THE EFFECT OF NUTRITIONAL LEVEL ON ADVANCING AGE AT PUBERTY IN NELORE HEIFERS

Marco Aurélio Romano[1]
Walquiria Hippolito Barnabe[2]
Antonio Euvídio Dias Feliciano e Silva[3]
Alfredo Ribeiro de Freitas[4]
Renata Morino Romano[5]

ASTRACT

The objective of this study was to verify the effect of nutritional level on advancing age at Renata puberty in Nelore heifers. Thirty-one animals at 6 months of age were randomly assigned in two groups: GI, or control, with 15 heifers that received a basal diet, and GII, or supplemented, with 16 heifers that received a diet with higher energy and crude protein levels. The development of the animals was accompanied by weighing them at 21-day intervals to verify the daily weight gain from 6 months to puberty. The diets were readapted when necessary. The reproductive status was monitored by ultrasound every two days; and by progesterone concentrations from blood samples drawn twice a week. The ovulation was determined when progesterone concentrations were higher than 1 ng/mL in three consecutive samples, and by ultrasound images of corpus luteum; and oestrous behavior in some animals. The GII group manifested puberty at 16.33 ±0.89 months of age and GI at 20.58 ±1.83 months of age ($p<0.05$). The mean weight at puberty was 302.33 ±21.31Kg for GI and 326.19 ±27.78Kg for GII. The daily weight gain was 0.647 ±0.08Kg for GII and 0.447 ±0.03Kg for GI ($p<0.05$). Progesterone levels at puberty were 2.7801 ±0.27 ng/mL for GI

[1] Universidade Estadual do Centro-Oeste, Departamento de Medicina Veterinária. Mailing Address of Romano: Rua Afonso Botelho, n. 1208 CEP 85012030. Guarapuava – PR – Brasil. Phone Number: 055 42 6263713. Fax Number: 055 42 6245955. E-mail: mromano@unicentro.br.
[2] Universidade de São Paulo, Faculdade de Medicina Veterinária e Zootecnia.
[3] EMBRAPA, Cenargen.
[4] EMBRAPA, CPPSE.
[5] Médica Veterinária Autônoma.

and 2.5747 ±0.26 ng/mL to GII. Only 13% of all heifers manifested oestrous behavior. The nutritional supplementation decreased the age of puberty onset in GII and both groups manifested puberty earlier than heifers feeding only on pasture in Subtropical conditions. It is possible to have an earlier puberty onset in Nelore heifers through nutritional supplementation, without negative effects on their reproductive physiology.

Key words: puberty; heifers; nelore

RESUMO

O objetivo deste trabalho foi verificar o efeito do nível nutricional na antecipação da idade à puberdade em novilhas nelore. Trinta e um animais de seis meses de idade foram aleatoriamente divididos em dois grupos: GI, ou testemunha, com 15 novilhas que receberam uma dieta basal, e GII ou suplementado, com 16 novilhas que receberam uma dieta com altos níveis de energia e proteína bruta. O desenvolvimento dos animais foi acompanhado por pesagens a cada 21 dias para verificar o ganho de peso diário. As dietas foram re-adaptadas quando necessárias. O estado reprodutivo foi monitorado por ultra-sonografia a cada dois dias, e por concentração de progesterona de amostras de sangue colhidas duas vezes por semana. A ovulação foi determinada quando as concentrações de progesterona forma maior que 1ng/mL em três amostras consecutivas, e por imagem ultra-sonográfica de corpo lúteo; e comportamento de estro em alguns animais. O grupo GII manifestou puberdade aos 16,33 ± 0,89 meses de idade e o GI 20,58 ± 1,83 meses de idade (p £ 0,05). A média do peso à puberdade foi 302,33 ± 21,31 Kg para o GI e 326,19 ± 27,78 Kg para o GII. O ganho médio diário foi de 0,647 ± 0,08 Kg para o GII e 0,447 ± 0,03 Kg para o GI (p £ 0,05). Os níveis de progesterona à puberdade foram 2,7801 ± 0,27 ng/mL para o G I e 2,5747 ± 0,26 ng/mL para o GII. Somente 13% das novilhas apresentaram manifestação de estro. A suplementação Nutricional retrocedeu a idade à puberdade no grupo GII e ambos os grupos manifestaram puberdade mais cedo que novilhas se alimentando somente em condições sub-tropicais de pastoreio. É possível antecipar o início da puberdade através da suplementação nutricional sem efeitos negativos na fisiologia reprodutiva de novilhas Nelore.

Palavras-chave: puberdade; novilhas; nelore

INTRODUCTION

An early onset of puberty is important to obtain a better reproductive performance and puberty is correlated with rate of body weight gain from the time of birth

(ARIJE; WILTBANCK, 1971; GARCIA; CALDERON, 1978; PLASSE et al., 1968). Although it's known that energy intake can affect ovarian function, the effects of nutritional levels on the growth pattern in cattle are not well known (MURPHY et al, 1991).

Some studies have shown that *Bos indicus* cattle reach puberty later than *Bos taurus*. The difference between them can reach twelve months (OYEDIPE et al, 1982; SILVA; ROMANO, 1991; WILTBANK et al, 1969) and it ultimately results in a delay in the age at first calving (HOLNESS et al, 1978; ROCHE; BOLAND, 1991; SAVIO et al, 1990).

The protein amount in pasture is low in tropical and subtropical regions, resulting in a poor diet for cattle (PFANDER, 1971). Probably, this is one of the main factors that influence growth, age at puberty and reproductive performance in these regions (OYEDIPE et al, 1982), due to a delay on the development of the endocrine system (DAY et al, 1986).

Regarding dietary levels, poor diets in post partum (BUTLER; SMITH, 1989) or pre partum (PERRY et al, 1991; SPICER et al, 1990; VILLA-GODOY et al, 1988) periods increase the interval between calving and next ovulation. Prolonged reduction in the energy intake in beef heifers suppresses the ovulation by alterations in the hypothalamic-pituitary-ovarian axis. The onset of ovarian acyclicity is preceded or accompanied by the loss of a positive feedback signal and progesterone priming ability (DAWUDA et al, 2002).

In contrast, heifers receiving a high-energy diet reach puberty at younger aged (372 ± 7days) and heavier body weight (263 ± 7 kg) than heifers fed only from pasture with low energy intake (435 ± 9 days and 221 ± 3 kg) (BERGFELD et al, 1994). The onset of puberty in animals with body weight gain of 1000 g per day was 12.9 mo-old and 379.6 kg, while heifers gaining 600 g per day reached puberty at 13.8 mo-old and 339.1 kg of body weight (FAJERSON; EDQVIST, 1993). Likewise, supplemented animals attained puberty earlier (19 moth) and heavier (207 kg) than animals receiving medium nutritional level (21.36 moth and 187 kg) or low nutritional level (23.37 moth and 161 kg) (OYEDIPE et al, 1982).

During the prepubertal period, the follicular development promotes the maintenance of basal blood estrogen levels, which inhibits LH secretion. Prior to puberty, there is an increase in estrogen levels and stimulation of LH secretion, resulting in the first ovulation (ADAMS et al, 1992; DAY et al, 1986; SCOTT et al, 1990; SUNDERLAND et al, 1994).

In heifers with adequate feed intake during the prepubertal period the LH secretion starts gradually increasing four months before puberty (DAY, 1984; SWANSON et al, 1972). In the absence of ovarian factors, in ovariectomized heifers, nutrition affects LH secretion. When the dietary energy intake increases, the level of LH secretion also

increases, denoting the influence of nutrition on the LH secretion pattern (MACKEY et al, 2000; SCOTT et al, 1990).

Based on these results, we believe that an earlier puberty onset can be attained by a better nutrition, by diets with higher energy levels than only pasture-based diets. The objective of the present experiment is to evaluate the effect of supplementary nutrition on the age and weight at puberty in Nelore heifers (*Bos indicus).* The hypothesis to be tested is that the onset of puberty in animals receiving high energy and protein diets occurs earlier than in animals receiving basal diets.

MATERIALS AND METHODS

ANIMALS

Thirty-one prepubertal 6 month-old heifers of Nelore breed weighing 129.58 ± 8.59 kg were allocated in individual stalls during 48 days, receiving individually fed. Each animal received a daily diet composed by stargrass (*Cynodon* spp.) ad libitum, coast cross (*Cynodon dactylon)* hay ad libitum and two kilograms of a concentrated mixture. The concentrated mixture was composed by 30% cracked corn grains, 7.7% soybean meal, 59.2% wheat meal, 2.4% limestone and 0.7% mineral mix to adapt to experimental conditions.

EXPERIMENTAL DESIGN

After experimental adaptation, the heifers were randomly assigned in two groups, one group (GI – or control) was composed of fifteen animals that received a basal diet with daily weight gain of 0.300 Kg per animal, and another group (GII – or supplemented) composed by 16 animals receiving a diet with higher energy and crude protein levels, with daily weight gain of 0.900 Kg per animal. The animals were allotted in four pastures, two for GI and two for GII. This experiment ran for 427 days, initiating in May and finishing in July of next year. The animals were weighed every 21 days, and the diets were readapted to maintain weight gain. All heifers were exposed to the teaser bull twice a day at 8:00am and 5:00pm to determine oestrous manifestations. The teaser bulls were alternated daily. When puberty was getting closer (estimated by weight) they stayed with the heifers all the time in the pasture. Blood samples were collected twice a week to determine progesterone concentrations (Coat-A-Count Progesterone, DPC, Medlab). Ovarian activity was monitored by ultrasound examination every two days to verify its development and structures like follicles and corpora lutea. The ovulation was determined by increasing levels of progestin starting at 1 ng/mL in three consecutive samples, by

ultrasound images of corpus luteum and by oestrous behavior in some animals. During all experimental period, the animals received veterinary care, like vaccination and endo and ectoparasites control.

DIETS

GI – stargrass, soybean meal and mineral salt ad libitum.

GII – stargrass ad libitum and mixed feed composed by 30% corn cracked grains, 7.7% soybean meal, 59.2% wheat meal, 2.4% limestone and 0.7% mineral mix.

Periodically, concentrated mixture, hay and pasture samples were collected and analyzed to determine total dry matter, crude protein, neutral detergent fiber and gross energy. The mean composition of foods is represented in Table 1.

Table 1. Mean composition in dry matter of foods supplied to Nelore heifers during experimental phase

Food	% DMB	% CP	% NDF	GE (cal/g)
Ration	86.60	17.05	31.22	4453.70
Coastcrosss hay	86.22	9.37	79.31	4417.70
Stargrass	25.89	9.30	81.43	4226.99
Soybean meal	88.31	49.60	7.79	4682.25

DMB: *dry matter basis; CP: crude protein; NDF: neutral detergent fiber; GE: gross energy.*

STATISTICAL ANALYSIS

The means of final weight (kg), total weight gain (kg), age at puberty (months), weight at puberty (kg) and daily weight gain (kg/day) were compared between the two groups by completed casually delineate to verify the efficiency of both treatments (procedure GLM and REG of SAS, SAS INSTITUTE, 1993).

RESULTS AND DISCUSSIONS

The supplementary feeding receded the age at puberty in GII (Table 2) and this group manifested puberty earlier than observed in Nelore animals (SILVA; ROMANO, 1991). The evidence of the earlier onset of puberty through supplementary diet showed the correlation of body weight gain from weaning to puberty and the pubertal age (ARIJE; WILTBANCK, 1971; GARCIA; CALDERON, 1978; PLASSE et al, 1968).

The results observed in GI confirmed the observations that puberty onset can be six to twelve months later in Zebu cattle than in European cattle (OYEDIPE et al, 1982;

WILTBANCK et al, 1969) and that the puberty on Nelore heifers feeding only on pasture occurs at 23.3 ± 4.0 months of age (SILVA; ROMANO, 1991).

Table 2. Initial and final body weight, total weight gain, daily weight gain, of GI (control) and II (supplemented) Nelore heifers

Animals	Initial weight (kg)	Final weight (kg)	Total weight gain (kg)	Daily weight gain (g/day)	Animals	Initial weight (kg)	Final weight (kg)	Total weight gain (kg)	Daily weight gain (g/day)
GI					GII				
1	116	302	186	0.392	2	131	391	260	0.681
3	130	268	138	0.291	4	127	360	233	0.610
5	126	343	217	0.568	6	121	364	243	0.636
8	128	321	193	0.505	7	120	374	254	0.665
10	128	333	205	0.432	9	136	395	259	0.678
14	130	317	187	0.490	11	138	372	234	0.613
15	135	323	188	0.386	12	133	381	248	0.649
17	142	326	184	0.387	13	112	351	239	0.626
18	140	318	178	0.375	16	126	386	260	0.681
20	136	355	219	0.573	19	123	398	275	0.693
21	135	311	176	0.460	23	131	381	250	0.655
22	112	309	197	0.516	24	119	359	240	0.628
28	131	341	210	0.550	25	132	377	245	0.641
29	134	320	186	0.392	26	135	355	220	0.576
31	130	312	182	0.383	27	128	381	253	0.662
					30	152	403	251	0.657
Means	130.2 (± 7,97)	319.93 (± 20.21)	189.73[a] (±19.71)	0.447[b] (± 0.08)		129 (±9.35)	376.75 (± 15.79)	247.75[a] (± 13.15)	0.647[b] (± 0.03)

a,b differ in column (p £ 0,05).
SEM = standard error of the mean.

Studies have shown that the age at first calving is later in Zebu cattle (HOLNESS et al, 1978; ROCHE; BOLAND, 1991; SAVIO et al, 1990), probably due to a delayed puberty onset in heifers under low nutritional levels (DAY et al, 1986) found in tropical and subtropical regions (PFANDER, 1971). This was also verified in this study given that animals in GI, kept under a low nutritional plan, had shown puberty later than GII. Maintaining a low nutritional level probably delays the maturation of the endocrine system (DAY et al, 1986) and slows down the first ovulation similarly as what happens during the post-partum period (BUTLER; SMITH, 1989; PERRY et al, 1991; SPICER et al, 1990; VILLA-GODOY et al, 1988).

Table 3. Age, weight and plasmatic progesterone at puberty in Nelore heifers of GI (control) and GII (supplemented)

Animals	Age at puberty (mo)	Weight at puberty (Kg)	Progesterone levels (ng/ml)	Animals	Age at puberty (mo)	Weight at puberty (Kg)	Progesterone levels (ng/ml)
GI				GII			
1	21	294	4.0382	2	17	333	3.0789
3	21	265	3.2712	4	17	320	1.2544
5	18	280	3.1121	6	16	286	1.5451
8	17	271	1.2628	7	15	289	2.0430
10	22	338	5.2513	9	19	370	1.0317
14	18	294	1.8721	11	19	344	3.2836
15	22	325	1.1312	12	17	330	3.2990
17	22	330	2.1000	13	18	352	2.8000
18	22	328	2.1901	16	15	291	2.4137
20	19	331	2.1376	19	19	369	3.2842
21	17	276	3.7693	23	16	312	3.7377
22	18	275	2.4997	24	17	322	4.2010
28	17	278	3.5918	25	19	356	3.7618
29	21	338	3.2919	26	16	300	2.0473
31	23	312	2.1827	27	16	304	1.7614
				30	16	341	1.6551
Means	19.87 [a] (± 0.47)	302.33 (± 27.31)	2.78 (± 0.27)		17.00 [b] (± 0.46)	326.19 (± 27.78)	2.57 (± 0.26)

a,b differ in column (p £ 0,05)
SEM = standard error of the mean

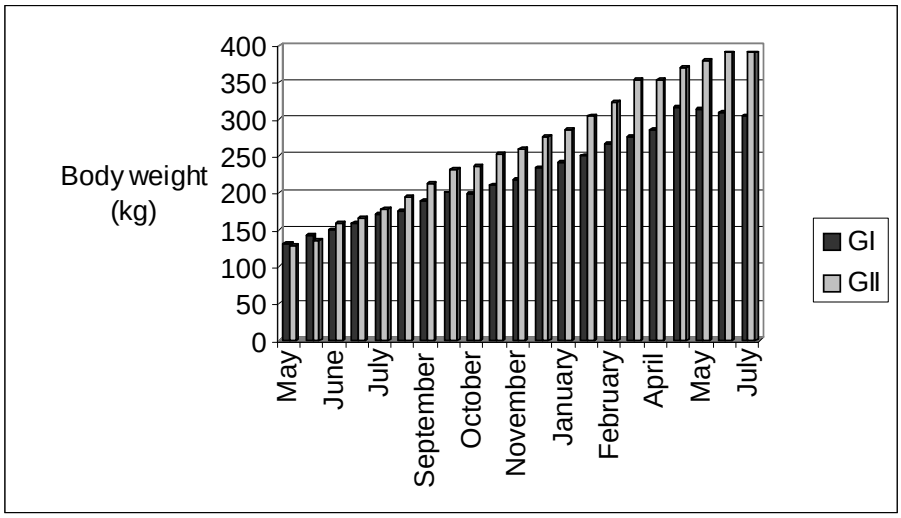
The mean weight at puberty did not change between groups GI and GII (Table 2) demonstrating that a minimum body weight is necessary to reach puberty (SUNDERLAND et al, 1994). However, the animals of GI took more time to achieve this weight. These results were in accordance with observations of body weigh gain in Nelore heifers kept on pasture (SILVA; ROMANO, 1991). Heifers in GII reached puberty heavier and younger (BERGFELD et al, 1994; FAJERSON; EDQVIST, 1993; OYEDIPE et al, 1982) than heifers in GI.

Only four animals (13%) manifested oestrous behavior at onset of puberty and homosexual behavior was not observed in any of the animals.

The levels of plasmatic progesterone at puberty in both groups are described on Table 3. These levels are variable between animals and groups, did not being observed

any correlation with the age, weight or treatments. The smaller level of plasmatic progesterone was 1.0317 ng/mL and bigger was 5.2513 ng/mL, and these values did not showed statistical differences.

Table 4. Mean weight evolution of two groups during experimental phase



The progression of the mean weight for both groups is shown on Table 4. This period may be divided in two segments, according to weather conditions observed in the Brazilian Southeast. In this region, rain is abundant from October to March, the photoperiod is large and the pasture conditions are better. The period from April to September is characterized by drought, with little precipitation; shorter photoperiod and poor pasture conditions.

As a result of dietary supplementation, animals in GII had grown during the whole observed period. They did not show loss of weight at any period. Animals in GI started to lose weight on May, when they were 18 month-old; because of the worse pasture conditions in this period and the increasing energy demand of the heifers.

In field conditions, the observed growth pattern in beef cattle is similar to that of GI – a result of seasonal pasture conditions. The seasonality and its consequent loss of weight are also responsible for delaying the first ovulation and increasing the calving intervals.

CONCLUSIONS

According to results and discussions presented, it is possible to reduce the age at puberty in Nelore heifers through nutritional supplementation, without negative effects on their reproductive physiology. However, a minimum body weight is necessary to puberty

manifestation in both nutritional levels. While some animals showed an unsatisfactory use of nutrients others showed a better one, suggesting a possible association to genetic characteristics. The homosexual behavior was not used to determine the onset of puberty in Nelore heifers. Non-supplemented animals will loss weight during seasonal variations and will have a delayed first ovulation.

REFERENCES

ADAMS, G.. P.; MATTERI, R. L.; GINTHER, O. J. Effect of progesterone on ovarian follicles, emergence of follicular waves and circulating follicle-stimulating hormone in heifers. *J. Reprod. Fertil.*, v. 95, p. 627-640, 1992.

ARIJE, O. F.; WILTIBANCK, J. N. Age and weight at puberty in Hereford heifers. *J. Anim. Sci.*, v. 33, p. 401-406, 1971.

BERGFELD, E. G. M.; KOJIMA, F. N.; CUPP, A. S.; et al. Ovarian follicular development in prepubertal heifers is influenced by level of dietary energy intake. *Biol. Reprod.*, v. 51, p. 1051-1057, 1994.

BUTLER, W. R.; SMITH, R.D. Interrelationships between energy balance and postpartum reproductive function in dairy cattle. *J. Dairy Sci.*, v. 72, p. 767-783, 1989.

DAWUDA, P. M.; SACIFE, J. R.; HUTCHINSON, J. S.; et al. Mechanisms linking under nutrition and ovarian function in beef heifers. *Anim. Reprod. Sci.*, v. 74, n. 1-2, p. 11-26, 2002.

DAY, M. L.; IMAKAWA, K.; GARCIA-WINDER, M.; et al. Endocrine mechanisms of puberty in heifers. Estradiol negative feedback regulation of luteinizing hormone secretion. *Biol. Reprod.*, v. 31, p. 332, 1994.

DAY, M. L.; IMAKAWA, K.; ZALESKY, D. D.; et al. Effects of restriction of dietary energy intake during the prepubertal period on secretion of luteinizing hormone and responsiveness of the pituitary to luteinizing hormone-releasing hormone in heifers. *J. Anim. Sci.*, v. 62, p. 1641-1648, 1986.

FAJERSON, P.; EDQVIST, E. E. Ultrasonography characterization of onset of puberty; first ovulation is followed by the development of a corpus luteum and a short luteal phase in Brown Swiss and Zebu heifers. *J. Anim. Sci.*, v. 71, suppl. 1, p. 205, 1993.

GARCIA, P. M.; CALDERON, V. M. Puberty of dairy calves in a herd on the coast in Peru. *Anim. Breed. Abstr.*, v. 46, p. 158, 1978.

HOLNESS, D. H.; HOPLEY, J. D. H.; HALE, D. H. The effects of plane of nutrition, live weight, temporary weaning and breed on the occurrence of oestrous in beef cows during the post-partum period. *Anim. Prod.*, v. 26, p. 47-54, 1978.

MACKEY, D. R.; WYLIE, A. R.; SREENAN J. M.; et al. The effect of acute nutritional change on follicle wave turnover, gonadotropin and steroid concentration in beef heifers. *J. Anim. Sci.*, v. 78, n. 2, p. 429-42, 2000.

MURPHY, M. G.; ENRIGHT, W. J.; CROWE, M. A.; et al. Effect of dietary intake on pattern of growth of dominant follicles during the oestrous cycle in beef heifers. *J. Reprod. Fertil.*, v. 92, p. 333-338, 1991.

OYEDIPE, E. O.; OSORI, D. I. K.; AKEREJOLA, O.; et al. Effect of level of nutrition on onset of puberty and conception rates of zebu heifers. *Theriogenology*, v. 18, n. 5, p. 525-539, 1982.

PERRY, R. C.; CORAH, L. R.; COCHRAN, R. C.; et al. Influence of dietary energy on follicular development, serum gonadotropins, and first postpartum ovulation in suckled beef cows. *J. Anim. Sci.*, v. 69, p. 3762-3773, 1991.

PFANDER, W. H. Animal nutrition in the tropics - problems and solutions. *J. Anim. Sci.*, v. 33, p. 843-849, 1971.

PLASSE, D.; WARNICK, A. C.; KOGER, M. Reproductive behavior of *Bos indicus* females in sub-tropical environment. I puberty and ovulation frequency in Brahman x British heifers. *J. Anim. Sci.*, v. 27, p. 94-100, 1968.

ROCHE, J. F.; BOLAND, M. P. Turnover of dominant follicle in cattle of different reproductive states. *Theriogenology*, v. 35, n. 1, p. 81-90, 1991.

SAS INSTITUTE. SAS/STAT User's guide: statistics, version 6, v2, 4.ed., Cary, 1993.

SAVIO, J. D.; BOLAND, M. P.; HYNES, N.; et al. Resumption of follicular activity in the early post-partum period of dairy cows. *J. Reprod. Fertil.*, v. 88, p. 569-579, 1990.

SCOTT, G. R.; DEYER, R. M.; HU, Y.; et al. Regulation of luteinizing hormone secretion in prepubertal heifers fed an energy deficient diet. *Biol. Reprod.*, v. 43, p. 450-456, 1990.

SILVA, A. E. D. F.; ROMANO, M. A. Alguns aspectos da maturidade de fêmeas da raça Canchim, Nelore e meio sangue Canchim e Nelore, Colégio Brasileiro de Reprodução Animal (Eds.) In: Congresso Brasileiro de Reprodução Animal, IX. *Anais*. Brasil, Belo Horizonte, MG, 1991.

SPICER, L. J.; TUCKER, W. B.; ADAMS, G. D. Insulin-like growth factor-I in dairy cows: relationships among energy balance, body condition, ovarian activity and estrous behavior. *J. Dairy Sci.*, v. 73, p. 929-937, 1990.

SUNDERLAND, S. J.; CROWE, M. A.; BOLAND, M. P. et al. Selection, dominance and atresia of follicles during the oestrous cycle of heifers. *J. Reprod. Fertil.*, v. 101, n. 3, p. 547-555, 1994.

SWANSON, L. V.; HAFS, H. D.; MOROW, D. A. Ovarian characteristics and serum LH, prolactin, progesterone and glucocorticoid from first oestrous to breeding size in Holstein heifers. *J. Anim. Sci.*, v. 34, p. 284, 1972.

VILLA-GODOY, A.; HUGHES, T. L.; EMERY, R. S.; et al. Association between energy balance and luteal function in lactating dairy cows. *J. Dairy Sci.*, v. 71, p. 1063-1072, 1988.

WILTBANK, J. N.; KASSON, C. W.; INGALLS, J. E. Puberty in crossbred and straightbred beef heifers on two levels of feed. *J. Anim. Sci.*, v. 29, p. 602-605, 1969.