224 Progesterone supplementation to dairy cows lacking a corpus luteum (CL) at the initiation of the Ovsynch protocol. R. S. Bisinotto*, N. Martinez, F. S. Lima, T. L. C. Pinto, R. S. Surjus, G. C. Gomes, E. S. Ribeiro, L. F. Greco, W. W. Thatcher, and J. E. P. Santos, *University of Florida, Gainesville.*

Objectives were to evaluate the effects of supplemental progesterone (P4) on fertility responses of Holstein cows lacking a CL at the initiation of the Ovsynch-56 program (d-10 GnRH, d-3 PGF_{2a}, h-16 GnRH, d0 AI). Cows had their ovaries evaluated by ultrasonography on d-10 and those without CL were assigned randomly to receive 0 (Control; n = 270) or 2 controlled internal drug-release (CIDR) inserts containing P4 from d-10 to d-3 (2CIDR; n = 261). Cows with CL on d-10 were used as positive controls (Diestrus; n = 756). Cows had their ovaries scanned on d-3 to detect newly formed CL. Blood was sampled on d-10, -9, -7, -5, -3, and 0 and P4 concentrations determined by RIA. Estrus was detected based on removal of tail chalk. Pregnancy was evaluated 32 and 60 d after AI. The LOGISTIC and GLIMMIX procedures of SAS were used to analyze binomial and continuous responses. The use of 2 CIDR inserts increased (P < 0.001) P4 concentrations between d-9 and -3 compared with Control (2.57 vs. 0.75 ng/mL), but concentrations were less than those of Diestrus (4.45 ng/mL). Ovulation to the first GnRH was greater (P < 0.001) for Control and 2CIDR compared with Diestrus (61.9, 57.1, and 35.9%, respectively), which resulted in a greater (P < 0.001) proportion of cows bearing a new CL on d-3 (71.5, 65.9, and 39.0%, respectively). Nonetheless, a greater proportion of Diestrus cows had CL on d-3 compared with Control and 2CIDR (89.6, 71.5, and 65.9%, respectively). Pregnancy per AI was less for Control than for Diestrus and intermediate for 2CIDR cows (Table 1). However, this trend was observed only in cows that were not in estrus at AI.

| Table 1. Fertility responses (% | %; no.) | to treatment |
|---------------------------------|---------|--------------|
|---------------------------------|---------|--------------|

| | Treatment | | | |
|---------------------|---------------------------|-------------------------|---------------------------|---------|
| | Control | 2CIDR | Diestrus | P-value |
| Estrus at AI | 59.3 (270) | 66.7 (261) | 55.8 (756) | 0.70 |
| Pregnant d 32 | | | | |
| Overall | 26.7 (270) ^{b,B} | 31.8 (261) ^A | 33.6 (756) ^a | 0.002 |
| Estrus at AI | 35.0 (160) | 36.2 (174) | 35.8 (422) | 0.93 |
| Not in estrus at AI | 14.6 (110) ^b | 23.0 (87) ^a | 30.8 (334) ^a | 0.001 |
| Pregnant d 60 | | | | |
| Overall | 23.0 (270) ^b | 27.7 (260) ^b | 31.5 (752) ^a | 0.002 |
| Estrus at AI | 29.4 (160) | 32.2 (174) | 33.4 (419) | 0.62 |
| Not in estrus at AI | 13.6 (110) ^b | 18.6 (86) ^B | 29.1 (333) ^{a,A} | 0.002 |
| Pregnancy loss | 13.9 (72) ^a | 12.2 (82) ^a | 5.2 (250) ^b | 0.03 |

Key Words: anestrus, progesterone, dairy cow

225 Resumption of postpartum ovarian cyclicity in dairy cows and its relationship with acute phase proteins, uterine health and lipolysis during the transition period. C. C. Brauner^{*1}, A. R. T. Krause¹, M. E. Lima¹, E. G. Xavier², A. Schneider¹, E. Schmitt³, E. Schwegler¹, M. M. Weschenfelder¹, P. Montagner¹, F. A. B. Del Pino¹, M. N. Corrêa¹, and L. F. M. Pfeifer³, ¹Universidade Federal de Pelotas, NUPEEC, Pelotas, RS, Brazil, ²Granjas 4 Irmãos S/A, Rio Grande, RS, Brazil, ³Empresa Brasileira de Pesquisa Agropecuária EMBRAPA, Porto Velho, RO, Brazil.

The aim of this study was to evaluate the resumption of postpartum cyclicity and its relationship with acute phase proteins, uterine health and lipolysis during the transition period of dairy cows. Twenty multiparous Holstein cows were enrolled in this study from a commercial dairy farm

with annual rolling herd average of $7,891 \pm 1.18$ kg of milk. To assess the cyclicity resumption, blood samples were collected weekly from 16 to 44 d during the postpartum period to evaluate the concentration of progesterone. Cows were classified as either ovulatory (OC group), consisting of cows that ovulated up to $44 \pm 2 d (n = 12)$ or anovulatory (AC group), those cows that did not ovulate in the same period (n = 8). Blood samples were collected weekly from day -21 relative to calving to 30 d postpartum aiming to evaluate the concentration of acute phase proteins (haptoglobin, paraoxonase and albumin), as well as, glucose and NEFA. Endometrial cytology was performed at 37 ± 2 postpartum days, to assess the uterus health considering the polymorphonuclear (PMN) cells count, using uterine low volume flushing. Data were analyzed by MIXED PROCEDURES of SAS. The OC had lower (P = 0.05) PMN percentage in endometrial cytology than AC with 26.3% vs. 53.4% PMN cells in the uterine flushing, respectively. The AC had lower (P = 0.03) concentrations of albumin during the prepartum (2.42 ± 0.07 vs. 2.64 \pm 0.05 g/dL) and postpartum period (2.22 \pm 0.09 vs. 2.52 \pm 0.07 g/dL P = 0.01), and higher (P = 0.04) concentrations of haptoglobin during the prepartun (0.69 ± 0.16 vs. 0.23 ± 0.13 g/L) and tended (P = 0.09) to have lower activity of paraoxonase during the postpartum period (87.81 \pm 9.11 U/L vs. 108.41 \pm 7.45 U/L) than the OC group. No differences (P > 0.05) were observed of NEFA and glucose concentrations during the transition period. In conclusion, cows that return earlier to estrous cyclicity have reduced concentrations of haptoglobin and proportion of PMN in endometrium, and increased concentrations of albumin, but no differences in blood NEFA were observed between OC and AC.

Key Words: inflammation, ovulation, NEFA

226 A missense mutation in growth differentiation factor-9 (GDF9) increases ovulation rate in sheep. M. P. Mullen* and J. P. Hanrahan, *Animal and Grassland Research and Innovation Centre, Teagasc, Athenry, Co. Galway, Ireland.*

The Finnish Landrace is a well-known high prolificacy sheep breed and has been used in many countries to increase fecundity of local breeds. All of the evidence adduced to date suggests that mutations with a large effect on ovulation rate are not responsible for the exceptional prolificacy of Finnsheep. The objectives of this study were 1) to ascertain if any of the 10 established mutations with large effects on ovulation rate in sheep or 2) if any other DNA sequence variants within the candidate genes GDF9 and BMP15, are implicated in the high prolificacy of the Finnish Landrace breed using material from lines developed by divergent selection on ovulation rate. Genotyping results showed that none of 10 known mutations, FecX^G, FecX^B, FecG^H, FecB^B, FecX^I, FecX^H, FecX^L, FecT^T, FecG^E or FecX^R, were present in the set of 108 Finnsheep tested and thus do not contribute to the exceptional prolificacy of Finnsheep. However, DNA sequence analysis of GDF9 identified a previously known mutation, V371M, segregating at significantly different frequencies between high and low ovulation rate lines. Subsequently analysis of Belclare sheep revealed a significant association V371M and ovulation rate (P < 0.001). Heterozygous carriers of V371M, a missense mutation in GDF9, exhibited increased ovulation rate (0.24 s.e. 0.084) relative to the wild type. This finding brings to 11 the number of mutations that exhibit large effects on ovulation rate in sheep and to 3, including FecB^B and FecG^E, the number of known mutations within the TGF β superfamily with a positive effect on prolificacy in the homozygous state without any associated sterile phenotype. These results further highlight the central role of members of the TGFB superfamily in the control of fertility in mammals.

Key Words: fecundity, growth differentiation factor-9 (GDF9), control of fertility