

Fertility in Angus cross beef cows following 5d CO-Synch + CIDR or 7d CO-Synch + CIDR estrus synchronization and timed artificial insemination

William D. Whittier^{1*}, John F. Currin¹, Holly Schramm¹, Sarah Holland¹, Ramanathan K. Kasimanickam²,

¹Department of Large Animal Clinical Sciences, Virginia Polytechnic Institute and State University, Blacksburg, VA 24061

²Department of Clinical Sciences, Washington State University, Pullman, WA 99163

This report was accepted for publication in the Elsevier Publication “Theriogenology” on 13 July 2013. A portion of the data was collected at the Powell River Project.

Abstract

The present study determined whether a 5d CO-Synch + CIDR protocol with a two doses of prostaglandin F_{2α} (PGF) would improve timed AI pregnancy rate compared to 7d CO-Synch + CIDR protocol in beef cows. Angus cross beef cows (N = 1817) at 12 locations were randomly assigned to 5d CO-Synch + CIDR or 7d CO-Synch + CIDR groups. All cows received 100 µg of gonadorelin diacetate tetrahydrate (GnRH) and a controlled internal drug release (CIDR) insert on Day 0. Cows (n=911) in the 5d CO-Synch + CIDR group received two doses of 25 mg PGF, the first dose given on Day 5 at CIDR removal and the second dose 6 h later, and 100 µg GnRH on Day 8 and were inseminated concurrently, 72 h after CIDR removal. Cows (n=906) in 7d CO-Synch + CIDR group, received 25 mg of PGF at CIDR removal on Day 7, and 100 µg GnRH on Day 10 and were inseminated concurrently, 66 to 72 h after CIDR removal. All cows were fitted with a heat detector aid at CIDR removal and were observed twice daily until insemination for estrus and heat detector aid status. Accounting for estrus expression at or prior to AI (P<0.0001) and body condition score (P<0.01), cows in the 5d CO-Synch + CIDR group had greater AI pregnancy rate compared to cows in the 7d CO-Synch + CIDR groups (58.1% vs. 55.1%; P = 0.04). More cows that exhibited estrus at or prior to AI became pregnant compared to cows that did not [65.7% (681/1037) vs. 44.5% (347/780); P<0.0001]. The AI pregnancy rate was lesser for cows with body condition ≤ 4 (≤ 4 – 49.3% (101/219), 5 to 6 – 57.9%; > 6 – 55.8%). The mean AI pregnancy rate difference between treatment groups and projected economic outcome varied among locations. In conclusion, cows synchronized with the 5d CO-Synch + CIDR protocol had greater AI pregnancy rate than those that received the 7d CO-Synch + CIDR protocol.

1. Introduction

Synchronization of estrus and/or ovulation has the potential to shorten the calving season, increase calf uniformity, and enhance the use of AI in beef cows [1]. Labor required to handle cattle and observe estrus, as well as the cost and difficulty in implementation of AI have limited

the wide spread adoption of estrous synchronization and AI in the beef industry [2,3]. In order to facilitate the use of estrous synchronization by beef producers, protocols need to limit expense as well as time and labor, which can be achieved by minimizing the number of times and frequency at which cows are handled during the process. There is also a considerable advantage to protocols that minimize or eliminate detection of estrus by employing timed AI (TAI). Several synchronization protocols using PGF 2α (PGF) and GnRH, with or without a progestin, have been developed that successfully synchronize estrus in beef cows [4-7].

In the CO-Synch protocol, PGF is administered 7 d after GnRH followed by a second GnRH injection and TAI 66 to 72 h later. The Ovsynch protocol is similar except that the second GnRH injection occurs at 48 h after PGF, and the TAI occurs 12 to 24 h later. These protocols resulted in an acceptable AI pregnancy rate. When a controlled internal device that releases progesterone (CIDR) was added to the protocol between GnRH and PGF [5,6], pregnancy rates to timed AI improved. Recently, it has been demonstrated that reducing the interval from GnRH administration and CIDR insertion to PGF administration from 7 to 5 Days and extending the interval from PGF to TAI from 60 to 72 h increased TAI pregnancy rates in suckled beef cows [8]. While the 5 d CO-Synch + CIDR protocol is effective, it increases the drug and labor cost and may limit adoption due to the necessity of administering two doses of PGF [9, 10] to induce luteolysis of 5 d corpus luteum, However, limiting handling number and/or frequency should not compromise pregnancy success and added pregnancy success may justify the extra investment in some situations. The comparison of the 5d CO-Synch + CIDR with 2 doses of PGF at CIDR removal with 4 handlings vs. the 7d CO-Synch + CIDR with 3 handlings in a large scale beef operation would allow determination of the program which will yield a better AI pregnancy rate. Based on the necessity of this comparison, a hypothesis was developed that the 5d CO-Synch + CIDR protocol would improve fertility of beef cows following timed AI compared to the 7d CO-Synch + CIDR protocol.

The objective of the study was to determine whether a 5d CO-Synch + CIDR with two doses of PGF (first dose at CIDR withdrawal and a second dose 6 h after the initial dose on Day 5) would improve timed AI pregnancy rate compared to the 7d CO-Synch + CIDR in beef cows.

2. Materials and methods

2. Methods

Angus cross beef cows (n = 1817) at twelve locations, inseminated at a fixed time during fall of 2010 (n=856) and spring of 2011 (n = 961) were included in this study. Within location, cows were randomly allocated to the 5d CO-Synch + CIDR or the 7d CO-Synch + CIDR groups and were given a body condition score (BCS; 1-emaciated; 9-obese) at the initiation of estrous synchronization protocol. All cows received 100 μ g of gonadorelin diacetate tetrahydrate (GnRH; Cystorelin®, Merial Animal Health, Duluth, GA) and a controlled internal drug release (CIDR; 1.38g of progesterone; Eazi-Breed™ CIDR® Cattle Insert; Pfizer Animal Health, New York, NY) insert on Day 0. Cows (n=911) in the 5d CO-Synch + CIDR group received two doses of 25 mg doses of dinoprost (PGF; 5mL Lutalyse® sterile solution; Pfizer Animal Health),

first dose at CIDR removal and second dose at 6 h later, on Day 5, and 100 µg GnRH on Day 8 and were inseminated concurrently, 70 to 73 h after CIDR removal. Cows (n=906) in the 7d CO-Synch + CIDR group, received 25 mg of PGF at CIDR removal on Day 7, and 100 µg GnRH on Day 10 and were inseminated concurrently, 66 to 72 h after CIDR removal. AI sires (Angus (n=8), Simmental (n=2)) were selected and assigned to cows based on sire traits and to avoid inbreeding.

All cows were fitted with a heat detector aid (Kamar Inc., Steamboat Springs, CO, USA) at CIDR removal. After CIDR removal, the cows were observed twice daily until insemination for estrus and Kamar status (estrus, activated Kamar = red Kamar, partial red Kamar and lost Kamar with mount marks vs. no estrus, intact Kamar = white Kamar) and estrus status (standing to be mounted) was recorded. A cow was determined to be in estrus if she was visually observed to stand for mounting or if she had an activated (color change from white to red), lost (with mount marks) or partially-activated Kamar. The timing of CIDR insertion, CIDR withdrawal, interval to the second PGF2 α injection and timed-AI was recorded for each animal. A Schematic representation of the synchronization protocol is shown (Fig. 1). One week later, intact Angus bulls were placed with cows (approximately 1:40 to 1:50), across treatments, for the remainder of the 60 to 70 d breeding season. Cows were examined for pregnancy status at 55 to 70 d after fixed-time AI, by ultrasonography (Aloka-500, (Sysmed Lab, Chicago, IL, USA) to identify time of conception .

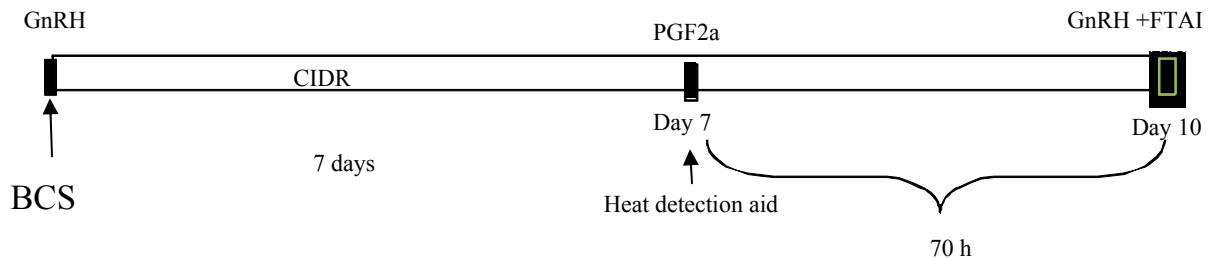
A partial farm budget type analysis for projected economic outcome was calculated based on mean AI pregnancy rate difference in each location and with the following assumptions.

1. \$49.14 advantage per AI pregnancy in the CO-Synch + CIDR treatments compared to a pregnancy from natural service [11],
2. One additional handling @ \$0.18/cow for cows in 5 day CO-Synch + CIDR group and
3. One additional PGF2a @ 2.07/dose for cows in 5 day CO-Synch + CIDR group

Because of the considerable variation in outcomes for the various locations this analysis becomes essentially a sensitivity analysis (how different AI pregnancy outcomes will impact economic outcomes under our conditions vs. a typical sensitivity analysis where regular intervals in outcomes are typically tested) for potential profit or loss for using the 5d CO-Synch + CIDR or 7d CO-Synch + CIDR protocol.

Mean AI pregnancy rate difference for each location was calculated by subtracting the mean AI pregnancy rate for the 7d CO-Synch + CIDR from the mean AI pregnancy rate for the 5d CO-Synch + CIDR.

7d CO-Synch + CIDR



5d CO-Synch + CIDR

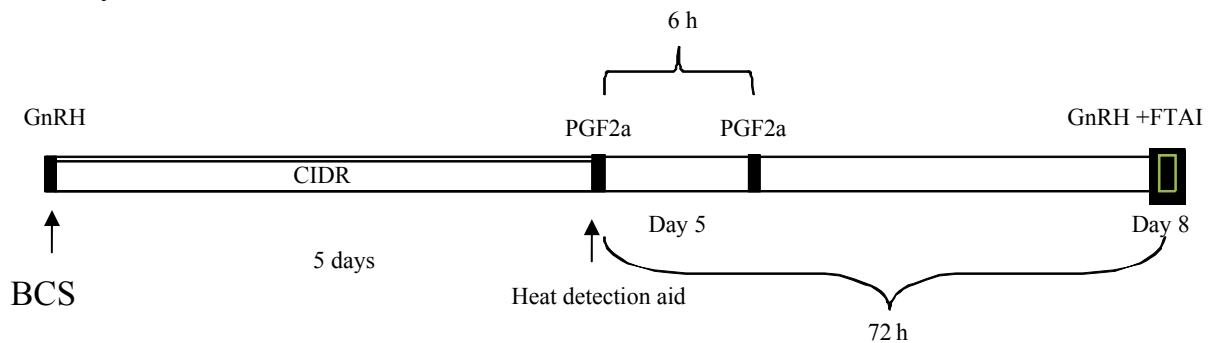


Figure 1. Schematic presentation of synchronization protocol

Briefly, Angus cross beef heifers ($n = 1817$) at 12 locations were randomly assigned to the 5d CO-Synch + CIDR or the 7d CO-Synch + CIDR groups. All cows received $100 \mu\text{g}$ of gonadorelin diacetate tetrahydrate (GnRH) and a controlled internal drug release (CIDR) insert on Day 0. Cows ($n = 911$) in the 5d CO-Synch + CIDR group received two doses of 25 mg PGF, the first dose given on Day 5 at CIDR removal and the second dose 6 h later, and $100 \mu\text{g}$ GnRH on Day 8 and were inseminated concurrently, 72 h after CIDR removal. Cows ($n = 906$) in the 7d CO-Synch + CIDR group, received 25 mg of PGF at CIDR removal on Day 7, and $100 \mu\text{g}$ GnRH on Day 10 and were inseminated concurrently, 72 h after CIDR removal. All cows were fitted with a heat detector aid at CIDR removal and were observed twice daily until insemination for estrus and heat detector aid status.

2.1 Statistical Analyses

Data were analyzed with a statistical software program (SAS Version 9.3 for Windows, SAS Institute, Cary, NC, USA). Differences in the mean body condition score between treatments were analyzed using one-way ANOVA (PROC GLM of SAS). Differences in mean interval (h) from CIDR insertion to CIDR withdrawal, interval from CIDR removal and time of

insemination between treatments were analyzed by ANOVA; the Bartlett test was used to assess homogeneity of variance (PROC GLM of SAS). Because variances for the mean interval were heterogeneous between treatments, a log₁₀ transformation was performed for testing differences. However, all tables and figures are presented with nontransformed values.

The PROC MIXED procedure of SAS [12] was used to examine the effect of explanatory variables influencing the estrus expression. Models included treatment (5d CO-Synch + CIDR vs. 7d CO-Synch + CIDR), days postpartum at initiation of synchronization (0 to 30, 31 to 60, 61 to 80, 81 to 100 and > 101 days), body condition scores (≤ 4 , 5 and 6 and > 6), season, location and appropriate interactions. Locations were offered as a random effect in the model. A *P* value of ≤ 0.05 was considered significant. For model reduction, the *P* value was set at ≤ 0.1 for inclusion and > 0.1 for exclusion until the model contained only significant main and interaction effects. The final model had treatment, location and days postpartum at initiation of synchronization.

The PROC MIXED procedure of SAS [11] was used to examine the effect of treatments (5d CO-Synch + CIDR vs. 7d CO-Synch + CIDR) on timed AI and breeding season pregnancy rates. Models included treatment (5d CO-Synch + CIDR vs. 7d CO-Synch + CIDR), AI-Sires, days postpartum at initiation of synchronization (30 to 60, 61 to 80, 81 to 100 and > 101 days), body condition scores (≤ 4 , 5 and 6 and > 6), estrus status prior to or at fixed time AI (yes or no), season, location and appropriate interactions. Locations and AI sires were offered as a random effect in the model. A *P* value of 0.05 was considered significant. For model reduction, the *P* value was set at ≤ 0.1 for inclusion and > 0.1 for exclusion until the model contained only significant main and interaction effects. The final model had treatment and estrus status.

It was hypothesized that the AI pregnancy rate difference between treatment groups will be 6.5%. To detect similar difference in the AI pregnancy rate, with adequate statistical power ($1-\beta = 0.8$) and statistical significance ($\alpha = 0.05$), the study needed a sample size of 900 cows per treatment group.

3. Results

There were 296, 178, 85, 494, 51, 61, 123, 61, 210 170, 49 and 39 cows from location 1 to location 12, respectively. Age, BCS and days postpartum at initiation of synchronization for cows assigned to the 2 treatments were not significantly different. The age, BCS and days postpartum at initiation of synchronization differed among locations (Table 2; $P < 0.05$). The time interval between CIDR insertion and removal did not differ between treatment groups ($P > 0.1$), however, the time interval differed between locations (Table 1; $P < 0.05$). In locations 2, 3, 4, 5 and 6 the mean time interval between CIDR removal and AI was 67.7 h.

Estrus expression rate differed between the 5d CO-Synch + CIDR and the 7d CO-Synch + CIDR groups ($P < 0.0001$), among days postpartum categories ($P < 0.01$) and body condition score categories ($P < 0.05$; Figure 2). A greater proportion of cows in the 7d CO-Synch + CIDR group expressed estrus compared to cows in the 5d CO-Synch + CIDR group, 61.1% (554/906) vs. 52.9% (482/911) respectively. A lesser proportion of cows calving late expressed estrus (30

to 60 - 49.6%; 61 to 80 – 59.5%; 81 to 100-59.2%; > 101 – 69.1%). Estrus expression rate ranged from 37.7 to 71.4% among locations (Figure 2). The estrus expression rate was lesser for cows with body condition score > 6 (≤ 4 – 58.9%, 5 to 6 – 57.8%; > 6 – 51.6%). There were no significant effects for treatment by body condition score or for treatment by days post calving interactions on estrus expression.

Table 1. Mean \pm SE¹ age, body condition score, days postpartum time interval, time intervals between CIDR² insertion and removal, between 1st and 2nd PGF³, and between CIDR removal and insemination for cows received 5- and 7-day CO-Synch + CIDR protocol[‡].

Factors	5d CO-Synch + CIDR	7d CO-Synch + CIDR
n	911	906
Age (y)	5.74 \pm 0.1	5.82 \pm 0.15
Body condition score ⁴	5.53 \pm 0.03	5.55 \pm 0.03
Days postpartum at initiation of synchronization	72.48 \pm 0.68	72.72 \pm 0.64
Interval from CIDR insertion to removal (h)	120.48 \pm 0.24	166.70 \pm 0.27
Interval between 2 PGF (h)	6.01 \pm 0.02	-
Interval from CIDR removal to AI (h)	72.13 \pm 0.44 ^a	69.58 \pm 2.19 ^b

ab- Different superscripts were statistically different P<0.05;

¹SE – Standard error;

²CIDR – Controlled internal drug release;

³PGF – Prostaglandin F2 α ;

⁴Refer to Figure 1 for treatment protocol;

⁵Body condition score: 1-emaciated; 9- obese

Adjusting for estrus expression at or prior to AI (P<0.0001) and body condition score (P<0.01) in the model, cows in the 5d CO-Synch + CIDR group had a greater AI pregnancy rate compared to cows in the 7d CO-Synch + CIDR group (58.1% vs. 55.1; P = 0.04). More cows that exhibited estrus at or prior to AI became pregnant compared to cows that did not [65.7% (681/1037) vs. 44.5% (347/780); P<0.0001]. The estrus expression rate was lesser for cows with body condition score ≤ 4 compared to other BCS categories (≤ 4 – 49.3%, 5 to 6 – 57.9%; > 6 – 55.8%). The AI pregnancy rate ranged from 37.3 to 70.5 among locations (Figure 3; P<0.01). There were no significant effects for treatment by body condition score and treatment by estrus at or prior to AI interactions on AI pregnancy rate. Mean AI pregnancy rate differences between treatment groups for different locations are shown in Figure 4.

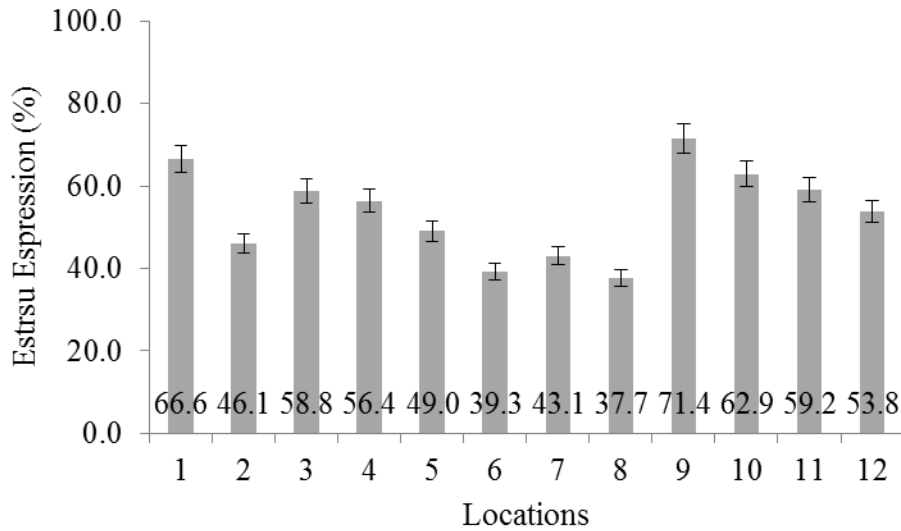


Figure 2. Percent estrus expression at or before timed AI in Angus cross beef cows in different locations synchronized with either the 5d or the 7d CO-Synch + CIDR protocol[‡] (P<0.0001)

[‡]Refer Figure 1 for treatment protocol

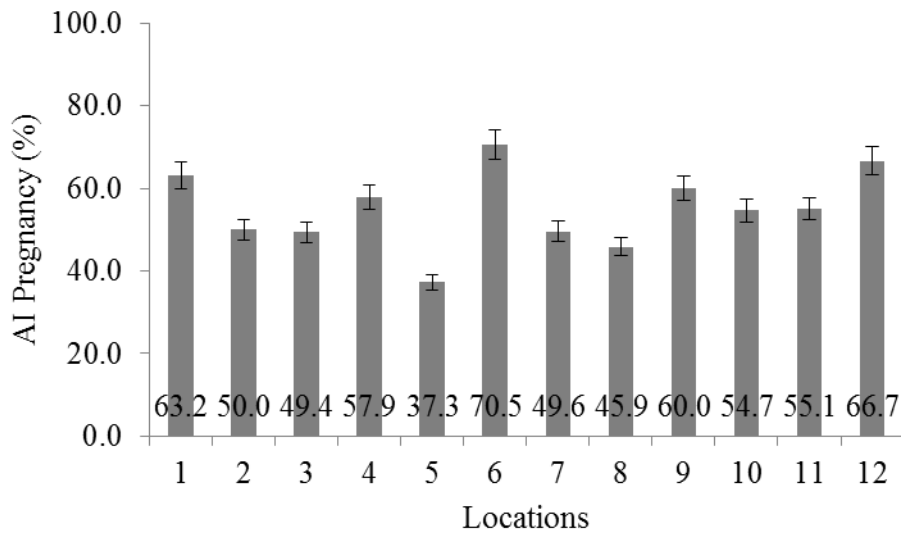


Figure 3. Percent AI pregnancy in Angus cross beef cows in different locations synchronized with either the 5d or the 7d CO-Synch + CIDR protocol[‡] (P<0.0001).

[‡]Refer Figure 1 for treatment protocol

Table 2. Variables influencing estrus expression by Angus cross beef cows synchronized with either 5-d or 7d CO-Synch + CIDR protocol¹

Effect	DF	F Value	P > F
Treatment ¹	1	14.83	0.0001
Body condition score ³	2	3.05	0.0476
Days post calving ⁴	4	4.30	0.0018

¹Refer to Figure 1 for treatment protocol;

²DF- Degrees of freedom;

³Body condition score categories, (<4, 5 to 6; >6);

⁴Days post calving categories (0 to 30, 31 to 60; 61 to 80, 81 to 100, >100);

Note: Location (1 to 12) was treated as random effect;

Table 3. Effect of treatment, body condition score and whether cows exhibited estrus at or prior to estrus on AI pregnancy in Angus cross beef cows synchronized with either 5-d or 7d CO-Synch + CIDR protocol¹

Effect	DF ²	F Value	P > F
Treatment ¹	1	4.07	0.0437
Body condition score ³	2	3.37	0.0347
Estrus at or prior to AI ⁴	1	64.39	<.0001

¹Refer Figure 1 for treatment protocol;

¹ DF-Degrees of freedom;;

³Body condition score categories, (<4, 5 to 6; >6)

⁴Cows visually observed to stand for mounting or had an activated (color change from white to red), lost (with mount marks) or partially-activated Kamar;

Note: Location (1 to 12) and AI Sires were treated as random effect;

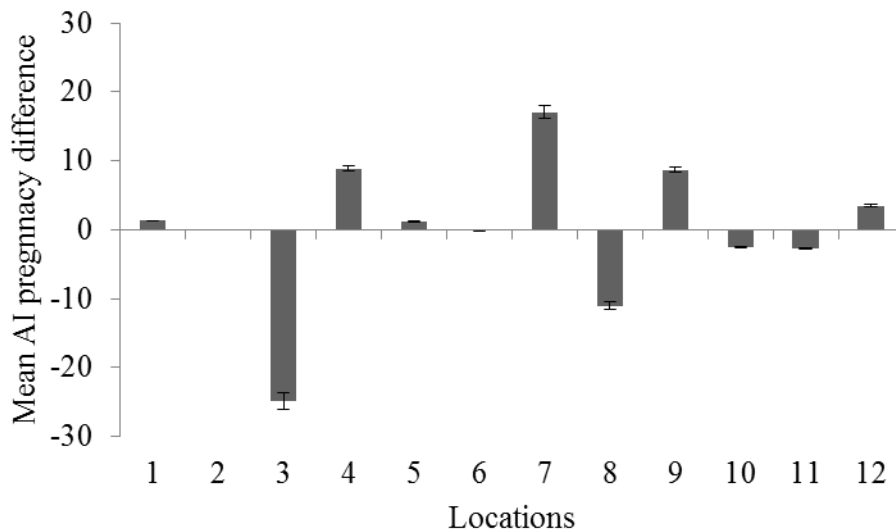


Figure 4. Mean AI pregnancy rate difference* between treatment[‡] groups in different locations

*Mean AI rate pregnancy difference = Mean AI pregnancy rate for the 5d CO-Synch + CIDR – Mean AI pregnancy rate the 7d CO-Synch + CIDR.

[‡]Refer Figure 1 for treatment protocol.

The analysis of projected economic outcomes for 100 cows for implementing 5 day CO-Synch protocol instead of 7 day CO-Synch protocol based on the mean AI pregnancy rate difference is presented in table 4. Three herds would have experienced an economic benefit and 9 herds would have had a lesser economic return with the results from this study.

Table 4. Economic analysis¹ per cow (based on analysis for 100 cows) - Economic benefit or loss (\$ US) from implementing the 5 day CO-Synch+CIDR program instead of the 7 day Co-Synch+CIDR program.

Locations	Mean AI pregnancy difference (%)	Gain or loss per AI pregnancy (\$) ²	Increased or decreased additional cost (\$) ³	Benefit per cow (\$) ⁴
1	1.3	0.64	2.25	-1.61
2	0	0.00	2.25	-2.25
3	-24.9	-12.24	2.25	-14.49
4	8.9	4.37	2.25	2.12
5	1.2	.59	2.25	-1.66
6	-0.2	-0.10	2.25	-2.35
7	17.1	8.40	2.25	6.15
8	-11.1	-5.45	2.25	-7.70
9	8.7	4.28	2.25	2.03
10	-2.6	-1.28	2.25	-3.53
11	-2.7	-1.33	2.25	-3.57
12	3.5	1.72	2.25	-0.53
Overall	3.0	1.47	2.25	-0.78

¹ How different AI pregnancy rates will impact economic outcomes under a given set of parameters;

² \$49.14 US advantage per AI pregnancy in the CO-Synch + CIDR treatments compared to a pregnancy from natural service [11];

³ Additional cost = 1 extra handling @ \$0.18/cow + 1 additional PGF2a @ 2.07/dose for cows in 5 day CO-Synch + CIDR group;

⁴ Benefit for the herd = US dollar gain or loss per AI pregnancy - Additional cost per cow;

4. Discussion

Beef cows inseminated following the 5d CO-Synch + CIDR protocol yielded greater AI pregnancy rate (3.0%) compared to cows inseminated after the 7d CO-Synch + CIDR protocol.

Limitation in the success of synchronizing estrus and achieving acceptable pregnancy rates in suckled beef cows is the fact that a significant proportion of cows are anestrus at the onset of the breeding season [5,6,7]. Numerous estrous synchronization protocols using PGF, GnRH, and (or) a progestin have been developed that induce cyclicity and successfully synchronize estrus in suckled beef cows [5,6,13]. In this study, 57% of cows exhibited estrus at or prior to AI which was lesser than the previous studies [7,14]. Cyclicity of cows in this study ranged from 38% to 71% at 12

locations. The previous studies indicate that there is large variability in the proportion of cycling females among beef cattle operations at the beginning of the breeding season [5,7,13,14].

Days post partum at the initiation of synchronization, body condition, as well as parity, can impact the proportion of cows that have resumed estrous cycle activity post-calving [7,13,14]. In this study, a lesser proportion of cows calving late in the calving season expressed estrus compared to cows that calved early in the preceding calving season (synchronization initiation at 30 to 60 dpp - 49.6% vs > 60 dpp – 60.1%). Overall, 28.8% (524/1817) of cows were in the category that had synchronization initiated at 30 to 60 dpp. Previous studies have demonstrated that primiparous cows are more likely to be anestrous at the initiation of the breeding season when compared to multiparous females [7, 13-16]. Body conditions score categories did affect the estrus expression in this study but age did not. More cows in the 7d CO-Synch + CIDR group exhibited estrus compared to cows in the 5d CO-Synch + CIDR group. It should be noted that the duration of the 7d CO-Synch + CIDR protocol is 10 days compared to 8 days for the 5d CO-Synch + CIDR group. Hence, cows in 7d CO-Synch + CIDR group had more time for the follicular growth after the GnRH resetting of the follicular wave and it is possible that the cows in 7d CO-Synch + CIDR group had bigger follicles which produced more estrogen and resulted in more cows exhibiting estrus. The variables days postpartum, estrus expression at or prior to AI and body conditions were accounted for in the effect of treatment on AI pregnancy rates.

In this study, cows synchronized with the 5d CO-Synch + CIDR protocol had a greater AI pregnancy rate compared to cows synchronized with 7d CO-Synch + CIDR protocol (58.1% vs. 55.1). In several previous studies, though not in contemporaneous comparisons, the AI pregnancy rates for the 5d CO-Synch+CIDR protocol ranged from 54.9 to 69% with a mean of 56.6% [17,18] and AI pregnancy rates for the 7d CO-Synch + CIDR protocol ranged from 50.4 to 58.8 % with a mean of 53.4% [19-21]. Cows that exhibited estrus had a greater pregnancy rate compared to cows that did not. It is interesting to note that more cows in the 7d CO-Synch + CIDR group exhibited estrus compared to cows in the 5d CO-Synch + CIDR group; nevertheless a greater proportion of cows in the 5d CO-Synch + CIDR group become pregnant. There was no treatment by estrus expression interaction for AI pregnancy rate ($P>0.1$). Location greatly influenced AI pregnancy rate ($P<0.0001$), which may have been a result of differing management, nutrition, genetics, production goals, and environment.

In this study, the 5d CO-Synch + CIDR protocol included 2 doses of PGF. Although one exogenous dose of PGF can induce rapid luteolysis in the mid-luteal phase, the early CL (up to d 5 post estrus) is somewhat refractory to the luteolytic action of PGF. In this study PGF administration occurred 5 days following the first GnRH injection. If the first GnRH injection induced ovulation and/or luteinization, then the CL would have been 5 days old at PGF administration which may be the reason for decreased luteolysis to a single dose of PGF. While variable responses in luteolysis have been noted when a single dose of PGF was given on Day 5 of the estrous cycle, the results from the previous studies indicated that two doses of PGF will overcome the refractoriness of the early CL, induce luteolysis and result in a greater AI pregnancy [17]. In this study the interval between first and second PGF was 6 h. This interval was chosen based on the result from previous study in which we have shown that cows receiving

a second PGF dose between 4 and 5 h and between 6 and 7 h resulted in 63.4% and 60.8% timed AI pregnancy rates, respectively, which was greater than any other interval examined [18].

The recommended time of insemination for the 7d CO-Synch+CIDR is 60 to 66 h after removal. In this study the mean time interval is 69.6 h (range 66.7 to 72.0 h) after CIDR removal. Studies that performed AI at 60 to 66 h from CIDR removal reported 54% (45 to 68%) AI-PR [22-27], whereas AI performed at 72 h from CIDR removal resulted in 51% AI-PR [28]. Busch et al., (2008) observed pregnancy rates from FTAI following CO-Synch + CIDR at 66 h were greater than those resulting from FTAI at 54 h [25]. Dobbins et al., (2006) determined conception rates after altered timing of AI associated with the CO-Synch + CIDR protocol. The conception rates at 48, 56, 64, and 72 h produced quadratic curves that peaked ($P < 0.01$) between 56 and 64 h [28]. Interestingly, the mean time interval from CIDR removal to AI in 6 locations was 67.7 h and 71.5 h for the other 6 locations. In the current study, even though there was difference in estrus expression between these groups ($P < 0.05$), no difference in AI-PR was observed for these two location groupings ($P > 0.05$).

In 6 locations, 5 day CO-Synch+CIDR protocol resulted in an increased AI pregnancy rates compared to the 7 day CO-Synch+CIDR protocol (1.3% to 17.1%; Mean AI pregnancy rate difference = Mean AI pregnancy rate for the 5d CO-Synch + CIDR – Mean AI pregnancy rate for the 7d CO-Synch + CIDR 1), whereas the 7 day CO-Synch+CIDR protocol resulted in greater AI pregnancy in 5 locations compared to the 5 day CO-Synch+CIDR protocol (0.2 to 24.9%) (Figure. 4). In one location there was no difference between treatment groups. The economic sensitivity analysis presented in Table 5 indicates that, given the mean difference in AI pregnancy rates between treatment groups in different locations from this study, substantially different profit/loss outcomes would result. For a producer to implement the 5 day CO-Synch+CIDR protocol rather than of the 7 day CO-Synch+CIDR protocol in 100 cows, the economic outcome could be as great as \$1448.59 less or up to \$615.29 more. The wide range in differences in pregnancy outcome for different treatments at these locations makes this a type of economic sensitivity analysis. It should be noted that the herd size varied widely for locations included in this study. So it is reasonable to expect that the random effects on economic benefit may vary depending upon the herd size. In order to have a similar breakeven, given the additional costs incurred, the 5-d CO-Synch + CIDR required a 5% greater pregnancy compared to the 7-d CO-Synch + CIDR protocol. Even though the economic sensitivity analysis was based on 100 cows, in locations where the herd size was less than 100, cows experienced greater loss (Mean: \$-505.10; range: -53.01 to -1448.59) compared to locations with herd size was more than 100 cows (Mean: \$48.55; range: -352.76 to 615.29).

Cows that received timed insemination following the 5d CO-Synch + CIDR synchronization protocol had 3% greater AI pregnancy rate than cows that received timed insemination following the 7d CO-Synch + CIDR synchronization protocol. In 6 out of 12 locations, cows that received the 5d CO-Synch + CIDR protocol had greater AI pregnancy rates than cows that received the 7d CO-Synch + CIDR protocol and, in 3 out of 12 locations, this

resulted in economic benefit. Due to vast variations in the economic benefit it would be prudent to consider the resources available before implementing a synchronization protocol.

Acknowledgements

The authors thank Staff of Virginia Department of Corrections and the Powell River Project for their cooperation and express their gratitude to Select Sires Inc., for their partial financial support.

References

- [1] Dziuk PJ, Bellows RA. Management of reproduction in beef cattle, sheep and pigs. *J Anim Sci* 1983;57(Suppl. 2):355-379.
- [2] http://www.aphis.usda.gov/animal_health/nahms/bee5Dowcalf/downloads/beef0708/Beef0708_dr_PartII.pdf (Pg. 18)
- [3] http://www.aphis.usda.gov/animal_health/nahms/bee5Dowcalf/downloads/beef0708/Beef0708_dr_PartII.pdf (Pg. 21)
- [4] Thompson KE, Stevenson JS, Lamb GC, Grieger DM, Löest CA. Follicular, hormonal, and pregnancy responses of early postpartum suckled beef cows to GnRH, norgestomet, and prostaglandin F_{2α}. *J Anim Sci* 1999;77:1823-1832.
- [5] Lamb GC, Stevenson JS, Kesler DJ, Garverick HA, Brown DR, Salfen BE. Inclusion of an intravaginal progesterone insert plus GnRH and prostaglandin F_{2α} for ovulation control in postpartum suckled beef cows. *J Anim Sci* 2001;79:2253-2259.
- [6] Stevenson JS, Johnson SK, Milliken GA. Symposium Paper: Incidence of postpartum anestrus in suckled beef cattle: Treatments to induce estrus, ovulation and conception. *Prof Anim Sci* 2003;19:124-134.
- [7] Larson JE, Lamb GC, Stevenson JS, Johnson SK, Day ML, Geary TW, et al., Synchronization of estrus in suckled beef cows for detected estrus and artificial insemination and time artificial insemination using gonadotropin-releasing hormone, prostaglandin F_{2α}, and progesterone. *J. Anim. Sci.* 2006;84:332-342.
- [8] Bridges GA, Helser LA, Grum DE, Mussard ML, Day ML. Decreasing the interval between GnRH and PGF_{2α} from 7 to 5 days and lengthening proestrus increases timed-AI pregnancy rates in beef cows. *Theriogenology* 2008;69:843-851.
- [9] Kasimanickam R, Day ML, Rudolph JS, Hall, Whittier WD. Two doses of prostaglandin improve pregnancy rates to timed-AI in a 5-day progesterone based synchronization protocol in beef cows. *Theriogenology* 2009;71:762-767.
- [10] Cruppe LH, Souto LA, Maquivar M, Gunn P, Mussard ML, Wolfenson D, et al., Use of two coincident doses of PGF_{2α} with the 5d CO-Synch + CIDR estrous synchronization program. *J Anim Sci* 2010;88 (Suppl. 1):849 (Abstr.).
- [11] Rodgers JC, Bird SL, Larson JE, Dilorenzo N, Dahlen CR, Dicostanzo A, et al., An economic evaluation of estrous synchronization and timed artificial insemination in suckled beef cows. *J Anim Sci* 2012 : 90:4055-4062.
- [12] Finger JD. Using SAS PROC MIXED to fit multilevel models, hierarchical models and individual growth models. *J Educ Behav Stat* 1998;24:323-355.
- [13] Stevenson J S, Thompson KE, Forbes WL, Lamb GC, Grieger DM, Corah LR. Synchronizing estrus and (or) ovulation in beef cows after combinations of GnRH, norgestomet, and prostaglandin F_{2α} with or without timed insemination. *JAnim Sci* 2000;78:1747-1758.
- [14] Stevenson JS, Lamb GC, Johnson SK, Medina-Britos MA, Grieger DM, Harmony KR, et al., Supplemental norgestomet, progesterone, or melengestrol acetate increases pregnancy rates

in suckled beef cows after timed inseminations. *J Anim Sci* 2003;81:571-856.

- [15] Short RE, Adams DC. Nutritional and hormonal interrelationships in beef cattle reproduction. *Can J Anim Sci* 1988;68:29-39.
- [16] Short RE, Bellows RA, Staigmiller RB, Berardinelli JG, Custer EE. Physiological mechanisms controlling anestrus and infertility in postpartum beef cattle. *J Anim Sci* 1990;68:799-816.
- [17] Kasimanickam R, Day ML, Rudolph JS, Hall JB, Whittier WD. Two doses of prostaglandin improve pregnancy rates to timed-AI in a 5-day progesterone-based synchronization protocol in beef cows. *Theriogenology* 2009;71:762-767.
- [18] Whittier WD, Kasimanickam RK, Currin JF, Schramm HH, Vlcek M. Effect of timing of second prostaglandin F2 alpha administration in a 5-day, progesterone-based CO-Synch protocol on AI pregnancy rates in beef cows. *Theriogenology* 2010;74:1002-1009.
- [19] Bucher A, Kasimanickam R, Hall JB, Dejarnette JM, Whittier WD, Kähn W, et al., Fixed-time AI pregnancy rate following insemination with frozen-thawed or fresh-extended semen in progesterone supplemented CO-Synch protocol in beef cows. *Theriogenology* 2009;71:1180-1185.
- [20] Kasimanickam R, Whittier WD, Collins JC, Currin JF, Inman B, Hall JB, et al., A field study of the effects of a monovalent *Leptospira borgpetersenii* serovar Hardjo strain hardjobovis vaccine administered with oxytetracycline on reproductive performance in beef cattle. *J Am Vet Med Assoc* 2007;231:1709-1714.
- [21] Kasimanickam R, Hall JB, Currin JF, Whittier WD. Sire effect on the pregnancy outcome in beef cows synchronized with progesterone based Ovsynch and CO-Synch protocols. *Anim Reprod Sci* 2008;104:1-8.
- [22] Stevenson JS, Lamb GC, Johnson SK, Medina-Britos MA, Grieger DM, Harmony KR, et al. Supplemental norgestomet, progesterone, or melengestrol acetate increases pregnancy rates in suckled beef cows after timed inseminations. *J Anim Sci* 2003;81:571-586.
- [23] Kasimanickam R, Collins JC, Wuenschell J, Currin JC, Hall JB, Whittier DW. Effect of timing of prostaglandin administration, controlled internal drug release removal and gonadotropin releasing hormone administration on pregnancy rate in fixed-time AI protocols in crossbred Angus cows. *Theriogenology*. 2006;66:166-172.
- [24] Larson JE, Lamb GC, Stevenson JS, Johnson SK, Day ML, Geary TW, et al. Synchronization of estrus in suckled beef cows for detected estrus and artificial insemination and timed artificial insemination using gonadotropin-releasing hormone, prostaglandin F2alpha, and progesterone. *J Anim Sci* 2006;84:332-342.
- [25] Busch DC, Wilson DJ, Schafer DJ, Leitman NR, Haden JK, Eilersieck MR, et al. Comparison of progestin-based estrus synchronization protocols before fixed-time artificial insemination on pregnancy rate in beef heifers. *J Anim Sci* 2007;85:1933-1939.
- [26] Bridges GA, Helser LA, Grum DE, Mussard ML, Gasser CL, Day ML. Decreasing the interval between GnRH and PGF2alpha from 7 to 5 days and lengthening proestrus increases timed-AI pregnancy rates in beef cows. *Theriogenology* 2008;69:843-851.
- [27] Kasimanickam R, Hall JB, Currin JF, Whittier WD. Sire effect on the pregnancy outcome in beef cows synchronized with progesterone based Ovsynch and CO-Synch protocols. *Anim Reprod Sci* 2008;104:1-8.
- [28] Dobbins CA, Tenhouse DE, Eborn DR, Harmony KR, Johnson SK, Stevenson JS. Conception rates after altered timing of AI associated with the CO-Synch + CIDR protocol. *J Anim Sci* 2006;84(Suppl. 1):50.