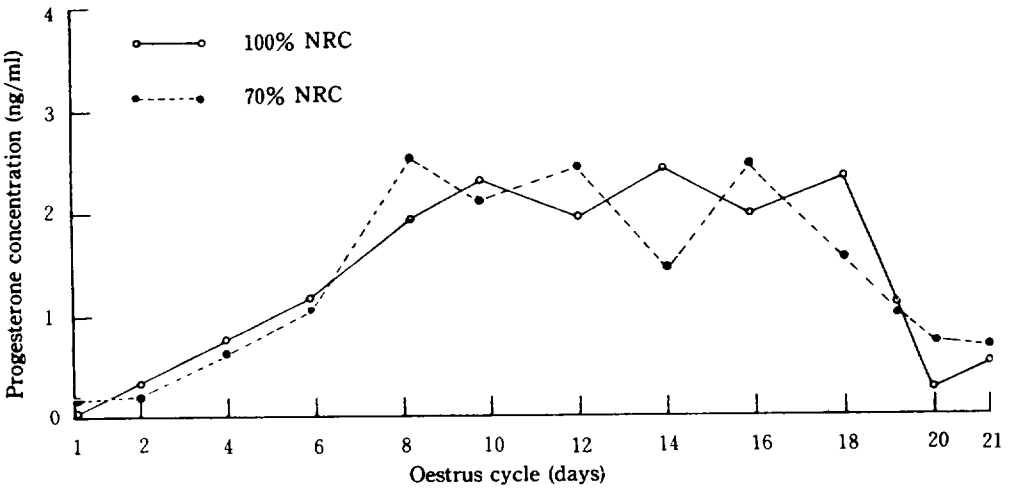

Fig 2. Plasma progesterone concentratration during oestrus cycle (ng/ml)

Summary

Studies were conducted to investigate the relationship between hormone leveles and nutritional levels for improving performance of Cheju native cattle.

In June 1984 a trial was initiated using 8 Cheju native calves after weaning, fed at two supplementary feeding leveles (NRC 100% and 70%). The body weight, breeding performence, change in progesterone level during pregnancy and estrus cycle were evaluated.

Mean body weight at 6 months of age was 155kg when fed 100% NRC ration but it was only 137kg when heifers reveived the 70% NRC ration. At 10, 15 and 20 months of age the body weight was 66,

160 and 115kg, respectively, showing that heifers fed the standard ration gained weight rapidly ($p<0.01$).

Average size of the left ovary in the standard group was 2.1 x 1.6cm and right ovary was 2.6 x 1.8cm. However in the restricted feeding group the ovaries were found to be smaller. Diameter of graffian follicles showed a similar tendency to ovarian size in the two groups.

The first oestrus in the standard feeding group appeared at 14.6 months when body weight weight was 265kg. Age at first calving was on average 28.9 months at a body weight of 436kg. On the other hand when heifers were fed the restricted ration the first oestrus appeared at 23.0 months at a body weight of 250kg. Average age at first calving was 38.9 months which was 10 months later than the average in the standard feeding group ($p<0.01$).

In standard feeding group the progesterone level was 2.0ng/ml at two weeks after pregnancy and gradually increased up to 14 weeks and peaked at 18 weeks. This peak(6.4-6.5ng/ml) was maintained up to 24 weeks when progesterone level decreased until it reached 2.1ng/ml at the end of pregnancy. In the restricted group progesterone level up to 16 weeks followed a similar pattern to the standard group but there was a tendency in the restricted group to have lower progesterone levels ($p<0.01$).

The standard and srstricted groups showed similar patterns of progesterone concentration during the oestrus cycle. There were no statistically significant differences in progesterone levels between standard and restricted groups but there was variation between individual animals.

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