## Physiology and Endocrinology: Estrous Cycle Manipulation–Dairy

**175 Ovulatory responses to withdrawal of progesterone feedback during the early and late luteal phase.** G. E. Mann<sup>\*1</sup> and R. S. Robinson<sup>2</sup>, <sup>1</sup>University of Nottingham, School of Biosciences, Division of Animal Sciences, Sutton Bonington Campus, Loughborough, UK, <sup>2</sup>University of Nottingham, School of Veterinary Medecine and Science, Sutton Bonington Campus, Loughborough, UK.

Synchronization protocols aim to optimize timing of ovulation following removal of the inhibitory action of progesterone on the final stages of ovulatory follicle development. This study investigated the effects of blocking progesterone action during different stages of the luteal phase on endocrine and ovulatory responses in cyclic ewes. Ewes (n = 4 pergroup) were treated during the Early (d 3-5) or Late (d 12-14) luteal phase with progesterone antagonist (100 mg i.m.  $2 \times$  daily; onapristone ZK98299, Schering AG) or Control vehicle and slaughtered on d 17 to determine ovarian morphology. Hormone changes were analyzed by ANOVA for repeated measured and ovulation rate by ANOVA. Early treatment resulted in increased (P < 0.01) plasma LH within 6h (Control  $1.8 \pm 0.3$ ; Treated  $5.7 \pm 0.3$  ng/mL), with LH then rising to surge like levels within  $25.5 \pm 5.1$  h (range 12–36 h). Following Late treatment, an initial modest (P < 0.1) increase in LH (Control 0.9 ± 0.1; Treated  $2.0 \pm 0.4$  ng/mL) was followed by surge-like levels  $51.0 \pm 3.9$  h after the start of treatment (range 42-60 h). During Late treatment plasma estradiol (Treated 2.0  $\pm$  0.1, Control 1.2  $\pm$  0.1 pg/mL; P < 0.001) and progesterone (Treated 3.3  $\pm$  0.2, Control 2.1  $\pm$  0.2 ng/mL; P < 0.01) were elevated. In contrast, Early treatment was not associated with any increase in either hormone. In Control ewes ovulation rate was 2.0  $\pm$ 0.0. In the Late group there were  $2.0 \pm 0.0$  mature corpora lutea and  $1.5 \pm 0.3$  accessory corpora lutea (total  $3.5 \pm 0.3$ ) per ewe. In the Early group it was not possible to clearly differentiate between the original and accessory corpora lutea but the total number  $(5.3 \pm 0.5)$  was higher (P < 0.05) than that seen in the Late group. Blocking progesterone feedback during the early luteal phase resulted in a more rapid induction of an LH surge and higher rate of accessory ovulation than treatment later in the luteal phase. This differential response of the endogenous reproductive architecture, presumably reflecting differences in both follicle populations and hypothalamic-pituitary responsiveness at the time of treatment, is often overlooked in the development of ovulatory synchronization programs.

Key Words: progesterone, ovulation, LH surge

**176** Estrus behavior and fertility responses in lactating grazing dairy cows after a timed AI program using estradiol cypionate. M. N. Correa<sup>\*1</sup>, M. E. Lima<sup>1</sup>, C. C. Brauner<sup>1</sup>, A. R. T. Krause<sup>1</sup>, E. G. Xavier<sup>2</sup>, E. Schmitt<sup>1</sup>, A. Schneider<sup>1</sup>, and F. A. B. Del Pino<sup>1</sup>, <sup>1</sup>Universidade Federal de Pelotas, NUPEEC, Pelotas, RS, Brazil, <sup>2</sup>Granjas 4 Irmaos S/A, Rio Grande, RS, Brazil.

The objective of this study was to determine the estrus behavior influence during an estrus synchronization program (Heatsynch) on fertility responses in lactating grazing dairy cows. Four hundred and 70 primiparous (n = 229) and multiparous (n = 241) Holstein cows were enrolled in a artificial synchronization program consisting of a controlled internal drug-releasing (CIDR) containing 1.9 g of progesterone, either a new one or previously used once (7 d), plus 25  $\mu$ g i.m. injection on d 0 (85 + 6 DIM) and all cows were given 25 mg PGF2 $\alpha$  i.m. upon CIDR removal 7 d later (d 7) followed by an injection of 1 mg of estradiol cypionate (ECP) i.m. at d 8. Cows were then observed for signs of estrus

for at least 2 h (twice daily at 7:00 and 18:00) for 3 d following CIDR removal. Those cows detected in estrus were inseminated 12 h later. Based on the detection of estrus, cows were divided into 3 groups: estrus behavior before TAI (Estrus AI) and inseminated 48-60 h after CIDR insert removal (n = 143); estrus behavior at the TAI (Estrus TAI) (n =127); and no estrus detection and timed inseminated (TAI) 72 h after CIDR removal (n = 200). A single technician was responsible for the estrus detection and inseminations. Pregnancy status was determined by ultrasonography at 30 to 35 d and 60 to 65 d after AI. Data were analyzed by the LOGISTIC procedure of SAS. Estrus behavior had effect on fertility responses (P < 0.01), where both groups that showed estrus (Estrus AI and Estrus TAI) had greater pregnancy rates than the TAI group at 30-35 d (45.5 vs 46.5 vs 28.5), and 60-65 d (36.4 vs 35.4 vs 15.5). Pregnancy losses also were affected (P < 0.01) by the estrus behavior (20.0 vs 23.7 vs 45.6), respectively for Estrus AI, Estrus TAI and TAI groups. There was a correlation (P = 0.02) among fertility responses and parity, where primiparous cows had better results than multiparous cows. Estrus behavior was similar (P > 0.05) among primiparous and multiparous cows. Estrus behavior during a Heatsynch program is beneficial to improve reproductive performance in lactating grazing dairy cows.

Key Words: estradiol cypionate (ECP), timed AI, pregnancy

**177** Effect of reusing CIDRs on estrus behavior and fertility responses in a Heatsynch protocol of grazing dairy cows. C. C. Brauner<sup>\*1</sup>, M. E. Lima<sup>1</sup>, A. R. T. Krause<sup>1</sup>, E. G. Xavier<sup>2</sup>, A. Schneider<sup>1</sup>, E. Schmitt<sup>1</sup>, F. A. B. Del Pino<sup>1</sup>, and M. N. Correa<sup>1</sup>, <sup>1</sup>Universidade Federal de Pelotas, NUPEEC, Pelotas, RS, Brazil, <sup>2</sup>Granjas 4 Irmaos S/A, Rio Grande, RS, Brazil.

The objective of this study was to determine the effect of reusing intravaginal progesterone inserts (CIDR) on estrus behavior and fertility responses of grazing dairy cows in a Heatsynch protocol. Three hundred and 90 5 primiparous (n = 183) and multiparous (n = 212) Holstein cows were enrolled in a Heatsynch program consisting of a CIDR containing 1.9 g of progesterone, either a new one or previously used once (7d use), plus 25  $\mu$ g GnRH i.m. injection on d 0 (82 + 6 DIM) and all cows were given 25 mg PGF2 $\alpha$  Å ± i.m. upon CIDR removal 7 d later (d 7) followed by an injection of 1 mg of estradiol cypionate (ECP) i.m. at d 8. Used CIDRs had been thoroughly rinsed with a mild disinfectant solution, airdried, and stored in a dry, enclosed container after first use. Cows were then observed for signs of estrus (twice daily at 7:00 and 18:00) for 3 d following CIDR removal. Cows observed in estrus were inseminated 12 h later. Based on the detection of estrus, cows were divided into 3 groups: estrus behavior before TAI (Estrus AI) and inseminated 48-60 h after CIDR insert removal (n = 115); estrus at the TAI (Estrus TAI) (n = 98); and no estrus detection and timed inseminated (TAI) 72 h after CIDR removal (n = 152). A single technician was responsible for the estrus detection and inseminations. Pregnancy status was determined by ultrasonography at 30 to 35 d and 60 to 65 d after AI. Data were analyzed by the LOGISTIC procedure of SAS. There were no difference (P > 0.05) among CIDR uses on pregnancy at 30–35 d (38.6% vs 40.1%); and 60-65 d (24.7% vs 30.0%) and pregnancy losses (36.1% vs 25.3%), respectively for CIDR use 1 and 2. We could not find any difference (P > 0.05) of estrus behavior and CIDR uses, as well as across cows parity. However, percentage of pregnancy losses in the primiparous group was great (P = 0.04) for the first CIDR use than the second one (42.3 vs 21.1). In conclusion, estrus behavior and fertility responses in a heatsynch protocol of grazing dairy cows are not affected by CIDR uses. Primiparous cows seems to have greater pregnancy losses (up to 65 d) when first CIDR was used.

Key Words: estradiol cypionate (ECP), timed AI, reproduction

**178** Effect of intrauterine administration of GnRH on LH secretion in lactating dairy cows. S. Bas\*<sup>1</sup>, M. L. Day<sup>2</sup>, and G. M. Schuenemann<sup>1</sup>, <sup>1</sup>Department of Veterinary Preventive Medicine, The Ohio State University, Columbus, <sup>2</sup>Department of Animal Sciences, The Ohio State University, Columbus.

The objective of this study was to evaluate the release of LH following the intrauterine (i.u.) administration of GnRH (Gonadorelin) in lactating dairy cattle. Cows (n = 22) were presynchronized with 2 injections of PGF2 $\alpha$  given 14 d apart (starting at 26 ± 3 DIM) followed by Ovsynch [OV; d –9 GnRH, d –2 PGF2 $\alpha$ , d 0 GnRH (h 0)] 12 d later. Ovarian structures were recorded and blood samples were collected on d –9 and d –2. Only cows presenting a CL ≥ 15 mm and at least one follicle ≥ 10 mm in diameter at the initial GnRH were used in the study (n = 13). At

the time of the second GnRH of OV (h 0), cows were blocked by parity and randomly assigned to 1 of 3 groups: 1) control group (CON; n =4) received 2 mL i.m. of sterile water, 2) intramuscular group (IM; n = 4) received 100  $\mu$ g i.m. of GnRH, and 3) intrauterine group (IU; n = 5) received 200 µg i.u. of GnRH with the addition of glycerol (7% v/v). Blood samples for determination of LH serum concentrations were collected at h -0.5, 0, 0.5, 1, 1.5, 2, 3, 4 and 5. Ovulation was determined 48 h after the second GnRH of OV (d 0) by transrectal ultrasonography. Progesterone and LH data were analyzed using mixed procedures of SAS. Serum progesterone concentrations at h 0 did not differ (P >0.05) between groups. The area under the curve for LH was greater for (P < 0.05) IM cows compared with IU or CON cows. Most of the cows ovulated by 48 h relative to d 0 in IM (4/4) and IU (3/5) groups compared with CON cows (0/4). Although IU administration of GnRH resulted in lower serum concentrations of LH than IM group, more IU cows ovulated by h 48 compared with CON. These findings support our previous findings, and further, suggests that i.u. administration may be an alternative route of delivery for GnRH to synchronize ovulation in estrous synchronization programs.

Key Words: intrauterine GnRH, LH, dairy cow