

Physiology and Endocrinology: Estrous Cycle Manipulation - Dairy

101 Ovarian follicular development, luteal function, and fertility in lactating Holstein cows treated with 14dCIDR_PGF or 2xPGF_Ovsynch56 for first insemination timed AI (TAI). R. C. Escalante*, S. E. Pooock, D. J. Mathew, W. R. Martin, E. M. Newsom, J. L. Denbigh, E. C. Adkins, and M. C. Lucy, *University of Missouri-Columbia, Columbia*.

Progesterone-containing devices (CIDR) inserted for 14 d can be used to presynchronize the estrous cycle in heifers before TAI ("14-day CIDR-PG" program; <http://beefrepro.unl.edu>). The objective was to test a similar program for lactating dairy cows. Holstein cows ($n = 71$; 35 to 60 d postpartum) were assigned to 2xPGF_Ovsynch56 (Control program; PGF_{2 α} , 14 d, PGF_{2 α} , 12 d, GnRH, 7 d, PGF_{2 α} , 56 h, GnRH, 16 h, TAI; $n = 34$) or 14dCIDR_PGF (14dCIDR_PGF program; CIDR in, 14 d, CIDR out; 19 d; PGF_{2 α} , 56 h, GnRH, 16 h, TAI; $n = 37$) that began on d 0 and ended on d 36 with TAI. Ultrasound exams and blood sample collections were performed on d 0, 14, 19, 26, 28, 33, and 35 and blood sampling alone was done on d 42 to monitor follicular development, ovulation and the corpus luteum. After the presynchronization step (CIDR or 2xPGF; d 0 to 14), the percentages of cows observed in estrus (47%) and having ovulation within 5 d (73%) were similar for 14dCIDR_PGF and control. The interval to estrus after CIDR removal/PGF injection (d 14), however, was less (50 ± 5 and 75 ± 5 h; $P < 0.001$) and the size of the largest follicle (LF) at CIDR removal/PGF_{2 α} injection (d 14) was greater (20.4 ± 0.8 and $16. \pm 8.8$ mm; $P < 0.01$) for 14dCIDR_PGF compared with control. Plasma progesterone concentrations (P4) increased ($P < 0.001$) after the presynchronization step from 0.9 ± 0.3 (d 19) to 7.0 ± 0.3 (d 33) ng/mL. There was a tendency ($P < 0.10$) for 14dCIDR_PGF cows to have greater P4 than control from d 26 to d 33 (6.6 ± 0.6 and 5.2 ± 0.6 ng/mL for 14dCIDR_PGF and control, respectively). Diameters of the LF at PGF_{2 α} (d 33; 15.6 ± 0.7 mm) and before the GnRH injection (d 35; 17.0 ± 0.6 mm) were similar for 14dCIDR_PGF and control. Percentages of cows ovulating (89%) and becoming pregnant after TAI (48%) were similar for 14dCIDR_PGF and control. Conclusions were that the 14dCIDR_PGF program was effective for synchronizing lactating dairy cows for TAI. Greater P4 during the luteal phase before TAI (theoretically advantageous to fertility) in 14dCIDR_PGF may be explained by the ovulation of a persistent follicle after CIDR withdrawal.

Key words: estrous synchronization, dairy, cow

102 Prostaglandin F_{2 α} and GnRH administration increase progesterone, luteal number, and proportion of dairy cows with corpora lutea before a timed AI program. J. S. Stevenson*, S. L. Pulley, and H. I. Mellieon Jr., *Kansas State University, Manhattan*.

The objective was to increase the proportion of cows having a functional corpus luteum (CL) and elevated progesterone (P4) at the onset of the Ovsynch protocol. Postpartum Holsteins in 1 herd were stratified by parity at calving (Sep. 2009 to Oct. 2010) and assigned randomly to 2 treatments: 1) PRE ($n = 134$): 2 25-mg injections of PGF_{2 α} (PG) 14 d apart (Presynch); and 2) PG3 ($n = 134$) one 25-mg injection of PG 3 d before 100 μ g GnRH (PreGnRH) with the PG injection administered at the same time as the second PG in the PRE treatment (10 d before Ovsynch). Cows were enrolled in the Ovsynch protocol (injection of GnRH 7 d before [GnRH-1] and 56 h after [GnRH-2] PG with AI 16 to 18 h after GnRH-2) 10 d after the last or only PG injection. Blood samples for P4 analysis (103 cows per treatment) were collected at d -34, -31, -20, -17, -10, -3, 0 (GnRH-2), and d 1.

Ovarian structures were measured by ultrasonography on d -17, -10, -3, 0, and 7 to determine ovulation and follicle diameters. Although P4 concentration did not differ between treatments before PreGnRH injection, number of CL per cow and proportion of cows having at least 1 CL were greater for PG3 than PRE cows, and more cows ovulated after PreGnRH than ovulated spontaneously in PRE. At GnRH-1, P4 concentration, number of CL per cow, and proportion of cows with at least 1 CL were greater for PG3 than PRE. Neither follicle diameter nor percentage of cows ovulating after GnRH-1 differed between treatments. At PG injection during the week of AI, P4 concentration tended to be greater for PG3 than PRE and PG3 had more CL per cow than PRE. Pregnancy rates per AI at d 32 for PG3 vs. PRE cows were 58.2 vs. 50.0% for 103 and 106 cows inseminated during nonsummer months ($P = 0.28$) and 7.7 vs. 8.8% for 39 and 34 cows inseminated during summer ($P = 0.28$), respectively. We concluded that the PreG treatment effectively increased ovulation and luteal function 7 d before the onset of Ovsynch resulting in improved follicular synchrony and potentially predisposing improved pregnancy rates per AI in lactating dairy cows.

Table 1.

Item	PG3	PRE	P-value
Energy-corrected milk at AI, kg	48.1 \pm 1.3	47.2 \pm 1.2	
PreGnRH			
CL per cow, no.	0.6 \pm 0.08	0.2 \pm 0.08	0.002
Cows with CL, %	49.5	35.9	0.007
Ovulation, %	79.6	50.1	0.002
Follicle size, mm	12.6 \pm 0.6	14.3 \pm 0.5	0.041
GnRH-1			
P4, ng/mL	3.5 \pm 0.3	2.5 \pm 0.3	0.018
CL per cow, no.	1.5 \pm 0.1	0.8 \pm 0.1	0.001
Cows with CL, %	94.2	76.7	0.001
Breeding week PG			
P4, ng/mL	5.0 \pm 0.4	4.1 \pm 0.4	0.130
CL per cow, no.	2.1 \pm 0.1	1.6 \pm 0.1	0.001

Key words: ovulation, luteal function, presynchronization

103 Evaluation of LH release after the intrauterine administration of gnRH in lactating dairy cattle. S. Bas*, C. G. Pinto, M. L. Day, and G. M. Schuenemann, *The Ohio State University, Columbus*.

The purpose of this study was to determine the preovulatory release of LH and ovulatory response after the intrauterine (i.u.) administration of GnRH (Gonadorelin) in lactating dairy cattle. Lactating cows ($n = 23$) were presynchronized with 2 injections of PGF_{2 α} given 14 d apart (starting at 26 ± 3 DIM) followed by Ovsynch (OV; GnRH-7 d-PGF_{2 α} -56 h-GnRH 16 h-timed-AI; TAI) 12 d later. Ovarian structures were recorded and a blood sample collected for each animal at the time of first GnRH of OV. Only those cows presenting a CL ≥ 15 mm and at least one follicle ≥ 10 mm in diameter remained in the study. Additionally, blood samples were collected and ovarian structures recorded at the time of PGF_{2 α} of OV. 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; $n = 7$) received 2 mL, i.m., of sterile water, 2) intramuscular group (IM; $n = 8$) received 100 μ g, i.m., of GnRH, and 3) intrauterine group (IU; $n = 8$) received 100 μ g, i.u.,

of GnRH. Blood samples for determination of LH serum concentrations were collected at h 0, 0.5, 1, 1.5, 2, 3 and 4. Furthermore, ultrasonography was performed twice daily (12 h interval) from h 0 to 60 for determination of ovulation. Serum progesterone concentrations at h 0 did not differ ($P > 0.05$) between groups. Concentrations of LH were greater ($P < 0.05$) in the IM than IU and CON groups at h 0, 0.5, 1, 1.5, 2, and 4 h but not at h 3 between the IM and IU group. Cows in IU started increasing LH concentrations at 1 h reaching maximum levels at 2–3 h post GnRH while LH concentrations did not increase during this period in CON cows. The proportion of cows that ovulated by h 60 was greater ($P < 0.05$) for the IM (8/8) and IU (7/8) groups as compared with CON cows (2/7). Administration of GnRH i.u. resulted in lower serum concentrations of LH than in the IM group, but the proportion of cows that ovulated by h 60 did not differ between treatments. In summary, these findings provide evidence that i.u. administration may be an alternative route of delivery for treatment with GnRH to synchronize ovulation in estrous synchronization programs.

Key words: dairy cow, intrauterine GnRH, ovulation

104 Effect of presynchronization strategy prior to ovsynch on fertility at first service in lactating dairy cows. A. Keskin¹, G. Yilmazbas-Mecitoglu^{*1}, E. Karakaya¹, A. Alkan², H. Okut³, A. Gumen², and M. C. Wiltbank⁴, ¹Department of Obstetrics and Gynecology, Faculty of Veterinary Medicine, University of Uludag, Bursa, Turkey, ²Tarfaz Company, Bursa, Turkey, ³Biometry and Genetics, Faculty of Agriculture, University of Yuzuncu Yil, Van, Turkey, ⁴Department of Dairy Science, University of Wisconsin-Madison, Madison.

The aim of this study was to evaluate the effect of presynchronization with or without detection of estrus on first service Pregnancy per Artificial Insemination (P/AI) and Ovsynch outcome in lactating dairy cows. A total of 511 cows were divided randomly but unevenly into 3 treatment groups at 44–50 d in milk (DIM). Ovsynch was started at the same time in all 3 groups (69–75 DIM). Cows in the Ovsynch group ($n = 126$) received no presynchronization before Ovsynch and all cows were bred by TAI. Cows in Presynch with estrus detection (PED) and Presynch with only TAI (PTAI) groups received 2 doses of PGF_{2α} 14 d apart starting at 44–50 -DIM- and Ovsynch was initiated 11 d after the last PGF_{2α} treatment. Cows in PED ($n = 267$) received AI if estrus was detected after either PGF_{2α} injection and cows that were not detected in estrus after PGF_{2α} received Ovsynch and TAI. Cows in PTAI ($n = 118$) were not inseminated to estrus with all cows receiving TAI after Ovsynch. Ovulatory response to the first GnRH of Ovsynch was different ($P = 0.002$) among treatment groups (83.1% in PTAI, 72.6% in PED, and 62.7% in CON). However, ovulatory response to the second GnRH of Ovsynch did not differ among treatments. A total 132 of 267 PED cows (49.4%) exhibited estrus and were inseminated. The P/AI at the 31 d pregnancy diagnosis was not different between cows with AI after estrus (37.8%; 50/132) or Ovsynch (34.1%; 46/135) in the PED group. The P/AI for the Ovsynch group (46.8%; 59/126) was greater than the PED group ($P < 0.05$) with Ovsynch greater ($P = 0.04$) than PED cows receiving TAI but not than PED cows bred to estrus ($P = 0.16$). The cows in PTAI had greater P/AI (55.9%; 66/118) at the 31 d pregnancy diagnosis than PED ($P < 0.01$; either estrus or TAI) and tended to be greater ($P = 0.08$) than Ovsynch. Thus presynchronization with PGF_{2α} (PTAI) increased ovulatory response to Ovsynch and improved P/AI in dairy cows. Interestingly, breeding of cows to estrus during Presynch reduced fertility to the TAI. These results indicate that maximal fertility is obtained when all cows receive TAI after the Presynch protocol.

Key words: dairy cow, presynchronization, Ovsynch

105 Effects of presynchronization (PRE) and length of proestrus (LP) on pregnancy per AI (P/AI) of grazing dairy cows subjected to the 5d-Cosynch protocol. E. S. Ribeiro^{*}, A. P. A. Monteiro, F. S. Lima, R. S. Bisinotto, H. Ayres, L. F. Greco, M. Favoreto, R. S. Marsola, W. W. Thatcher, and J. E. P. Santos, *University of Florida, Gainesville.*

Objectives were to compare the effects of method of PRE and LP on fertility of grazing dairy cows subjected to the 5d-Cosynch protocol. Lactating cows ($n = 1,754$) were blocked by breed, parity, and d postpartum, and randomly assigned to 1 of 2 PRE protocols: Presynch, 2 injections of PGF given 14 d apart, on study d -24 and -10, and starting the timed AI protocol (TAI) 10 d later; or Double-Ovsynch (DO), study d -17 GnRH, d -10 PGF, d -7 GnRH, and starting the TAI protocol 7 d later. The TAI protocol consisted of GnRH on d 0, PGF on d 5 and 6, and GnRH+AI either at 58h (COS58) or 72h (COS72) after the d5 PGF. Ovaries were scanned twice before enrollment in the study. Blood was sampled and analyzed for estradiol on the day of AI. The P/AI was determined 30 and 65 d after TAI. Data were analyzed using PROC GLIMMIX. Presynch increased ($P = 0.02$) estrus at AI compared with DO (25.9 vs. 20.8%), but it did not affect estradiol concentration at AI (6.0 vs. 7.1 pg/mL), or P/AI on d 30 (59.1 vs. 56.8%, $P = 0.39$) and 65 (51.2 vs. 51.7%, $P = 0.30$) after insemination. The COS72 increased estrus (28.5 vs. 10.8%, $P < 0.01$) and estradiol concentration at AI (7.2 vs. 5.8 pg/mL, $P = 0.04$) compared with COS58. The LP did not affect P/AI on d 30 (58.7 vs. 56.1%, $P = 0.20$), but COS72 was superior ($P = 0.04$) than COS58 on d 65 (52.8 vs. 48.1%). This difference was caused by a tendency ($P = 0.08$) for interaction between PRE and LP, in which COS58 resulted in smaller P/AI in Presynch than DO cows (43.9 vs. 52.4%), whereas COS72 resulted in greater P/AI in Presynch than DO cows (54.2 vs. 51.4%). Pregnancy loss was greater for Presynch than DO (12.7 vs. 8.3; $P = 0.01$) and for COS58 than COS72 (13.5 vs. 9.4%; $P = 0.03$). Anovular cows had smaller ($P < 0.01$) P/AI than cyclic cows on d 30 (35.4 vs. 61.6%) and 65 (30.4 vs. 54.8%), but no interaction ($P > 0.50$) between cyclic status and either PRE or LP treatments were detected. Presynch and DO resulted in similar fertility, but extending the LP to 72h improved fertility in the 5-d Cosynch protocol, primarily in cows receiving Presynch.

Key words: cow, presynchronization, proestrus

106 Two- and three-wave estrous cycles in dairy cows, investigated with a mechanistic mathematical model. M. Boer^{*1,3}, S. Röblitz², C. Stötzel², R. Veerkamp¹, B. Kemp³, and H. Woelders¹, ¹Animal Breeding and Genomics Centre, Wageningen UR Livestock Research, Lelystad, the Netherlands, ²Computational Systems Biology Group, Zuse Institute Berlin, Berlin, Germany, ³Adaptation Physiology Group, Department of Animal Sciences, Wageningen University, Wageningen, the Netherlands.

A normal bovine estrous cycle contains 2 or 3 waves of follicle development. However, it is unknown how the number of waves per cycle is regulated. Some studies report a better fertility of 3-wave cycles compared with 2-wave cycles, suggesting that older and larger ovulatory follicles in cycles with 2 waves contain oocytes of less quality than cycles with 3 waves. A better understanding of the endocrine mechanisms regulating follicle development is important to obtain more precise control of the estrous cycle, which can help to improve pregnancy rates. Our aim was to investigate which mechanisms are likely can-

didates for regulation of the number of waves. A mechanistic mathematical model that describes the dynamics of the bovine estrous cycle, using a set of 13 linked differential equations with 57 parameters, was developed. The model includes the processes of follicle and corpus luteum development and the working of key hormones that interact to control these processes. In the bovine, the follicle that is dominant at the moment of corpus luteum regression ovulates. Therefore, it was hypothesized that the number of follicle waves in a cycle is determined by follicle growth rate and time point of corpus luteum regression. Ten parameters related to these mechanisms were tested, of which 6 had an effect on the number of waves. The model output changed from 3 to 2 waves in a cycle when the effect of follicle stimulating hormone or of progesterone on follicle growth was changed, or when the time course

of the luteal phase was changed. In the simulations, 2-wave cycles had a shorter cycle length compared with 3-wave cycles. Depending on the parameterization, the model simulated repeated as well as alternated cycles with 2 or 3 waves. Intermediate values of parameters related to follicle growth rate frequently resulted in irregularities, while gradual shifts in the length of the luteal phase often still resulted in a regular cycle. Therefore, these simulation results suggested that a cycle has a 'default' number of waves based on follicle growth rate, which can be influenced by the time point of corpus luteum regression.

Key words: bovine estrous cycle, follicle waves, mathematical model