Physiology and Endocrinology: Animal Physiology

523   The "immunocrit," a simple measure of passive transfer, is a useful predictor of nursing ability and preweaning mortality of piglets.   J. L. Vallet*, J. R. Miles, L. A. Rempel, and L. A. Kuehn, *USDA, ARS, U.S. Meat Animal Research Center*, *Clay Center, NE*.

Initiation of lactation and newborn piglet nursing ability are 2 factors that can influence preweaning mortality. We have developed the "immunocrit" that can assess both lactation initiation and neonatal piglet nursing ability based on the transfer of immunoglobulin G (IgG) from the sow to the piglet. To perform an immunocrit, 50 μL serum was mixed with 50 μL 40% ammonium sulfate, the mixture was loaded into a hematocrit capillary tube and centrifuged for 5 min. The result was the ratio of the mm precipitate to mm solution in the tube. To test the immunocrit, the smallest piglet in 205 litters was sacrificed, blood was collected and full and emptied stomachs were weighed to obtain the weight of stomach contents. Blood was analyzed for IgG by precipitation of serum samples with protein A-Sepharose followed by SDS-PAGE. Densitometry of the heavy chain of IgG was used to quantify IgG. To test use in cattle, blood samples from 96 calves were obtained 24 h after birth and IgG was measured (Bovine IgG radial immunodiffusion kit; VMRD, Inc.). Piglet and calf blood samples were also analyzed by immunocrit. For piglets, the correlation between densitometry and immunocrit values was 0.83. For calves, the correlation between kit IgG and immunocrit values was 0.90. Piglet immunocrit values were also correlated (r = 0.44; $P < 0.01$) with stomach contents at 24 h, indicating that immunocrits could be used to screen for piglet nursing ability. To assess the influence of immunocrit values on piglet preweaning survival, immunocrits were performed on every piglet in 48 1st and 68 2nd parity litters and survival to weaning was recorded. Preweaning survival was independently associated ($P < 0.01$) with birth weight of the piglet and immunocrit values. Second parity sows had greater litter average immunocrit values compared with 1st parity sows (0.133 ± 0.003 and 0.123 ± 0.004, respectively; $P < 0.05$). These results indicate that the immunocrit is a useful tool to monitor colostrum intake in piglets and calves.

Key Words: colostrum, immunoglobulin, lactation

524   Influence of temperament on stress hormone and IgG concentrations in Brahman calves.   N. C. Burdick*[1], D. A. Neuendorff[2], R. C. Vann[3], J. G. Lyons[1], T. H. Welsh Jr.[1], and R. D. Randel[2], [1]*Texas AgriLife Research*, *College Station*, [2]*Texas AgriLife Research*, *Overton*, [3]*MAFES, Mississippi State University*, *Raymond*.

This study was designed to determine the influence of temperament on cortisol, epinephrine (EPI), norepinephrine (NE), and IgG concentrations in Brahman calves. Calves from crops in 2006 and 2007 were selected based on temperament score measured 28 d before and at weaning. Based on temperament score the 10 calm, intermediate, and temperamental calves from each sex (bulls and heifers) were selected from each calf crop (n = 120). Blood was collected 28 d before weaning, at weaning, and 28 and 56 d post-weaning to determine serum cortisol and IgG, and plasma EPI and NE concentrations. Data were analyzed using the MIXED procedure of SAS specific for repeated measures. Sources of variation included temperament, sex, day, and year. Cortisol ($P < 0.01$) was, and EPI tended ($P = 0.08$) to be, greater in 2006 than 2007. Cortisol was affected by temperament with calm calves having lower cortisol (14.4 ± 0.7 ng/mL; $P < 0.01$) than intermediate (18.6 ± 0.7 ng/mL) and temperamental (30.8 ± 0.7 ng/mL) calves. Heifers had greater cortisol than bulls ($P < 0.01$; 22.9 ± 0.6 and 19.6 ± 0.6 ng/mL, respectively). Cortisol increased from d0 (18.5 ± 0.8 ng/mL) through d+56 ($P < 0.01$; 25.2 ± 0.8 ng/mL). Calm calves had lower EPI (149 ± 18 pg/mL) than intermediate (203 ± 18 pg/mL) and temperamental calves ($P < 0.01$; 381 ± 18 pg/mL). Heifers had greater EPI ($P < 0.01$; 278 ± 14 pg/mL) than bulls (212 ± 15 pg/mL). Concentrations of EPI decreased from d-28 (326 ± 20 pg/mL) through d+56 ($P < 0.01$; 169 ± 22 pg/mL). Calm calves had lower concentrations of NE than intermediate and temperamental calves ($P < 0.04$; 510 ± 72, 732 ± 74, and 719 ± 72 pg/mL, respectively). Concentrations of NE changed over time ($P = 0.01$) and were not affected by sex ($P = 0.98$). Concentrations of IgG were not affected by temperament ($P = 0.43$) or time ($P = 0.22$) but tended to be affected by sex with heifers (26.7 ± 2.2 mg/mL) having greater IgG than bulls (21.6 ± 2.2 mg/mL; $P = 0.11$). In summary, temperamental calves have greater stress hormone concentrations. Additionally, cortisol, EPI, and IgG concentrations can vary depending on sex of the calf.

Key Words: cattle, temperament, stress

525   Effect of cytochrome P450 and aldo-keto reductase inhibitors on progesterone decay in primary bovine hepatic cell cultures.   C. O. Lemley* and M. E. Wilson, *West Virginia University*, *Morgantown*.

Progesterone is required for maintenance of pregnancy and peripheral concentrations of progesterone are affected by both production and inactivation. Hepatic cytochrome P450 (CYP) and aldo-keto reductases (AKR) play a pivotal role in the first step of steroid inactivation. The current objectives were to discern the proportional involvement of hepatic progesterone catabolic enzymes on progesterone decay using specific enzyme inhibitors. Liver biopsies were taken from 6 lactating dairy cows and dissociated using a non-perfusion technique. Confluent wells (n = 12/treatment) were preincubated for 4 h with enzyme inhibitor and then challenged with progesterone for one hour. Cell viability was unaffected ($P > 0.50$) by inhibitor treatment and averaged $84 \pm 1\%$. In control wells 50% of the progesterone had been inactivated after a one hour challenge with 5 ng/ml progesterone. Preincubation with curcumin (CYP and AKR inhibitor), ticlopidine (CYP2C inhibitor) or naproxen (AKR inhibitor) caused the greatest reduction ($P < 0.001$) in progesterone decay compared with controls and averaged 77, 39 or 37%, respectively. Hydroxylation of 4-nitrophenol to 4-nitrocatechol in intact cells was inhibited by 65% ($P < 0.001$) after treatment with curcumin or ticlopidine. However, phase II glucuronidation of phenol red or 4-nitrocatechol in intact cells was inhibited ($P < 0.01$) by treatment with curcumin, dicumarol or naproxen, showing a lack of specificity in phase I enzyme inhibition (CYP and AKR). The contribution of CYP2C and CYP3A enzymes to progesterone decay in bovine hepatic cell cultures appeared to be 40 and 15%, respectively. Depending on the inhibitor used it would appear that the AKR enzymes contributed 40% to the observed progesterone decay; however, a portion of this loss may be due to glucuronosyltransferase (phase II enzyme) inhibition. A greater understanding of these steroid biotransformation pathways in the dairy cow could help researchers modify the bioavailability of progesterone.

Key Words: cytochrome P450, aldo-keto reductase, progesterone decay

526   Residual feed intake selection and its effects upon pre- and postpartum changes in NEFA concentrations and body weight and condition in Brahman females.   A. K. Poovey*[1,2], A. N. Loyd[1,2], A. W. Lewis[1], D. A. Neuendorff[1], S. L. Morgan[1,2], L. C. Caldwell[2],

T. D. A. Forbes[3], T. H. Welsh Jr.[2], and R. D. Randel[1], [1]*Texas AgriLife Research*, *Overton*, [2]*Texas AgriLife Research, College Station,* [3]*Texas AgriLife Research*, *Uvalde*.

Residual feed intake (RFI) is one method to identify efficient animals based upon the relationship of predicted to actual individual feed intake. Nonesterified fatty acid (NEFA) concentrations are the products of catabolism of triglycerides and are negatively correlated with energy balance and body weight in females. The objective of this study was to examine the relationship among RFI status with pre- and postpartum NEFA concentrations in beef females. Based upon prior post-weaning RFI evaluations, Brahman females (n = 93) were classified as having either a negative RFI (efficient) or a positive RFI (inefficient). Body condition score (BCS), body weight (BW), and serum samples were collected at weekly intervals beginning 5 wk before and continuing through 5 wk after calving. Pre-calving, NEFA concentrations did not differ between low and high RFI groups ($P > 0.3$). There was an interaction between NEFA concentrations and prepartum time period ($P = 0.02$). However, during this time, RFI ($P = 0.8961$) and age group ($P = 0.11$) did not affect change in BW although there tended to be differences in BW between 2 year old (YO) heifers and cows 4 years and older ($P = 0.05$). Similarly, change in BCS was not affected by RFI ($P = 0.88$); however, age group tended to differ ($P = 0.06$). Specifically, 2 YO cows lost more BCS than 3 YO ($P = 0.02$) and 3 YO tended to lose more BCS than cows 4+ YO ($P = 0.07$). Postpartum NEFA concentrations changed over time ($P = 0.02$) and were affected by sampling week ($P = 0.02$). Neither age nor RFI influenced changes in BW ($P = 0.75$; $P = 0.98$, respectively) or BCS ($P = 0.29$; $P = 0.79$, respectively). Circulating concentration of NEFA in pre- and postpartum females was not related to RFI previously evaluated when the females were heifers. Changes in BW and BCS did not differ due to previous evaluation for RFI as heifers during the same peri-partum periods. Age of cow was the only factor found to be related to serum NEFA concentrations or changes in BW and BCS.

Key Words: feed efficiency, nonesterified fatty acids, peri-partum cows

527   Ruminal degradability and intestinal release of different vitamin A formulations.   D. P. Preveraud* and P. A. Geraert, *Adisseo France SAS*, *Antony, France*.

The most convenient way to provide vitamin A (VA) to ruminants is to include it with concentrate mixtures. Feed industry usually formulates a VA ester emulsified into a gelatin beadlet to ensure good stability and biological availability. The aim of this study was to evaluate the rumen degradability and intestinal release of different VA formulations, including the double emulsion (Microvit A1000 Supra, Adisseo), by using a Mobile Nylon Bag (MNB) methodology. Two dairy cows equipped with ruminal and duodenal cannulas were used in a 2-period crossover experimental design. The 9 tested formulations were incubated in the rumen in nylon bags (6 replicates) for 6, 10 or 18h and residues from these bags were then incubated in a pepsin HCl solution (38.5°C, pH 2) for 2.5h to simulate abomasal digestion, and finally inserted through the duodenal cannula to be recovered from feces after 24h. Nylon bags were then analyzed for their VA content and results were expressed as a percentage of residual VA compared with initial content of non-incubated product (see table). Microscopic photographs were also taken to follow the degradation of the different formulations. The overall results of this experiment indicated that these VA sources could be divided into 3 groups according to their rumen stability and in relation with their formulation: stable (double emulsion, Microvit), intermediate (products A, B, C, G and H) and sensitive (products D, E and F). Despite a low

(14%) but significant loss of the double emulsion formulation after 18h incubation in rumen, 81% of the initial amount finally reaches the intestine where VA is totally released. Among the sources of vitamin A being studied, the double emulsion formulation seems to be more bioavailable to cattle. This should be confirmed later on by a greater tissue concentration of VA.

Table 1. VA residues[1] at each step of the MNB technique (% of initial content)

	6-h rumen	10-h rumen	18-h rumen	abomasum	feces
Microvit A1000 Supra	102[l]	97[l]	86[ijk]	81[hi]	ND
product A	76[ghi]	57[f]	23[d]	2[a]	ND
product B	82[hij]	72[gh]	20[cd]	ND	ND
product C	70[g]	45[e]	10[abc]	ND	ND
product D	26[d]	6[a]	ND	ND	ND
product E	24[d]	4[a]	ND	ND	ND
product F	18[bcd]	18[bcd]	19[cd]	7[ab]	3
product G	51[ef]	20[cd]	ND	ND	ND
product H	76[ghi]	70[g]	53[ef]	24[d]	ND

[1]Values are LS means; ND: Non Detectable; letters indicate statistical difference within columns (Fisher Test: $P < 0.05$; α = 5%)

Key Words: vitamin A formulation, rumen stability, intestinal release

528   Poisson analysis of number of services per conception for Iranian Holstein cows.   H. Farhangfar*[1] and F. Bahri[2], [1]*Birjand University, Birjand, Iran*,

[2]*Ferdowsi University of Mashhad, Mashhad, Iran*.

To evaluate the effects of some environmental factors on the number of services per conception (NSC) in Iranian Holstein cows, a total of 38,074 records obtained from 10,726 cows at different parities (1985–2009) was utilized. All records were collected from a very large dairy herd comprising of 2 separate units. The units were different from each other from dairy farm management point of view. Average number of services per conception was 2.29 in the whole data set. A generalized statistical linear model was applied to analyze NSC. Poisson distribution was assumed for NSC and a log link function was applied for the response variable. In the model, fixed effects of period of years (in 3 levels), month of insemination, technician, parity and unit were included. Based upon the year of insemination, 3 year periods were defined as follows: 1985–1990, 1991–2000 and 2001–2009. The statistical model was run with the use of GLMMIX procedure add in program implemented in SAS software. The results indicated that year period and unit were the only significant factors that influenced NSC ($P < 0.01$). Least squares means of NSC (in original scales) were 2.1776, 2.2807 and 2.2805 for year periods of 1985–1990, 1991–2000 and 2001–2009, respectively. The least squares means of NSC (in original scales) of farms units were obtained to be 2.1707 and 2.3234 for units 1 and 2, respectively revealing slightly different reproductive efficiency between 2 units of the dairy herd. Among different parities, the largest and lowest NSC least squares means were found for parity 7 and 10, respectively. However, no significant differences were revealed among the parities. From the results of this research it could be concluded that the reproductive efficiency of Iranian Holstein cows has been deteriorating over the past decades suggesting that a sophisticated genetic selection program

should be considered for improving the traits associated with reproduction performance.

529 Effects of continuous infusion of tumor necrosis factor-alpha (TNFα) into adipose tissue on glucose and fatty acid metabolism in lactating dairy cattle. C. A. Martel*[1], L. K. Mamedova[1], E. J. Minton[1], M. L. Jones[2], J. A. Carroll[3], and B. J. Bradford[1], [1]*Department of Animal Sciences and Industry, Kansas State University, Manhattan,* [2]*Veterinary Medical Teaching Hospital, Kansas State University, Manhattan,* [3]*Livestock Issues Research Unit, ARS-USDA, Lubbock, TX.*

Late-lactation Holstein cows (n = 9/treatment) were used to evaluate effects of TNFα administration on glucose and fatty acid (FA) metabolism. Cows were blocked by feed intake and milk yield and randomly assigned within block to 1 of 3 treatments: control, TNFα, and pair-fed control. Treatments (4 mL saline or 14 μg/kg TNFα in 4 mL saline) were infused continuously over 7 d via 2 osmotic pumps in the adipose layer in the tailhead region. Plasma, milk samples, milk yield, and DMI data were collected daily. On d 7, pumps were removed and liver and contralateral tailhead adipose samples were collected. Results were modeled with fixed effect of treatment and random effect of block; P values >0.10 were considered non-significant. TNFα did not alter adipose or liver TNFα mRNA abundance, plasma TNFα, IL-4, IL-6, or interferon-γ concentrations, DMI, or rectal temperature. Milk fat and lactose concentrations decreased with TNFα ($P < 0.05$), but milk yield was unchanged and treatments did not alter the proportion of short vs. long-chain FA in milk on d 7. Treatments did not alter plasma NEFA concentration, liver triglyceride content, or adipose mRNA abundance for hormone-sensitive lipase or perilipin. Plasma glucose turnover rate, as measured by disappearance of U-^{13}C-glucose bolus, was not altered by treatment, nor was liver mRNA abundance for phosphoenolpyruvate carboxykinase or pyruvate carboxylase. However, TNFα tended to decrease adipose TNFα mRNA abundance ($P = 0.09$) and increase liver IL-10 mRNA abundance ($P = 0.05$) compared with controls. This TNFα delivery protocol may have allowed for an adaptive anti-inflammatory response to suppress systemic inflammation, which may account for the lack of metabolic responses, in contrast with previous responses to daily subcutaneous TNFα injections.

530 Reproductive rate of semi-free ranging Bison (*Bison bison*) at the National Bison Range. M. J. Borgreen*[1,2], T. J. Roffe[2], E. M. Berry[1], R. B. McCosh[1], and J. G. Berardinelli[1], [1]*Montana State University, Bozeman,* [2]*US Fish and Wildlife Service, Bozeman, MT.*

Recruitment of calves at the National Bison Range (NBR) near Moiese, Montana, has dropped from the historic average of 87 to 33 calves per 100 breeding-age cows in 2008. The purpose of monitoring the NBR bison pregnancy rate (PR) and calf recruitment is to determine where in the reproductive cycle NBR female bison fail to recruit calves. The reproductive cycle was divided into 3 stages: conception to early embryonic development; maintenance of pregnancy during the second and third trimesters; and, calving to recruitment. In 2008, transrectal ultrasonography was used to determine PR in cows (ages 4 to 12 yr) in October; 28 of 41 cows (68%) were pregnant. Pregnant cows were painted with a unique bleach number. Fecal samples were collected in Oct., Jan., Mar. and Apr. until the bleach number was illegible. Fecal samples were analyzed for progesterone (P4). Pregnancy rates estimated by fecal P4 concentration decreased ($P < 0.01$) from 100% (n = 28) in Oct. to 53% (n = 15) in Apr. The percentage lost continuously decreased throughout the second stage, with the largest percent decrease between first and second trimester (17%). Of the original 28 pregnant cows the PR in April was 53%; a reduction of 47%. This closely matched calf recruitment of the herd at the 2009 roundup (71 calves for 126 cows; 56%): indicating that the accuracy for estimating pregnancy using fecal P4 was 94.7%. In 2009, PR was determined by ultrasonography of 89 cows, including 38 of the 41 cows from 2008. Pregnancy rate for 2009 was 63%, which was similar to PR in 2008 (68%). Radio collars were secured to 27 pregnant and 10 non-pregnant cows. These animals will be monitored throughout the rest of the reproductive cycle to determine calf production using fecal P4 assay. Fecal P4 assays appear to give an accurate estimate of PR in semi-free ranging bison. In conclusion, it appears that the decrease in calf recruitment at the NBR can be, at least in part, due to fetal losses during gestation.

J. Anim. Sci. Vol. 88, E-Suppl. 2/J. Dairy Sci. Vol. 93, E-Suppl. 1/Poult. Sci. Vol. 89, E-Suppl. 1

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