

THE EFFECT OF BULL EXPOSURE ON THE EARLY POSTPARTUM REPRODUCTIVE PERFORMANCE OF SUCKLING ANGUS COWS

Efecto de la Exposición al Toro sobre el Desempeño Reproductivo Postparto Temprano de Vacas Angus Amamantando

Antonio Landaeta-Hernández^{1,2*}, Pedro Meléndez¹, Julián Bartolomé¹, Owen Rae¹ y Louis F. Archbald¹

¹Department of Large Animal Clinical Sciences, College of Veterinary Medicine, University of Florida. Gainesville, P.O. Box 100136, FL 32610-0136. ²Unidad de Investigaciones Zootécnicas, Dpto. de Producción e Industria Animal, Facultad de Ciencias Veterinarias, Universidad del Zulia. Maracaibo, Venezuela. *E-mail: landaetaa@yahoo.es

ABSTRACT

The present study tested the effect of biostimulation by bull exposure on the early postpartum reproductive performance of well fed and permanently suckled Angus cows. At 1 week postpartum, 90 Angus cows with no history of calving problems and their calves were allocated by parity, body weight, and body frame into 3 isolated groups of 30 cows each. Cows from groups A and B were permanently exposed to mature bulls (BE) while cows from Group C (NE) served as a control. Analyzed variables included; intervals from calving to resumption of ovarian activity (ICR), and first behavioral estrus (ICE), length of the first estrous cycle (ECL), number of cows showing estrus/group, and number of estrus periods (NEP) occurred per cow during the 90 days previous to the breeding season. The ICR (38.1 ± 3.4 vs 51.0 ± 4.4 ; $P < 0.02$) and ICE (38.1 ± 3.4 vs 59.9 ± 2.5 ; $P < 0.001$) were shorter in BE than in NE cows, and no differences between bull-exposed groups were found. More BE cows resumed reproductive cyclicity with normal ECL than NE cows (A= 16/53%, B=16/53%, C=8/26.6%; $P < 0.01$). During trial, more BE cows showed behavioral estrus than NE cows (A: 29/30= 97%, B: 30/30 =100%, and C: 24/30 = 80%; $P < 0.002$). Similarly, NEP recorded in BE cows was greater than that of NE cows (A=70, B=68, and C=42; $P < 0.0002$). More BE cows ($P < 0.001$) had 3 estrous periods during trial. It was concluded that, even under conditions involving adequate nutrition and permanent suckling, there was a positive effect of biostimulation on early reproductive performance in postpartum Angus cows.

Key words: Male-effect, biostimulation.

RESUMEN

El presente estudio examinó el efecto de la bioestimulación por exposición al toro sobre el desempeño reproductivo durante el postparto temprano de vacas Angus bien alimentadas y sujetas a amamantamiento permanente. A una semana postparto, 90 vacas Angus sin historia de problemas al parto y sus becerros fueron distribuidos según el número de partos, peso y talla corporal en 3 grupos aislados de 30 vacas cada uno. Las vacas de los grupos A y B fueron expuestas permanentemente a toros maduros (BE), en tanto que las vacas del grupo C (NE) sirvieron como control. Las variables analizadas fueron: intervalos parto-reanudación de la actividad ovárica (ICR) y parto-primero celo expresado (ICE), longitud del primer ciclo estral (ECL), número de vacas mostrando celo por grupo y número de celos por vaca durante los 90 días previos a la época de monta (NEP). El ICR ($38,1 \pm 3,4$ vs $51,0 \pm 4,4$; $P < 0,02$) y el ICE ($38,1 \pm 3,4$ vs $59,9 \pm 2,5$; $P < 0,001$) fueron más cortos en BE que en NE, y no se hallaron diferencias entre grupos expuestos a toro. Más BE reanudaron la ciclicidad ovárica con ciclos normales en comparación con las NE (A=16/53%, B=16/53%, C=8/26,6%; $P < 0,01$). Durante el experimento, más vacas BE mostraron celo que las NE (A: 29/30= 97%, B: 30/30 =100%, C: 24/30= 80%; $P < 0,002$). Así mismo, el NEP observado en vacas BE fue mayor que en las vacas NE (A=70, B=68, y C=42; $P < 0,0002$). Más ($P < 0,001$) vacas BE tuvieron 3 celos durante el estudio. Se concluyó que, aun bajo condiciones de nutrición adecuada y amamantamiento permanente, hubo un efecto bioestimulador positivo sobre el desempeño reproductivo temprano de las vacas Angus postparto.

Palabras clave: Efecto macho, bioestimulación.

INTRODUCTION

The major influences on reproductive performance are exerted by environmental and management factors. Thus, reproductive performance can be enhanced by applying rational management strategies. One of the greatest constraints with increasing management level is finding an even point between profitability and level of management to be applied, including the use of recent technologies. This is especially true in tropical and underdeveloped countries, in which the relationship cost: benefit is often a limitation for applying technology.

Biostimulation by bull exposure represents a relatively inexpensive management tool often underestimated. Likely reasons for such underestimation may be the beliefs that in presence of good nutrition [51, 76] and calf suckling [28, 82], the beneficial effects of biostimulation can be either minimized or suppressed. Moreover, teaser bulls are associated with an increased risk of diseases transmission.

Although the evidence of the positive effect of biostimulation appears to be clear in both *Bos taurus* [15, 83] and *Bos indicus* [58, 72] beef breeds, most research reports have tested the effect of biostimulation on reproductive function without appropriate handling of aspects related to nutrition and suckling [20, 34, 49, 59, 72]. In addition, despite its potential repercussions on subsequent fertility, there is a lack of information on the influence of biostimulation on the early postpartum performance previous to the breeding season.

Therefore, objectives in this study were to test the effect of biostimulation by bull exposure on the early postpartum performance (e.g., intervals from calving to resumption of ovarian activity and first behavioral estrus, length of the first estrous cycle, and number of estrus periods occurred per cow during the 90 days previous to the breeding season) in well fed postpartum Angus cows subjected to permanent suckling.

MATERIALS AND METHODS

This field study was conducted during the Spring season at the Santa Fe Beef Research Unit of University of Florida (subtropical North-Central Florida). Since the reproduction of this herd is totally synchronized, at 1 week postpartum, 90 An-

gus cows without any history of calving problems and their calves were allocated by parity, body weight, and body frame into 3 groups of 30 cows each. To account for possible differences due to bulls, cows in groups A and B were placed in separated pens with one mature epididymectomized [63] Angus bull each. Cows in Group C were not exposed to any bulls, and were located and handled as to not receive any type of stimuli from the bull-exposed groups. The arrangement of bulls and cows provided a bull:cow ratio of 1:30 which is normally recommended for commercial beef cattle operations [12].

The elapsed time period within which reproductive performance was monitored in this study included from one week postpartum to 90 days postpartum. Thus, since in this synchronized herd all the recorded dependent variables were almost restricted to the first 60 days, it was globally considered an indicator of early reproductive performance.

As indicated in the baseline comparison (TABLE I), there were no differences in parity, body frame, postpartum body weight, and gestation length among groups. With the exception of calf weight at 90 d (107 ± 2 vs $101.2 \pm$ kg; $P < 0.09$), no other differences were found between calves from biostimulated and control groups with respect to calf birth weight (31.9 ± 1.1 vs 30.7 ± 1.1 kg, respectively) and calf sex distribution (34 and 13 vs 26 and 17 males and females respectively). Since milk yield is a largely reported effect on reproductive performance, it was indirectly estimated by monitoring calf weight. Thus, calves were weighed at the beginning and at the end of the experiment and values used as independent random variable.

All cows received daily prepartum and postpartum feed supplementation. Feed consisted of a 2 kg mix of corn, oilseed meal, coconut meal, citrus pulp and minerals. In addition, during the study all cows in each group were fed Bahia grass (*Paspalum notatum*) hay and water *ad libitum*. No grass was available in any of the three assigned pens (0.25 hs approx. dimension). All calves were left with their dams to suckle throughout the study. In this way, all cows were subjected to similar management scheme with regard to nutrition and suckling of calves. Animals were handled according policies of the Institutional Animal Care and Use Committee from University of Florida.

TABLE I
BASELINE COMPARISONS. LEAST SQUARE MEANS \pm STANDARD ERROR (LSM \pm SE) / COMPARACIONES DE BASE. MEDIAS POR MINIMOS CUADRADOS \pm ERROR ESTANDAR

Group	Parity	W1(k) ^a	BCS1	Frame		
				L	M	S
A*	2.6 \pm 0.3	512 \pm 12	4.8 \pm 0.1	3	18	9
B*	2.3 \pm 0.3	521 \pm 12	4.6 \pm 0.1	5	15	10
C**	2.4 \pm 0.3	524 \pm 12	5.0 \pm 0.1	3	18	9
p-value	P > 0.47	P > 0.74	P > 0.47	P > 0.88		

W1= Initial body weight ; BCS1= Initial body condition score * = Biostimulated groups; ** Control. Parity= N^a partos, W1= Peso inicial en kgs. BCS1= Condición corporal inicial. Frame = Talla: L= Large, M=Middle, S=Small.

All 90 cows were used to determine the effect of biostimulation by bulls on the expression of the first postpartum estrous period. However, a subset of 30 cows (10 cows from each group) allocated by parity, was used to determine the effect of biostimulation on time for resumption of ovarian activity (first rise of progesterone above 1 ng) and monitoring nutritional status (non-esterified fatty acids -NEFA-) by jugular bleeding.

To assess the interval from calving to resumption of ovarian activity, plasma concentration of P4 was determined using a solid-phase, non-extraction radioimmunoassay (Coat-a-Count® kit) previously described [73]. The sensitivity of the assay kit is 0.02 ng/mL as indicated by manufacturer. The coefficient of variance intra-assay was less than 10%. All samples were processed in one batch, thus, the coefficient of variance inter-assay was 0. Once a rise of progesterone above 1 ng/ml was determined and a luteal structure was observed by ultrasonography, an interval from calving to the corresponding day of the progesterone rise was calculated. Plasma concentrations of NEFA were weekly obtained to assess nutritional status during the study. Determination of NEFA plasma concentration was performed at the Peri-parturient Disease Laboratory of the National Animal Disease Center, Ames, Iowa. It was expected to find concentrations of NEFA between 0.2-0.3 mmol/liter in all cows during the entire experiment as an indicator of appropriate energy balance [62, 65].

To assess the interval from calving to first postpartum estrus, data of mounting activity were collected by HeatWatch® mount detectors (Ddx Inc Denver, CO, USA) fitted to all cows in the study at the initiation of the experiment. The occurrence of at least 3 mounts within a period of 4 h [56] was considered the start of an estrous period. Once HeatWatch® detected the first postpartum estrus, the interval from calving to such first estrus was calculated. For those cows which not showed behavioral estrus, the duration of the experiment (90 d) was assigned as a value for this variable. A similar criterion was applied to cows that did not have a rise of progesterone above 1 ng/mL. during the experiment. Bias was prevented by labeling vials containing blood samples with serial numbers instead of the cow number and by using HeatWatch® mount detectors to collect the information regarding mounting activities.

Statistical analysis of plasma NEFA concentrations was performed by ANOVA with repeated measures using PROC MIXED of SAS [64]. The statistical model initially comprised the effect of cow, group, and week as independent class variables. Independent random variables were initial body weight (W1), body condition score (BCS1), calf birth weight (CBW) and final calf weight (CW2), while tested interaction was group x week. Cow within group was the error term. The final model comprised the effects of group, week, group x week, and cow within group.

The statistical analysis of the interval from calving to resumption of ovarian activity was performed by ANOVA using

PROC GLM of SAS [64]. The statistical model initially comprised the effects of group and treatment as independent class variables. Independent random variables were W1, BCS1, CBW, CW2 and duration of first estrus and the interactions of group x treatment. Since no difference between bull-exposed groups was found, data were pooled. Due to the lack of significance of several dependent variables, the final statistical model only included treatment.

As determined by HeatWatch®, the interval from calving to first behavioral estrus was performed by ANOVA using PROC MIXED of SAS [64]. The statistical model comprised the effects of treatment and group as independent class variables. Independent random variables were W1, BCS1, CBW and CW2 and the interaction group x treatment. Since no difference between groups exposed to bulls was determined, data of Groups A and B were pooled. Due to the lack of significance of several dependent variables, the final model included treatment, W1, and CW2.

The length of the first postpartum estrous cycle was determined as the time interval between the first and second estrous periods as detected by HeatWatch®. The length of the first postpartum estrous cycle was examined as an indicator of type of cyclicity postpartum. According to the information provided by HeatWatch®, estrous cycles were classified as normal (18-24 d), long (≧ 25 d), and short (● 17 d) length [61]. Comparisons between groups (e.g. biostimulated and non-biostimulated) regarding the length of the first postpartum estrous cycle were performed by Chi Square procedure (Epi-Info®).

Information regarding number of cows showing estrus within groups, number of estrous periods per group and number of estrous periods per cow during the first 90 d postpartum was obtained from the data recorded by HeatWatch®. Both, the number of cows showing estrus per group and number of estrus per group during the trial were compared by Chi-square procedure (Epi-Info®). With regard the number of estrous periods per cow during the first 90 d postpartum, this information was statistically analyzed with a model that initially included group and treatment as independent class variables. Independent random variables were W1, BCS1, CBW, CW2, interval from calving to first postpartum estrus, duration of first postpartum estrus. Examined interaction was group x treatment. Since no differences due to group were observed, data from biostimulated groups were pooled and further analysis comprised a statistical model which included treatment and interval from calving to first postpartum estrus. The statistical approach to analyze these data was analysis of variance using PROC MIXED of SAS [64].

In the present study, a p-value of $P < 0.05$ was used to define statistical significance of every test, while p-values above $P < 0.05$ and below $P < 0.10$ were considered tendencies.

RESULTS AND DISCUSSION

Intervals from calving to resumption of ovarian activity and to first behavioral estrus

As shown in TABLE II, the intervals from calving to resumption of ovarian activity and to first behavioral estrus were shorter in bull-exposed than in non-exposed cows, and no differences between bull-exposed groups were found. In addition, as a marginal finding of this study, it was observed that those cows which had the heaviest calves by 90 d postpartum had longer ($P < 0.002$) intervals to first behavioral estrus than the others.

Despite the finding agrees with most of available reports, in several studies [1, 20, 25, 51, 52, 66, 67], discrepancies are found with regard the effect of bull-exposure on the intervals from calving to resumption of ovarian activity and to first behavioral estrus. Probably reasons for such discrepancies include lack of consensus with regard the magnitude of the progesterone rise indicating resumption of postpartum ovarian activity [1, 14, 15, 19, 20, 25, 51, 67, 72, 76], intensity of the biostimulatory effect [e.g., 20,67], reduced sample size [1], nutrition [7, 34, 76], ignoring nutritional interactions with genotype and parity [25, 51, 52, 67], differences on nutritional demands between dairy and beef breeds [66, 67], and the negative effect of high milk yield on the expression of behavioral estrus [66, 67].

In a recent study, the response to the biostimulatory effect of bull-exposure was found to be conditioned by the frequency, duration and quantity of stimuli in primiparous *Bos taurus* beef cows [4]. The effect of parity on the intervals from calving to resumption of ovarian activity and first postpartum estrus has been largely reported [2, 68] and it is strongly related to differences on nutritional requirements between first calving and mature cows [47, 48]. Meanwhile, the effect of

breed probably relies upon the differences in frame and body weight (body mass) between small and large breeds which may be associated to differences in nutritional requirements, especially during the early postpartum period. The impact of cow body size on reproductive efficiency has been reported in both *Bos taurus* and *Bos indicus* cattle [11, 53, 81]. High milk yield has been found in association with a poor estrus expression [40, 46] which leads to limited detection of estrus. Physical and/or mechanics difficulties derived from udder volume may be considered another factor limiting expression of estrus in lactating dairy cows [80].

Knowledge about mechanisms by which bull-exposure may reduce the intervals from calving to resumption of reproductive cyclicity and to first postpartum estrus remains limited. A pheromone signal expelled in body fluids or excretory products has been pointed out to be responsible for triggering the biostimulatory effect in cattle [3,58]. Although pheromone signals from urine and sebaceous glands have been reported in other species [31, 38, 41, 54], in cattle, both the route of emission and chemo-signal remain unknown. Some researchers have suggested urine [39, 42] and odors emanating from sebaceous glands in perineal zone and hindquarters [6, 79]. Independently of the specie and source or signal, class antigens of the major histocompatibility complex (MHC) seem to be responsible for these odor signals [69, 70]. The reception and transport of the pheromone signal is probably initiated via olfaction [6] through the vomeronasal organ [29, 36]. However, mechanisms involving the posterior internalization and translation of the pheromone signal into the endocrine system are unknown. Enhanced functionality of early luteal tissue and sub-luteal and transient progesterone levels have been suggested to be in relationship with the biostimulatory signal [45].

With regard the influence of suckling on the intervals from calving to resumption of ovarian activity and first estrus,

TABLE II
EFFECT OF BIOSTIMULATION ON THE INTERVAL FROM CALVING TO RESUMPTION OF OVARIAN ACTIVITY (ICR) AND FIRST BEHAVIORAL ESTRUS (ICE) IN POSTPARTUM ANGUS COWS. LEAST SQUARE MEANS ± STANDARD ERROR (LSM ± SE) / EFECTO DE LA BIOESTIMULACION SOBRE EL INTERVALO PARTO-REANUDACION DE LA ACTIVIDAD OVÁRICA Y PRIMER CELO EXPRESADO. MEDIAS POR MINIMOS CUADRADOS ± ERROR ESTANDAR

Group	N	ICR		ICE	
		LSM + SE (d)	N	LSM + SE (d)	N
A (Bio)*	10	39.7 ± 4.5 ^a	29	45.1 ± 2.5 ^f	
B (Bio)*	10	36.3 ± 5.0 ^{ab}	30	41.7 ± 2.5 ^f	
C (Control)	10	51.0 ± 4.5 ^c	24	59.9 ± 2.5 ^g	
Treat (Bio)*	20	38.1 ± 3.4 ^d	59	38.1 ± 3.4 ^h	
Control	10	51.0 ± 4.4 ^e	24	59.9 ± 2.5 ⁱ	

(a,c)= Means in a column lacking common superscript differ ($P < 0.08$)
 (b,c)= Means in a column lacking common superscript differ ($P < 0.03$)
 (d,e)= Means in a column lacking common superscript differ ($P < 0.02$)
 (f,g)= Means in a column lacking common superscript differ ($P < 0.001$)
 (h,i)= Means in a column lacking common superscript differ ($P < 0.001$)
 *= Exposure to bulls from one week postpartum.

studies investigating the effect of suckling on reproductive function have concluded that inhibition of LH pulses is a major reason for delayed resumption of ovarian activity [28, 82]. However, as with previous reports [24], this study suggest that biostimulation seems to override the negative influence of suckling. Nevertheless, since cows suckled by the heaviest calves at 90 d exhibited elapsed intervals from calving to resumption of ovarian activity, our findings also suggest a cut-off point for the biostimulatory effect by either milk yield or number of suckling periods. Previous studies have shown a negative effect of growth rate, milk consumed and number of suckling periods [30, 68, 74]. However, the effect of biostimulation was not assessed in parallel with suckling in those studies. To the authors's knowledge, there are no studies determining the limits at which biostimulation can overrides or not the negative effect of suckling or milk yield on intervals from calving to resumption of ovarian activity and to first estrus.

Although scarcely reported, differences in the biostimulatory effect due to variation between bulls appear unlikely. Testing a different hypothesis, a previous study [14] did not find differences in the interval from calving to first estrus between *Bos taurus* beef cows exposed to either young or mature bulls. In this study, no differences due to bulls were found on the intervals from calving to first estrus or resumption of ovarian activity.

First postpartum estrous cycle length.

Biostimulation influenced the length of the first postpartum estrous cycle. As shown in TABLE III, more bull-exposed cows resumed reproductive cyclicity with estrous cycles of normal length (18-24 d) than non-exposed cows (P<0.01). Length of estrous cycle could not be determined in 47% of non-exposed cows because they did not show estrus, or did not have a second estrus during the 90 days of the experiment.

A normal estrous cycle is considered to last 18-24 days [61, 75]. Those cycles with an extent below or above this range are called short and long cycles, respectively [61]. Although with some conflictive reports, variability of the estrous cycle

length has been reported to occur due to parity [27], lactation [27], individual variations [8, 32], genetics [33, 44], genotype x environment interactions [21], embryo mortality [17], environmental factors [16, 43, 60], and postpartum endocrinology [27, 37, 55].

In both dairy and *Bos taurus* and *Bos indicus* beef cows, resumption of ovarian activity postpartum is usually followed by estrous cycles of short length [27, 37, 77]. Luteal structures of limited functionality have been related to the occurrence of short luteal phases and subsequent short estrous cycles, which are considered by several researchers to be important for establishment of normal cyclicity [22, 27, 77]. Nevertheless, as with others [5, 34], the findings suggest that biostimulation or administering some external source of progestagens [10, 23, 57, 71] at early postpartum stage may induce cows to resume cyclicity with normal estrous cycles. However, discussions are limited because studies exploring the influence of biostimulation on the length of the first postpartum estrous cycle are uncommon.

During early postpartum stages, the uterine environment is dominated by the presence of oxytocin and PGF_{2α} [9, 18, 78]. Meanwhile, other events include; a sub-optimal preovulatory secretion of estradiol [50], reduced concentrations of receptors for progesterone in uterus [84], and limited capability of early luteal structures to decrease the concentration of uterine receptors for oxytocin [23, 35] and to cope with the high levels of postpartum PGF_{2α}. All together, these events set the scenario for luteolysis and a short estrous cycle, because regulation of uterine receptors for progesterone is controlled by both estradiol and progesterone [13]. Estradiol promotes an increase of receptors for progesterone while progesterone down-regulates its own receptors [13].

It appears possible that the early increase in plasma progesterone observed in male-exposed beef cows [15, 45] and sheep (*Ovis aries*) [26] may represent the priming effect from a corpus luteum with enhanced ability to suppress the PGF_{2α} effect as suggested by others [23, 35]. Such an enhanced ability

TABLE III
TYPE OF ESTROUS CYCLE AT RESUMPTION OF CYCLICITY AMONG GROUPS. FREQUENCY DISTRIBUTION /
TIPO DE CICLO ESTRAL AL REINICIO DE LA CICLICIDAD ENTRE GRUPOS. DISTRIBUCIÓN DE FRECUENCIAS

Type of Cycle	n (%)*		
	Group A ¹	Group B ¹	Group C (control)
Normal (18-24 d)	16 (53%) ^a	16 (53%) ^a	8 (26.6%) ^b
Short (< 17 d)	8 (26.6%)	5 (16.6%)	3 (10%)
Long (≧ 25 d)	3 (10%)	5 (16.6%)	5 (16.6%)
Anestrus	1 (3%)	–	6 (20%)
No 2nd estrus**	2 (7%)	4 (13%)	8 (26.6%)

¹ = Exposure to bulls from one week postpartum.
 * = Number and percentage of cows within group.
 ** = Lack of a second estrus prevents knowing type of cycle.
 (a,b) Values in a row lacking common superscript differ (P<0.01).

of the corpus luteum to suppress the PGF_{2α} effect may have led to cows in this study to resume cyclicity with estrous cycles of normal length.

Number of estrous periods per cow and per group during trial

As shown in TABLE IV, more cows from bull-exposed groups (A: 29/30= 97% and B: 30/30= 100%) showed behavioral estrus than non-exposed cows (C: 24/30= 80%; P<0.002). Similarly, the total number of estrous periods recorded in cows from bull-exposed groups (70 and 68) during the trial was greater (P<0.0002) than that of cows in the control group (42). No statistical difference (P>0.05) was determined between biostimulated groups regarding number of cows showing estrus and number of estrous periods observed per group. More bull-exposed cows (P< 0.001) had 3 estrous periods during the 90 d following parturition than non-exposed.

The findings of greater proportion of bull-exposed than non-exposed cows showing behavioral estrus during the trial and the greater number of estrous periods per cow observed in bull-exposed groups compared with controls is in agreement with others [19, 58]. Likely reason for these findings relies upon

the biostimulatory effect of bull-presence to resume cyclicity early. Having resumed cyclicity early, chances for additional estrous periods are greater for bull-exposed cows compared to non-exposed. Cows having several estrous periods before re-breeding may represent a positive implication, especially for those cattle operations with an established breeding season. To some extent, as the number of postpartum estrous periods increase, fertility also increases. Thus, it might be expected that bull-exposed cows become pregnant.

Non-esterified fatty acids (NEFA)

The nutritional status of cows was assessed by measuring plasma NEFA's concentration. As shown in TABLE V, overall plasma NEFA concentration was greater in bull-exposed than non-exposed cows (P< 0.002). However, as previously indicated [62, 65] it was within the expected margin for postpartum cows (0.2-0.3 mmol/l). Because of calves from bull-exposed groups were heavier at the end of the study, such differences in plasma NEFA concentrations more likely occurred because bull-exposed cows were producing more milk than non-exposed cows. However, to the authors knowledge, studies assessing the effect of bull-exposure on milk yield at early stages of lactation are not available.

TABLE IV
NUMBER OF ESTROUS PERIODS WITHIN GROUPS DURING TRIAL. FREQUENCY DISTRIBUTION / NUMERO DE CELOS DENTRO DE CADA GRUPO DURANTE EL ESTUDIO. DISTRIBUCION DE FRECUENCIAS

N° Estrous	n (%)*	n (%)*	n (%)*
	Group A ¹	Group B ¹	Group C (control)
1	2 (7%)	4 (13%)	8 (26.6 %)
2	14 (47%)	14 (47%)	14 (47%)
3	12 (40%) ^a	12 (40%) ^a	2 (6.6%) ^b
4	1 (3%)	–	–
0	1 (3%)	–	6 (20%)
2	27 (90%) ^c	26 (87%) ^c	16 (53%) ^d

Number and percentage of cows within group.

1= Exposure to bulls from one week postpartum.

(a,b) Values in a row lacking common superscript differ (P<0.001).

(c,d) Values in a row lacking common superscript differ (P<0.0002).

TABLE V
PLASMA NEFA¹ CONCENTRATION. LEAST SQUARE MEANS ± STANDARD ERROR (LSM ± SE) / CONCENTRACIONES PLASMATICAS DE NEFA. MEDIAS POR MINIMOS CUADRADOS ± ERROR ESTANDAR

Group	N	NEFA (mmol/l)
A (Bio)*	10	0.32 ± 0.02 ^a
B (Bio)*	10	0.30 ± 0.02 ^a
C (Control)	10	0.18 ± 0.02 ^b

(a,b) Means in a column lacking common superscript differ (P<0.01)

1= Non-esterified fatty acids.

*= Exposure to bulls from one week postpartum.

CONCLUSIÓN

It was concluded that, the biostimulatory effect of bull-exposure was not suppressed by adequate nutrition and permanent suckling. Cows exposed to bulls at early postpartum periods showed reduced intervals from calving to resumption of ovarian activity and first behavioral estrus, resumed cyclicity with estrous cycles of normal length, and had more estrous periods during the 90 days previous to the breeding season.

ACKNOWLEDGMENT

USDA Animal Health Funds at the College of Veterinary Medicine, University of Florida. Employees of University of Florida's Santa Fe Beef Unit. National Animal Disease Center, USDA-ARS, Ames, Iowa. USDA-ARS, Brooksville. Cow Chips Inc, Denver, CO.

BIBLIOGRAPHIC REFERENCES

- [1] ALBERIO, R.H.; SCHIERSMANN, G.S.; CAROU, N.; MESTRE, J. Effect of a teaser bull on ovarian and behavioral activity of suckling beef cows. **Anim. Reprod. Sci.** 14: 263-272. 1987.
- [2] ARANGUREN-MENDEZ, J.A. Reproductive indexes in 5/8 Brahman, 5/8 Holstein and 5/8 Brown Swiss cross-bred cows. **Rev. Científ. FCV-LUZ.** VI (3): 141-147. 1996.
- [3] BERARDINELLI, J.G.; JOSHI, P.S. Initiation of postpartum luteal function in primiparous restricted-suckled beef cows exposed to a bull or excretory products of bulls or cows. **J. Anim. Sci.** 83: 2495-2500. 2005.
- [4] BERARDINELLI, J.G.; TAUCK, S.A. Intensity of the biostimulatory effect of bulls on resumption of ovulatory activity in primiparous, suckled, beef cows. **Anim. Reprod. Sci.** 99: 24-33. 2006.
- [5] BLASCO, I.; REVILLA, R., Characterization of progesterone profiles in postpartum beef cows. **Proc. 12th International Congress on Animal Reproduction.** The Hague, The Netherlands. 1: 27-29. 1992.
- [6] BLAZQUEZ, N.B.; FRENCH, J.; LONG, S.E.; PERRY, G.C.; STEVENS, K. Bovine oestrus odours: behavioural and histological investigations. **Proc. 11th Int Congress Anim Reprod. and AI.** Dublin, Ireland 4: 550. 1988.
- [7] BOLAÑOS, J.M.; FORSBERG, M.; KINDAHL, H.; RODRIGUEZ-MARTINEZ, H. Biostimulatory effects of estrous cows and bulls on resumption of ovarian activity in postpartum anoestrous zebu (*Bos indicus*) cows in the humid tropics. **Theriogenol** 49: 629-636. 1998.
- [8] BONDAR, A.A. Improving the visual method of oestrus diagnosis in cows and designing the insemination centre. **Molochno M'yasne Skotarsvo Ukraine-Russia** 82: 17-21. 1993.
- [9] BONDURANT, R.H. Inflammation in the bovine female reproductive tract. **J. Anim. Sci.** 77 (Suppl 2) / **J. Dairy Sci** 82 (Suppl 2): 101-110. 1999.
- [10] BREUEL, K.F.; LEWIS, P.E.; INSKEEP, E.K.; BUTCHER, R.L. Endocrine profiles and follicular development in early-weaned postpartum beef cows. **J. Reprod. Fertil.** 97: 205-212. 1993.
- [11] BUTTRAM, S.T.; WILLHAM, R.L. Size and management effects on reproduction in first, second, and third parity beef cows. **J. Anim. Sci.** 67: 2191-2196. 1989.
- [12] CHENOWETH, P.J. Bull behavior, sex-drive and management. Chapter 23. In: Fields, M.J, and Sands, R.S. (Eds.). **Factors Affecting Calf Crop.** C.R.C. Press, Boca Raton, FL. USA. 319-330 pp. 1994.
- [13] CLARKE, C.L. Cell-specific regulation of progesterone receptor in the female reproductive system. **Mol. Cell Endocrinol.** 70: C29-C33. 1990.
- [14] CUPP, A.S.; ROBERSON, M.S.; STUMPF, T.T.; WOLFE, M.W.; WERTH, L.A.; KOJIMA, N.; KITOK, R.J.; KINDER, J.E. Yearling bulls shorten the duration of postpartum anoestrus in beef cows to the same extent as do mature bulls. **J. Anim. Sci.** 71: 306-309. 1993.
- [15] CUSTER, E.E.; BERARDINELLI, J.G.; SHORT, R.E.; WEHMAN, M.; ADAIR, R. Postpartum interval to estrus and patterns of LH and progesterone in first-calf suckled beef cows exposed to mature bulls. **J. Anim. Sci.** 68: 1370-1377. 1990.
- [16] DOBSON, H.; KAMONPATANA, M. A review of female cattle reproduction with special reference to a comparison between buffaloes, cows and zebu. **J. Reprod. Fertil.** 77: 1-36. 1986.
- [17] DROST, M.; THATCHER, W.W. Heat stress in dairy cows, its effect on reproduction. **Vet. Clin. N.A.: Food Anim. Pract.** 3: 609-618. 1987.
- [18] EDQVIST, L.E.; KINDAHL, H.; STABENFELDT, G. Release of prostaglandin F_{2α} during the bovine periparturition period. **Prostagland.** 43: 1379-1388. 1978.
- [19] FERNANDEZ, D.L.; BERARDINELLI, J.G.; SHORT, R.E.; ADAIR, R. The time required for the presence of bulls to alter the interval from parturition to resumption of ovarian activity and reproductive performance in first-calf suckled beef cows. **Theriogenol.** 39: 411-419. 1993.
- [20] FIKE, K.E.; BERGFELD, E.G.; CUPP, A.S.; KOJIMA, F.N.; MARISCAL, V.; SANCHEZ, T.S.; WERHMAN, M.E.; KINDER, J.E. Influence of fenceline bull exposure on duration of postpartum anoestrus and pregnancy rate in beef cows. **Anim. Reprod. Sci.** 41: 161-167. 1996.

- [21] GALINA, C.S.; ARTHUR, G.H. Review on cattle reproduction in the tropics. Part 4. Oestrus cycles. **Anim. Breed. Abstr.** 58: 697-707. 1990.
- [22] GARVERICK, H.A.; PARFET, J.R.; LEE, C.N.; COPELIN, J.P.; YOUNGQUIST, R.S.; SMITH, M.F. Relation of pre- and post-ovulatory gonadotropin concentrations to subnormal luteal function in postpartum beef cattle. **J. Anim. Sci.** 66: 104-111. 1988.
- [23] GARVERICK, H.A.; ZOLLERS, W.G.; SMITH, M.F. Mechanisms associated with corpus luteum life span in animals having normal or subnormal luteal function. **Anim. Reprod. Sci.** 28: 111-124. 1992.
- [24] GAZAL, O.S.; GUZMAN-VEGA, G.A.; WILLIAMS, G.L. Effects of time of suckling during the solar day on duration of the postpartum anovulatory interval in Brahman x Hereford (F1) cows. **J. Anim. Sci.** 77: 1044-1047. 1999.
- [25] GIFFORD, D.R.; D'OCCHIO, M.J.; SHARPE, P.H.; WEATHERLY, T.; PITTAR, R.Y.; REEVE, D.V. Return to cyclic ovarian activity following parturition in mature cows and first-calf beef heifers exposed to bulls. **Anim. Reprod. Sci.** 19: 209-216. 1989.
- [26] GODFREY, R.W.; GRAY, M.L.; COLLINS, J.R. The effect of ram exposure on uterine involution and luteal function during the postpartum period of hair sheep ewes in the tropics. **J. Anim. Sci.** 76: 3090-3094. 1998.
- [27] GONZALEZ-STAGNARO, C.; RAMÍREZ, L.N.; MADRID-BURY, N.; MEDINA, D. Caracterización del ciclo estrual y de la actividad luteal al inicio del postparto en vacas mestizas. **Rev. Arg. Prod. Anim.** 15: 1043-1044. 1995.
- [28] GRIFFITH, M.K.; WILLIAMS, G.L. Roles of maternal vision and olfaction in suckling-mediated inhibition of luteinizing hormone secretion, expression of maternal selectivity, and lactational performance of beef cows. **Biol. Reprod.** 54: 761-786. 1996.
- [29] HART, B.L. Roles of the olfactory and vomeronasal systems in behavior. **Vet. Clin. N.A.: Food Anim. Pract.** 3: 463-475. 1987.
- [30] HENAO, H.; OLIVERA-ANGEL, M.; MALDONADO-ESTRADA, J.G. Follicular dynamics during postpartum anestrus and the first estrous cycle in suckled or non-suckled Brahman (*Bos indicus*) cows. **Anim. Reprod. Sci.** 63: 127-136. 2000.
- [31] HILLYER, G.M. An investigation using a synthetic porcine pheromone and the effect on days from weaning to conception. **Vet. Rec.** 98: 93-94. 1976.
- [32] HOLROYD, R.G. Differences between *Bos taurus* and *Bos indicus* cattle concentrating on female reproductive performance. **Proc. Beef Cattle Practice** Queensland University, Australia. 9: 31- 46. 1986.
- [33] HOLROYD, R.G.; ENTWISTLE, K.W.; SHEPERD, R.K. Effects on reproduction of estrous cycle variations, rectal temperatures and live weights in mated Brahman cross heifers. **Theriogenol** 40: 453-464. 1993.
- [34] HORNBUCKLE II, T.; OTT, R.S.; OHL, M.W.; ZINN, G.M.; WESTON, P.G.; HIXON, J.E. Effects of bull exposure on the cyclic activity of beef cows. **Theriogenol** 43: 411-418. 1995.
- [35] HUNTER, M.G. Characteristics and causes of the inadequate corpus luteum. **Gentlemen. Reprod. Fertil.** (Suppl 43): 91-99. 1991.
- [36] HURNICK, J.F.; KING, G.J.; ROBERTSON, H.A. Estrous and related behaviour in postpartum Holstein cows. **Appl. Anim. Ethol.** 2: 55-68. 1975.
- [37] INSKEEP, E.K. Factors that affect fertility during oestrus cycles with short or normal luteal phases in postpartum cows. **J. Reprod. Fertil.** 49: 493-503. 1995.
- [38] IWATA, E.; WAKABAYASHI, Y.; KAKUMA, Y.; KIKUSUI, T.; TAKEUCHI, Y.; MORI, Y. Testosterone-dependent primer pheromone production in the sebaceous gland of male goat. **Biol. Reprod.** 62: 806-810. 2000.
- [39] IZARD, M.K.; VANDENBERG, J.G. The effects of bull urine on puberty and calving date in crossbred beef heifers. **J. Anim. Sci.** 55: 1160-1169. 1982.
- [40] JEZIERSKI, T.; PRZYBYLSKA, H. Intensity of oestrus behavior in Black and White and Red and White cows, and its relation to milk performance. **Prace-I-Materialy Zootechniczne Poland** 42: 49-55. 1992.
- [41] KNIGHT, T.W.P.; LYNCH, P.R. The source of ram pheromones that stimulate ovulation in the ewe. **Anim. Reprod. Sci.** 3: 133-138. 1980.
- [42] KONONOV, V.P.; NURAKHMETOV, ZH. Physical and chemical properties of bull sex pheromones. **Soviet Agric. Sci.** 3: 58-61. 1993.
- [43] LAMOTHE-ZAVALA, C.; FREDRIKSSON, G.; KINDAHL, H. Reproductive performance of zebu cattle in Mexico. 1. Sexual Behavior and seasonal influence on estrous cyclicity. **Theriogenol** 36: 887-896. 1991.
- [44] LANDAETA-HERNANDEZ, A.J. Social, genetic, physiological and environmental factors affecting estrous duration, estrous intensity, and follicular dynamics in beef cows. Dpt Animal Sciences, University of Florida. Gainesville. Master thesis. Pp 213. 1999.
- [45] LANDAETA-HERNANDEZ, A.J.; GIANGRECO, M.; MELENDEZ, P.; BARTOLOME, J.; BENNET, F.; RAE, D.O.; HERNANDEZ, J.; ARCHBALD, L.F. Effect of biostimulation on uterine involution, early ovarian activity and first postpartum estrous cycle in beef cows. **Theriogenol** 61: 1521-1532. 2004.

- [46] LOPEZ, H.; SATTER, L.D.; WILTBANK, M.C. Relationship between level of milk production and estrous behavior of lactating dairy cows. **Appl. Anim. Behav. Sci.** 81: 209-223. 2004.
- [47] LUCY, M.C.; SAVIO, J.D.; BADINGA, L.; DE LA SOTA, R.L.; THATCHER, W.W. Factors that affect ovarian follicular dynamics in cattle. **J. Anim. Sci.** 70: 3615-3626. 1992a.
- [48] LUCY, M.C.; THATCHER, W.W.; STAPLES, C.R. Postpartum function: nutritional and physiological interactions. Chapter 16 In: Van Horn HH, and Wilcox CJ (Eds.) **Large Dairy Herd Management**. American Dairy Science Association, Champaign, IL, USA. Pp 135-145. 1992b.
- [49] MACMILLAN, K.L.; ALLISON, A.J.; STRUTHERS, G.A. Some effects of running bulls with suckling cows or heifers during pre-mating period. **N.Z.J. Exp. Agr.** 7: 121-124. 1979.
- [50] MANN, G.E.; LAMMING, G.E. The role of sub-optimal preovulatory oestradiol secretion in the aetiology of premature luteolysis during the short oestrus cycle in the cow. **Anim. Reprod. Sci.** 64: 171-180. 2000.
- [51] MONJE, A.R.; ALBERIO, R.; SCHIERSMANN, G.; CHEDRESE, J.; CAROU, N.; CALLEJAS, S.S. Male effect on the postpartum sexual activity of cows maintained on two nutritional levels. **Anim. Reprod. Sci.** 29: 145-156. 1992.
- [52] NAAZ, C.D.; MILLER, H.L. Effects of bull exposure on postpartum interval and reproductive performance in beef cows. **Can. J. Anim. Sci.** 70: 537-542. 1990.
- [53] OLSON, T.; VARGAS, C.A.; CHASE Jr., C.C.; HAMMOND, A.C. Relación entre tamaño corporal y fertilidad en Ganado Brahman. **Proc. Congres. Prod. Carne**. Asunción, Paraguay. 8/25-26. 35-49 pp. 1998.
- [54] PANDEY, S.D.; PANDEY, S.C. Effect of intranasal irrigation with Zinc sulphate on male-induced oestrus in the wild mouse, *Mus musculus domesticus*. **Anim. Reprod. Sci.** 8: 295-300. 1985.
- [55] PENY, R.C.; CORAH, L.R.; KIRACOFÉ, G.H.; STEVENSON, J.S.; BEAL, W.E. Endocrine changes and ultrasonography of ovaries in suckled beef cows during resumption of postpartum estrous cycles. **J. Anim. Sci.** 69: 2548-2555. 1991.
- [56] RAE, D.O.; KUNKLE, W.E.; CHENOWETH, P.J.; SAND, R.S.; TRAN, T., Relationship of parity and body condition score to pregnancy rates in Florida beef cattle. **Theriogenol** 39: 1143-1152. 1993.
- [57] RAMIREZ-GODINEZ, J.A.; KIRACOFÉ, G.H.; MCKEE, R.M.; SCHALLES, R.R.; KITOK, R.J. Reducing the incidence of short estrous cycles in beef cows with norgestomet. **Theriogenol** 15: 613-623. 1981.
- [58] REKWOT, P.I.; OGWU, D.; OYEDIPE, E.O. Influence of bull biostimulation, season and parity on resumption of ovarian activity of zebu (*Bos indicus*) cattle following parturition. **Anim Reprod. Sci.** 63: 1-11. 2000.
- [59] REKWOT, P.I.; OGWU, D.; OYEDIPE, E.O.; SEKONI, V.O. The role of pheromones and biostimulation in animal reproduction. **Anim Reprod. Sci.** 65: 157-170. 2001.
- [60] RHODES, F.M.; FITZPATRICK, L.A.; ENWISTLE, K.W.; DE'ATH, G. Sequential changes in ovarian follicular dynamics in *Bos indicus* heifers before and after nutritional anoestrus. **J. Reprod. Fertility** 104: 41-49. 1995.
- [61] ROBERTS, S.J. Parturition and physiology of female reproduction. Chapters VI and XII. In: Roberts SJ. (Ed.) **Veterinary Obstetrics and Genital Diseases**. Roberts, S.J. Ithaca, N.Y. USA. Pp 201-221 and 343-369. 1971.
- [62] RUKKWANSUK, T.; WENSING, T.; KRUIP, T.A.M. Relationship between triacylglycerol concentration in the liver and first ovulation in postpartum dairy cows. **Theriogenol** 51: 1133-1142. 1999.
- [63] SALDIVIA, M.; SILVA, M.O.A.; VASQUEZ, A.L.A. Sección del ligamento dorsal del pene y epididectomía en la preparación de toros detectores de celo. **Rev. Cientí. FCV-LUZ**. II (2): 7-10. 1992.
- [64] STATISTICAL ANALYSIS SYSTEM INSTITUTE (SAS). User's Guide: Statistics, Version 8. Cary, NC. USA. 2000.
- [65] SCHRICK, F.N.; SPITZER, J.C.; GIMENEZ, T.; HENRICKS, D.M.; JENKINS, T.C.; PLAYER, T.G. Effect of dietary energy restriction on metabolic and endocrine responses during the estrous cycle of the suckled beef cows. **J. Anim. Sci.** 68: 3313-3321. 1990.
- [66] SHIPKA, M.P.; ELLIS, L.C. No effects of bull exposure on expression of estrous behavior in high-producing dairy cows. **Appl. Anim. Behav. Sci.** 57: 1-7. 1998.
- [67] SHIPKA, M.P.; ELLIS, L.C. Effects of bull exposure on postpartum ovarian activity of dairy cows. **Anim. Reprod. Sci** 54: 237-244. 1999.
- [68] SHORT, R.E.; BELLOWES, R.A.; STAIGMILLER, R.B.; BERARDINELLI, J.G.; CUSTER, E.E. Physiological mechanisms controlling anestrus and infertility in postpartum beef cattle. **J. Anim. Sci.** 68. 799-816. 1990.
- [69] SINGH, P.B.; BROWN, R.E.; ROSER, B. MHC antigens in urine as olfactory recognition cues. **Nature**. 327: 161-164. 1987.
- [70] SINGH, P.B. Chemosensation and genetic individuality. **Reprod** 121: 529-539. 2001.
- [71] SMITH, V.G.; CHENAULT, J.R.; MCALLISTER, J.C.; LAUDERDALE, J.W. Response of postpartum beef cows

- to exogenous progestogens and gonadotropin releasing hormone. **J. Anim. Sci.** 64: 540-551. 1987.
- [72] SOTO-BELLOSO E.; RAMIREZ-IGLESIAS, L.; GUEVARA, L.; SOTO-CASTILLO, G. Bull effect on the reproductive performance of mature and first calf-suckled zebu cows in the tropics. **Theriogenol** 48: 1185-1190. 1997.
- [73] SRIKANDAKUMAR, A.; INGRAHAM, R.H.; ELLWORTH, M.; ARCHBALD, L.F.; LIAO, A.; GODKE, R.A. Comparison of a solid-phase, no-extraction radioimmunoassay for progesterone with an extraction assay for monitoring luteal function in the mare, bitch and cow. **Theriogenol** 26: 779-793. 1986.
- [74] STAGG, K.; SPICER, L.J.; SREENAN, J.M.; ROCHE, J.F.; DISKIN, M.G., Effect of calf isolation on follicular wave dynamics, gonadotropin and metabolic hormone changes, and interval to first ovulation in beef cows fed either of two energy levels postpartum. **Biol. Reprod.** 59: 777-783. 1998.
- [75] STEVENSON, J.S. Clinical reproductive physiology of the cow. Chapter 32. In: Youngquist RS. (Ed.) **Current Therapy in Large Animal Theriogenology**. W. B. Saunders Co., Philadelphia. 257-267 pp. 1997.
- [76] STUMPF, T.T., WOLFE, M.W., WOLFE, P.L., DAY, M.L., KITTOK, R.J., KINDER, J.E. Weight changes prepartum and presence of bulls postpartum interact to affect duration of postpartum anestrus in cows. **J. Anim. Sci.** 70: 3133-3137. 1992.
- [77] TEGEGNE, A.; GELETO, A.; KASSA, T. Short luteal phases and ovulations without oestrus in primiparous Borana (*Bos indicus*) cows in the central highlands of Ethiopia. **Anim. Reprod. Sci.** 31: 21-31. 1993.
- [78] THATCHER, W.W.; GUILBAULT, L.A.; DROST, M. Normal uterine physiology and involution. **Proc. Am. Assn't. Bovine Practitioners**. Des Moines, IA, Nov 27-30. 75-80 pp. 1984.
- [79] UMEMURA, K.; SUGAWARA, K.; ITO, I. The source of odour emitted from oestrus cows. **Jap J. Zoot. Sci.** 59: 779-786. 1988.
- [80] VAN VLIET, J.H.; VAN EERDENBURG, F.J.C.M. Sexual activities and oestrus detection in lactating Holstein cows. **Appl. Anim. Behav. Sci.** 50: 57-69. 1996.
- [81] VARGAS, C.; OLSON, T.A.; CHASE, C.C.; HAMMOND, A.C.; ELZO, M.A. Influence of frame size and body condition score on performance of cattle. **J. Anim. Sci.** 77: 3140-3149. 1999.
- [82] WILLIAMS, G.L.; GRIFFITH, M.K. Sensory and behavioral control of gonadotropin secretion during suckling-mediated anovulation in cows. **J. Reprod. Fertil.** (Suppl. 49): 463- 475. 1995.
- [83] ZALESKY, D.D.; DAY, M.L.; GARCIA-WINDER, M.; IMAKAWA, K.; KITTOK, R.J.; D'OCCHIO, M.J.; KINDER, J.E. Influence of exposure to bulls on resumption of estrous cycles following parturition in beef cows. **J. Anim. Sci.** 59: 1135-1139. 1984.
- [84] ZOLLERS, W.G.; GARVERICK, H.A.; SALFEN, B.E.; MOFFAT, R.J.; YOUNGQUIST, R.S.; SMITH, M.F. Receptor concentrations for progesterone and oxytocin in endometrial tissue from postpartum beef cows expected to have a short or normal estrous cycle. **Biol. Reprod.** 44 (Suppl 1):160 (Abst). 1991.