# Plasma Luteinizing Hormone and Testosterone Concentrations in Different Breeds of Young Beef Bulls in the Tropics

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## Abstract

Plasma concentrations of luteinizing hormone (LH) and testosterone were measured at 3, 8 and 11 months of age in 48 Africander cross (AX), 24 Brahman cross (BX), 21 Hereford–Shorthorn, selected (HSS) and 14 Hereford–Shorthorn, random-bred (HSR) bulls. In all breeds plasma LH was lower (P<0.01) at 8 months (1.7 ng/ml) than at 3 months (2.6 ng/ml) or at 11 months (2.6 ng/ml). Over all ages there were no differences among breeds in mean plasma LH (AX 2.4, BX 2.4, HSS 1.8, HSR 2.2 ng/ml) and no breed  $\times$  age interactions. In contrast, plasma testosterone increased significantly (P<0.01) with age and at a faster rate in the AX breed, resulting in a significant (P<0.05) breed  $\times$  age interaction. Testosterone concentrations, though similar among breeds at 3 months of age (0.45 ng/ml), were much higher (P<0.01) by 11 months in AX (2.56 ng/ml) than in BX (1.30 ng/ml), HSS (0.78 ng/ml) or HSR (0.66 ng/ml) bulls.

Although LH did not differ among the breeds studied, the more pronounced increase in testosterone with age in the Africander cross bulls is consistent with the higher level of fertility commonly observed in this breed when compared to Brahman cross and Hereford–Shorthorn breeds during natural mating in Queensland.

## Introduction

During several years of natural mating of beef cattle at the CSIRO Tropical Cattle Research Centre, Rockhampton, the reproductive performance of Brahman cross (BX) and Hereford–Shorthorn cross (HS) breeding groups has been inferior to that of the more highly fertile Africander cross (AX) breed (Seebeck 1973). Single-sire matings demonstrated significant differences in performance between bulls in the BX and HS breeds, where variation was high, but not in the AX breed. Reciprocal matings in which AX and BX bulls were joined with both breeds of cows have shown that males contribute almost as much as females to the lower fertility of the BX breed (Seifert *et al.* 1980). Chenoweth and Osborne (1975) have reported lower libido and higher incidence of testicular hypoplasia in young BX and HS bulls compared to AX bulls. Christensen and Seifert (1976) found that BX bulls produced a lower proportion of live sperm and a higher proportion of abnormal sperm than AX bulls.

In view of these findings relative to breed differences in reproductive performance of bulls, it was of interest to investigate differences in reproductive hormone concentrations. In the present study, differences in plasma concentrations of luteinizing hormone (LH) and testosterone between breeds of bulls during the first year of life were examined. This study was undertaken with relatively large numbers of animals under field conditions which prevented intensive blood sampling in individual animals. It was a preliminary investigation designed to assess the likely value of more intensive studies of relationships between endocrine measurements and reproductive performance in tropical beef bulls.

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# Materials and Methods

Animals

One hundred and seven bulls of the following breed groups were studied:

- (1) 48 Africander cross (AX)-50% Africander, 25% Hereford, 25% Shorthorn;
- (2) 24 Brahman cross (BX)-50% Brahman, 25% Hereford, 25% Shorthorn;
- (3) 21 Hereford-Shorthorn, selected (HSS)-50% Hereford, 50% Shorthorn;
- (4) 14 Hereford-Shorthorn cross, random (HSR)-50% Hereford, 50% Shorthorn.

The AX, BX and HSS groups have been selected over previous years for high growth rate under prevailing conditions of high summer temperatures, high tick and worm levels, and poor winter nutrition. The HSR breed group was not selected but served as a random-bred control for the HSS breed group.

All bulls were born between mid-October and mid-December. Single blood samples were collected during the morning on three occasions, February, July and October, when the bulls were approximately 3, 8 and 11 months of age. Weaning occurred at 6 months of age. During the entire study the bulls grazed improved pastures of Siratro, green panic and buffel grass.

#### Analytical Methods

Blood samples were collected from the jugular vein into heparinized vacuum tubes and immediately chilled. Plasma was separated and stored at  $-20^{\circ}$ C until analysed for LH and testosterone by radioimmunoassay. The method used for LH was the Goding *et al.* (1969) solid-phase radioimmunoassay. The rabbit anti-ovine LH antiserum, the iodinated LH tracer and details of the assay sensitivity, validity, precision and specificity have been described by Gidley-Baird and Bindon (1976). In the present work iodinated NIH-LH-S16 was used as tracer (specific activity  $7 \cdot 4 \text{ MBq}/\mu g$ ) and NIH-LH-B3 used as the LH standard, run in quadruplicate at twofold dose intervals from  $0 \cdot 25$  to  $32 \cdot 0$  ng. Under the conditions of the assay [antiserum used at a dilution of 1 in 30 000; tracer mass equivalent to 0.5-0.6 ng (50 000 cpm) per tube; duplicate 0.3 ml aliquots of bovine plasma unknowns] the sensitivity of the method was 0.3 ng NIH-LH-B3 per tube or  $1 \cdot 0$  ng/ml plasma. Two ovine reference plasma samples were run in each assay: for the low-activity sample (4.95 ng/ml) the within-assay coefficient of variation was 8.9% (n = 20). For the high-activity reference sample (57.34 ng/ml) the within-assay variation was 15.2% (n = 16). Between-assay variation for these reference samples varied between 10 and 12%. All samples from a particular experiment were run in the one assay to avoid complications arising from between-assay variation.

In the testosterone assay described previously (Bindon *et al.* 1976) a rabbit antiserum (Calbiochem) cospecific for testosterone and dihydrotestosterone was used. In our bulls plasma concentration of dihydrotestosterone isolated by chromatography on celite and measured by radioimmunoassay are less than 1% of those of testosterone (Wong *et al.* 1977). Duplicate 0.1 ml aliquots of plasma were tested. The sensitivity of the methods was 10 pg per tube or 0.1 ng/ml. Coefficients of variation were less than 10% both within and between assays.

#### Statistical Methods

The analysis of variance of the main body of data was compiled using least squares in fitting a linear regression model for testing the general effects of breed, age, and breed  $\times$  age interaction. Birth day, initially included as a covariate, was subsequently deleted when found to be insignificant. The mean square within breeds was used to test breed differences and the mean square for error to test interaction and age effects. For specific tests of differences between breeds at each age and of differences between ages in each breed, the multiple range test of Duncan (1957) was used. In accordance with this method standard errors per observation were used to derive the shortest significant ranges for testing differences between means.

## Results

The mean concentrations of LH and testosterone in the four breed groups at the three ages are shown in Figs 1 and 2. Summaries of the overall analyses of variance of the LH and testosterone data are presented in the following tabulation:

Source of variation	d.f.	Mean squares	
		LH	Testosterone
Breeds	3	5.375	17 247**
Animals within breeds	103	2.373	3 404
Age	2	21 · 482**	20.727**
Breed $\times$ age	6	1.076	5 · 547*
Error	206	$2 \cdot 100$	1.615
* <i>P</i> <0.05; ** <i>P</i> <0.01.			

Concentrations of LH varied little among breeds (AX  $2 \cdot 4$ , BX  $2 \cdot 5$ , HSS  $1 \cdot 8$ , HSR  $2 \cdot 2$  ng/ml). The marginally lower level in the HSS breed was not significant. The outstanding feature of the LH results was the significantly lower level ( $1 \cdot 7$  ng/ml) at 8 months of age compared to mean levels of  $2 \cdot 6$  ng/ml at both 3 and 11 months.

Unlike LH, plasma testosterone generally increased with age, but the rate and pattern of increase varied among breeds resulting in a significant breed  $\times$  age interaction (P < 0.05). In AX bulls the increase in testosterone was linear from 3 to 11 months. In other breeds testosterone increased from 3 to 8 months but changed little from 8 to 11 months. Though



Fig. 1. Means and standard errors (vertical bars) for plasma concentrations of LH in 48 AX, 24 BX, 21 HSS and 14 HSR bulls at 3, 8 and 11 months of age.

testosterone concentrations were similar among breeds at 3 months of age (0.45 ng/ml), by 11 months of age they were significantly higher (P < 0.01) in AX (2.56 ng/ml) than in BX (1.30 ng/ml), HSS (0.78 ng/ml) or HSR (0.66 ng/ml) bulls.

#### Discussion

The major objective of this study was to look for breed differences in LH and testosterone in young bulls reared according to local practice in northern Australia. In this preliminary study using large numbers of bulls, but relatively few samples per animal, the intention was to obtain a broad view of possible breed differences in reproductive hormone concentrations that might reflect known breed differences in fertility. Sampling times were chosen according to convenience corresponding to such management events as branding and weighing. Since all the bulls were born at the same time of the year (early summer), in accordance with the practice of taking the fullest advantage of high summer nutrition, possible seasonal influences on hormone concentrations could not be separated from age effects. This was not considered a handicap since the primary aim was to observe hormone

concentrations under normal existing conditions rather than to separately characterize age and seasonal effects.

The most significant finding under the conditions of this study was that, in spite of similarities among breeds in concentrations of LH, plasma testosterone increased much more rapidly with age in the AX breed than in the BX, HSS and HSR breeds. Using much smaller numbers of bulls sampled more frequently, Schams *et al.* (1981) obtained similar results in other breeds. Testosterone increased much more rapidly with age in four Brown Swiss bulls in Munich than in four British Friesian bulls in Kenya, though there appeared to be no marked differences between breeds in LH. FSH and prolactin appeared lower in the Kenya bulls, suggesting that these hormones may be of importance in modifying the testosterone response to LH.



Fig. 2. Means and standard errors (vertical bars) for plasma concentrations of testosterone in 48 AX, 24 BX, 21 HSS and 14 HSR bulls at 3, 8 and 11 months of age.

Another significant finding in the present study was that LH was lower at 8 months than at 3 or 11 months of age. More detailed studies by others of LH changes in *Bos taurus* bulls up to 1 year of age have not given consistent results. LH has been reported to:

- (1) Remain unchanged with age (Karg et al. 1976);
- (2) Be elevated with large fluctuations from 1 to 5 months of age and then to decrease and remain steady up to 1 year (Lacroix *et al.* 1977);
- (3) Be biphasic with broad peaks extending from 2 to 5 months and from 11 to 12 months with an intervening trough (Rawlings *et al.* 1972);
- (4) Increase linearly from 7 to 13 months of age (Lunstra et al. 1978); and
- (5) Increase from birth to 3 months of age, decrease to a nadir at 8 months, and then rise over the next 4 months (Schams *et al.* 1981).

Our own LH results based on fewer ages of sampling tend to agree with the patterns reported by Rawlings *et al.* (1972) and Schams *et al.* (1981). A general inspection of the results of all the reported studies of LH patterns offers no evidence that a critical age for evaluating possible breed differences in LH was missed by our limited sampling frequency.

With regard to testosterone, Karg *et al.* (1976) reported that in young Brown Swiss bulls there was a trough or plateau at about the time of puberty separating earlier and later peaks. This pattern was independent of season. Studies using other breeds of *Bos taurus* bulls (Rawlings *et al.* 1972; Secchiari *et al.* 1976; Lacroix *et al.* 1977) were in general agreement but suggested that the timing of the pubertal trough may vary with breeds or conditions of management. Under the conditions existing at this laboratory, other studies

(T. B. Post, unpublished observations) involving more frequent sampling over a greater range of ages, indicated that none of our bulls reached a trough before 12–13 months of age. It is therefore unlikely that the observed differences in testosterone between breeds at 11 months were due to the BX, HSS and HSR breeds having fallen into a pubertal trough earlier than the AX breed.

On the basis of these breed comparisons, then, this study suggests an association between pre-pubertal levels of testosterone, but not of LH, and the reproductive performance of different *breeds* of bulls in the tropics. The results justify further studies to answer the more pertinent question of whether or not the testosterone status of an *individual* young bull is predictive of future reproductive performance. Such studies will require more careful evaluation of individual bulls than the single-sample technique used in this study.

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