## Physiology and Endocrinology: Synchronization of Estrus in Cattle

713 ASAS Centennial Presentation: Development of cattle estrus and breeding management. J. W. Lauderdale\*, *Lauderdale Enterprises, Inc, Augusta, MI.* 

McKenzie's laboratory (U. MO; 1930s) described the estrous cycle of domestic animals. Brownell (Cornell; 1936) reported AI and bull bred pregnancy rates were similar. Between 1938 (first, EJ Perry, NJ) and 1940, 54 AI organizations in 21 states were formed. Ulberg, Christian and Casida (1951) controlled time of ovulation in cows with progesterone (P) injected daily. Trimberger and Hansel (1955) controlled time of ovulation in cows with P injected daily but conception rate was reduced. Zimbelman (1963) reported the dose-response for MAP in cattle, leading to the first orally active progestogen to synchronize estrus in cattle. Participants at the Brook Lodge Ovarian Regulatory Mechanisms Conference (1965) reviewed P for cattle estrus synchronization; discussions launched discovery and development of prostaglandins (PGF2alpha) for regressing corpora lutea (CL) in cattle. Laboratories led by Thorburn, Niswender, Scaramuzzi, Henricks and Hansel (1969-1972) reported profiles of reproductive hormones, data crucial to development of successful breeding management programs. Rowson et al., Lauderdale, and Liehr et al. (each 1972) reported PGF2alpha to be luteolytic in cattle. Lauderdale (1974) reported PGF2alpha estrus synchronization programs and AI, including AI at a specifically designated day and time (TAI). Hansel and Fortune (1978) reported use of GnRH plus PGF2alpha enhanced estrus synchronization and pregnancy rate over PGF2alpha alone in cattle. The Ginther laboratory pioneered and developed transrectal ultrasonography (1982-1998), leading to characterization of follicular waves in cattle (Fortune et al. and Ginther et al., each 1988). Harms, Wiltbank and Randel (1984) reported pulsatile release of LH in cattle, leading to management of follicular waves with GnRH. Knowledge of the estrous cycle of cattle, including follicular waves, and commercially available progestogen, PGF2alpha and GnRH products led to development of multiple cattle breeding management programs by numerous ASAS members. TAI pregnancy rates of 60%-70% are achieved routinely under commercial cattle breeding management programs using PGF2alpha, GnRH and progestogen products.

Key Words: Cattle, Breeding Management, History

714 Identification of differential gene expression during transition of bovine corpus luteum from early to mid–phase and their potential role in acquisition of luteolytic sensitivity to prostaglandin F2 alpha. M. P. Gorvanahally\*, M. Salem, J. Yao, K. Inskeep, and J. A. Flores, *West Virginia University, Morgantown.* 

Prostaglandin F2 alpha (PGF<sub>2α</sub>) brings about regression of the bovine corpus luteum (CL). This luteolytic property of PGF<sub>2α</sub> is used in the beef and dairy cattle to synchronize estrus. A limitation of this protocol is an insensitivity of the early CL to luteolytic actions of PGF<sub>2α</sub>. The mechanisms underlying luteal sensitivity (LS) are poorly understood. The early CL has maximum number of PGF<sub>2α</sub> receptors; therefore differences in signaling events might be responsible for LS. Hence differential gene expression at two developmental stages, days 4 (D-4) and 10 (D-10) post estrus, might account for differences in signal transduction pathways associated with LS. This possibility was examined in these studies. Microarray analysis (n=3 per stage) identified 180 genes that were differentially expressed (P<0.05). These were categorized

into genes involved in cell signaling (13%), metabolism (10%), protein degradation (5%), RNA processing and transcription regulation (15%), protein biosynthesis and modification (19%), extracellular matrix and cvtoskeletal proteins (6%), DNA replication and modulation (2%), antioxidant property (3%), miscellaneous (17%), and unknown functions (10%). Real-time PCR confirmed the differential expression of 9 randomly selected genes, including protein kinase C inhibitor protein-1 (KCIP-1) and regulator of G-protein signaling 2 (RGS2), observed in microarray. Further, the in vivo effect of exogenous  $PGF_{2\alpha}$  (n=3 per stage) on selected genes that were found differentially expressed during this developmental transition was examined.  $PGF_{2\alpha}$  increased the expression of a guanine nucleotide binding protein beta polypeptide 1 (GNB1) in D-4 CL and Ca2+/calmodulin dependent kinase kinase 2 beta (Camkk2) in D-10 CL. Therefore, GNB1, Camkk2, KCIP-1, and RGS2 are candidate genes that might play significant role in acquisition of luteal sensitivity to PGF2a.

Key Words: Corpus Luteum, PGF<sub>2α</sub>, Corpus Luteum Insensitivity

**715** Synchronizing new follicular wave emergence in *Bos indicus*influenced heifers with estradiol benzoate: Role of the magnitude of the acute increase in progesterone. J. D. Pack\*1.<sup>2</sup>, I. C. Velez<sup>1.2</sup>, M. Amstalden<sup>1,2</sup>, and G. L. Williams<sup>1,2</sup>, <sup>1</sup>*Texas AgriLife Research, Beeville, TX*, <sup>2</sup>*Texas A&M University, College Station.* 

The hypothesis was that incremental changes in circulating progesterone (P4) are an important factor in the ability of estradiol benzoate (EB) to successfully synchronize new follicular wave emergence (NFWE) and ovulation. Eight pubertal Braford (F-1) heifers were used in a twice replicated Latin square design with four treatments designed to provide differing maximum concentrations of plasma P4: 1) EB; 2.5 mg EB i.m. only; 2) EB + CIDR; EB and new CIDR; 3) EB + AC-CIDR; EB and new autoclaved CIDR; 4) EB + AC-CIDR + P4; EB, new autoclaved CIDR and 500 mg P4 i.m. at CIDR insertion. Heifers received treatments in random order between d 3 and 6 after ovulation (d 0), with a 2 to 3-wk washout period between each experimental period. On d 7, CIDR inserts were removed and all cows were treated with 25 mg prostaglandin F2-alpha. Transrectal ultrasonography was used to monitor ovarian structures throughout. Jugular blood samples were collected at 0, 30, 60, 120, 240, and 360 min, and at 6-h intervals through 60 h for hormone assays. Treatments 2 through 4 increased (P < 0.01) circulating P4 concentrations compared to treatment 1 (EB only); however, treatment 4 (EB + AC-CIDR + P4) created the greatest increase (2-h peak = 42.5ng/mL) of longest duration (at least 60 h). Groups 2 and 3 (EB + CIDR and EB + AC-CIDR) were intermediate (4-h peaks = 10.7 ng/mL) and did not differ. Plasma progesterone in group 1 (EB only) remained < 5 ng/mL. Suppression of plasma FSH was greatest (P<0.001) in group 4 (EB + AC-CIDR + P4), with mean 60-h concentrations  $(1.5 \pm 0.4 \text{ ng/mL})$ less than in all other groups  $(1.8 \pm 0.4 \text{ ng/mL})$ . Mean concentrations of LH ( $0.4 \pm 0.1$  ng/mL), frequencies of occurrence of NFWE ( $87.5 \pm 7.2$ %) and ovulation (84.4  $\pm$  6 %), and intervals to NFWE (5.0  $\pm$  0.3 d) did not differ among treatments. Elevating circulating P4 above that created by a corpus luteum or corpus luteum + CIDR, delayed FSH resurgence but failed to alter timing or occurrence of ovarian events.

Key Words: Synchronization, Bos indicus, Progesterone

**716** Effect of PRID<sup>®</sup> administered 5 to 11 days post–insemination on serum progesterone concentrations in lactating dairy cows. S. J. Scott\*, R. B. Walsh, S. J. LeBlanc, J. Woodward, J. S. Walton, and K. E. Leslie, *University of Guelph, Guelph, ON, Canada*.

A high rate of hepatic metabolism could decrease progesterone concentrations important for the maintenance of the developing conceptus, leading to early embryonic loss. The effects of a Progesterone-Releasing Intravaginal Device (PRID<sup>®</sup>) administered at d 5-11 post-insemination, on circulating serum progesterone concentrations was studied in 222 lactating dairy cattle on 4 farms. Cows were randomly assigned to receive PRID or PID (Placebo Intravaginal Device). Blood was collected at both insertion and removal of the device. Serum progesterone concentrations were determined by ELISA analysis (Immulite). At the time of insertion, circulating progesterone levels were not significantly different between cows administered PRID, versus control ( $2.0 \pm 1.42$ ng/mL). There tended to be a treatment by time interaction such that the differences in progesterone concentrations between PRID and control cows was significant only for cows that received the device 9 - 11 d post-insemination (P < 0.08) (Table 1). At d 12 – 18 post-insemination the serum progesterone concentration in cows that became pregnant on the previous service was 4.0 ng/mL and 3.1 ng/mL, in cows that were and were not diagnosed pregnant, respectively. For cows that were diagnosed pregnant the change in progesterone from d 5 - 11 to d 12 - 18 was 1.8  $\pm$  1.6 ng/mL, versus 1.1  $\pm$  1.9 ng/mL in cows that were not diagnosed pregnant (P < 0.005). These results indicate that the administration of PRID approximately 1 wk post-insemination will increase progesterone levels, potentially affecting reproductive success.

## Table 1: Circulating progesterone concentrations at insertion and removal of device.

Days post-AI	n	Mean $P_4$ at insertion (ng/mL)	SD	Mean P <sub>4</sub> at removal (ng/mL)	SD
5 – 8 PID	58	1.6	1.1	3.7	1.9
5 – 8 PRID	75	1.7	1.3	3.5	1.9
9 – 11 PID	48	2.5	1.6	3.1	2.2
9 – 11 PRID	41	2.5	1.5	3.7	2.0

Key Words: PRID, Progesterone, Post-Insemination

**717** Comparison of long-term CIDR-based protocols to synchronize estrus in beef heifers. N. R. Leitman\*, D. C. Busch, D. A. Mallory, D. J. Wilson, M. R. Ellersieck, M. F. Smith, and D. J. Patterson, *University of Missouri, Columbia.* 

Two experiments evaluated modifications to a long-term CIDR-based protocol to synchronize estrus and compared differences among protocols on the basis of their potential to facilitate fixed-time AI in beef heifers. In Exp. 1 estrous cycling beef heifers (n = 85) were assigned to 1 of 2 treatments by age and BW. Heifers in T1 received a CIDR from d 0 to 14, GnRH on d 23, and PG on d 30. Heifers in T2 received a CIDR from d 2 to 16, GnRH on d 23, and PG on d 30. Ovaries were scanned by ultrasonography on d 23 to characterize follicular dynamics and d 25 to determine response to GnRH. In Exp. 2 beef heifers (n = 353) were assigned within reproductive tract scores (2 or 3 = prepubertal; 4 or 5 = estrous cycling) by age and BW to 1 of 4 treatments. Heifers in

T1 and T2 received the same treatments described in Exp. 1. Heifers in T3 and T4 received the same treatments as T1 and T2, respectively, minus the addition of GnRH. All heifers were fitted with HeatWatch transmitters for estrus detection and AI was performed 12 h after estrus onset. In Exp. 1 heifers assigned to T1 had larger dominant follicles at GnRH compared to T2 (P < 0.01; 10.9 vs. 9.5 mm, respectively) but response to GnRH, estrous response after PG, mean interval to estrus and variance for interval to estrus after PG did not differ (P > 0.05). AI conception and final pregnancy rate were similar (P > 0.05). In Exp. 2 estrous response after PG did not differ (P > 0.10). Differences in mean interval to estrus and variance for interval to estrus (P < 0.05) differed based on the 3-way interaction of treatment length, GnRH, and estrous cyclicity status. AI conception and final pregnancy rates were similar (P > 0.05). In summary, the 2 d schedule modification failed to improve synchrony of estrus. In Exp. 1 no differences in estrous response or synchrony of estrus were detected between T1 and T2. In Exp. 2 the 3-way interaction suggests that further evaluation of long-term CIDR-based protocols is required with and without the addition of GnRH to determine the efficacy of these protocols for use in facilitating fixed-time AI. This research was supported by USDA-NRI grant 2005-55203-15750.

Key Words: Beef Heifer, Estrus Synchronization, Progestin

**718** Timing of artificial insemination in beef cows following the CO-Synch + CIDR protocol. D. C. Busch<sup>\*1</sup>, D. J. Schafer<sup>2</sup>, D. J. Wilson<sup>1</sup>, D. A. Mallory<sup>1</sup>, N. R. Leitman<sup>1</sup>, J. K. Haden<sup>2</sup>, M. R. Ellersieck<sup>1</sup>, M. F. Smith<sup>1</sup>, and D. J. Patterson<sup>1</sup>, <sup>1</sup>University of Missouri, Columbia, <sup>2</sup>MFA Inc., Columbia, MO.

The experiment was designed to compare pregnancy rates in postpartum beef cows resulting from fixed-time AI (FTAI) after administration of the CO-Synch + CIDR protocol. Cows (n = 851) at two locations over two years (yr 1; n = 218, 206: yr 2; n = 199, 228) were stratified by age, BCS and days postpartum (DPP) to 1 of 2 FTAI intervals. Cows were administered GnRH (100 µg, i.m.) and equipped with a controlled internal drug release (CIDR) insert (1.38 g progesterone, d 0). CIDR inserts were removed 7 d later at the time PG (25 mg, i.m.) was administered (d 7). Continuous estrus detection was performed at Location 2 using HeatWatch. Transmitters were fitted at the time of PG and removed at the time of AI. Artificial insemination was performed at predetermined fixed-times [54 h (FTAI 54; n = 424) or 66 h (FTAI 66; n = 427) after PG] and all cows were administered GnRH (100 µg, i.m.) at AI. Two blood samples were collected between 8 to 10 d and immediately prior to treatment initiation to determine pre-treatment estrous cyclicity status of cows [progesterone  $\geq$  0.5 ng/mL; (FTAI 54, 288/424, 68%; FTAI 66, 312/427, 73%; P = 0.07]. At Location 2, more cows exhibited estrus prior to FTAI 66 than FTAI 54 (P < 0.01; 107/216, 50% and 57/218, 26%, respectively). Pregnancy rates were higher (P < 0.01) among cows that exhibited estrus than those that did not (123/163, 76% and 150/270, 56%, respectively). There were no treatment by location interactions within year (P > 0.10) for age, DPP, or BCS; thus the results were pooled for the respective treatments. Fixed-time AI pregnancy rates were higher for FTAI 66 than FTAI 54 (P = 0.05; 286/426, 67%) and 257/424, 61 %, respectively). Pregnancy rates resulting from FTAI did not differ between year (P = 0.09), farm (P = 0.80), AI sires (P =0.11) or technicians (P = 0.64). There was no difference between FTAI pregnancy rates based on pre-treatment estrous cyclicity status (P =0.30), and no difference between treatments in final pregnancy rates

(P = 0.77). In summary, fixed-time AI following CO-Synch + CIDR at 66 h resulted in significantly greater FTAI pregnancy rates compared to FTAI at 54 h.

Key Words: Artificial Insemination, Estrus Synchronization

**719** Substitution of estradiol benzoate for GnRH in the Select Synch + CIDR protocol with or without temporary calf removal in *Bos indicus*-influenced cattle. J. D. Pack\*<sup>1,2</sup>, I. C. Velez<sup>1,2</sup>, M. Amstalden<sup>1,2</sup>, and G. L. Williams<sup>1,2</sup>, <sup>1</sup>*Texas AgriLife Research, Beeville, TX*, <sup>2</sup>*Texas A&M University, College Station.* 

The hypothesis was that estradiol benzoate (EB) would be more effective than GnRH in synchronizing new follicular wave emergence (NFWE) and subsequent ovulation in Brahman-influenced cows subjected to a controlled intravaginal drug release insert (CIDR)-based treatment protocol. Sixty-four postpartum (PP) Braford (F-1) cows (79 % cyclic) in 2 replicates (32 cows/replicate) were stratified by parity, d PP and body condition score (BCS), and assigned randomly to a 2 x 2 factorial arrangement of groups that contrasted hormone treatments (GnRH vs EB) at time 0 and post-CIDR suckling status (weaned vs suckled). Treatments were: 1) Select-Synch + CIDR; n=16; 2) Select-Synch + CIDR with 48h calf removal; n=16; 3) E-Synch + CIDR; n=16; or 4) E-Synch + CIDR with 48h calf removal; n=16. All females were a minimum of 50 d PP and had a BCS of at least 5 (1 to 9 scale) at treatment onset. On d 0, cows in all groups received a CIDR, with those in groups 1 and 2 receiving a single injection of 100 µg GnRH i.m., and those in groups 3 and 4 receiving a single injection of 2 mg EB i.m. CIDR inserts were removed on d 7 and all cows received a single injection of prostaglandin F2-alpha (25 mg i.m.), with 48-h calf removal in groups 2 and 4. Transrectal ultrasonography was performed daily for a maximum of 144 h after CIDR removal. Proportions of GnRH- and EB-treated cows exhibiting (75 vs 68.9%) and intervals ( $4.1 \pm 0.3$  vs  $3.8 \pm 0.3$  d) to NFWE did not differ. Proportions of GnRH- and EBtreated cows ovulating (68.8% vs 78.1%) and their respective intervals to ovulation (88.6  $\pm$  6.3 vs 94  $\pm$  6 h) after CIDR removal did not differ, nor did post-CIDR suckling status affect ovulation frequency (77.4 vs 69.7) or interval to ovulation  $(87.3 \pm 6.5 \text{ vs } 95 \pm 5.8 \text{ h})$ . However, ovulation before 72 h was greater (P < 0.07) in Select-Synch + CIDR (4/32) compared to E-Synch + CIDR (0/32). Although minor, the latter effect could contribute to a reduction in asynchronous follicle maturity in TAI protocols.

Key Words: Synchronization, Estradiol Benzoate, Bos indicus

720 Ovarian and fertility responses of Holstein heifers after GnRH, progesterone, and PGF<sub>2 $\alpha$ </sub> at five stages of the estrous cycle. J. S. Stevenson\*, *Kansas State University, Manhattan.* 

Estrus and ovulation were manipulated in 283 heifers by administering GnRH, progesterone (P4), and PGF at 5 stages of the estrous cycle. Estrus was presynchronized with a P4 insert (CIDR) for 7 d before PGF was administered 24 h before insert removal. Successive groups of 10 heifers were assigned to treatments (2 heifers/treatment) beginning on d 2 of the cycle; then groups of treatments on d 5, 10, 15, and 18. Treatments were a CIDR insert placed intravaginally (d 0) for 7 d plus: 1) 25 mg of PGF on d 7 at insert removal (PGF); 2) 100 µg of GnRH on d 0

+ PGF on d 7; 3) PGF 24 h before insert removal (early PGF); and 4) 100 µg of GnRH + early PGF. Controls received no insert (GnRH on d 0 and PGF on d 7). Ovaries were scanned ultrasonically on d 0, 2, 7, 9, and 11 to assess follicular diameters and ovulation. Blood was collected on d 0, 2, 6, 7, 8, and 9 to quantify serum concentrations (ng/mL) of P4. Insemination occurred after detected estrus or by timed AI (TAI) at 64 h after insert removal. Only 15.6% of 141 GnRH-treated heifers ovulated by 48 h after GnRH, with twice as many ovulating when treatment was initiated on d 5 (26%) compared with other cycle days (12.4%). Stage of cycle at treatment influenced serum P4; it was greater in all CIDRtreated heifers on d 2 and 6 of treatment. Progesterone tended (P = 0.08) to be greater in GnRH-treated heifers on d 6. Treatment effects on P4 on d 7 and 9 occurred because of the early decrease in P4 in response to PGF 24 h before insert removal. Follicle diameters at 48 h after insert removal (d 9) were unaffected by treatment, but were less (P < 0.05) in heifers when treatment was initiated on d 5 ( $12.0 \pm 0.4$  mm) compared with other stages of the cycle (13.9  $\pm$  0.3 mm). Pregnancies per AI (P/ AI) were unaffected by stage, but tended (P = 0.10) to be less after TAI (43.5%) than after detected estrus (56.1%) and in control compared with CIDR treatment. Treatments initiated on d 10 of the cycle produced the greatest P/AI (65.4%) compared with other stages (d 2 = 38.9%; d 5 =37.7%; d 15 = 47.4%; or d 18 = 41.5%). Compared with control, more P4-treated heifers ovulated by d 11.

## Table 1.

		Treatment					P <	
Trait	PGF	GnRH	Early PGF	GnRH + early PGF	Control	Trt	Stage	$\mathbf{T}\times\mathbf{S}$
Heifers (n)	47	48	47	45	48			
Ov. by d 2, %	0.0	18.8	0.0	28.9	29.2	0.001	0.06	0.52
P4, d 2	6.1	6.4	6.5	6.4	3.7	0.001	0.001	0.71
P4, d 6	5.2	5.4	4.8	5.6	4.3	0.04	0.001	0.67
P4, d 7	5.1	5.0	2.4	2.3	4.2	0.001	0.001	0.001
P4, d 8	0.9	0.8	0.7	0.7	0.9	0.24	0.001	0.44
P4, d 9	0.6	0.5	0.4	0.5	0.9	0.001	0.01	0.99
Follicle diam. (d 9), mm	13.3	13.3	13.3	14.1	13.6	0.001	0.01	0.33
Ov. by d 11, %	89.4	89.6	91.5	88.9	68.8	0.001	0.01	0.33
P/AI, %	52.7	52.5	43.6	44.6	36.4	0.35	0.31	0.74
(n)	(55)	(59)	(55)	(56)	(55)	0.55	0.51	0.74

Key Words: Progesterone, GnRH, Heifers

**721** Relationship between uterine pH at fixed-time AI and pregnancy success in beef cattle. S. F. Lares\*, S. D. Fields, B. L. Perry, D. G. Chen, and G. A. Perry, *South Dakota State University, Brookings.* 

Research has shown that cows in estrus within 24 h of fixed-time AI had greater pregnancy rates compared to cows not in estrus. Furthermore, uterine pH decreased at the initiation of estrus and pH has been reported to influence sperm motility and longevity. Our objective was to determine the relationship between uterine pH at fixed-time AI and pregnancy success. Lactating beef cows in two herds were synchronized with the CO-Synch protocol [100  $\mu$ g GnRH on d -9; 25 mg PG on d -2; and 100  $\mu$ g GnRH plus timed AI on d 0 (48 h after PG); n = 126] or the CO-Synch+CIDR protocol [100  $\mu$ g GnRH and CIDR insertion on d -10; 25 mg PG and removal of CIDR on d -3; 100  $\mu$ g GnRH with timed AI on d 0 (60 h after PG); n = 97]. Uterine pH was determined

at fixed-time AI (n = 80 and 63 for herd 1 and 2, respectively). Cows were determined to be in standing estrus by activation of an EstroTect estrus detection aid, determined at timed AI and approximately 18 h later. Pregnancy was determined 60 to 70 d after AI. At time of AI, cows that had initiated estrus had decreased (P = 0.01) uterine pH compared to cows not in estrus ( $6.78 \pm 0.03$  and  $6.89 \pm 0.03$ , respectively). In addition, cows that had initiated estrus by 18 h after AI had decreased (P < 0.01) uterine pH at time of AI compared to cows that did not initiate estrus (6.78  $\pm$  0.02 and 6.96  $\pm$  0.04, respectively). Cows that initiated estrus prior to AI had increased (P = 0.05) pregnancy success (52% vs. 38%) compared to cows that had not initiated estrus by AI. Cows that initiated estrus by 18 h after AI tended to have increased (P = 0.078) pregnancy success (43% vs. 32%) compared to cows that did not initiate estrus. Furthermore, uterine pH at AI had an approximately linear effect on pregnancy success within the observed pH range. As uterine pH increased pregnancy success decreased (P = 0.076, logistics regression). In summary, uterine pH at time of AI was decreased in cows that exhibited estrus and uterine pH tended to have a linear effect on pregnancy success.

Key Words: Uterine pH, Pregnancy Rate, Fixed-Time AI

**722** Comparison of pregnancy rates in beef cattle after fixedtime AI using semen processed with two different extenders. D. C. Busch\*<sup>1</sup>, N. R. Leitman<sup>1</sup>, D. A. Mallory<sup>1</sup>, D. J. Wilson<sup>1</sup>, J. F. Bader<sup>2</sup>, J. L. Martin<sup>3</sup>, M. F. Smith<sup>1</sup>, and D. J. Patterson<sup>1</sup>, <sup>1</sup>University of Missouri, Columbia, <sup>2</sup>Merial Limited, Fulton, MO, <sup>3</sup>Accelerated Genetics, Baraboo, WI.

This experiment was designed to compare pregnancy rates in beef heifers and postpartum beef cows after fixed-time AI (FTAI) with semen processed with two different extenders. A single ejaculate of semen was collected from each of 4 bulls. Semen was processed using either a control (C) or Affirm<sup>TM</sup> (A) semen extenders. The semen was frozen and stored in liquid nitrogen. Heifers (n = 53) at one location were stratified by age and BCS and cows (n = 968) at 7 locations were stratified by age, BCS, and days postpartum (DPP) to 1 of 2 treatments involving the two semen extenders (C or A). Heifers were synchronized using the CIDR Select protocol and cows were synchronized using the CO-Synch + CIDR protocol. Fixed-time AI was performed at 72 and 66 h, respectively, after treatment administration for the two age groups of females. There were no interactions involving semen extender X location (P > 0.10) for age, DPP, or BCS; thus the results were pooled for the respective treatments. There was no difference in the resulting FTAI pregnancy rates between heifers and cows inseminated with C (58%) or A (54%, P = 0.20) and there was no difference between sires (P = 0.47); however, there was an interaction involving semen extender X sire (P = 0.02). One sire exhibited a reduction (-18%, P < 0.01) in FTAI pregnancy rates while another exhibited an improvement (+19%, P = 0.10) in FTAI pregnancy rates with A versus C. The other two sires exhibited no differences (P > 0.70) between C and A. Furthermore, there was an effect (P < 0.05) of AI technician, age, BCS, and DPP on FTAI pregnancy rates. Cows  $\ge 7$  yr of age had lower pregnancy rates (P < 0.05) than cows 3 to 6 yr of age but were similar to 2 yr- old cows. Pregnancy rates were higher (P < 0.01) for cows that calved  $\ge 50$  d before FTAI (59%) than those that calved  $\le 50$  d before FTAI (41%). In summary, there was no difference in pregnancy rates resulting from FTAI between the C and A semen extenders; however, there were significant differences involving the interaction of sire X semen extender.

Key Words: Artificial Insemination, Estrus Synchronization, Semen

**723** The effect of an opioid antagonist on dairy cattle fertility after insemination. V. Fuentes- Hernandez\*, A. Bernal-Canseco, P. I. Fuentes-Castro, and R. Orozco-Hernandez, *Universidad de Guadalajara*, *Mexico*.

With the objective of studying the effect of naloxone administered im at the onset on estrus in dairy cattle, and to observe the effect of this opioid antagonist on conception rate after insemination, a small cattle holding with 120 cows was selected. There was no discrimination regarding to age and number of parturitions. A double blind experiment was designed to avoid bias in the results. The veterinarian was instructed to inject the drug using multiple dose flasks with no label. When a cow showed the first signs of estrus, im injections of 5 mL containing either saline or 10 mg naloxone were administered at 12 hr intervals and continued for 24 hr after insemination. Note was taken of conception at first insemination after treatment, and the number of inseminations required for positive pregnancy in cows repeating estrus. The experiment was carried out from January 2007 to January 2008. An accumulated total of 43 cows were treated during this period. It was observed that cows treated with naloxone (n = 21) required  $1.25 \pm 0.25$  inseminations and insemination rate in saline treated was  $2.5 \pm 0.5$ . After an ANOVA if was observed that conception rate in naloxone-treated cows was higher ( $P \le 0.01$ ) compared with controls. It was concluded that naloxone treatment during the estrous period in dairy cattle reduced insemination rate and gives further support to endogenous opioids as important modulators of reproduction in cows.

Key Words: Opioids, Cow, Fertility