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The use of PGF2α as ovulatory stimulus for timed artificial insemination in cattle

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ABSTRACT

The objective of this study was to evaluate the effect of a PGF2α-analogue (PGF) on ovulation and pregnancy rates after timed artificial insemination (TAI) in cattle. In experiment 1, crossbred dual-purpose heifers, in a crossover design (3 × 3), were given an intravaginal progesterone-releasing insert (controlled internal drug release [CIDR]) plus 1 mg estradiol benzoate (EB) intramuscularly (im) and 250 μg of a PGF-analogue im on Day 0. The CIDR inserts were removed 5 days after follicular wave emergence, and the heifers were randomly divided into three treatment groups to receive the following treatments: (1) 1 mg of EB im (EB group, n = 13); (2) 500 μg of PGF im (PG group, n = 13); or (3) saline (control group, n = 13), 24 hours after CIDR removal. Ovulation occurred earlier in EB (69.81 ± 3.23 hours) and PG groups (73.09 ± 3.23 hours) compared with control (83.07 ± 4.6 hours; P = 0.01) after CIDR removal. In experiment 2, pubertal beef heifers (n = 444), 12 to 14 months of age were used. On Day 0, the heifers were given a CIDR insert plus 2 mg EB im. On Day 9, the CIDR was removed and the heifers were given 500 μg of PGF im. Heifers were randomly assigned into one of three treatment groups: (1) 1 mg of EB (EB group; n = 145); (2) 500 μg of PGF (PG group; n = 149), both 24 hours after CIDR removal; or (3) 600 μg of estradiol cypionate (ECP group; n = 150) at CIDR removal. Timed artificial insemination occurred 48 hours after CIDR removal in the ECP group and 54 hours in the PG and EB groups. The percentage of heifers ovulating was higher in the PG group compared with the other groups (P = 0.08). However, the pregnancy rates did not differ among groups (47.6%, 45%, and 46.6%, for EB, PG, and ECP, respectively; P = 0.9). In experiment 3, 224 lactating beef cows, 40 to 50 days postpartum with 2.5 to 3.5 of body condition score were treated similarly as described in experiment 2, except for the ECP group, which was excluded. The treatments were as follows: 1 mg EB (EB group; n = 117) or 500 μg PGF (PG group; n = 107), 24 hours after CIDR removal. The calves were temporarily separated from their dams from Days 9 to 11. No difference was detected on the pregnancy rate between the EB and PG groups (58.1% vs. 47.6%, respectively; P = 0.11). Taken together, the combined results suggested that PGF2α could be successfully used to induce and synchronize ovulation in cattle undergoing TAI, with similar pregnancy rates when compared with other ovulatory stimuli (ECP and EB).

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1. Introduction

Recently, considerable research efforts have focused on developing technologies to induce synchronized ovulation for timed artificial insemination (TAI) in beef [1–3] and dairy cattle [4–6]. Treatments developed to control both luteal and follicular function have permitted the exciting possibility of controlling and synchronizing the time of ovulation and artificial insemination without the need for estrus detection [7]. Consistent TAI protocols have been extensively used by South American beef and dairy industries, and the usual protocol basically consists of an intravaginal progesterone insert, estradiol benzoate (EB) to induce follicular wave emergence, PGF2α analogues (PGFs) to induce luteolysis, and an ovulatory stimulus of either an ester of estradiol (benzoate or cypionate) or a GnRH analogue. This standard protocol and its modifications provided satisfactory fertility in commercial Brazilian herds, independent of breed (*Bos indicus*, *Bos taurus*, or crossbreds).

In cattle, hormonal-based estrus synchronization protocols use PGF to regress a CL that might be functional by the end of the protocol. It is known that prostaglandin is a biologically potent substance with multiple applications in the control of reproduction [8]. In cattle, the most common uses are because of its luteolytic properties, which are useful for estrous synchronization, regression of persistent CL, and induction of abortion or parturition [9]. Although PGF and its analogues are primarily used as luteolysins, they have also been reported to have an effect on ovulation, embryo implantation, pregnancy maintenance, and postpartum physiology [8]. Moreover, PGF, which is primarily used for synchronization of estrus alone or in association with progestins, estrogens, and GnRH, has also been reported to act as an ovulatory stimulus in prepubertal heifers [10,11]. Prostaglandin F2α analogue increases pituitary responsiveness to GnRH, thereby enhancing the release of LH [12] in a process that results in ovulation. In our previous study, PGF treatment induced first ovulation in prepubertal heifers, independent of whether heifers were pretreated with exogenous progesterone [11]; however, its effectiveness as an ovulatory inducer in TAI programs has not yet been studied.

On the basis of these considerations, the objectives of this study were: 1) to evaluate whether exogenous administration of PGF induces synchronized ovulations; and 2) to study the efficacy of PGF as an ovulatory stimulus for TAI in cattle. We tested the hypotheses that exogenous PGF 1) induces ovulation in a synchronized manner, and 2) when used as an ovulatory stimulus, provides an efficiency similar to esters of estradiol, which are usually used in TAI programs in cattle.

2. Materials and methods

The Committee for Ethics in Animal Experimentation from the Universidade Federal de Santa Maria approved all of the procedures performed in these experiments.

2.1. Experiment 1

The experimental design is shown (Fig. 1A). This study was performed at the experimental research farm of Embrapa Rondônia (Brazilian Agricultural Research Corporation, Rondônia, Brazil; 08°48′12″ S, 63°50′56″ W). Thirteen dual-purpose crossbred pubertal heifers (Gyr × Holstein), 14 to 18 months and 290 to 360 kg body weight were used. The heifers were kept in an outdoor grazing system (*Brachiaria brizantha* pasture) with ad libitum access to mineral salt and water.

All heifers were treated with an intravaginal progesterone-releasing insert (1.9 g, controlled internal drug release [CIDR], Pfizer Animal Health, São Paulo, Brazil) plus 1 mg estradiol benzoate (EB; Bioestrogen, Biogénesis-Bagó, Curitiba, Brazil) intramuscularly (im), and 250 μg im of a PGF analogue, D-cloprostenol (Croniben, Biogénesis-Bagó, Curitiba, Brazil) on Day 0. The CIDR inserts were removed 5 days after follicular wave emergence (i.e., approximately 9 days after CIDR insertion) and all heifers received an additional 250 μg im dose of D-cloprostenol at this time point. Twenty-four hours after CIDR removal, the heifers were randomly divided into three treatment groups: 1) 1 mg EB (EB group, n = 13) im; 2) 500 μg PGF (PG group, n = 13) im; and 3) saline (control group, n = 13) im. The experiment was performed in a crossover design (3 × 3) in which all heifers participated in all treatments groups (e.g., 13 heifers per group). The interval between replicates was 1 month.

2.2. Experiment 2

This experiment was performed at a commercial farm located in the southern region of Brazil (32°20′46″ S and 52°32′39″ W). Pubertal crossbred Aberdeen Angus heifers (n = 444), 12 to 14 months and 313 ± 25 kg body weight were enrolled in this experiment.

The experimental design is shown (Fig. 1B). On Day 0, the heifers were treated with CIDR and 2 mg EB (Gonadiol; MSD Saúde Animal, São Paulo, Brazil) im. On Day 9, the CIDRs were removed and the heifers received a 500 μg im injection of a PGF analogue (sodium cloprostenol, Sincrosin; Vallée, Montes Claros, Minas Gerais, Brazil); and were then randomly assigned into one of the following three treatments: (1) 1 mg EB (EB group, n = 145); (2) 500 μg sodium cloprostenol (PG group, n = 149) im; both groups treated at 24 hours after CIDR removal (Day 10); and (3) 600 μg estradiol cypionate (ECP group, n = 150; ECP; Hertape Calier Saúde Animal, Juatuba, Minas Gerais, Brazil) im at the same moment of CIDR removal (Day 9; Fig. 1B). Timed artificial inseminations were performed 48 ± 2 hours after CIDR removal in the ECP group and 54 ± 2 hours after CIDR removal in PG and EB groups. Two inseminators performed all inseminations and semen straws from four bulls of proven fertility were used for TAI.

2.3. Experiment 3

Experiment 3 was performed on the same commercial farm as experiment 2 (Fig. 1C). Multiparous suckling crossbred Aberdeen Angus cows, 40 to 50 days postpartum with a body condition score (BCS) ranging from 2.5 to 3.5 (from 1 = emaciated to 5 = obese) were enrolled in this experiment. Of the 341 cows initially available for the experiment, 224 (65.7%) cows were considered suitable, on

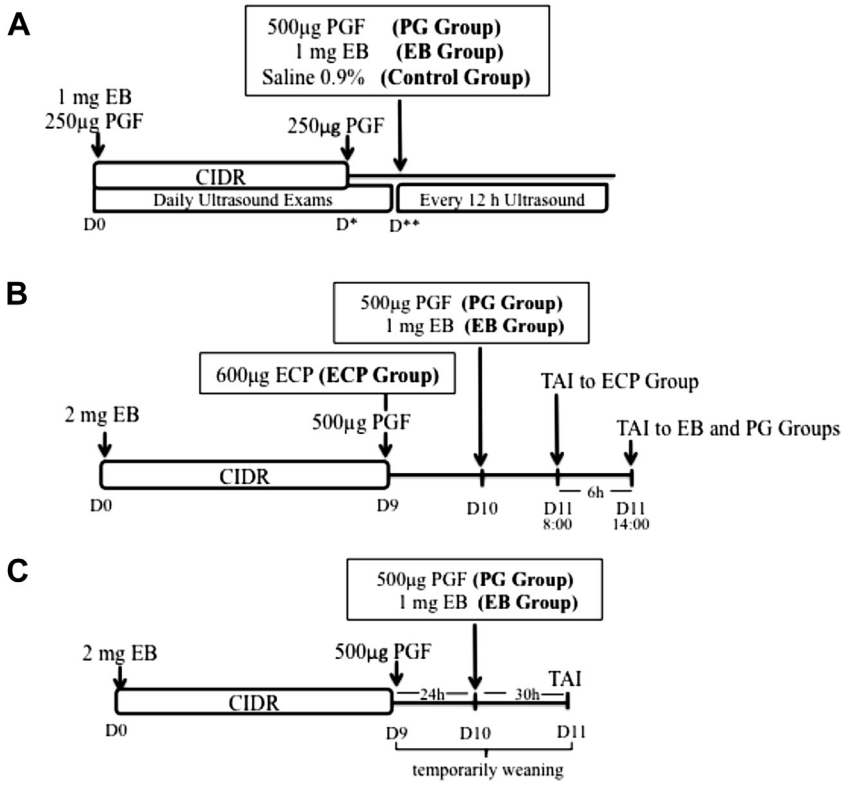

Fig. 1. Experimental design for (A) heifers in experiment 1, (B) heifers in experiment 2, and (C) suckled cows in experiment 3. CIDR, controlled internal drug release; D*, Day 5 of the follicular wave; D**, Day 6 of the follicular wave; EB, estradiol benzoate; ECP, estradiol cypionate; PGF, prostaglandin; TAI, timed artificial insemination.

the basis of an adequate BCS and the presence of a CL as detected using ultrasound examinations. All animals were maintained on a pasture and with ad libitum access to mineral salt and water.

The cows were treated and randomly allocated into treatment groups, similar to the heifers in experiment 2, except for the ECP group, which was absent this time: (1) EB group (n = 117) and (2) PG group (n = 107). All calves were temporarily separated from their dams from CIDR removal (Day 9) to TAI (Day 11). During this 54 hour-interval, the calves were kept in pens, which were out of the sight of their dams, and had ad libitum access to water. The TAI was performed 54 ± 2 hours after CIDR removal (Fig. 1C). Two inseminators performed all inseminations and semen straws from two bulls of proven fertility were used for TAI.

2.4. Ultrasonographic examinations

Before the initiation of the experiments, the cattle were examined twice using transrectal ultrasonography, 7 to 11 days apart, to confirm the presence of the CL and to evaluate the degree of uterine tonus, to ensure that all heifers were pubertal and the cows had resumed postpartum ovarian activity. Only cattle in which a CL was detected in at least one of the examinations were included in this study.

In experiment 1, the heifers were scanned daily using B-mode transrectal ultrasonography (Weld, equipped with a 5 MHZ linear-array transducer; Beijing, China) to monitor ovarian follicular development from Day 0 to Day 9, and subsequently, twice daily until ovulation or in the absence of ovulation, up to 5 days after CIDR removal. At each scanning session, a sketch of each ovary was generated and the diameter and location of follicles greater than or equal to 3 mm in diameter were recorded according to methods described elsewhere [13]. Ovulation was defined as the disappearance (from one scanning session to the next) of a previously identified follicle greater than or equal to 8 mm in diameter [14]. The day of wave emergence was defined retrospectively as the day when the dominant follicle was first detected at a diameter of 4 to 5 mm [13]. In heifers that ovulated, ultrasonographic examinations were performed 7 days later to determine the CL diameter.

In experiments 2 and 3, transrectal ultrasonography (Honda HS-1500V, equipped with a 7.5 MHz linear-array transducer; Toyohashi, Aichi, Japan) was performed in a subset of 204 heifers (experiment 2) and 128 cows (experiment 3) on Days 9 and 11 to determine the diameter of the largest follicle, and also on Day 18 to determine the presence of the CL. Ultrasound examination was also performed on Day 38 in all cattle to detect pregnancy.

2.5. Statistical analysis

All statistical analyses were performed using the SAS 9.0 software (2006; SAS Institute Inc., Cary, NC, USA). In experiment 1, single-point outcome variables (e.g., time of

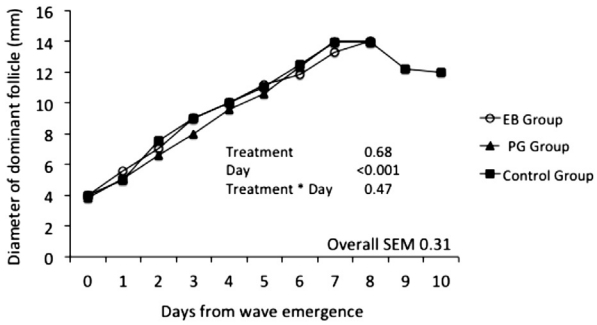

Fig. 2. Comparison of the pattern of dominant follicle growth in heifers treated with EB group, PG group, and nontreated controls. Exogenous EB and PG were given on Day 6 of the follicular wave. EB, estradiol benzoate; PG, prostaglandin F2α.

ovulation, diameter of dominant follicle on Days 9 and 11, and diameter of CL on Day 17) were analyzed using one-way analysis of variance and the means were compared among groups using Tukey's post hoc test. Growth of the dominant follicle from emergence to ovulation was compared among groups using the MIXED procedure for repeated measures to evaluate the main effects of treatment and time (days) and their interactions [15]. In experiments 2 and 3, in the initial analyses of the single-point outcome variables (i.e., maximum diameter of the ovulatory follicle at CIDR removal and at TAI, interval to ovulation, growth rate of the ovulatory follicle, diameter of the CL), the statistical model included the effect of the bull, inseminator, body weight, age, BCS, lactation number (cows), and days postpartum (cows). The variables bull, inseminator, body weight, age, lactation number, days postpartum, and BCS had no significant effect on the model and were therefore excluded from the final statistical model. Thus, single-point outcome variables were analyzed using one-way analysis of variance and means were compared among groups using Tukey's post hoc test. Proportions with dichotomous outcomes (yes or no), such as ovulation and pregnancy rates were analyzed using the Chi-square test. Differences among groups were considered statistically significant when the probability (P-value) was less than or equal to 0.05.

3. Results

3.1. Experiment 1

Overall, progesterone inserts were removed 8.7 ± 0.3 days after insertion (range, Day 7–Day 10). The ovarian follicular growth profile is shown in Figure 2. All heifers ovulated within 5 days after CIDR removal. The interval from CIDR removal to ovulation did not differ between the EB and PG groups (69.82 ± 3.23 hours vs. 73.1 ± 3.23 hours, respectively; $P > 0.05$). However, heifers in the control group did ovulate later compared with the other two groups (83.07 ± 4.6 hours; $P = 0.01$). Only one heifer ovulated at 84 hours after CIDR removal in the PG group. The distribution and percentage of heifers ovulating after CIDR removal is shown in Figure 3.

The diameter of the ovulatory follicle ($P = 0.36$) and the CL ($P = 0.46$) did not differ among groups. Considering all groups, the heifers ovulated a follicle with 14.8 ± 1.9 mm in diameter and formed a Day-7 CL of 22.9 ± 2.9 mm in diameter.

3.2. Experiment 2

The ovarian responses, ovulation and pregnancy rates in heifers are summarized in Table 1. The percentage of heifers ovulating tended to be higher in the PG group compared with the EB and ECP groups ($P = 0.08$). There was no difference in the pregnancy rates among groups ($P = 0.9$). The diameter of the dominant follicle on Day 9 was also not different among groups ($P = 0.18$). Nevertheless, heifers treated with PGF had the largest dominant follicle at TAI; conversely, heifers treated with ECP had the smallest dominant follicles at TAI ($P < 0.01$).

3.3. Experiment 3

The ovarian responses, ovulation and pregnancy rates in cows are summarized in Table 1. There was no difference in the diameter of the dominant follicle at TAI, ovulation and pregnancy rates among groups ($P > 0.05$). However, dominant follicle on Day 9 tended to be greater in cows treated with EB compared with cows treated with PGF ($P = 0.07$).

4. Discussion

The results of this study supported the initial hypothesis that treatment with PGF would induce ovulations in a synchronized manner. Moreover, we demonstrated that exogenous PGF could be successfully used as an ovulatory stimulus for TAI in heifers and cows. Although previous studies have demonstrated the ability of PGF to induce ovulation in prepubertal heifers [10,11], to the best of our knowledge, this is the first report to determine that PGF *per se* can be used as an ovulatory stimulus for TAI. Most importantly, this study demonstrated that if used as an

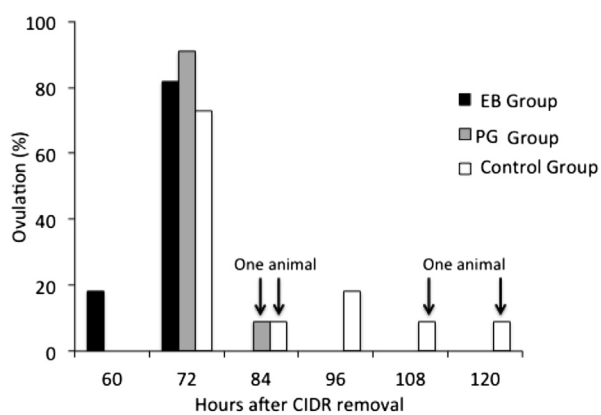

Fig. 3. Distribution and percentage of heifers ovulating after CIDR removal in the EB group, PG group, and nontreated controls. EB, estradiol benzoate; PG, prostaglandin F2α.

Table 1
Percentages of heifers (experiment 2) and cows (experiment 3) that ovulated and became pregnant, and the mean (±standard error of the mean) diameter of the dominant follicle on Days 9 and 11 in cattle treated with EB group, PG group, or ECP group as ovulatory stimuli.

Characteristics	Heifers (experiment 2)				Cows (experiment 3)		
	PG	EB	ECP	P-value	PG	EB	P-value
Ovulation rate	95.6% (66/69)	85.1% (57/67)	85.3% (58/68)	0.08	85.4% (53/62)	92.4% (61/66)	0.20
Pregnancy rate	45% (67/149)	47.6% (69/145)	46.6% (70/150)	0.9	47.6% (51/107)	58.1% (68/117)	0.11
Dominant follicle diameter (mm) on Day 9	10.2 ± 0.2	12.1 ± 1.6	9.9 ± 0.2	0.18	10.7 ± 0.2	11.2 ± 0.23	0.07
Dominant follicle diameter (mm) at TAI	13.1 ± 0.3[a]	12.2 ± 0.2[b]	11.5 ± 0.2[c]	<0.001	13.6 ± 0.33	13.2 ± 0.39	0.41

Abbreviations: EB, estradiol benzoate; ECP, estradiol cypionate; PG, prostaglandin F2α; TAI, timed artificial insemination.
[abc] Values without a common superscript differed between groups.

ovulation inducer, PGF provides pregnancy rates in heifers and suckled cows similar to that achieved using estradiol-based ovulation inducers.

Although PGF tended to increase the ovulation rate in heifers, no differences in pregnancy rates were detected in cattle when PGF, EB, or ECP were used in the TAI protocol. Prostaglandin F2α–induced synchronized ovulations in cattle, thus providing satisfactory fertility in heifers and cows subjected to TAI (given an expected pregnancy rate of ~50%). This was a remarkable achievement, particularly because the current results were similar to that reported by previous studies using treatments with EB and ECP to induce ovulation in heifers [16–18] and postpartum cows [18–21], which achieved pregnancy rates ranging from 34% to 70%. One could speculate that the results of the present study were potentially associated to an indirect effect of PGF (by inducing luteolysis, progesterone decrease, and subsequent LH surge) rather than a direct effect of PGF on the follicle to induce ovulation. Nevertheless, it was clearly demonstrated in experiment 1 that PGF-induced ovulations occurred in a pattern similar to that of EB-induced and the cattle ovulated approximately 10 hours earlier compared with controls. Martinez, et al. [22] reported that when an ovulatory inducer was not used in association with the CIDR-based protocol, ovulation occurs up to 108 hours after CIDR removal, an interval that is most likely detrimental to the pregnancy rate. For optimal fertility to TAI, ovulations must occur synchronously (within a few hours) in most of the treated cattle, with the least number of failures. In experiment 1, the interval from insert removal to ovulation (median = 72 hours) was highly synchronized in heifers treated with either PGF or EB 24 hours after CIDR removal. Other studies have demonstrated a similar interval from CIDR removal to ovulation using EB (~74 hours) [14,23], and LH (~74 hours) [24] as ovulatory stimuli in heifers. In contrast, ECP apparently causes a delay in ovulation (~81 hours) [16] when compared with EB and PGF in heifers. In another study, in which heifers were not exogenously induced to ovulate, ovulations occurred on average at 82 hours after insert removal [22], with an interval similar to that of the control group in the present study. With regard to fertility, although there were numerically less ovulations in both EB and ECP groups in experiment 2 in this study, the pregnancy rates were not negatively affected by this finding.

In the present study, we included a third treatment group in experiment 2 (ECP), which aimed to compare the most commonly adopted TAI protocol used by the South American beef industry [2] with EB and PGF. Pregnancy rates were similar among all ovulatory inducers in heifers. In contrast, in experiment 3, PGF-treated suckled cows had, albeit not statistically significant, less pregnancies (~10%) than EB-treated cows. Although PGF-treated cows presented satisfactory pregnancy rates (47.6%) considering their stage of lactation (50 days postpartum), EB-treated females presented a pregnancy rate above these expectations (~60%). Although the present experiment was not designed to address the potential variations in the moment of ovulation between the PGF- and EB-treated suckled cows, previous studies have reported that suckled cows treated with EB ovulated, on average, 70 hours after progesterone insert removal [20,21]. Our results on the time of ovulation in EB-treated heifers (experiment 1) were consistent with those of previous reports. Thus, further studies are required to determine the moment of ovulation in suckling cows treated with PGF. If ovulation is delayed in PGF-treated lactating cows, but still occurs in a synchronized fashion, an appropriated TAI schedule may further be adjusted to a PGF-based protocol, and consequently, pregnancy rates may increase and reach percentages that are similar to that of EB-based protocols.

In the present study, there was a difference in the time of PGF injections, during progesterone treatment, among experiments (1 vs. 2 and 3). Two injections of PGF, on Day 0 and at CIDR removal, were used in experiment 1 to rapidly decrease progesterone concentrations, which potentially improved the synchronization of ovulation. However, experiments 2 and 3 were performed at commercial beef operations and, because of the logistic of these farms, the standard TAI protocol commonly used was adopted in that commercial herd. For this reason, we injected a full dose (500 μg) of PGF on Day 9 instead of dividing it into two injections. The requirement to follow the farm's standard management protocols was also the reason for the use of temporarily calf removal in postpartum cows (experiment 3). Regardless of these small differences between experiments, the groups (PGF vs. EB vs. ECP) were compared within each experiment, in which all cattle received the same treatment. Thus, we believe that the small differences between experimental procedures because of management did not affect the value of the responses regarding the use of PGF to induce ovulation in TAI protocols reported here. Indeed, these variations provided supporting evidence of PGF effects on ovulation induction under different management regimens.

Differences in the diameter of the largest follicle before TAI detected in experiment 2 may be attributed to the

mechanism of action of each ovulation inducer. Exogenous administration of esters of estradiol (EB and ECP) resulted in ovulation because of the induction of a synchronous LH surge [25]. Nevertheless, the PGF mechanism of action to induce ovulation most likely allowed the growth of the follicle for a longer period compared with EB. In experiment 2, heifers treated with PGF exhibited dominant follicles that grew approximately 3 mm from Days 9 to 11, whereas it did not grow during the same period in those heifers treated with EB. This lack of growth of the dominant follicle in heifers induced to ovulate with EB was most likely because of an earlier increase in LH secretion, which interrupted follicular growth and initiated cascading events that eventually resulted in ovulation. The results of the present study and those of previous reports [10,11] suggest that PGF promotes ovulation via distinct mechanisms other than only the indirect effect of luteolysis induction.

Several studies have attempted to understand how PGF acts on inducing ovulation [12,26–28]. Previous reports have demonstrated an increased frequency of LH release occurring 6 hours after treatment with a PGF analogue in anestrous cows [12]. In addition, it has been suggested that PGF2α exerts a direct effect on the anterior pituitary [8], which increases pituitary responsiveness to GnRH and thereby enhances the release of LH [12]. In bulls, a subcutaneous injection of 20 mg of PGF2α increased concentrations of LH [29]. However, the effect of PGF on LH secretion is controversial. Although PGF caused ovulation in anestrous cows [30] and ewes [31], it was only associated with LH release in cows. In addition to the central effect (pituitary–gonadal axis) previously described, PGF also appears to play a local role in the ovary. Prostaglandin secreted by the preovulatory follicle was closely linked to the ovulatory process [26]. Prostaglandins (PGE2 and PGF2α) are produced by the granulosa cells [32,33], and act directly on the preovulatory follicle. Considering this finding, ovulation induced by PGF could have an effect on sensitizing GnRH receptors in the pituitary [12] or by direct action on target cells of the preovulatory follicles. Prostaglandins are putative intraovarian mediators of several periovulatory events related to the ovulatory gonadotropin surge [26]. Moreover, prostaglandins appear to play a critical role in follicle rupture. Cyclooxygenase COX-2 (also known as prostaglandin endoperoxide synthase and prostaglandin G/H synthase), which catalyzes the rate-limiting step in the production of prostaglandin [34], is present in the mammalian ovary. Although the interval between the onset of the ovulatory gonadotropin surge and follicle rupture varies in rodents (14 hours), cows (28–30 hours), and horses (36–48 hours), COX-2 expression on the granulosa cells and elevated PG concentrations occurs in each of these species approximately 10 hours before follicle rupture [35]. In addition, deletion of COX-2 gene expression in knockout mice decreased the number of ovulations sites per ovary [36]. Thus, these reports provided strong evidences that PGF acts locally in the ovary during periovulatory events. Moreover, PGF secretion increases significantly in the follicle as ovulation approaches; thus, giving a pharmacologic dose of PGF might hasten ovulation.

Apparently, every prostaglandin analogue has the potential to induce ovulation in many species. Many commercial PGF analogues have been described to be associated to periovulatory events in cattle: alfaprostol [29], dinoprost [9], and sodium cloprostenol [10,11]; buffaloes: cloprostenol [37,38]; and ewes: D-cloprostenol [31]. Accordingly, both PGF analogues used in the present study were effective to induce ovulation: D-cloprostenol in experiment 1 and sodium cloprostenol in experiments 2 and 3. However, further studies on the effect of each single PGF analogue on ovulation are still necessary to properly evaluate specific properties of each compound.

Overall, the present study provides important data on the use of PGF as an ovulatory stimulus for TAI programs in cattle. From a commercial perspective, the major significance of this study was the possibility of developing a new method to induce ovulation in cattle. Interestingly, PGF has no withdrawal period, whereas estradiol esters do have a long withdrawal period or are even prohibited in several countries. Depending on the formula and the pharmaceutical laboratory, an ester of estradiol may have a withdrawal period of up to 30 days for milk and meat. Furthermore, since 2008, the use of estradiol-17β has been banned in farm animals by the European community because it has been deemed a complete carcinogen by the Committee on Veterinary Measures Relating Science to Public Health.

Thus, we strongly believe that PGF has a great potential to induce ovulation in TAI programs, and especially in substitution for estradiol or be used in GnRH-based protocols. Prostaglandins could enhance the response to GnRH or replace it, thereby reducing the costs of TAI protocols, particularly in countries where the use of estradiol is prohibited [39,40]. Although the TAI protocols used in this study did not completely remove estradiol from treatments, we abolished the second estradiol injection without decreasing ovulation rate, resulting in 9 fewer days in the withdrawal period, which would be cost-effective for the farmer. Further studies are required to investigate whether PGF could replace GnRH as a less expensive alternative in ovsynch-type protocols, or provide additional options for incorporating PGF-based protocols to cattle reproductive management programs. Our group is currently working on experiments aimed to appropriately respond to these challenges, and new hypotheses are being tested in an attempt to answer the above mentioned research questions.

4.1. Conclusions

Prostaglandin F2α analogue can be successfully used to induce ovulation in a synchronized fashion in heifers. In addition, the effectiveness of PGF as an ovulatory stimulus for TAI was confirmed in both heifers and cows on the basis of satisfactory ovulation and pregnancy rates.

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