Fixed-Time Artificial Insemination (TAI) in Suckled Beef Cows in Response to Equine Chorionic Gonadotropin (eCG)

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Administration of equine chorionic gonadotropin (eCG) at the time of prostaglandin (PGF) and CIDR removal of the CO-synch + CIDR protocol does not enhance pregnancy rates. Administration of eCG tended to increase follicle diameter at the time of insemination and increased progesterone concentration and corpus luteum volume seven days after AI.

Summary

Two experiments were conducted to evaluate whether eCG at the time of PGF injection had an effect on estradiol concentration in blood, dominant follicle diameter at timed artificial insemination (TAI), concentrations of progesterone (P4) and corpus luteum (CL) volume 7 d after TAI, and improves pregnancy rates. In Experiment 1, suckled beef cows (n = 513; pure- and crossbred Angus, Simmental, and Hereford) were enrolled in a 7-d CO-Synch + CIDR protocol (100 μg gonadotropin releasing hormone [GnRH] at controlled internal drug release device [CIDR] insertion [d -7]; 25 mg prostaglandin $F_{2\alpha}$ [PGF] at CIDR removal [d 0]; and 100 μg GnRH at TAI 66 h after PGF [d 3]) at three locations. Cows were randomly assigned to be controls or receive eCG (400 IU) at the time of PGF injection and CIDR insert removal. Pregnancy was diagnosed by transrectal ultrasonography at median d 35 and 67 after TAI. Serum P4 concentration was determined in blood samples collected on d -17, -7, 0, 3, and at both pregnancy diagnoses (pregnant cows only) to determine cycling status, luteolysis, and potential differences in CL function after TAI. Unadjusted pregnancy rates (PR) on d 35 was 42.9 vs. 49.8% for eCG vs. controls, respectively. Cycling cows were 1.5 times more (P = 0.05) likely to conceive than noncycling cows. Control cows were 1.5 times more (P = 0.04) likely to conceive than those treated with eCG. In Experiment 2, 35 cows were stratified by days postpartum and body condition score (BCS) and assigned to one of four treatments. Treatment 1 (control, CO; n = 9) and 2 (eCG; n = 9) were the same as described in Experiment 1; treatment 3 (COCR; n = 9) was the same as CO but calves were temporarily removed for 72 h between PGF and TAI; treatment 4 (eCGCR; n = 8) was the same as treatment 3 plus an additional injection of eCG (400 IU) was given at the time of CIDR removal. Ultrasonography was performed on d 0, 3, and 10 to evaluate follicle development and ovulation, CL regression between d 0 and 3 and CL volume on d 10. Blood samples were collected on d 0, 3, and 10 to measure P4 concentrations. Blood samples were collected every 12 h between d 0 and 3 to measure estradiol concentration in blood. Progesterone concentration on d 10 was greater (P < 0.01) for all the treatments compared with CO (1.4 vs 2.7 vs 2.7 and 2.8 ng/ml for CO, COCR, eCG, and eCGCR respectively). Follicle diameter tended (P = 0.09) to be greater for eCG than CO 48 h after eCG injection while COCR and eCGCR were intermediate (12.9, 13.1, 15.3, and 14.4 mm, for CO, COCR, eCG and eCGCR, respectively). Corpus luteum volume was greater (P < 0.05) on d 10 for eCGCR compared with CO and COCR (3.8 vs 2.1 vs 1.6 cm^3, for eCGCR, CO and COCR, respectively)

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while eCG was intermediate (2.9 ± 0.5 cm^3) but also greater (P < 0.05) than COCR. Estradiol concentration was greater (P < 0.05) for eCG treatment compared with control and eCGCR at 24 h (1.5 vs 0.5 vs 0.9 pg/ml, respectively) and 36 h (1.5 vs 0.4 vs 0.6 pg/ml, respectively) while COCR was intermediate (1.1 and 1.0 pg/ml, at 24 and 36 h, respectively). We conclude that eCG tended to increase dominant follicle diameter at TAI and increase concentrations of P4 and CL volume 7 d after TAI, however it did not improve pregnancy rates.

Introduction

Evidence exists that eCG and estradiol administration are important for a desirable pregnancy outcome in protocols for anestrous cows. Because no legal estrogen product is available in the U.S. market for use in cattle, we believed we could duplicate the estrogen effect by administering an appropriately small dose of eCG during the latter part of follicular maturation to stimulate more estradiol biosynthesis without inducing follicular proliferation (super-ovulation). We hypothesized that administering a small dose of eCG would stimulate greater estradiol secretion associated with follicular maturation that would mimic the pro-fertility effects of estradiol administration in beef cows. Support for this hypothesis is founded in studies where 400 IU of eCG were administered to cattle at the time of progesterone insert removal and ovulation was induced by estradiol benzoate before timed AI (Baruselli et al., 2003). Pregnancies per AI were increased in response to eCG (38.9 vs. 54.7%). Their hypothesis was that increased fertility resulted from stimulated luteal function in the CL that resulted from ovulation of follicles exposed to eCG. Using a similar protocol, it was reported that despite a similar-sized preovulatory follicle, both the CL size and concentrations of progesterone in blood plasma post-estrus were increased after eCG (Bergamaschi et al., 2005). The effects on eCG on circulating concentrations of estradiol were not quantified, but were likely increased in response to eCG-stimulated estradiol biosynthesis by the dominant follicle. Therefore, potentially increasing concentrations of estradiol by eCG administration after dominant follicle selection and near the time of induced luteolysis may duplicate the profertility effects in lactating cows that have been demonstrated by administering estrogen 24 to 36 h after induced luteolysis. The objective of Experiment 1 was to evaluate if eCG administered at time of PGF and CIDR insert removal will improve pregnancy rates in suckled beef cows. The objective of Experiment 2 was to evaluate if eCG would alter follicle diameter at the time of insemination and increases concentrations of P4 and CL volume seven days after timed AI.

Materials and Methods

Experiment 1

Suckled beef cows (n = 513; pure- and crossbred Angus, Simmental, and Hereford) were enrolled in a 7-d CO-Synch + CIDR protocol (100 µg GnRH at CIDR insertion [d -7]; 25 mg $PGF_{2\alpha}$ (PGF) at CIDR removal [d 0]; and 100 µg GnRH at AI 66 h after PGF [d 3]) at three locations. Cows were randomly assigned to be Controls or receive eCG (400 IU im) at the time of PGF injection and CIDR insert removal (Figure 1). Pregnancy was diagnosed by transrectal ultrasonography at median d 35 and 67 after AI. Serum P4 concentration (ng/mL) was determined in blood samples collected on d -17, -7, 0, 3, and at both pregnancy diagnoses (pregnant cows only) to determine cycling status, luteolysis, and potential differences in CL function after AI.

Experiment 2

Cows were stratified by days postpartum and body condition score and randomly assigned to one of four treatments: 1) received 100 µg GnRH and a CIDR insert (d -7), followed in 7 d by 25 mg PGF2α and CIDR removal (d 0), followed in 72 h by GnRH and AI (d 3); (Control; n = 9), 2) same as Control but calves were removed from their dams for 72 h between d 0 and d 3 (COCR; n = 9), 3) same as Control but cow received 400 IU of eCG on d 0 (eCG; n = 9), 4) same as COCR but cows received an aditional 400 IU of eCG on d 0 (eCGCR; n = 8). Transrectal ultrasonography and blood sample collection was performed every 12 h between PGF and TAI to follow follicle development and blood concentration of P4 and estradiol. Blood sample and ultrasonography was performed 7 d after TAI to measure concentration of P4 in blood and CL volume.

For both Experiment 1 and 2, the MIXED procedure of SAS was used to determine if differences existed for continuous variables: concentrations of hormones, follicle diameter, and corpus luteum volume, whereas the GLIMMIX procedure of SAS was used to determine difference in binary data of pregnancy and conception rate.

Results and Discussion

Experiment 1

Pretreatment cycling status differed ($P < 0.01$) among locations (locations 1 = 76.5%; 2 = 54.3%; and 3 = 27.4%). For cows having elevated (>1 ng/mL) P4 at CIDR insert removal, 97.4% had luteolysis, with 17.3% of cows having low (<1 ng/mL) P4 at insert removal and at TAI, and 1.2% with increasing P4 from insert removal to TAI. Progesterone did not differ on d 35 of pregnancy (6.0 ± 0.3 and 6.4 ± 0.4 ng/ml) or d 67 (6.6 ± 0.4 and 6.4 ± 0.3 ng/ml) for eCG and Controls, respectively. Unadjusted PR on d 35 was 42.9 vs. 49.8% for eCG vs. controls, respectively. Herd, cycling status, technician, and treatment influenced PR. Cycling cows were 1.5 times more ($P = 0.046$) likely to conceive than non-cycling cows. Control cows were 1.5 times more ($P = 0.036$) likely to conceive than those treated with eCG (Table 1). Cows in location 3 were 1.8 to 3.5 times more ($P = 0.004$) likely to conceive than cows at other locations. Pregnancy loss to d 67 did not differ ($P > 0.05$) between treatments (3.7 vs. 2.3% for eCG vs. Controls), respectively.

Experiment 2

The results of follicle diameter, P4 concentration, and CL volume are summarized in Table 2. Follicle diameter tended to be greater ($P = 0.09$) 48 h after PGF for eCG compared to Control, while COCR and eCGCR were intermediate (12.9, 13.1, 15.3, and 14.4 mm, for Control, COCR, eCG and eCGCR, respectively). Progesterone concentration on d 10 was greater ($P < 0.01$) for all the treatments compared with Control (Figure 2). Corpus luteum volume was greater ($P < 0.05$) on d 10 for eCGCR compared with Control and COCR (3.8 vs 2.1 vs 1.6 cm^3, respectively) while eCG was intermediate (2.9 ± 0.5 cm^3) but also greater ($P < 0.05$) than Control (Figure 3). Estradiol concentration was greater ($P < 0.05$) for eCG treatment compared with Control and eCGCR at 24 h (1.5 vs 0.5 vs 0.9 pg/ml, respectively) and 36 h (1.5 vs 0.4 vs 0.6 pg/ml, respectively) while COCR was intermediate (1.1 and 1.0 pg/ml, at 24 and 23 h, respectively; Figure 4). Our data from Experiment 1 demonstrate that 400 IU of eCG injected at time of CIDR insert removal did not improve pregnancy rates. Experiment 2 demonstrated that eCG increased estradiol blood concentration during the first 36 h after injection compared with the other treatments except from COCR. Injection of eCG tended to increase follicle diameter 48 h later and also elevated P4 concentration in serum and CL volume 7 d after ovulation.

Literature Cited

Baruselli et al.. 2003. Theriogenology 59:214.

Bergamashci et al. 2005. In: Anais do Congresso Latinoamericano de Buiatria 23; Jornadas Chilenas de Buitria 7, 2005, Valdivia, Chile, pp 154.

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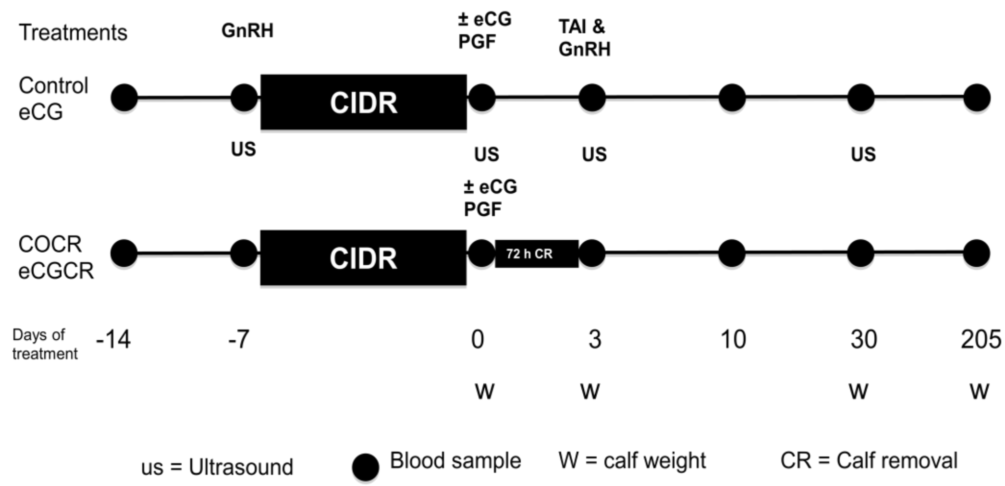

Figure 1. Schematic of experimental design for cows treated with or without eCG (eCG and Control, Exp. 1 and 2) and with or without eCG ± CR (eCGCR and COCR, Exp. 2).

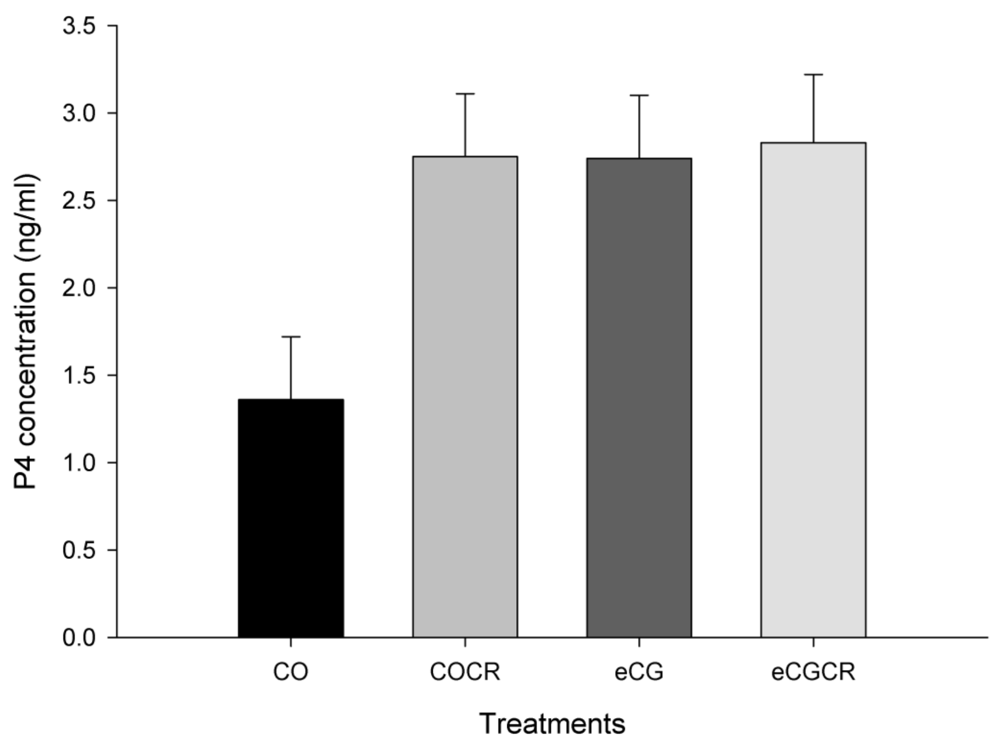

Figure 2. Concentration of progesterone (P4) on d 10 associated with cows assigned to one of four eCG or calf removal treatments on d 0. (Exp. 2) *Means differ (P < 0.01).

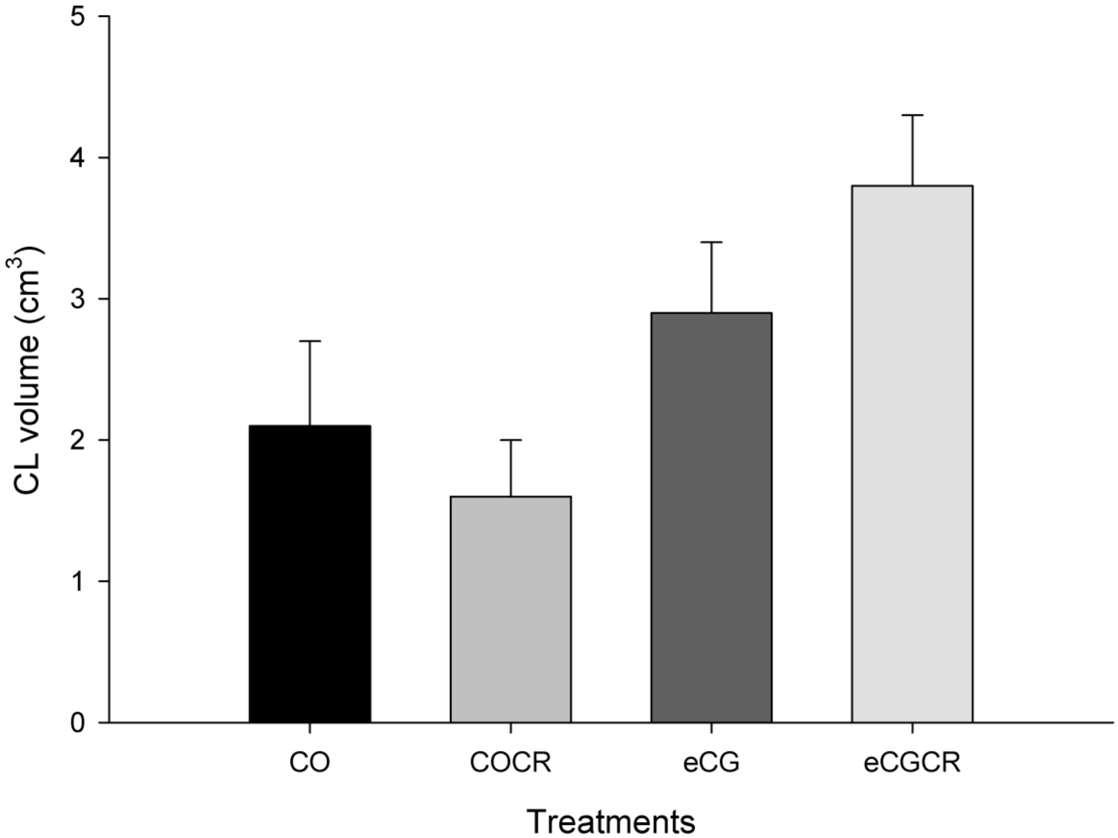

Figure 3. Corpus luteum (CL) volume on d 10 associated with cows assigned to one of four eCG or calf removal treatments on d 0. (Exp. 2) [abc]Means differ (P < 0.05).

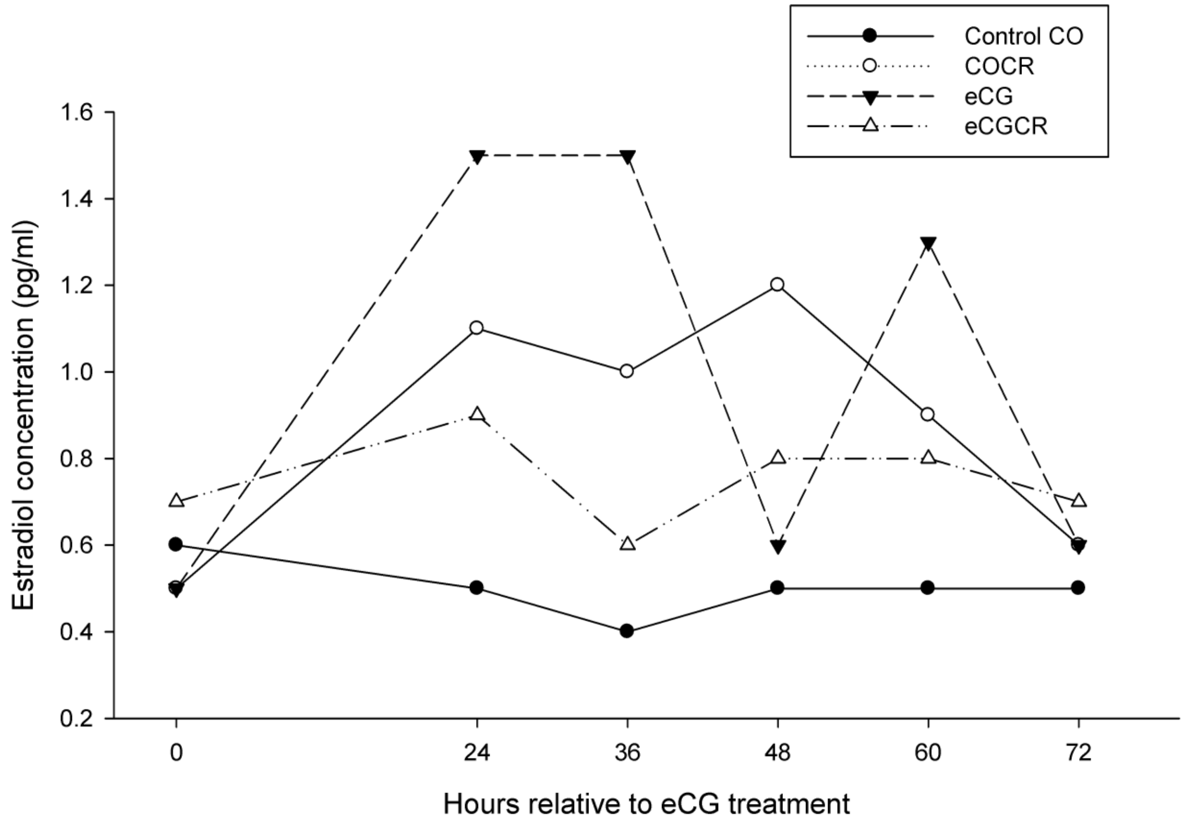

Figure 4. Concentration of estradiol relative to hours after treatment with or without eCG and/or calf removal (CR; Exp. 2). *Means differ (P < 0.05).

Table 1. Pregnancy rates and pregnancy loss in cows after fixed-time artificial insemination and either no treatment (Control) or treatment with 400 IU of eCG at CIDR removal (eCG; Experiment 1).

Treatment	Pregnancy rate[1]	Pregnancy loss[2]
	---------%---------	
eCG	42.9	3.7
Control	49.8[x]	2.3

[x]Control cows were 1.5 times more (P = 0.004; 95% CI = 1.1 to 5.6) likely to conceive than eCG cows.
[1]Detected on d 35 post-timed AI.
[2]Between d 35 and 67 of pregnancy.

Table 2. Follicle diameter, concentrations of progesterone (P4), and corpus luteum (CL) volume relative to equine chorionic gonadotropin (eCG) or 72-h temporary calf removal treatments (Experiment 2).

	Treatment with or without eCG or CR on d 0[1]			
Item	Control	COCR	eCG	eCGCR
Follicle diameter d 2, mm	12.9 ± 0.9[v]	13.1 ± 0.9[vw]	15.3 ± 0.9[w]	14.4 ± 1.0[vw]
P4 on d 10, ng/ml	1.4 ± 0.4[x]	2.7 ± 0.4[y]	2.7 ± 0.4[y]	2.8 ± 0.4[y]
CL volume on d 0, cm^3	2.1 ± 0.6[xy]	1.6 ± 0.4[xz]	2.9 ± 0.5[yz]	3.8 ± 0.5[y]

[1]CR = 72-hour temporary calf removal; eCG = treatment with 400 IU equine chorionic gonadotropin at CIDR removal; eCGCR = 72-hour temporary calf removal and treatment with 400 IU equine chorionic gonadotropin at CIDR removal.
[vw] Tendency for diameter or volume to differ (P = 0.10).
[xyz] Means within a row differ (P < 0.05).