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Short-term exposure of high body weight heifers to testosterone-treated steers increases pregnancy rate during a late autumn-early winter natural service

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Abstract

Exposure to bulls seem to stimulate cyclic activity in postpartum beef cows, hasten puberty and increase pregnancy rate in heifers. The aim of this experiment was to determine if short-term exposure of pubertal beef heifers to testosterone-treated steers prior to late autumn-early winter bull breeding would increase pregnancy rates. An additional objective was to determine if body weight at the beginning of biostimulation affects the response to the testosterone-treated steers. Hereford heifers (n = 246, 17-22 months of age and 240 to 360 kg) were isolated from bull contact 30 days before the onset of the experiment and assigned to two experimental groups. One hundred and twenty seven heifers were joined with nine testosterone-treated steers (Days 0-15), whereas the other 119 heifers remained isolated from any contact with males. On Day 15, all heifers were joined with 10 bulls for 30 days (Day 45). Pregnancy was determined on Days 60 and 75. On Day 60, no differences were observed between groups in the frequency of pregnant heifers (18.2 vs 7.7% pregnant). On Day 75, more heifers in the biostimulation group that had an initial body weight ≥310 kg were pregnant (61.1 vs 32.4%, for stimulated and non-stimulated heifers respectively, P = 0.016), whereas there were no significant differences in heifers with lower initial body weight (17.9 vs 30.3% pregnant, for stimulated and non-stimulated heifers respectively). In conclusion, pregnancy rate was increased in high body weight heifers during late autumn-early winter natural service following pre-breeding exposure of the heifers to testosterone-treated steers.

Keywords: biostimulation, male effect, puberty, socio-sexual signals.

Introduction

Beef productivity is directly related to age at puberty, especially in seasonal breeding systems (Ferrell, 1982). The main determinants of age at puberty are body weight and weight gain (Wolfe *et al*., 1990). In Uruguay, beef heifers managed under grazing conditions begin cyclic activity between 15 and 17 months of age, if body weight is between 278 and 295 kg (Quintans *et al*., 2004, 2007). In extensively grazing-based systems, as those that predominate in our region, Hereford X Aberdeen Angus heifers reach puberty at approximately 15 months and approximately 300 kg only if high body weight gains (0.65 kg/d) are attained during the previous winter (Quintans *et al*., 2004). However, from May (mid-autumn), and during winter grazing heifers lose weight and in most cases cyclic activity ceases (Quintans *et al*., 2003).

The effect that the presence of males has on inducement of the female estrus and ovulation through various signals (genital stimulation, pheromones or other less defined external cues), is widely known as biostimulation. The continuous presence of bulls stimulates cyclic activity in postpartum cows (Zalesky *et al*., 1984; Rekwot *et al*., 2000a; Landaeta-Hernandez *et al*., 2004; Berardinelli and Joshi, 2005; Miller and Ungerfeld, 2008). Biostimulation may also be used to advance puberty in heifers. Although in some trials exposure to bulls did not have any positive effect on the onset of puberty (MacMillan *et al*., 1979), other authors reported an earlier puberty onset in exposed Bos taurus (Roberson *et al*., 1991; Assis *et al*., 2000), crossbreed (Hereford x Nelore; Quadros and Piva-Lobato, 2004) and Bos indicus (Oliveira *et al*., 2009) heifers. Bastidas *et al*. (1999) also reported that bull exposure promoted ovarian follicular growth rate in prepubertal Brahman heifers. Advancement of heifers' puberty using biostimulation may be of considerable commercial significance if it increases the proportion mated or inseminated and conceiving during the first 3 weeks of a breeding program (MacMillan *et al*., 1979).

The aim of this experiment was to determine if short-term exposure of pubertal beef heifers to testosterone-treated steers prior to a late autumn-early winter bull breeding would increase pregnancy rates. An additional objective was to determine if body weight at the beginning of biostimulation affects the response to testosterone-treated steers.

Materials and Methods

Animals and location

The experiment was carried out at a commercial farm in Soriano, Uruguay (33º S) between May and June (late autumn - early winter) with 246 heifers selected from a herd of 276 Hereford heifers, 17-22 months old. Heifers with initial body weight <240 or >360 kg were excluded from the experiment.

Ten Angus bulls (4-6 years) were selected according to a breeding soundness evaluation performed one month before the start of the experiment and previous breeding experience. Evaluation included a general physical examination and a particular reproductive examination of testes, epididymydes and accessory sexual glands. Collection and evaluation of semen was not done.

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Management

Heifers were isolated from bull contact (minimum distance = 4000 m) during the 30 days before the onset of the experiment. On May 2nd (Day 0), heifers were weighed and allocated to three categories according to their body weight: Low Weight: 240-269.5 kg; Medium Weight: 270-309.5 kg; and High Weight: 310-360 kg.

Within each weight range, heifers were randomly allocated to two experimental groups. Whereas heifers in biostimulated group were joined with nine testosterone-treated steers, heifers in the isolated group remained isolated from any contact with males until Day 15. Steers received 1 g testosterone propionate im (Testosterona Ultra Lenta Fuerte, Dispert, Montevideo, Uruguay) on Days -10, 0, and 7.

On Day 15, steers were removed and both groups of heifers were joined with the bulls and remained together as a single group until Day 45. On Days 60 and 75, pregnancy was determined by transrectal ultrasonography (Aloka 500 with a 5 MHz transducer, Tokyo, Japan). Most heifers were weighed again on Day 61, although some of them were not weighed due inclement weather. Numbers of heifers per group, and initial and final body weight for each category are summarized (Table 1).

Table 1. Number of animals, initial body weight, and body weight (BW) loss in heifers that were biostimulated with testosterone-treated steers or that remained isolated from males. Heifers were grouped as low body weight (LW, 240-269.5 kg), medium body weight (MW, 270-309.5 kg) or high body weight (HW, 310-360 kg).

	Biostimulated			Isolated			Overall BW Loss (kg)
	n	Initial BW (kg)	BW Loss (kg)	n	Initial BW (kg)	BW Loss (kg)	
LW	35	253.6 ± 1.4	13.9 ± 2.3	30	257.0 ± 1.4	14.8 ± 1.4	13.7 ± 1.2 [a]
MW	56	290.0 ± 1.4	8.5 ± 1.7	55	287.7 ± 1.4	10.3 ± 1.6	8.6 ± 1.2 [b]
HW	36	330.7 ± 2.4	6.8 ± 1.4	34	331.5 ± 2.7	9.0 ± 1.6	8.6 ± 1.1 [b]

a vs b: $P < 0.01$.

Statistical analyses

The change of body weight (Days 0 to 61) for each category was compared with a two-way ANOVA, in which the main effects were the categories and the experimental group. The accumulated frequency of pregnant heifers in each body weight category on Days 60 and 75 was compared using a Chi-square test.

Results

There were no differences ($P > 0.05$) between biostimulated and isolated heifers for either initial or final body weight (Table 1). However, Low Weight heifers lost more weight than Medium Weight and High Weight heifers (13.7 ± 1.2, 8.6 ± 1.2, and 8.6 ± 1.1 kg respectively, $P < 0.01$).

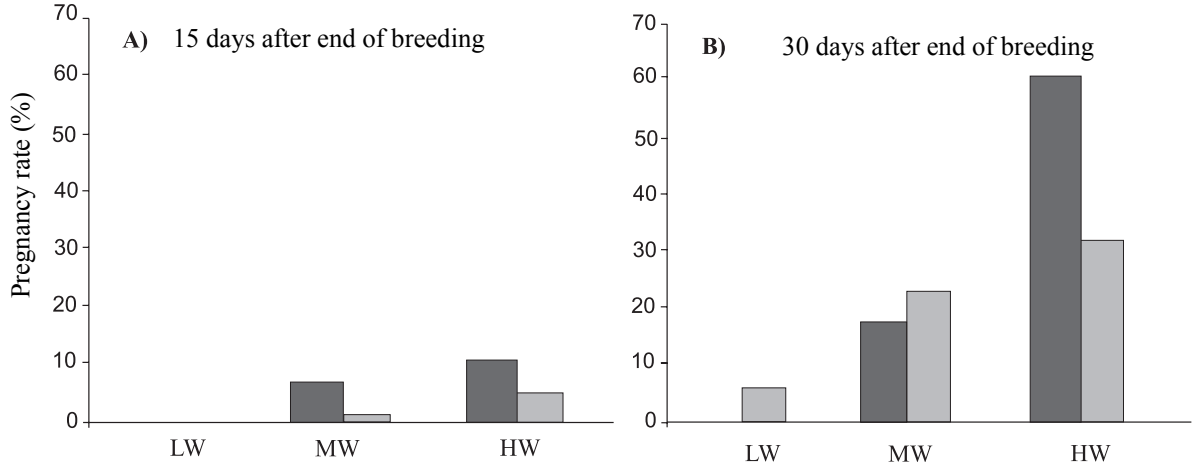

Figure 1. Pregnancy rate in heifers that were in contact with testosterone-treated steers (black bars) or that remained isolated from males (grey bars) for 15 days prior to a late autumn-early winter bull breeding for 30 days. Pregnancy was determined 15 (A) and 30 (B) days after the end of breeding. Prior to allocation to treatment groups, heifers were grouped as low body weight (LW, 240-269.5 kg), medium body weight (MW, 270-309.5 kg) or high body weight (HW, 310-360 kg). Pregnancy rates were similar among group 15 days after the end of breeding. However, 30 days after the end of breeding (B) pregnancy rates were higher ($P < 0.2$) in biostimulated heifers of the HW group.

On Day 60, there were no significant differences in pregnancy rates between treatment groups or among body weight categories (Fig. 1A). However, on Day 75, for High Weight heifers, pregnancy rate was higher in biostimulated than isolated heifers (22/36 vs 11/34 respectively, P = 0.016; Fig. 1B), with no other significant differences between the two treatments for either of the two remaining weight categories.

Discussion

In the present study, High Weight heifers exposed to testosterone-treated steers prior to natural service had a higher pregnancy rate. In previous experiments in prepubertal heifers, there was an increase in the proportion of cyclic heifers related to the prolonged presence of bulls (Rekwot *et al*., 2000b; Oliveira *et al*., 2009). In contrast to sheep and goats, in which anestrous females were induced to ovulate 2-3 days after the introduction of the males (see reviews: Ungerfeld *et al*., 2004; Ungerfeld, 2007), heifers ovulated during a more prolonged interval. However, as little as 2 weeks of male exposure may be enough to induce changes in the mechanisms controlling heifers' reproductive physiology and increase pregnancy rates, at least in conditions as those of this experiment.

Although biostimulation may be provoked by bulls or by androgen-treated cows (Burns and Spitzer, 1992), no published information was found regarding the use of testosterone-treated steers. It has also been reported that there is not an LH response to untreated steers (Tauck, 2008).. Similarly to what has been previously reported in sheep (Fulkerson *et al*., 1981) and goats (Mellado and Hernandez, 1996), these findings suggests that, in cattle, biostimulating signals from males are also under androgen control. As bulls are not present on all farms, the use of testosterone-treated steers may be an easy and effective technique to include in practical management to stimulate non-cyclic cattle, at least in countries in which the application of androgens is allowed.

In previous experiments performed in similar conditions, it has been reported that during late autumn-early winter grazing heifers lost body weight and many animals became anestrus (Quintans *et al*., 2003; López-Mazz *et al*., 2008). Although there were no data available regarding previous cyclic activity in the experimental animals, probably High Weight and at least some of Medium Weight heifers were cycling during summer-autumn. However, probably many of them lapsed into anestrus when they began to lose weight. Perhaps exposure to testosterone-treated steers favored cyclic activity only in heifers with higher body weights. Since the transition into anestrus was not immediate, the differences could only be detected during the second pregnancy diagnosis. This means that it is important not only the amount of body weight loss, which was similar between Medium Weight and High Weight heifers, but also the body weight that heifers had when testosterone-treated steers were introduced. It may also be speculated that the presence of steers may have prevented the biostimulated High Weight heifers from returning to a non-cyclic condition for a short period, which coincided with the breeding period and resulted in pregnant heifers.

Another complementary explanation is that while High Weight heifers were in adequate physiological condition to respond to biostimulation, more Low and Medium Weight heifers may still have been acyclic, reducing their ability to respond. Small *et al.* (2000) observed positive responses to biostimulation in terms of timed AI pregnancy rates in older heifers, but detrimental effects of bull presence in prepubertal heifers. Due to the experimental design, we do not know if non-stimulated heifers were previously cycling, returned faster to a non-cyclic condition, or were still cycling but did not become pregnant.

Lower Weights heifers lost more weight than Medium Weight and High Weight heifers. This may also be related with hierarchical relationships, as in cattle social rank was related with body weight (Stricklin *et al.*, 1980), and that social tension was greater in low ranked individuals (Bowes and Wood-Gush, 1986). Therefore, as happens in other ruminants (Jørgensen *et al*., 2007), the lower body weight loss may be a consequence of better access to feed resources by high ranked individuals.

In conclusion, pregnancy rate was increased in high body weight heifers with pre-breeding exposure of the heifers to testosterone-treated steers during late autumn-early winter natural service..

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