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Short Communication

# Follicular dynamics of induced estrus of females from the Pantaneira breed

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## ABSTRACT

The aim of this study was to evaluate the performance of heifers from the Pantaneira breed in FTAI programs and characterize follicular dynamics after induced estrus synchronization programs. The pregnancy rate observed in an FTAI program (36,4%) followed by the AI of estrus return was 45.5%. The study of follicular dynamics revealed that females from this breed, on average, show ovulation 77 hours after the withdrawal of the progesterone device, being therefore later than in other bovine breeds. The results suggest that the late ovulation of the females submitted to estrus synchronization programs may negatively influence their pregnancy rates.

#### **INTRODUCTION**

Pantaneira cattle are at risk of extinction, with less than 1000 animals in the effective herd. The loss of a single breed irreversibly prevents access to their genes in breeding programs and future industrial crosses (EGITO; MARINANTE; ALBUQUERQUE, 2002).

The Pantaneira breed has high ability to survive and reproduce in rudimentary environments (SANTOS et al., 2005). There are records that such animals are resistant to trypanosomiasis, myiasis, worms and ticks, being able to survive in extreme ecological conditions, in a region where climatic conditions range from floods to drought throughout the year (DANI; OLIVEIRA, 2013).

Reproductive biotechnologies are essential tools for effective breed multiplication and conservation

It is known that animals from different bovine breeds have different physiological characteristics, including the duration of estrus which is 18 hours for *Bos taurus* (LOPEZ; SATTER; WILTBANK, 2004) and 12 hours for *Bos indicus* (PIRES et al., 2003). Such differences directly influence biotechnologies such as FTAI and ETFT (embryo transfer at fixed time), since the time required for the animals to respond to hormonal protocols will

programs. However, preliminary results of the use of Fixed Time Artificial Insemination (FTAI) and Multiple Ovulation and Embryo Transfer (personal communication) were below expectations when compared to other bovine breeds. Based on this information, it is believed that differences in the reproductive physiology of these animals may justify the lower efficiency of such biotechniques.

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also be different (FERREIRA et al., 2012; SALES et al., 2015; SOUZA, 2012).

According to our research this was the first scientific study that characterized the ovarian dynamics and progesterone concentrations in bovine females from the Pantaneira breed submitted to hormonal treatment. So far, there were only data from books whose reports had been obtained from the pantaneiro population (MAZZA et al., 1994).

The aim of this study was to report the reproductive performance of the Pantaneira cattle in the FTAI program and to characterize the follicular dynamics induced by estrus synchronization programs.

#### MATERIAL AND METHODS

Eleven heifers from the Pantaneira breed, from a contemporary 24-month-old, cyclical, single and with a mean body score of 3.5 batch, were submitted to a standard FTAI protocol for bovines, where: on day zero (D0) the introduction of the progesterone implant (CIDR®) with 1.9 mg of progesterone and the intramuscular application of 2 mg of estradiol benzoate (BE) was carried out. On day eight (D8) the CIDR® was withdrawn and 400UI of Eauine Chorionic Gonadotrophin (eCG; Novormon®, MSD Animal Health) and 0.15 mg of d-cloprostenol were applied. On day nine (D9) 1 mg of EB was administered intramuscularly and 48-54 hours after the withdrawal of the progesterone implant, the FTAI was done. The inseminations of return were initiated 18 days after the procedure. In order for this to be carried out, estrus was observed for 7 days, and after 12 hours of their detection, the animals were inseminated again. The inseminations were performed with doses of semen coming from a single bull belonging to one of the conservation groups of the breed. The semen was evaluated according to CBRA standards (1998).

In order to evaluate the follicular dynamics, fifteen cyclic and with a body score of 3.5 (scale 1 - 5) Pantaneira heifers were used, they underwent estrus hormonal induction twice at a 28-day interval; thus obtaining 30 experimental units. The following synchronization protocol was used: In the D0 the ultrasound evaluation of the ovaries was made, followed by the introduction of CIDR® and the application of 2 mg EB and 25 mg of dinoprost tromethamine (Lutalyse®, Zoetis) bv intramuscular injection. On day 7 (D7) CIDR® was withdrawn, then we began the ultrasonographic evaluation and the blood collection every 24 hours. From day 9 (D9) the evaluations and collections of blood began to be performed every 12 hours until the moment of ovulation or until the D12, the last evaluation day.

Each ultrasonographic evaluation was performed counting the follicles present and measuring the diameter of the largest follicle present in each ovary. The measurements were made with the aid of the resources from the ultrasound equipment itself. The presence of corpus luteum was also recorded. The following parameters were evaluated: Daily follicular growth rate, ovulatory follicle diameter, the interval between the withdrawal of the progesterone device and ovulation and serum progesterone levels (P4). The daily growth rate (mm / day) was calculated by the formula: (Mfol mfol)/Nd, being Mfol: Diameter of the largest follicle found; mfol, Diameter of the smallest follicle found; Nd: Number of observation days (COUTINHO et al., 2007; URIBE-VELÁSQUEZ; SOUZA; OSORIO, 2011). The evaluation of serum P4 concentrations was done by Radioimmunoassay, using the P4 Kit (TKP Genese®).

All the procedures performed in order for this study to be carried out were approved by the Animal Ethics Committee under the protocol 015/2014.

The results are expressed as means followed by standard deviation.

#### **RESULTS AND DISCUSSION**

The pregnancy rate obtained with the FTAI management was 36.4% (4/11). At the end of the program, 45.5% (5/11) of pregnancy was obtained after the inseminations of the return estrus.

The FTAI protocol used was the standard for beef cattle and for which an average of 50% (43,2% a 67%) pregnancy rate is expected (HILL et al., 2014; SALES et al., 2015). In the present study, the pregnancy rate of FTAI and return insemination (Table 1) was below desired, as observed for other cattle breed. In general, pregnancy rates above 70% are expected when FTAI and AI are associated (CAMPOS et al., 2013; ESCALANTE et al., 2013). Similar results to the ones of this study have been reported by researchers and collaborators of other conservation groups of the breed (unpublished data).

We attempted to explain the low conception rates obtained in FTAI programs by studying the follicular dynamics of two induced estrus from Pantaneira females. The P4 profile over the experimental period is shown in figure 1, and it clearly indicates that when concentrations of P4 reached concentrations less than or equals to 1 ng / ml, the dominant follicle grew to its maximum size (9.95  $\pm$  0.34), considered the preovulation size.





Table 1 shows the parameters of the follicular dynamics (Daily follicular growth rate, ovulatory follicle diameter and the interval between the withdrawal of the progesterone device and ovulation) of an induced follicular wave in Pantaneira heifers.

Table 1. Parameters of follicular dynamics of an induced follicular growth wave in Pantaneira heifers.

Parameters	Results
Ovulation after withdrawal of P4 (hours)	77±13,2
Daily growth rate (mm / day)	2,19 ±0,25
Preovulatory follicle diameter (mm)	9,95±0,34

The result of the interval between the withdrawal of the progesterone implant and the ovulation was the most expressive of this study (77 hours), because it is longer than those described in the literature for other breed: 73 hours for the Nelore breed (SALES et al., 2015) and 72.2 for Holstein (FERREIRA et al., 2012). Souza (2012) reported that, under the same environmental conditions and management of this study, Girolando cows ovulated 63 hours after the withdrawal of P4.

Considering that the greatest fertility period of bovine oocytes has an average of six hours after ovulation (RIVIEIRA, 2009) and that the spermatozoids require approximately 8 hours for their capacitation (RIVIEIRA, 2009) having a mean viability of 24 hours and their maximum fertility being in the first hours, an average delay of 5 hours in ovulation, as observed in this breed, can significantly interfere the pregnancy rates. An ovulation delay of 6 to 12 hours in regularly cyclical females, is considered a pathology because oocytes from these ovulations are considered incapable of being fertilized (FISCHER et al., 2002). In the case of the Pantaneira breed, it is believed that the upper interval between the withdrawal of the progesterone implant and the ovulation is not a pathology but a characteristic of the breed in response to hormonal treatments. It is noteworthy that the herd in this study were ex sito, and there are reports that females of this breed were submitted to natural mating, the birth rate (above 70%) was satisfactory for animals raised in Pantanal (MAZZA et al., 1989).

It is notable, on the other hand, that the standard deviation of this variable demonstrates great individual variability (60 and 108 hours), which may be related to the fact that the above mentioned breed was not subjected to genetic selection over the years. Despite this, the result cannot be disregarded since the difficulty to obtain satisfactory results with the artificial insemination has been shared between the nuclei of conservation of the breed.

#### CONCLUSION

The evaluation of the follicular dynamics of an estrus hormonal induced from Pantaneira breed demonstrated that the interval between the withdrawal of the progesterone device and the ovulation (77 hours) is superior to that of other breeds, which may have influenced negatively the pregnancy rates of females submitted to the FTAI programs.

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