

mentation of AB or YC, but slightly decreased in AB added treatments. Supplemental of YC increased the population of yeast and AB was also helpful to increase the population of yeast in feces ( $P < 0.05$ ). Fecal pH was lower in AB added treatments than other treatments ( $P < 0.05$ ). The AB added treatments also had a lower ammonia concentration than other treatments in an artificial slurry ( $P < 0.05$ ). The more ammonia reduction was observed when AB and YC were supplemented together ( $P < 0.05$ ). These results indicated that supplement of AB and YC could increase the population of yeast and decrease pH in feces. They were also helpful to reduce the ammonia emission from slurry.

#### Effect of supplemental mixed yeast culture and antibiotics on fecal characteristics of weaned pigs

	0%		0.2%		0.1%		0.2%		SEM	Effect
AB										
YC	0%	0.1%	0.2%	0%	0.1%	0.2%				
Microflora (CFU/g of feces)										
Coliform	11.6	11.2	10.3	9.7	10.1	10.1	0.2			
Yeast	6.9 <sup>b</sup>	7.2 <sup>b</sup>	7.6 <sup>b</sup>	7.1 <sup>b</sup>	8.3 <sup>a</sup>	8.3 <sup>a</sup>	0.2			AB, YC
Fecal pH	8.3 <sup>a</sup>	8.2 <sup>a</sup>	8.2 <sup>a</sup>	7.6 <sup>b</sup>	8.0 <sup>ab</sup>	7.8 <sup>ab</sup>	0.1			AB
Ammonia concentration (mM/g of artificial slurry)										
0 h	21.3 <sup>a</sup>	21.1 <sup>ab</sup>	19.8 <sup>ab</sup>	19.5 <sup>ab</sup>	19.4 <sup>ab</sup>	16.1 <sup>b</sup>	0.7			AB
8 h	70.2 <sup>a</sup>	58.8 <sup>b</sup>	59.1 <sup>b</sup>	58.1 <sup>b</sup>	35.0 <sup>c</sup>	56.8 <sup>b</sup>	3.0			AB, YC, AB × YC
24 h	91.1 <sup>a</sup>	67.5 <sup>b</sup>	70.6 <sup>b</sup>	79.2 <sup>ab</sup>	47.0 <sup>c</sup>	66.6 <sup>b</sup>	4.0			AB, YC

<sup>a,b</sup> $P < 0.05$

**Key Words:** Yeast Culture, Feces, Weaned Pig

**M111 Evaluation of yeast culture concentrates in weaning pig diets.** A. Balfagon<sup>\*1</sup>, M. D. Lindemann<sup>1</sup>, G. L. Cromwell<sup>1</sup>, and G. Keller<sup>2</sup>, <sup>1</sup>University of Kentucky, Lexington, <sup>2</sup>Varied Industries Corporation (Vi-Cor), Mason City, IA.

Two experiments involving a total of 78 crossbred weaning pigs were conducted to compare two yeast culture concentrates (YCC) in nursery diets. A basal diet was formulated in both Phase I (Wk 1 & 2) and Phase II (Wk 3 & 4) to which the products (YCC-1: Diamond V XP<sup>TM</sup>, Diamond V Mills Inc., Cedar Rapids, IA and YCC-2: A - Max<sup>TM</sup> concentrate, Varied Industries Corp., Mason City, IA) were included at 1%. In Exp. 1, six pens of pigs (21 d of age and 7.6 kg BW) were used in a preference test. The feeder location was switched three times each week to obviate behavioral or pen location effects on feed selection. The ratio of diets consumed during each phase and for the total experiment was calculated and analyzed using an unpaired t-test with Welch's correction procedure. While there was a slight numerical favor for the YCC-2 during Phase I (53.0% vs. 47.0%, respectively) and for the YCC-1 during Phase II (55.9% vs. 44.1%), there was no difference ( $P > 0.10$ ) between treatments, with each yeast

product accounting for about 50% of the feed intake for the individual and overall periods (YCC-1: 52.7%; YCC-2: 47.3%). In Exp. 2 (the performance test), 50 pigs (21 d of age and 7.1 ± 0.7 kg BW) were allotted in a randomized complete block basis (five pens/trt with five pigs/pen). The performance for the total experiment was: ADG - 410 and 425 g/d; ADFI - 621 and 619 g/d; and F/G - 1.514 and 1.467, respectively, for YCC-1 and YCC-2. There were no significant differences ( $P > 0.10$ ) between products for any weekly period or for the total experimental period. In conclusion, there were no manifest preference or performance differences between the two YCC products.

**Key Words:** Preference, Weaning Pig, Yeast Culture

**M112 Digestibility of CP, AA, and energy in a novel yeast product by pigs.** H. H. Stein<sup>\*1</sup>, M. L. Gibson<sup>2</sup>, M. G. Boersma<sup>1</sup>, and C. Pedersen<sup>1</sup>, <sup>1</sup>South Dakota State University, Brookings, <sup>2</sup>Dakota Gold Research Association, Sioux Falls, SD.

Two experiments were conducted to measure the digestibility of CP, AA, and energy in a novel yeast product that was produced by extraction from ethanol by-product streams. In Exp. 1, eight barrows that were equipped with a T-cannula in the distal ileum were randomly allotted to a 2-period switch-back design and fed a yeast-based diet and a N-free diet. Both diets were provided in daily amounts equivalent to three times the energy requirement for maintenance. Ileal digesta were collected from the cannulae and the standardized ileal digestibility coefficients (SID) for CP and AA were calculated. Results of this experiment showed that the SID for CP was 74.8%. The SID for Lys, Met, Thr, Trp, Ile, Leu, and Val were 82.2, 88.6, 71.1, 82.2, 79.5, 84.0, and 74.5%, respectively. The average SID for all indispensable AA was 81.4% while the average for the dispensable AA was 75.5%. Exp. 2 was designed to measure the concentration of DE and ME in the yeast product. Six growing barrows were placed in metabolism cages and randomly allotted to a 2-period switch-back design. A corn-based diet (98% corn, 2% vitamins and minerals) was formulated. A second diet consisting of 40% yeast and 60% of the corn-based diet was also formulated. Both diets were supplied in a daily amount equivalent to 2.5 times the energy requirement for maintenance. Collections of feces and urine were performed and the energy balance for each of the two diets was calculated. The energy concentration in the corn was calculated from the corn-based diet while the energy concentration in the yeast was calculated from the corn-yeast diet using the difference method. Results of this experiment showed that the concentration of DE and ME in yeast (5,600 and 5,350 kcal per kg DM, respectively) is higher ( $P \leq 0.001$ ) than in corn (4,071 and 3,992 kcal per kg DM, respectively). It is concluded that the yeast product extracted from ethanol by-product streams has a high digestibility of AA and a high concentration of energy. This product may be well suited as an energy and AA source in diets for swine.

**Key Words:** AA Digestibility, Pigs, Yeast

## Physiology and Endocrinology I

**M113 Desert climatic effects on freezability and some biochemical constituents of Barki ram semen.** M. Zeitoun<sup>\*1</sup> and K. El-Bahrawy<sup>2</sup>, <sup>1</sup>Alexandria University, Alexandria, Egypt, <sup>2</sup>Mariout Research Station, Desert Research Center, Ministry of Agriculture, Alexandria, Egypt.

This study utilized 12 mature fertile Barki rams located in the Maryout Research Station. Semen was collected during June â€“ August (summer season, 1999) and during December â€“ March (winter season, 1998). Semen was collected using an artificial vagina with 0.5 ml Tris buffer in the collection tubes (1:1 dilution). Semen sampler were diluted and packed in straws (0.25ml) and frozen (-196°C) in liquid nitrogen. Data on physical characteristics of semen were recorded (volume, motility, % crsosome integrity, % dead and live, pH, concentration and % abnormality). In addition, seminal plasma of both seasons was harvested and Na<sup>+</sup>, K<sup>+</sup>, free amino acids and total protein were determined. Also, SDS-PAGE was conducted to characterize the peptide fractions of

seminal plasma of both seasons. Results indicated higher ( $p < 0.05$ ) post-thaw (0h) motility in winter (44.1%) than in summer (17.2%) ejaculates, whereas at 4 hrs. post-thaw the percent intact acrosome approached 72.3% and 65.9% for summer and winter ejaculates, respectively. Moreover, percent dead and abnormal spermatozoa were higher ( $p < 0.05$ ) in post-thaw spermatozoa of summer than winter ejaculates. Sodium concentration was not different between summer and in winter, however K<sup>+</sup> concentrations was higher ( $p < 0.05$ ) in winter (71.7 ppm) than in summer (47.3 ppm) ejaculates. This resulted in different K<sup>+</sup>/Na<sup>+</sup> ratio between the two seasons. Total protein was found to be as much twice (14.0g/dL) in summer as in winter (7.7g/dL). The glutamic acid and glycine were higher in winter than summer semen. The SDS-PAGE exhibited two more peptide fractions (330 and 24 kDa) in winter than summer seminal plasma. The total number of peptide fractions was 14 in winter and 12 in summer.

**Key Words:** Climate, Semen, SDS-PAGE Protein

**M114 The effects of Pulsatilla miniplex® administrations on some blood values in dairy cows.** F. S. Hatipoglu\*<sup>1</sup>, M. S. Gulay<sup>1</sup>, M. Findik<sup>2</sup>, S. Aslan<sup>2</sup>, C. Altinsaat<sup>2</sup>, and G. Atintas<sup>2</sup>, <sup>1</sup>Akdeniz University, Antalya, Turkey, <sup>2</sup>Ankara University, Ankara, Turkey.

The experiment was designed to evaluate the effects of Pulsatilla miniplex® (Pm), a female constitution substance with hormone like effect for treatment of sterility, on red blood cell count (RBC), packed cell volume (PCV), hemoglobin (Hb), erythrocyte sedimentation rates (ESR), leukocyte count (WBC), WBC profiles and mean corpuscular hemoglobin concentrations (MCHC). Multiparous Holstein cows (3 to 5 year old) were assigned to control (C; n=21) and experimental (TRT; n=21) groups after the first blood sampling at 30 d prior to expected calving days and kept under the same management conditions. The first subcutaneous Pm application (10 ml) was done at the postpartum 2nd hr (d 0) to cows in TRT group, whereas no Pm was administered to cows in C group. Consecutive blood samples were collected from vena jugularis immediately before each Pm administrations on 0, 15, 25, 35 days of postcalving. No significant differences were detected in mean PCV (C=30.5 ± 0.6 vs. TRT=30.1 ± 0.6 %), Hb (C=10.8 ± 0.3 vs. 10.9 ± 0.3 g %), ESR (17.3 ± 0.9 vs. TRT=17.5 ± 0.9 mm/h), lymphocyte (C=55.9 ± 2.7 vs. TRT=50.7 ± 2.7 %), monocyte (C=2.43 ± 0.4 vs. TRT=2.52 ± 0.4 %), neutrophile (C=38.7 ± 2.7 vs. 40.1 ± 2.7 %) or MCHC (C=35.5 ± 0.9 vs. TRT= 36.3 ± 0.9 g/100ml). On the other hand, mean RBC (C=6.0 ± 0.3 vs. 6.3 ± 0.3X10<sup>6</sup>/μL; P<0.09), WBC (C=9.06 ± 0.37 vs. 8.15 ± 0.38 x 10<sup>3</sup>/μL; P<0.1), basophile (C=0.27 ± 0.14 vs. TRT=0.62 ± 0.14 %; P<0.01), and eosinophile (C=2.75 ± 0.9 vs. TRT=6.19 ± 0.9%; P<0.01) were significant. Overall, the parameters examined remained within the physiological range in both groups. In conclusion, Pm injections after calving did not cause any negative effect on the blood parameters tested in this study.

**Key Words:** Pulsatilla Miniplex, Blood Parameters, Dairy Cows

**M115 Estrogens and isoflavones affect porcine muscle satellite cell growth.** C. Rehfeldt\*, M. Mau, and T. Viergutz, *Research Institute for the Biology of Farm Animals, Dummerstorf, Germany.*

The role of estrogens and estrogen-like compounds, such as dietary phytoestrogens, in pig skeletal muscle growth is largely unknown. The aim of this study was therefore to investigate the effects of estrogens and isoflavones on porcine muscle satellite cell growth in vitro. Myogenic cells were derived from M. semimembranosus of new born piglets, typified for muscle cell specific proteins (desmin, N-CAM) and grown in culture. The effects of different concentrations of 17β-estradiol (0.1 nM; 1 nM; 1μM), estrone (1 nM; 1μM), and of genistein and daidzein (0.1; 1; 10; 100 μM) on DNA synthesis measured as 6 h-[<sup>3</sup>H] thymidine incorporation (dpm/DNA) were determined in serum-free growth medium. After 7 h (1+6 h) exposure both 17β-estradiol and estrone slightly decreased DNA synthesis (-4 to -7%). Slight decreases were also observed in response to 1 and 10 μM genistein (-5; -10%) and to 0.1, 1, 10, and 100 μM daidzein (-3 to -13%), whereas 100 μM genistein (-74%) substantially lowered DNA synthesis. Associated decreases in cell number (DNA) were observed with 100 μM genistein (-8%) and daidzein (-6%). No signs of apoptosis were observed by cell cycle analysis of cultures exposed to 1 and 10 μM genistein and 17β-estradiol (0.1 nM; 1 nM; 1μM). The results suggest that both estrogens and isoflavone phytoestrogens may directly affect porcine muscle cell growth with the effects being dose-dependent.

This study was supported by the Deutsche Forschungsgemeinschaft (DFG; Re 978-11).

**Key Words:** DNA Synthesis, Cell Culture, Myogenic Cells

**M116 Use of milk oestradiol in conjunction with milk progesterone analysis to quantify reproductive function in dairy cows.** D. V. Scholey, N. R. Kendall\*, A. P. F. Flint, and G. E. Mann, *University of Nottingham, Sutton Bonington Campus, Loughborough, UK.*

While milk progesterone measurement can be used to identify the timing of luteolysis and the postovulatory progesterone rise, it reveals nothing about the characteristics of the follicular phase. The general pattern of follicular phase events entails an increase in oestradiol followed by a fall, triggered by the LH surge, which preceded ovulation. Thus the follicular phase fall in milk oestradiol can be used as an indicator of the LH surge and subsequent ovulation. In this study we have developed a milk oestradiol assay and used it, in conjunction with milk progesterone analysis, to characterise the follicular phase in 24 multiparous lactating dairy cows. Milk samples were collected at daily intervals and analysed for progesterone by ELISA. Based on milk progesterone data, daily samples from the follicular phase period of low progesterone were then analysed for milk oestradiol. Milk was first defatted by centrifugation and surface fat removal. To extract oestradiol, defatted milk was passed through a C18 SepPak cartridge, washed with distilled water and the oestradiol eluted with acetone. The extract was then dried down and resuspended in buffer prior to radioimmunoassay. The mean interval from progesterone fall (to <3ng/ml) to progesterone rise (to >3 ng/ml) was 9.0±0.3 days (range 7-12 days). Within this 9 day period, the interval from luteolysis to the preovulatory oestradiol fall was 4.6±0.2 days (range 3-7 days) and from the oestradiol fall to the postovulatory progesterone rise 4.4±0.2 days (range 3-7 days). Peak milk oestradiol concentrations of 4.0±0.3pg/ml (range 2.0-6.6 pg/ml) were achieved within 3.0±0.2 days (range 2-6 days). Following this peak oestradiol fell to 1.9±0.2pg/ml (range 0.8-4.0pg/ml). We have successfully used milk oestradiol analysis to noninvasively characterise the timing of follicular phase events. Using this approach we are able to obtain far more information than is available following milk progesterone measurement alone.

**Acknowledgements:** Funded by Defra, Milk Development Council and Intervet under the Link Sustainable Livestock Programme.

**Key Words:** Milk Oestradiol, Milk Progesterone, Cow

**M117 Effect of dietary phosphorus on reproductive function and performance of Holstein cows.** S. K. Tallam\*, A. D. Ealy, K. A. Bryan, and Z. Wu, *Pennsylvania State University, University Park.*

The objective was to determine the effect of dietary P on postpartum resumption of luteal function, response to ovulation synchronization, and reproductive performance. Fifty-four multiparous Holstein cows were assigned at calving to diets containing 0.35 or 0.47% P. Resumption of normal ovarian function was monitored three times weekly by ultrasonography, beginning 10 d after parturition until the end of a 60-d voluntary waiting period. Cows were subsequently synchronized and bred using the Ovsynch protocol. Dietary P did not affect the number of days to first postpartum ovulation or corpus luteum development. Peak serum progesterone concentration for estrous cycles during the voluntary waiting period did not differ, although there was a tendency for greater luteal phase serum progesterone concentration for the 0.35% P group during the first estrous cycle. The ovulation synchronization rate was 61.5 and 69.2% for the 0.35 and 0.47% P groups, respectively, uninfluenced by diet. The overall pregnancy rate at 200 d of lactation (60.9 and 60.0%) and the number of services per pregnancy (2.1 and 1.9) did not differ between groups. Serum P concentration increased (from 6 to 7 mg/dl) with dietary P during the first 3 mo postpartum. Mean milk yield during the first 40 wk of lactation did not differ, averaging 40.5 and 39.0 kg/d for the two groups. Fecal P content measured during the first 16 wk of lactation averaged 0.63 and 0.89% for the 0.35% and 0.47% P groups, respectively. Varying dietary P from 0.35 to 0.47% did not affect resumption of luteal function, response to ovulation synchronization, or reproductive performance.

Item	0.35% P	0.47% P	SEM	P
Days to first progesterone increase (> 1 ng/ml)	35.0	40.5	4.4	0.38
Days to first progesterone increase (> 1 ng/ml)	35.0	40.5	4.4	0.38
Number of ovulations (1 to 60 DIM)	1.4	1.3	0.2	0.68
Number of ovulations (1 to 60 DIM)	1.4	1.3	0.2	0.68
Duration of estrous cycle, d	21.0	18.8	1.0	0.12
Duration of estrous cycle, d	21.0	18.8	1.0	0.12
Peak serum progesterone <sup>a</sup> , ng/ml	11.2	9.8	1.0	0.35
Peak serum progesterone <sup>a</sup> , ng/ml	11.2	9.8	1.0	0.35
Luteal phase progesterone <sup>b</sup> , ng/ml	8.1	5.6	1.1	0.12
Luteal phase progesterone, ng/ml	8.1	5.6	1.1	0.12

<sup>a</sup>For estrous cycles during 1-60 DIM; <sup>b</sup>First estrous cycle; <sup>c</sup>First service.

**Key Words:** Dairy Cows, Phosphorus Requirement, Reproductive Performance

**M118 Progesterone (P4) Concentrations and Ovarian Response after Insertion of a New or a 7 d Used Intravaginal P4 Insert (IPI) in Proestrous Lactating Cows.** R. L. A. Cerri\*, H. M. Rutigliano, R. G. S. Bruno, and J. E. P. Santos, *University of California, Tulare.*

Cyclic Holstein cows were synchronized with GnRH followed 7 d later by PGF2a, and randomly assigned to a New (1.38 g of P4; n=31) or a 7-d Used autoclaved IPI (n=31) inserted 48 h after PGF2a injection for 7 d. Blood was sampled at PGF2a injection, daily during IPI treatment, and at 0, 15, 30, 45, 60, 90, 120 and 180 min relative to IPI insertion and removal. Plasma was analyzed for P4 by a validated EIA assay and intra and inter-assay CV were 6.5 and 7.1%, respectively, with a sensitivity of 0.06 ng/mL. Ovaries were examined by ultrasonography at the GnRH and PGF2a injections, daily during IPI treatment, and after IPI removal, when the cow was in estrus and 48 h later. Progesterone concentrations and size of follicles were analyzed by ANOVA using the MIXED procedure of SAS, and estrus and ovulation by the Logistic procedure of SAS. Milk yield and BCS did not influence P4 concentrations. Plasma P4 concentrations increased from 0.18 to 0.78 ng/mL in the first 15 min after IPI insertion, but achieved a plateau (0.9 ng/mL) at 90 min. Concentrations of P4 in the first 180 min tended to be greater (P=0.10) for Used than New IPI (0.90 vs 0.77 ng/mL). Concentrations of P4 decreased daily, but the New IPI sustained slightly greater (P=0.04) P4 concentration (0.67 vs 0.78 ng/mL). On d 7, all cows had P4 < 1 ng/mL. After removal of IPI, P4 dropped to basal concentrations by 90 min and no difference was observed between treatments. During d 1 to 7, only 18.8 and 16.8% of the plasma samples from New and Used IPI cows, respectively, were greater than 1 ng/mL (P=0.62). Primiparous had consistently greater (P<0.05) P4 concentrations than multiparous throughout the study. Six New and 8 Used IPI cows ovulated in the first 48 h of insertion, likely as a consequence of a LH surge prior to insertion. After IPI removal, estrus, ovulation, double ovulation, days to estrus and ovulation did not differ between treatments (P>0.15). Both New and Used IPI resulted in similar, and mostly subluteal (<1 ng/mL), P4 concentrations in lactating dairy cows, but prevented ovulation during proestrus.

**Key Words:** Intravaginal Progesterone Insert, Dairy Cows

**M119 Behavioral and endocrine responses to estradiol-17b (E) in ovariectomized Holstein cows.** P. Reames\*, T. Hatler, and W. Silvia, *University of Kentucky, Lexington.*

Ovariectomized Holstein cows (n=7) were used to characterize responses to estradiol-17b (E) when infused at rates to maintain concentrations within physiological ranges. The experiment was conducted in 8 replicates using 3-7 cows/replicate. Cows were pretreated with CIDRs 6 days prior to the start of infusion

with E (d -6). CIDRs were removed after 72 h (d -3). E infusions were initiated on d 0. Each cow received an i.v. injection of E at one of 5 doses (designated doses 0, 1, 2, 3, 4): 0, 3, 6, 9 or 12 ng/kg bw, respectively. These doses were calculated to result in 0, 3, 6, 9 or 12 pg/ml in blood. Cows then received i.v. infusions of E at rates of 0, 6, 12, 18 or 24 ug/kg bw/h for 8 h to maintain the concentration. Additional cows (n=10/2/replicate) received an injection of E (500 µg; i.m.) for estrus detection. Cows were brought together for visual estrus detection (30 min) at 10, 12 h post infusion, then at 4 h intervals for 48 h. Estrus was defined as accepting a mount > 2 times during the observation period. Venous blood samples were collected at 2-h intervals throughout the experimental period to quantify LH. This experiment was considered a randomized design. Effects of cow, replicate and dose were tested for by ANOVA. Seven cows received dose 4 and all showed estrus. Five cows received dose 0 (one of them twice). Six cows received dose 1. None of these cows showed estrus. Five cows received each dose at least once. Three showed estrus at doses 2, 3 and 4. Two only showed estrus at dose 4. Thus, cows may differ in the amount of E needed to induce estrus. Of the cows that showed estrus, duration was less for dose 2 (8.8 h) than for dose 4 (17.1 h; P<0.05), indicating that the duration of estrus is dose dependent. The onset of estrus tended to be later for dose 2 (19.6 h) than for dose 4 (13.6 h; P<0.10). The magnitude of the LH surge was less for doses 2 and 3 than for dose 4 (P<0.05), however the timing of the LH surge (after start of infusion) was not different among doses (P>0.4). Thus, estrus in cows receiving dose 2 was shorter and delayed in onset, relative to the timing of the LH surge, compared to cows receiving dose 4.

**Key Words:** Cow, Estrus, LH

**M120 Bovine uterine temperature measured by novel IC thermometer placed in the uterotubal junction.** S. Kamimura\*<sup>1</sup>, E. Kurataki<sup>1</sup>, N. Roa Avila<sup>1</sup>, K. Hamana<sup>1</sup>, K. Morita<sup>1</sup>, and I. Shibata<sup>2</sup>, <sup>1</sup>Kagoshima University, Kagoshima, Japan, <sup>2</sup>Sanyo Electric, Tokyo, Japan.

Physio-environments in bovine uteri have not been adequately reported due to the limitations in methodology. Objective of the study was to measure bovine uterine temperature (UT) by novel IC thermometer (IC). Data was compared with conventional rectal (RT) and vaginal temperature (VT). IC is a chip housed in a button shaped stainless steel enclosure, 17 mm in length, 5mm in width with a range of +15 to +46 C in 0.125 C increments. It was calibrated with a standard mercury thermometer, and was also correlated with a rectal thermometer installed with a thermistor sensor probe (0.1 C precision). UT was measured in 20 min intervals for 4 weeks, while RT and VT was measured every 4 hr. Japanese Black cows, 8 cows in the summer and 3 cows in the winter were subjected to caesarian section (CS). IC was installed in the left uterine horn proximate to the uterotubal junction during CS by trans-lumbar laparotomy. After 4 weeks recording, IC was removed by 2<sup>nd</sup> CS and mounted on the chip reader in the PC. The accumulated data was analyzed by ANOVA for repeated measures. Ambient temperature (AT) was simultaneously recorded. In 5 cows, blood was daily collected for hormonal assay and underwent ultrasonography to monitor ovarian dynamics. Temperature after operation was temporarily elevated for 4 days (0.14 C) and excluded. Averages in UT, RT and VT in the summer (AT: 28.76 C) were 38.57±0.23 C, 38.67±0.23 C, 38.60±0.35 C, and those in the winter (AT: 14.46 C) were 38.63±0.21 C, 38.68±0.21 C, 38.67±0.20 C, respectively. UT was significantly lower than RT or VT, and UT in the summer was lower than in the winter (P<0.01). Diurnal rhythm was observed at all three temperatures, lowest at 08:00 and highest at 20:00. Upon ovulation, UT in the luteal phase was significantly higher (38.61±0.20 C) than in the follicular phase (38.51±0.22 C, P<0.01), whereas no difference was observed in RT or VT. In conclusion, IC placed in the uterotubal junction successfully measured UT and detected diurnal rhythm. UT fluctuation was stable, but showed luteal hyperthermia.

**Key Words:** IC Thermometer, Uterine Temperature, Cows

**M121 Cloning and characterization of adsf/resistin in korean native cow.** J. Park, H. Kang, C. Y. Lee, and Y. S. Moon\*, *Jinju Nation University, Jinju, Gyung Nam, Korea.*

Adipocyte-specific secretory factor (ADSF)/resistin is a small cysteine-rich protein secreted from adipose tissue and ADSF has been implicated in modulating adipogenesis in human and rodents. The objective of this study was to clone a gene encoding ADSF/resistin and to characterize its function in Korean Native Cow. The coding sequence was 330 base pairs and it encoded a protein of 109 amino acids. An NCBI BLAST-search revealed the cloned cDNA fragment shared significant homology (82%) with the cDNA encoding the human ADSF/resistin. The nucleotide sequence homology of the Korean cow was 68% and 64% for the rat and mouse, respectively. Homology between Korean cow and Genbank deposit sequences of bovine ADSF/resistin was 98% for the nucleotide sequence and 99% for the amino acid sequence. A high frequency of single nucleotide polymorphism was identified in intron 3 but not in any other exons or introns of ADSF. Further studies are required to analysis the association between the polymorphisms and carcass traits in the cow. Tissue distribution of ADSF mRNA was examined in liver, skeletal muscles (sirloin, rump), subcutaneous fat, and retroperitoneal fat by RT-PCR. ADSF mRNAs were detected in fat tissues but not in liver and muscles, suggesting that ADSF/resistin expression may be induced during adipogenesis. Although, the physiological function of ADSF/resistin in cow remains to be determined, these data indicate ADSF is related to the adipocyte phenotype and may have a possibly regulatory role in adipocyte function.

**Key Words:** Adsf/Resistin, Adipocyte, Cow

**M122 Repeatability estimate for embryo survival following insemination at PG-induced heats in beef heifers.** M. G. Diskin\* and J. M. Sreenan, *Teagasc Research Centre, Athenry, Co. Galway, Ireland.*

Embryo survival rate is a major determinant of reproductive efficiency in dairy and beef herds. Data from this laboratory and elsewhere shows that embryo death, before about day 16 of gestation, is the major cause of low cow conception rate. There is some evidence of repeatable differences between animals in their ability to establish and sustain pregnancy and of genetic variability for heifer pregnancy rate and dairy cow sustainability. However, the endocrine, molecular and genetic basis for these apparent differences in embryo survival rate have not been established. The initial objective of this study was to establish repeatability estimates for embryo survival rate in heifers. A total of 69 reproductively normal heifers was used. Each heifer was artificially inseminated (AI) on 4 occasions over a 9-month period using the following regimen. Initially estrus was synchronised using a 2-injection prostaglandin (PG) regimen and heifers were inseminated at 6-12 hours after observed standing estrus (day 0). Heifers were ultrasonically scanned for pregnancy on days 28 and 35. All pregnant heifers received PG on day 35 to induce embryo loss. Six weeks after the induced embryo loss all heifers were reprogrammed using a 2-injection PG-regimen, inseminated and scanned as described above and a similar schedule was followed for a further two rounds of AI. Frozen-thawed semen from one high-fertility bull was used for all AI. Repeatability estimates were derived from an analysis of variance as the intraclass correlation among records on the same individual in different rounds. Conception rate was similar ( $P>0.05$ ) for each round of AI with an overall rate of 63%. Based on results to-date a repeatability estimate of  $0.18 \pm 0.003$  for embryo survival following a PG-induced heat was recorded. This analysis indicates that embryo survival has low to moderate repeatability.

**Key Words:** Embryo, Survival, Repeatability

**M123 Reproductive performance following estrous synchronization of Angus, Brahman and Angus x Brahman crossbred cows.** L. Praharani\*, D. O. Rae<sup>2</sup>, and T. A. Olson<sup>2</sup>, <sup>1</sup>*Research Institute of Animal Production, Bogor, Indonesia,* <sup>2</sup>*University of Florida, Gainesville.*

This study evaluated reproductive parameters and timed-artificial insemination (TAI) responses in heifers and cows of differing proportions of Angus and Brah-

man breeding following estrous synchronization using Synchronate-B (SMB). It was conducted at the Beef Research Unit, University of Florida. Data from 564 heifers and 1,257 cows consisting of 276 Angus (A), 387 Brahman (B), 250 (75%A:25%B), 422 (50%A:50%B), 277 (25%A:75% B), and Brangus (62.5%A:37.5%B) from 1991 through 1997 were analyzed using the PROC GENMOD procedure. The model included the fixed effects of year of breeding, age of cow, body condition score, breed type of cow, and breed type of sire with days from calving to synchronization date as a covariate plus all two-way interactions. Year of breeding, body condition score, and breed type of cow all affected estrus expression rate, pregnancy rate, and calving rate ( $P<0.01$ ) and age of cow affected pregnancy rate ( $P<0.01$ ). Few Brahman cows exhibited estrus on the first day after implant removal ( $P<0.01$ ). The estrus expression, pregnancy and calving rates obtained from estrous synchronization using SMB and TAI were moderate to high. Results showed that the Brahman female reproductive performance was comparable to Angus and their crossbreds. Gestation length of cows was influenced by year of breeding, breed type of cow, and age of cow ( $P<0.01$ ). Gestation length ranged from 282.1 days to 289.4 days; Angus cows had the shortest gestation length ( $P<0.01$ ) and Brahman, the longest ( $P<0.01$ ). There was a positive trend between percentage of Brahman breeding in the cows and gestation length. The introduction of Brahman-bred, particularly in subtropical regions, is important in improving beef cattle productivity because of their subtropical adaptive traits. However, the tendency for high proportion Brahman-bred cows to have longer gestation lengths is of concern, despite reproductive performance similar to Angus cows.

**Key Words:** Reproductive Performance, Estrous Synchronization, Cows

**M124 The Crestar® protocol with estradiol benzoate, PGF2 $\alpha$ , PMSG or GnRH to control estrus cycle and ovulation in beef cows.** R. J. C. Moreira<sup>1</sup>, A. V. Pires<sup>1</sup>, D. Z. Maluf<sup>1</sup>, E. H. Madureira<sup>2</sup>, M. Binelli<sup>2</sup>, J. R. Gonçalves<sup>3</sup>, L. G. Lima<sup>3</sup>, and I. Susin<sup>1</sup>, <sup>1</sup>*ESALQ/University of São Paulo, Piracicaba, SP, Brazil,* <sup>2</sup>*FMVZ/University of São Paulo, Pirassununga, SP, Brazil,* <sup>3</sup>*FEALQ, Londrina, PR, Brazil.*

The objective of this study was to evaluate the effects of using PMSG, GnRH, estradiol benzoate or PGF2 $\alpha$  in combination with Crestar® protocol and AI at fixed time on ovulation of beef cows. Three hundred and forty-eight multiparous cows, crossbreed Nelore (*Bos taurus indicus*) X Charolais (*Bos taurus taurus*), were divided in two groups: 179 suckling cows and 169 non-suckling cows. Cows received the Crestar® protocol for follicular growth synchronization consisting of a subcutaneous implant with 3mg of norgestomet and 3mg of norgestomet plus 5mg of estradiol valerate injection (day of implant insert). Implants were removed after nine days. Cows were submitted to one of five treatments for pharmacological control of ovulation and were artificially inseminated at fixed time. Experimental treatments were: T1 (n=70): injection of physiological solution 48h after implant removal (D12); T2 (n=68): 0.75mg of estradiol benzoate 24h after implant removal (D11); T3 (n=70): 150 $\mu$ g of PGF2 $\alpha$  at same day of implant removal (D9) and 0.75mg of estradiol benzoate 24h after implant removal (D11); T4 (n=70): 500 UI of PMSG at implant removal (D10) and T5 (n=70): 500 $\mu$ g of GnRH 48h after implant removal (D12). Cows were artificially inseminated 54-56h after implant removal. Pregnancy rate was analyzed by logistical regression program. There were no differences ( $P>0.05$ ) on pregnancy rate among treatments (35.7, 31.4, 22.0, 37.0 and 42.8% for T1, T2, T3, T4 and T5, respectively).

**Key Words:** Nelore, Pregnancy Rate

**M125 Conception rates and serum progesterone concentration in dairy cattle administered gonadotropin releasing hormone five days after artificial insemination.** J. M. Howard\*<sup>1</sup>, R. Manzo<sup>1</sup>, J. C. Dalton<sup>2</sup>, and A. Ahmadzadeh<sup>1</sup>, <sup>1</sup>*University of Idaho, Moscow,* <sup>2</sup>*University of Idaho, Caldwell.*

The objective of this study was to determine the effect of administration of exogenous GnRH five days after artificial insemination (AI) on serum progesterone concentration and conception rates in dairy cattle. In experiment 1, 23 Holstein cows were synchronized using the Ovsynch protocol. Five days after

AI (d 0) cows were assigned randomly to receive either saline (CON; n = 11) or 100 ug GnRH (GnRH; n = 12). To examine ovarian structures, ultrasonography was performed on d -1 and every other day beginning on d 5 until d 14. On d 5 and d 14 blood samples were obtained to measure serum progesterone concentrations. All cows in the GnRH-treated group developed an accessory corpus luteum (CL), whereas cows in the CON group did not. Mean serum progesterone concentrations did not differ between GnRH and CON groups on d 5 ( $1.64 \pm 0.46$  ng/ml vs.  $2.04 \pm 0.48$  ng/ml). On d 14 serum progesterone concentrations were higher ( $P < 0.05$ ) in the GnRH group compared to CON ( $5.22 \pm 0.46$  ng/ml vs.  $3.36 \pm 0.48$  ng/ml). In Exp. 2, 542 lactating cows, at two different commercial dairies, were used to test the effect of administering GnRH 5 d after AI on conception rates. Cows were synchronized and detected for estrus according to tail chalk removal. Cows detected in estrus received AI immediately. Five days after AI, cows were assigned randomly to receive either GnRH (n= 266) or saline (n = 276). Pregnancy status was determined by palpation per rectum approximately 40 d after AI. Conception rates did not differ between GnRH and CON groups at either location (26.7% GnRH vs. 24.3% CON). Regardless of treatment, days in milk, parity, milk yield, and number of services had no effect on the odds ratio of pregnancy. In summary, the results of this study indicated that GnRH administered 5 d after AI increased serum progesterone by developing an accessory CL but did not improve conception rates in dairy cattle.

**Acknowledgements:** Authors would like to express their appreciation to Meril for the product support.

**Key Words:** GnRH, Progesterone, Conception Rate

**M126 Evaluation of progestagen implants reutilization on pharmacological control of estrus cycle and ovulation in beef cows.** D. Z. Maluf<sup>1</sup>, A. V. Pires<sup>\*1</sup>, R. J. C. Moreira<sup>1</sup>, E. H. Madureira<sup>2</sup>, M. Binelli<sup>2</sup>, J. R. Gonçalves<sup>3</sup>, L. G. Lima<sup>3</sup>, and I. Susin<sup>1</sup>, <sup>1</sup>ESALQ/University of São Paulo, Piracicaba, SP, Brazil, <sup>2</sup>FMVZ/University of São Paulo, Pirassununga, SP, Brazil, <sup>3</sup>FEALQ, Londrina, PR, Brazil.

Two hundred and twenty-one (78 suckling by 40 to 90 days) Nelore (*Bos taurus indicus*) X Charolais (*Bos taurus taurus*) cows were used to evaluate the reutilization of progestagen implants to control pharmacologically the estrus cycle and ovulation. Cows were randomly assigned to one of three protocols for estrous synchronization and artificial insemination in pre-fixed time. In treatment 1 (T1; n=73) cows were implanted with Crestar<sup>®</sup> (3 mg de norgestomet); in treatment 2 (T2; n=75) cows were implanted with Crestar<sup>®</sup>, already used in a previous synchronization; and in treatment 3 (T3; n=73) cows received two Crestar<sup>®</sup> implants, also previously used, both placed side by side in the same ear. All cows were injected with an intramuscular (i.m.) dose of 2 mL of progesterone (25mg/mL) + estradiol benzoate (1mg/mL) at the time of implants insertion (D0). Implants were removed after 8 days (D8) and an i.m. dose of Preloban<sup>®</sup> (150µg of D-cloprostenol) was administered. Twenty-four hours after implants removal, cows were i.m. injected with a dose of Estrogin<sup>®</sup> (1mg of estradiol benzoate). All cows were artificially inseminated at 54-56 h after implants removal. Ninety percent of semen used in the experiment was from only one bull, the remaining 10% were equally distributed among treatments. Artificial insemination was performed by only one technician. Suckling cows had a body condition score of 5-6, in a scale of 1-9, and the non-suckling cows were 6-7. Statistical analysis was accomplished by using Statistical Analysis System 8.0 and logistic regression. There was no difference ( $P > 0.05$ ) on cow's pregnancy rate among treatments. Pregnancy rates were 39.72, 34.21 and 36.98% for T1, T2 and T3, respectively. Progestagen implants reutilization did not affect pregnancy rate in beef cows ready for reproduction.

**Key Words:** Nelore, Reproduction

**M127 Myostatin inhibits the differentiation of bovine preadipocyte.** S. Hirai\*, H. Matsumoto, H. Kawachi, T. Matsui, and H. Yano, *Kyoto University, Kitashirakawa-oiwake, Sakyo-ku, Kyoto, Japan.*

Myostatin (growth differentiation factor 8, GDF-8) is a member of the transforming growth factor- $\beta$  superfamily, and a key critical regulator of skeletal muscle development. In cattle, mutation of myostatin gene results in increasing skeletal muscle, which is known as double-muscling. The expression of myostatin mRNA is found primarily in skeletal muscle, but it is also detected in the adipose tissue. Myostatin was reported to inhibit the differentiation of 3T3-L1 preadipocyte. However, the action of myostatin on bovine preadipocyte has not been studied. We investigated the effect of myostatin on the differentiation of preadipocyte in stromal vascular (SV) cells derived from bovine adipose tissue. SV cells were subcultured in DMEM with 5% FBS and 100  $\mu$ M vitamin C. After confluence (day 0), differentiation was induced by 0.5 mM 1-methyl-3-isobutyl-xanthine, 0.25  $\mu$ M dexamethasone, 2.5  $\mu$ g/ml insulin and 5  $\mu$ M troglitason for 2 days. Then, the medium was changed to DMEM with FBS, vitamin C, insulin, and troglitason, and the cells were cultured for 6 days. The cells were treated with myostatin (0, 100, 300 ng/ml) throughout the differentiation period (from day 0 to day 8) or during the early phase of differentiation (from day 0 to day 2) with 4 cultures each per treatment, and harvested for the measurement of glycerol-3-phosphate dehydrogenase (GPDH) activity at the end of the differentiation period. GPDH activity was significantly reduced by the each dose of myostatin treatment throughout the differentiation period ( $p < 0.01$ ). However, the high dose of myostatin was required for the significant reduction of GPDH activity ( $p < 0.01$ ) when the cells were treated during the early phase of differentiation. These results suggest that myostatin is a negative regulator of bovine preadipocyte differentiation. Myostatin secreted from muscular and/or adipose tissues probably regulates the differentiation of bovine preadipocyte.

**Key Words:** Myostatin, Bovine, Preadipocyte

**M128 Interrelationships among parity, body condition score (BCS), milk yield, AI protocol, and cyclicity with embryonic survival in lactating dairy cows.** H. M. Rutigliano\* and J. E. P. Santos, *University of California, Tulare.*

Lactating Holstein cows, 6,123, from 9 studies in 5 dairy farms were evaluated to determine the relationships among parity, BCS, milk yield, cyclicity (cyclic or anovular), and AI protocol (inseminated at estrus or timed AI) on pregnancy rates (PR) and embryonic survival at the first postpartum AI. Cows were inseminated following a pre-synchronized (2 PGF2a 14 d apart) ovulation (timed AI) or estrous synchronization (inseminated at estrus) protocol initiated 12 to 14 d after the pre-synchronization. Blood was sampled and analyzed for progesterone twice, 12 to 14 d apart, to determine cyclicity. Cows were scored for body condition (1-5 scale) after calving, and again at AI, between 63 and 75 d postpartum. Pregnancy was diagnosed at  $30 \pm 3$  and  $50 \pm 8$  d after AI. Data were analyzed by multivariate logistic regression controlling for study, dairy, parity, BCS, BCS change, milk yield, cyclicity, and insemination protocol. More multiparous than primiparous were cyclic (81.8 vs 69.4%;  $P < 0.001$ ). In addition to parity, cyclicity was also influenced ( $P < 0.001$ ) by BCS at calving and at AI, BCS change, and milk yield. However, milk yield, BCS at calving and AI protocol had no effect ( $P > 0.10$ ) on PR at 30 and 58 d after AI or pregnancy loss. More cyclic than anovular cows were pregnant at 30 (40.1 vs 28.1%;  $P < 0.001$ ) and 58 (34.3 vs 22.0%;  $P < 0.001$ ) d after AI, and anovulation increased pregnancy loss (14.4 vs 18.7%;  $P = 0.05$ ). Pregnancy loss was highest (21.9 vs 16.6 vs 12.1%;  $P < 0.01$ ) and PR at d 58 lowest (22.3 vs 30.7 vs 35.6%;  $P < 0.01$ ) in cows that lost 1 unit of BCS than those that lost  $< 1$  or experienced no change in BCS from calving to AI. Likewise, a higher BCS at AI (3.75 vs 3.0 to 3.5 vs  $< 2.75$ ) increased ( $P < 0.01$ ) PR at 30 (46.3 vs 40.7 vs 34.2%) and 58 (41.8 vs 35.1 vs 28.5%) d after AI. Minimizing losses of BCS after calving and improving cyclicity early postpartum are expected to increase PR because of enhanced embryonic survival. However, AI protocol and milk yield do not affect pregnancy and embryonic survival after the first postpartum AI.

**Acknowledgements:** NRICGP USDA

**Key Words:** Embryo Survival, Cyclicity, Dairy Cows

**M129 Ontogeny of hypothalamic gene expression during prepuberal development in the gilt.** C. R. Barb<sup>\*1</sup>, R. L. Richardson<sup>1</sup>, R. Rekaya<sup>2</sup>, R. R. Kraeling<sup>1</sup>, and G. J. Hausman<sup>1</sup>, <sup>1</sup>USDA-ARS, Athens, GA, <sup>2</sup>University of Georgia, Athens.

The molecular mechanisms that regulate hypothalamic development in the pig are complex. To understand physiological pathways controlling age related changes in hypothalamic development, a custom microarray was utilized to profile differential gene expression. Total hypothalamic RNA was isolated from gilts at 90, 150 and 210 days (d) of age and used to prepare dye labeled cDNA probes, which were hybridized to arrays representing about 600 pig genes involved in growth and reproduction. Mixed linear model was used to analyze log transformed intensities in both channels. Sixty three genes were differentially expressed ( $P < 0.01$ ) from 90 to 210 d of age, which included genes involved in feed intake regulation, steroid binding, growth hormone secretion and intracellular signaling. The gene, AGRP and melanocortin-3-receptor involved in feed intake were up-regulated at 150 d and 210 d, respectively. Somatostatin (SS) was up-regulated and SS receptor-1 was down-regulated at 150 and 210 d, respectively. The progesterone receptor, steroid membrane binding protein, janus kinase-1 and MAPK3 were up-regulated at 210d. Neuronal protein, NP25, involved in brain development was also up-regulated at 210d. These results demonstrate, for the first time, differentially expressed hypothalamic genes in the prepuberal pig. The ontogeny of expression of key hypothalamic genes that regulate development, appetite and reproductive function will lead to a more detailed understanding of the molecular mechanisms controlling growth and the onset of puberty in the pig.

**Key Words:** Pig, Hypothalamus, Gene Expression

**M130 Effect of heat stress on the response to superovulation, embryo quality and survival, and the fertility of recipient cows in commercial dairy herds in Mexico.** R. Lozano<sup>\*1</sup>, M. Aspron<sup>3</sup>, C. Vasquez<sup>2</sup>, and E. Gonzalez-Padilla<sup>2</sup>, <sup>1</sup>INIFAP-Mexico, Aguascalientes, Mexico, <sup>2</sup>UNAM, Mexico D.F., <sup>3</sup>Private consultant, Queretaro, Mexico.

To study the effect heat stress on dairy cows and embryos produced through MOET, two trials were conducted. Forty-two cows with estimated 305d milk production of  $10,756 \pm 444$  l (P305D) and  $111.4 \pm 6.7$  days in milk (DIM), were superovulated with the same lot of FSH during the temperate (T,  $n = 20$ ) or the hot (H,  $n = 22$ ) seasons. In a second experiment, embryos collected and frozen during T or H were transferred to cows at the peak of lactation during the T or H months, resulting four groups: TT ( $n=26$ ); TH ( $n=25$ ); HT ( $n=28$ ) and HH ( $n=28$ ). Within group, embryos and recipients were randomly assigned. The Temperature-Humidity indexes (THI) differed during collection ( $70.5 \pm 0.3$  and  $78.0 \pm 0.3$ ) and transference ( $70.2 \pm 0.5$  and  $76.5 \pm 0.5$ ) for T and H, respectively. T and H were similar in cows responding (85 and 86 %), fertilization rate (72 and 80%), and number of CL (11.9 and 11.7), respectively, and different ( $P < 0.01$ ) in recovery rate of embryos+ova (89 and 54%), number of embryos/cow ( $7.47 \pm 1.28$  and  $4.47 \pm 1.21$ ) and number of ova/cow ( $3.17 \pm 0.77$  and  $1.63 \pm 0.72$ ), respectively. During T and H, 11.8% and 36.8% of the cows collected yielded 0 to 2 embryos+ova, respectively ( $P < 0.1$ ). Morulas and early blastocysts of excellent quality were more common during T ( $P < 0.01$  and  $P < 0.11$ , respectively). After transference, gestation percentage (GEST) was higher for T embryos (29.8 vs. 17.4%,  $P < 0.14$ ) and was also higher for T recipients (33.3 vs. 14.0%,  $P < 0.05$ ). There was an interaction, since TT was higher (45.0%) than the other groups ( $P < 0.05$ ) and HT (21.5%) higher ( $P < 0.05$ ) than TH (14.5%) and HH (13.4%). GEST was greater for excellent quality embryos as compared to good (30.4 vs. 16.8%,  $P < 0.11$ ). Heat stress reduced the viability of the embryos, and GEST in recipient cows; however there was not reduction in ovarian response or fertilization rate. The reduction of recovery rate during H could be related to ovulation failure or abnormal capture or transit of ova through the oviduct.

**Acknowledgements:** Financed by CONACYT, project 31457-B and dairy producers of Aguascalientes, Mexico

**Key Words:** Heat Stress, Embryo Transfer, Stress-Reproduction

**M131 Relationship between milk lactoperoxidase, progesterone and estradiol concentrations during estrus in dairy cows.** A. Ahmadzadeh<sup>\*1</sup>, M. L. Silber<sup>2</sup>, and J. C. Dalton<sup>1</sup>, <sup>1</sup>University of Idaho, Moscow, <sup>2</sup>Washington State University, Pullman.

The objective of the study was to characterize the relationship between milk lactoperoxidase (LP), serum progesterone (P4), estradiol (E2), and behavioral estrus in lactating Holstein cows. Ten cows, 7 with induced estrus and 3 exhibiting spontaneous estrus, were used in the study. Cows were synchronized using two injections of prostaglandin F2 $\alpha$  (PGF) 14 d apart. Blood samples were collected for five consecutive days starting with the second PGF injection (d 14) and on d 28. To quantify LP in milk, fresh milk samples were collected twice daily, from d 14 to 19 and on d 28. In cows with spontaneous estrus, blood and milk samples were collected on the day of estrus and 9 d after estrus. To verify estrus, all cows were continuously monitored for behavioral estrus by the HeatWatch<sup>®</sup> estrus detection system. Four of 7 cows exhibited estrus. There was a strong association between milk LP activity and behavioral estrus. Milk LP activity increased ( $P < 0.05$ ), compared to the day of the second injection of PGF (d 14) and the day before estrus. Milk LP activity decreased ( $P = 0.07$ ) during the luteal phase and was similar to the levels observed before estrus. Mean P4 concentrations were lower ( $P < 0.05$ ;  $0.7 \pm 0.55$  ng/ml) on the day of estrus compared to d 14 and d 28 ( $2.3 \pm 0.55$  and  $3.7 \pm 0.55$  ng/ml, respectively). In cows exhibiting spontaneous estrus, milk LP activity was more than two fold higher at the time of estrus compared to the luteal phase whereas serum P4 was lower ( $P < 0.05$ ) at the time of estrus ( $0.2 \pm 0.6$  ng/ml) when compared to the luteal phase ( $2.3 \pm 0.6$  ng/ml). Mean serum E2 was higher ( $P = 0.05$ ) at the time of estrus compared to the luteal phase ( $15.2 \pm 2.4$  vs  $7.8 \pm 2.4$  pg/ml). There was no change in LP or serum P4 in cows that did not exhibit estrus. This preliminary data appears to indicate that while milk LP is elevated during estrus, it was also negatively associated with blood P4 and positively associated with blood E2 during estrus.

**Acknowledgements:** Authors express their appreciation to NAAB for partial support of this study.

**Key Words:** Milk Lactoperoxidase, Estrus, Cattle

**M132 Assessing pregnancy status using digital infrared thermal imaging in Holstein heifers.** M. Jones<sup>\*1</sup>, A. Denson<sup>1</sup>, E. Williams<sup>1</sup>, A. Dos Santos<sup>1</sup>, K. Graves<sup>1</sup>, A. Kouba<sup>2</sup>, and S. Willard<sup>1</sup>, <sup>1</sup>Mississippi State University, Mississippi State, <sup>2</sup>Memphis Zoo, Memphis, TN.

Digital infrared thermal imaging (DITI) is a diagnostic technique for non-invasively monitoring body surface temperature (TEMP) gradients in animals as influenced by a change in physiology. The objective of this study was to determine if DITI could be used to detect pregnancy in Holstein heifers using DITI of the animal's side. DITI was conducted at 2-wk intervals alternating sides ( $n=8$  imaging sessions;  $n=4$  left/ $n=4$  right) of pregnant and non-pregnant heifers ( $n=20$ , respectively, each session). Ambient (AMB) TEMP, relative humidity (RH), and rectal TEMP (RT) were obtained at each imaging period. DITI images were acquired of the animal's side (shoulder to flank) and DITI TEMP minimum (MIN), maximum (MAX), average (AVG), and standard deviation (SD) were quantified. Mean AMB, RH, and THI were  $17.7 \pm 1.7^\circ\text{C}$ ,  $81.4 \pm 2.9\%$  and  $63.4 \pm 2.7$ , respectively. Results differed by sampling day ( $P < 0.05$ ) in MIN, MAX, AVG and SD TEMP from the left and right sides, which were conducted on different days. There was a day (side) x pregnancy status interaction ( $P < 0.05$ ). Therefore data were analyzed within side (left vs right) across days and pregnancy status for DITI measures. Heifer RT were greater ( $P < 0.05$ ) for non-pregnant ( $38.31 \pm 0.02^\circ\text{C}$ ) than pregnant ( $38.16 \pm 0.02^\circ\text{C}$ ) heifers regardless of sampling day; however there was no correlation ( $R=0.08$ ;  $P > 0.10$ ) between RT and day of gestation within pregnant heifers. Right-side DITI revealed a higher ( $P < 0.05$ ) MIN TEMP for pregnant vs. non-pregnant heifers across days, whereas MAX TEMP and SD did not differ ( $P > 0.10$ ) by pregnancy status when AMB was  $> 10^\circ\text{C}$ . Left-side DITI was variable across days for all DITI variables, and left-side DITI for MIN, MAX, AVG and SD were not as correlated to day of gestation ( $R=0.19, 0.26, 0.26$  and  $-0.23$ , respectively;  $P < 0.08$ ) as right-side DITI ( $R=0.48, 0.49, 0.49$  and  $-0.37$ , respectively;  $P < 0.01$ ). These data suggest that right-side DITI is a better predic-

tor of pregnancy than left-side DITI in Holstein heifers; however, the ability to discriminate pregnant from non-pregnant heifers is questionable, and greatly affected by ambient temperature.

**Acknowledgements:** Funded by the Conservation Action Network, Memphis Zoo

**Key Words:** Thermography, Pregnancy, Holstein

**M133 Thermography of the vulva in Holstein dairy cows: A comparison of estrus vs. diestrus.** M. Jones<sup>\*1</sup>, A. Denson<sup>1</sup>, S. Bowers<sup>1</sup>, K. Moulton<sup>1</sup>, E. Williams<sup>1</sup>, K. Graves<sup>1</sup>, A. Dos Santos<sup>1</sup>, A. Kouba<sup>1</sup>, and S. Willard<sup>1</sup>, <sup>1</sup>Mississippi State University, Mississippi State, <sup>2</sup>Memphis Zoo, Memphis, TN.

It has been suggested previously that thermal imaging (DITI) of the external genitalia (vulva) of female animals might be used to detect the timing of estrus non-invasively. To this end, a study was conducted in Holstein dairy cows (n=20) to determine if DITI of the vulva could be used to discriminate between estrus vs. diestrus phases of the bovine estrous cycle. Cows were synchronized (CIDR/PGF2 $\alpha$ ) and study measurements taken daily from the day after CIDR withdrawal (d 1) for 30 d as follows: ambient (AMB) temperature (TEMP), relative humidity (RH), rectal TEMP (RT), blood samples for serum concentrations progesterone (P4) determinations (analyzed by RIA), and vulva surface TEMP via DITI for minimum (MIN), maximum (MAX), average (AVG), and standard deviation (SD) of TEMP gradients. During this study it was noted that during the first estrus (ES-1) and early diestrus (DS-1) periods post-synchrony AMB TEMP (21.4  $\pm$  0.1 $^{\circ}$ C) was greater (P < 0.05) than during late diestrus (DS-2) and the return to estrus (ES-2; 12.1  $\pm$  0.2 $^{\circ}$ C). RT was also higher (P < 0.01) at ES-1/DS-1 (pooled: 24.5  $\pm$  0.05 $^{\circ}$ C) than ES-2/DS-2 (pooled 23.9  $\pm$  0.03 $^{\circ}$ C). Serum concentrations of P4 did not differ (P > 0.10) between ES-1 and ES-2 (pooled: 0.31  $\pm$  0.04 ng/ml), but were lower (P < 0.01) than DS-1 and DS-2 (pooled: 10.0  $\pm$  0.52 ng/ml), which also did not differ (P > 0.10). At an AMB TEMP of 21.4 $^{\circ}$ C, DITI MIN and SD revealed no TEMP difference (P > 0.10) between ES-1 and DS-1, whereas DITI MAX showed a greater (P < 0.01) TEMP at ES-1 (37.5  $\pm$  0.12 $^{\circ}$ C) compared to DS-1 (37.2  $\pm$  0.10 $^{\circ}$ C). At an AMB TEMP of 12.1 $^{\circ}$ C, MIN, MAX, AVG, and SD revealed no difference (P > 0.10) between ES-2 and DS-2. While DITI MAX TEMP revealed a 0.30 $^{\circ}$ C difference between ES-1 and DS-1, it is unclear whether this magnitude of a difference is physiologically relevant for the detection of estrus. Moreover, the ability to discriminate between estrus and diestrus was greatly influenced by ambient temperature.

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**Key Words:** Thermography, Estrus, Holstein

**M134 Activin inhibits the differentiation of bovine preadipocyte.** H. Matsumoto<sup>\*</sup>, S. Hirai, H. Kawachi, T. Matsui, and H. Yano, *Kyoto University, Kitashirakawa-oiwake, Sakyo-ku, Kyoto, Japan.*

The development of beef marbling is associated with an increase in adipocyte number within muscle, suggesting that the differentiation of preadipocyte could occur within the muscle during the formation of beef marbling. Thus, the regulation of adipogenesis would be beneficial for controlling intramuscular fat deposition. Activin, a member of TGF- $\beta$  superfamily was found in many tissues including muscle and fat, which increased the interest in local effects of activin. Recently, activin A was reported to inhibit the differentiation of a mouse preadipocyte cell line, 3T3-L1 preadipocyte (Hirai et al., *Mol. Cell Endocrinol.* in press). However, the action of activin A on the differentiation of bovine preadipocyte has not been studied. We investigated the effects of activin A on the differentiation of bovine preadipocyte. Stromal vascular (SV) cells were isolated from bovine adipose tissue and were cultured in DMEM with 5% FBS and 100  $\mu$ M vitamin C. After confluence, differentiation was induced by 0.5 mM 1-methyl-3-isobutyl-xanthine, 0.25  $\mu$ M dexamethasone, 2.5  $\mu$ g/ml insu-

lin, and 5  $\mu$ M troglitazone for 2 days. Then, the cells were cultured for 6 days in DMEM with FBS, vitamin C, troglitazone and insulin. Activin A was treated throughout the differentiation period. The cells were harvested for the measurements of differentiation markers such as glycerol-3-phosphate dehydrogenase (GPDH) activity, lipid accumulation, and the expression of adipocyte fatty acid-binding protein (aP2) mRNA. Activin A treatment reduced GPDH activity, lipid accumulation, and aP2 mRNA level. These results suggest that activin A inhibits the differentiation of preadipocyte in bovine SV cells. The differentiation of bovine preadipocyte was inhibited by the treatment with 5 ng/ml activin A. On the other hand, 20 ng/ml activin A was required to reduce the level of these adipocyte markers in 3T3-L1 cells. Therefore, bovine preadipocyte may be more sensitive to activin A than 3T3-L1 cells.

**Key Words:** Activin, Bovine, Preadipocyte

**M135 Effect of tumor necrosis factor  $\alpha$  on the development of *in vitro* derived bovine embryos.** L. Gast<sup>\*</sup> and C. Whisnant, *North Carolina State University, Raleigh.*

Tumor Necrosis Factor alpha (TNF-  $\alpha$ ) is a cytokine released by macrophage cells in response to local infection or disease. Mastitis, a common infectious disease in cattle, has been shown to increase the blood levels of TNF-  $\alpha$  mRNA. This immune response pathway leads to an increase in other cytokines and prostaglandins which can compromise oocyte maturation and embryo development. The objective of this study was to improve our understanding of the effects of TNF-  $\alpha$  on embryo development. Ovaries were obtained from a local abattoir and immature cumulus oocyte complexes (COC) were aspirated from 2-10mm diameter follicles. For maturation, COCs were placed in M199 including fetal bovine serum (FBS), LH, FSH, Estradiol, and Gentamycin additives and incubated at 39 $^{\circ}$ C for 18-20 hours. After maturation and fertilization, 3 groups of 20-25 presumptive zygotes were randomly placed in control developmental media (M199 plus FBS) or developmental media containing either 50ng/ml or 100ng/ml of TNF-  $\alpha$  and incubated at 39 $^{\circ}$ C. Cleavage rate was evaluated 72 hpi and was not affected by the treatments. Developmental data was collected by observation of blastocyst stages (compact morula, early blastocyst, mid-blastocyst, late blastocyst, or hatching blastocyst) at 168hpi and 196hpi. At 168hpi, embryos exposed to 50ng/ml TNF-  $\alpha$  developed similarly to the control embryos, unlike the embryos exposed to 100ng/ml TNF-  $\alpha$ , which had a 10% decrease in the number of blastocysts. The total percent of all blastocyst stages observed in the treatment groups at 168hpi are as follows: Control 32%, 50ng/ml TNF-  $\alpha$  32%, and 100ng/ml TNF-  $\alpha$  22%. At 196hpi, embryos treated with 100ng/ml TNF-  $\alpha$  exhibited a further 7% decrease in observed blastocysts. The total percent of all blastocyst stages observed at 196hpi were control 22%, 50ng/ml TNF-  $\alpha$  30%, and 100ng/ml TNF-  $\alpha$  15%. The control group showed a 10% decrease in total number of blastocysts observed at 196hpi and may be caused by degeneration of some developing blastocysts. In summary, higher doses of TNF-  $\alpha$  decreased bovine embryo development, whereas some lower doses can actually increase embryonic development *in vitro* (P<.05).

**Key Words:** Cytokine, Bovine, IVF

**M136 Effects of feeding yeast culture and propionibacteria on milk glucose, plasma glucose and plasma insulin concentrations in Holstein cows.** K. V. Lehloenya<sup>1</sup>, D. R. Stein<sup>1</sup>, M. M. Aleman<sup>\*1</sup>, D. T. Allen<sup>1</sup>, T. G. Rehberger<sup>2</sup>, D. A. Jones<sup>1</sup>, and L. J. Spicer<sup>1</sup>, <sup>1</sup>Oklahoma State University, Stillwater, <sup>2</sup>Agtech Products, Inc., Wakesha, WI.

To determine the effect of supplemental feeding of Diamond V-XP Yeast Culture (XPY) alone or in combination with Propionibacteria strain P169 on concentrations of glucose in milk and plasma and insulin in plasma, 31 primiparous (PP) and multiparous (MP) Holstein cows were fed one of three dietary treatments between 2 wk prepartum to 30 wk postpartum: 1) Control (n=10), fed a corn silage-based total mixed ration (TMR); 2) XPY (n=11), fed Control TMR plus XPY (at 56 g/head/d); and 3) XPY+P169 (n=10), received Control TMR plus XPY plus P169 (at 6 x 10<sup>11</sup>/head/d). Milk samples during sequential

p.m. and a.m. milkings were collected during a 2-wk period (wk 23 and 24 of lactation) from all cows, and blood samples were collected hourly during a 16-h post-feeding interval during wk 27 of lactation from only MP cows. Milk glucose was affected ( $P<0.01$ ) by dietary treatment such that both PP and MP cows fed XPY+P169 had 28% greater ( $P<0.05$ ) milk glucose levels ( $251\pm$  mg/dL) than Control cows and 32% greater milk glucose levels than XPY-fed cows. Diurnal plasma glucose concentration ( $59\pm 1$  mg/dL) was not affected by diet in MP cows. Plasma insulin levels were affected ( $P<0.01$ ) by dietary treatment and time such that plasma insulin levels in MP cows fed XPY+P169 ( $0.86\pm 0.05$  ng/mL) were 34% and 30% greater ( $P<0.01$ ) than in MP cows fed Control and XPY, respectively. Although 16-h post-feeding fluctuations in plasma glucose did not significantly differ among Control, XPY and XPY+P169 groups, the lack of a detectable increase in plasma glucose may be due, in part, to the fact that plasma insulin levels increased faster in XPY+P169 fed cows. Milk glucose and plasma insulin responses to XPY+P169 feeding suggest that XPY+P169 supplementation might have enhanced gluconeogenesis and increased glucose uptake by the mammary gland.

**Key Words:** Yeast Culture, Propionibacteria, Glucose

**M137 Supplemental feeding of propionibacteria to lactating dairy cows: Effects on plasma hormones and metabolites.** M. M. Alemán<sup>\*1</sup>, D. R. Stein<sup>1</sup>, D. T. Allen<sup>1</sup>, K. W. Gates<sup>1</sup>, K. J. Mertz<sup>2</sup>, T. G. Rehberger<sup>2</sup>, D. A. Jones<sup>1</sup>, and L. J. Spicer<sup>1</sup>, <sup>1</sup>Oklahoma State University, Stillwater, <sup>2</sup>Agtech Products, Inc., Waukesha, WI.

From 14 d prepartum to 175 d postpartum, multi- (MP) and primiparous (PP) Holstein cows were fed one of three dietary treatments: 1) Control (n=13), fed a

total mixed ration (TMR); 2) HP169 (n=11), fed TMR plus  $6 \times 10^{11}$ /head/d (high dose) of Propionibacterium Strain P169; or 3) LP169 (n=14), fed TMR plus  $6 \times 10^{10}$ /head/d (low dose) of P169. Blood samples were collected weekly for 25 wk and analyzed for plasma concentrations of glucose, insulin, insulin-like growth factor-I (IGF-I), leptin, nonesterified fatty acids (NEFA) and cholesterol (CHOL). Between wk 25 and 30 bovine somatotropin (bST) was given to all groups every 2 wk. Plasma glucose was affected by diet x parity ( $P<0.001$ ) such that glucose levels in LP169 MP cows ( $59.8\pm 1.1$  mg/dL) were 5.5% lower than in HP169 MP cows; LP169 PP cows ( $67.9\pm 0.9$  mg/dL) had 6% to 9% greater plasma glucose levels than HP169 and Control PP cows. Plasma insulin was affected by diet ( $P<0.001$ ) such that LP169 had less plasma insulin than HP169 and Control cows (during wk 13-25), and HP169 cows had greater insulin than Controls (during wk 1-12). Plasma IGF-I, NEFA and leptin levels did not differ ( $P>0.15$ ) among diet groups between wk 1 and 25, but PP cows had greater ( $P<0.02$ ) IGF-I and lower ( $P<0.01$ ) NEFA levels than MP cows. Plasma CHOL was affected by diet x parity ( $P<0.01$ ) such that LP169 MP cows ( $246\pm 11$  mg/dL) had 25% greater levels than HP169 and Control MP cows; CHOL levels in PP cows did not differ among diet groups. During bST, HP169 MP cows and LP169 PP cows had lower ( $P<0.01$ ) IGF-I levels than their respective Controls. Regardless of parity, LP169 cows had greater ( $P<0.10$ ) leptin levels than Controls cows, and HP169 cows had greater ( $P<0.01$ ) NEFA than Control cows. We conclude that P169 may hold potential as a direct-fed microbial to enhance metabolic efficiency during early and mid-lactation.

**Key Words:** Propionibacteria, Direct-fed Microbial, Hormones

## Production, Management and the Environment: Environment and Economics

**M138 Effects of population density on growth and vermicompost production of earthworms (*Eisenia spp.*).** J. Hernández<sup>\*1</sup>, S. Pietrosevoli<sup>1</sup>, W. Echeverría<sup>2</sup>, R. Palma<sup>2</sup>, A. Faria<sup>1</sup>, C. Contreras<sup>2</sup>, and A. Gomez<sup>1</sup>, <sup>1</sup>La Universidad del Zulia, Maracaibo, Zulia, Venezuela., <sup>2</sup>Proyecto FONACIT PSI-200000792, Maracaibo, Zulia, Venezuela.

During 84 days, in an area classified as dry forest of Zulia state, Venezuela; a medium scale experiment was performed in order to evaluate the effects of three population densities: low, medium and high (1000, 2000 and 4000 earthworm/m<sup>2</sup> respectively) on earthworm biomass and superficial vermicompost production. Initial biomass was  $166.53 \pm 11.34$  mg/earthworm. Experimental design was a completely randomized with five replicates. Earthworms were maintained in concrete bins with a precomposted mixture 50:50 of oil palm fiber and bovine manure. Total substrate offered was  $0.220 \text{ m}^3$  /bin. Every 21 days, biomass of the first 20 earthworms founded in the upper area of the bins was recorded; and free surface vermicompost was collected having its volume measured. Data was analyzed using STATISTIX software. Statistical differences were established among treatments for both earthworm biomass  $327.98 \pm 46.08$ ;  $278.9 \pm 47.8$  and  $198.4 \pm 31.4$  mg/earthworm and total vermicompost production  $0.039 \pm 0.004$ ;  $0.045 \pm 0.002$  and  $0.049 \pm 0.035 \text{ m}^3$  for low, medium and high density respectively. Tukey media test not showed differences between medium and low densities for biomass, and neither between medium and high density for vermicompost production. Population density of earthworms affected biomass and vermicompost production, with the lowest biomass/earthworm with high density treatment, which achieved the best vermicompost production.

**Key Words:** Population Density, Earthworms, Biomass and Vermicompost

**M139 Effects of feeding frequency on growth and reproduction of earthworms (*Eisenia spp.*).** J. Hernández<sup>1</sup>, S. Pietrosevoli<sup>\*1</sup>, A. Faria<sup>1</sup>, R. Palma<sup>2</sup>, and R. Canelón<sup>2</sup>, <sup>1</sup>La Universidad del Zulia., Maracaibo, Zulia, Venezuela., <sup>2</sup>Proyecto FONACIT PSI- 200000792, Maracaibo, Zulia, Venezuela.

During 105 days, in an area classified as dry forest of Zulia state, Venezuela; a medium scale experiment was performed in order to evaluate the effects of three feeding frequency: once (100% substrate), three (50, 25 and 25 % substrate) and four times substrate supply (25, 25, 25 and 25 % substrate) on earthworm biomass, total biomass/feeding frequency and cocoons production. Total substrate offered was  $0.220 \text{ m}^3$ /bin. Initial density and biomass were 1000 earthworm/m<sup>2</sup> and  $234.88 \pm 19.93$  mg/earthworm respectively. Experimental design was a completely randomized with five replicates. Earthworms were maintained in concrete bins with a precomposted mixture 50:50 of oil palm fiber and bovine manure. Every 21 days, biomass of three groups of 100 earthworms each was recorded. On day 42, cocoons founded on the upper 10 cm of the bins were collected. At the end of the trial after capturing all the specimens in each bin, total earthworm biomass was registered. Data was analyzed using STATISTIX software. Statistical differences were established among treatments for earthworm biomass  $136.49 \pm 12.29$ ;  $147.95 \pm 11.92$  and  $172.56 \pm 12.46$  mg/earthworm for once, three and four times feed supply respectively. Tukey media test not showed differences between once and three times supply. Cocoons production ( $345 \pm 155.37$ ;  $363.6 \pm 108.47$  and  $168 \pm 66.126$  cocoons for once, three or four time feed supply respectively) and total biomass ( $426.21 \pm 121.26$ ;  $383.51 \pm 59.13$  and  $342.15 \pm 190.32$  g/bin for once, three or four time feed supply respectively) were not influenced by treatments ( $p \leq 0.07$  and  $p \leq 0.06$ ). Feeding frequency affected earthworm biomass. The highest individual biomass was registered when feed was supplied four times. Benefits obtained in increasing feed frequency are not equilibrated with increasing of management task required.

**Key Words:** Feeding Frequency, *Eisenia* Earthworms, Biomass and Cocoons Production

**M140 Evaluation of advanced dairy systems shade tracker fans and korral kool coolers on a commercial dairy in Buckeye, Arizona.** M. VanBaale<sup>1</sup>, D. Ledwith<sup>1</sup>, R. Burgos<sup>\*1</sup>, R. Collier<sup>1</sup>, D. Armstrong<sup>1</sup>, J. Smith<sup>2</sup>, M.