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Antral follicular count is a tool that may allow the selection of more precocious Bradford heifers at weaning☆

R. Santa Cruz [a, *, 1], R.A. Cushman [b], C. Viñoles [a, 1]

[a] Programa Nacional de Investigación Producción Carne y Lana, INIA Tacuarembó, Ruta 5 km 386, 45000, Tacuarembó, Uruguay
[b] USDA, ARS, U.S. Meat Animal Research Center, Clay Center, NE, USA

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ABSTRACT

Although antral follicle count is a repeatable parameter across life that is positively associated with fertility, its use at weaning as a tool to discard less fertile heifers has not been extensively evaluated. The hypotheses of this work are: 1) maximum antral follicle count (MAFC) is repeatable between weaning and pre breeding evaluations, allowing selection of more fertile heifers at an early age, 2) heifers with high MAFC have growth and development parameters linked to an earlier puberty and pregnancy, 3) MAFC has a positive correlation with AMH concentrations, so that both could be used inter changeably. In this study, Hereford (n = 42 and n = 50) and Braford (n = 40 and n = 50) females were used in years 1 and 2; respectively, in a completely randomized experimental design. Heifers were examined for five to ten days at two different moments (post weaning and pre service), to determine MAFC. The concentrations of Anti müllerian hormone (AMH) were evaluated on the day of MAFC assessment. Growth and development parameters were evaluated post weaning and pre service. The repeatability of MAFC between post weaning and pre service evaluations was poor in three cases (Hereford Year 1 = 0.36 and 2 = 0.39 and Braford, Year 2 = 0.32) but it was high for Braford in Year 2 (0.72). The AMH repeatability between post weaning and pre service evaluations was high in one case (Braford Year 2 = 0.72) and moderate in the others (Year 1, Hereford = 0.50 and Braford = 0.52 and Year 2, Hereford = 0.50). In Year 2, Braford heifers with greater MAFC attained puberty at an earlier age ($r^2 = 0.129$; $P = 0.0196$). Also, diminished MAFC corresponded with decreased growth and development, thus less Braford heifers with low MAFC were inseminated (2/16), compared to those with medium (12/17) and high MAFC (7/17; $P < 0.01$). Moreover, Braford heifers with low AFC had less progesterone in the cycle post insemination but pregnancy rate was not affected. In Braford heifers in Year 2, there was a high correlation between MAFC and AMH concentrations (0.85 $P < 0.001$). The results of these experiments indicate that post weaning MAFC and AMH concentrations may be applied to select those Braford heifers that attain puberty at an early age, but these tools are not useful in Hereford heifers.

1. Introduction

Age at puberty and first calving affect the reproductive performance of breeding cows and sheep during the remainder of their productive life [1,2]. Heifers that conceive in the first 21 days of the breeding season, stay in the herd longer and wean heavier

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* Corresponding author.
E-mail address: rodrigo.santacruz@poloagroforestal.edu.uy (R. Santa Cruz).
[1] Present address: Polo Agroforestal, Estación Experimental Bernardo Rosengurtt, Facultad de Agronomía, Universidad de la República, Ruta 26 km 408, 37000, Cerro Largo, Uruguay.

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calves during their productive life [3]. Increased age, live weight and body condition improve the probability of attaining puberty and conceiving early [4]. Other parameters of growth and development such as hip height and body composition, have been used successfully because of their high association with live weight [5]. However, puberty is a complex process involving a series of events. Nutrition, age and genetics influence puberty, mainly as regulators of the maturation of the hypothalamic – pituitary - ovarian axis that must occur for the initiation of normal oestrous cycles [6]. The complexity of the onset of puberty explains that selecting replacement heifers based on body weight may not be enough. More recently, antral follicle count (AFC) and Anti müllerian (AMH) hormone concentration have been reported to be phenotype markers of fertility in heifers at mating [7]. Heifers that have greater AFC also have greater concentrations of AMH and progesterone and they become pregnant earlier than heifers with lower AFC [8–10]. Similarly as occurs in ewe lambs where an earlier attainment of puberty have been associated with increased circulating concentrations of AMH [11]. Greater concentrations of progesterone have a positive impact on embryo development [12], thus contributing to an earlier conception. Moreover, an earlier attainment of puberty allows the occurrence of more oestrous cycles before breeding and a greater fertility at first service [13]. Although, AFC is a repeatable parameter across the life of a cow [7], its use at weaning as a tool to discard less fertile heifers has not been extensively evaluated [14].

The hypotheses of this work are: 1) MAFC is repeatable between weaning and pre breeding evaluations, allowing selection of more fertile heifer at an early age; 2) Heifers with high MAFC have growth and development parameters related to an earlier puberty that could be associated with an earlier pregnancy; 3) AFC has a high correlation with AMH concentrations, so that both parameters would be used inter changeably.

The objectives were to evaluate MAFC repeatability between weaning and pre breeding, its correlation with AMH, parameters of growth and development, age at puberty, progesterone concentrations at the insemination cycle and pregnancy at 13–15 months in Hereford and Braford heifers.

2. Materials and methods

2.1. Location and animals

Animal experimentation was approved by the Ethics Committee for the Use of Animals (CEUA; file number 2013.13). Hereford and Braford females were used in different years (2013 2014: Year 1 and 2015 2016: Year 2), in a completely randomized experimental design. The evaluation period started with the selection of calves at weaning and ended with pregnancy diagnosis, 30 days after bull removal from the herd. The number of animals used in Year 1 was 42 Hereford and 40 Braford and in Year 2, 50 animals of each breed. The calves were daughters of at least seven different bulls per breed per year. Hereford heifers were managed in the Experimental Unit "Glencoe" and Braford heifers in the Experimental Unit "La Magnolia" both from INIA Tacuarembó.

2.2. Live weight, age at the beginning of evaluations and feeding

In Year 1, the animals were selected at weaning in April, with 189 ± 2.2 kg and 189 ± 2.5 kg of live weight and 198 ± 3 days and 182 ± 3 days of age, for Hereford and Braford calves, respectively. From weaning until middle September calves grazed on *Campos grasslands* [15] with a forage allowance of 10 kg of dry matter per kg of live weight (kg DM/kg LW) [16] and offered supplement *ad libitum* in an automatic self feeder. Thereafter, heifers grazed 4 h per day on improved pastures with a forage allowance of 7 kg DM/kg LW and the supplement was adjusted to 1% of the LW. The supplement provided 14% of crude protein (PC) and 3 Mcal of metabolizable energy (ME)/kg DM. The average weight gain between weaning and mating was 0.700 ± 0.1 kg in Hereford and 0.681 ± 0.1 kg in Braford heifers, attaining mating (at 13–15 months) with an average weight of 356 ± 3.5 kg and 322 ± 3.9 kg, respectively. During the breeding season, all heifers grazed *Campos grasslands* with a forage allowance of 7 kg DM/kg LW.

In Year 2, calves were weaned in March (Hereford) and April (Braford), with 164 ± 2.5 kg and 168 ± 3.6 kg of live weight and 165 ± 2 days and 174 ± 3 days of age; respectively. Between weaning and mating, they grazed annual winter pastures (oats and ryegrass); with an average forage allowance of 1.1 kg DM/kg LW in both systems. All females were supplemented with corn DDGS (Dried Distillers Grains with Solubles) (22.6% digestible protein and 2.8 Mcal/kg DM ME), that was adjusted according to the average LW. Supplementation was offered at 0.7–1.2% of the LW. The average daily weight gain from weaning to service was 0.855 ± 0.1 kg in Hereford and 0.669 ± 0.1 kg in Braford, reaching the beginning of mating (at 13–15 months) with an average LW of 378 ± 3.8 kg and 312 ± 4.6 kg in Hereford and Braford, respectively. During the breeding season, all heifers grazed *Campos grasslands* and sorghum in Hereford and setaria in Braford, at a minimum allowance of 7 kg MS/kg LW and 1 kg MS/kg LW in natural and annual summer pastures, respectively.

2.3. Breeding

In Year 1, all heifers were synchronized using the Ovsynch protocol [17] plus a progesterone device. They were inseminated artificially at fixed time 52–60 h after Cloprostenol, using frozen semen from two bulls per breed with more than 60% motile sperm after thawing. Ten days after artificial insemination, a single sound bull was introduced into each herd for an additional period of 50 days.

In Year 2, heifers were synchronized with synthetic prostaglandin F2 alpha (Cloprostenol D, 150 mg i.m., Dalmaprost® Laboratory Fatro, Uruguay) and the insemination was performed following heat detection for 5 days, using frozen semen from two different bulls per breed with the same conditions as in Year 1. After the end of insemination, a sound bull was introduced in each herd for an additional period of 50 days. Inseminations were performed by two trained technicians in Hereford and Braford in both years.

2.4. Animal measurements

2.4.1. Antral follicle count (AFC)

The maximum antral follicle count (MAFC) was evaluated in both ovaries, recording diameter, number, and position of all follicles ≥ 2 mm on ovarian maps. The evaluation was carried out at two different ages (post weaning = 246 days of age on average and pre service = 385 days of age on average) using a real time, B mode scanner with a transrectal probe of 7.5 MHz (Aloka Co., Ltd., Tokio, Japón). The same operator performed all measurements in each year, but the technician was different between years of the study. In Year 1, the evaluations (post weaning and pre service) were carried out 6 times over 12 days, while in Year 2, they were done during five consecutive days.

In the post weaning evaluation, a rigid probe was used and manipulated externally applying the methodology described by Viñoles et al. [18], in which the ovaries were visualized using the bladder and the uterus as references structures and rotating the probe in a clockwise direction to locate the left ovary and in a counter clockwise direction to locate the right ovary.

For the pre service evaluation, heifers were synchronized with