Physiology Endocrinology and Metabolism

1397 Cloning and characterization of genomic fragments encoding a putative ovine epidermal growth factor puedogene. Sushil J. John* and Sylvie Bilodeau-Goeseels, Agriculture and Agri-Food Canada, Lethbridge Research Centre, Lethbridge, Alberta, Canada.

Mature epidermal growth factor (EGF) is a 53 amino acid polypeptide, known to stimulate cell proliferation as well as DNA synthesis in many cell types. In humans, it is processed from a 1200 amino acid precursor protein encoded by 24 exons within a 127 kb gene. The mature region is encoded by exons 20 and 21. EGF has been a potential feed additive since it has been shown to improve nutrient uptake and reduce intestinal infection in animals systems. Despite this economic promise, EGF sequence has not been reported from any ruminant sp. In this study, we report the first isolation and sequence characterization of genomic fragments encoding the mature region of a putative ovine EGF. We have isolated using PCR, two fragments spanning exons 20 and 21 respectively of the ovine EGF gene. Analysis of the nucleotide sequence from the two fragments shows 70% and 62% homology respectively with the published human EGF gene sequence. The exon-intron junctions are also conserved between the two species. Comparison of the deduced protein sequence of the ovine EGF with known EGF proteins from other species showed a homology varing from 38-40%, indicating an unique nature of ovine EGF protein. Although, some critical amino acids important for its function are conserved, presence of 3 stop codons in position 22, 33 and 41 are aspects that indicate this gene encodes a truncated, non-functional protein. In conclusion, we have isolated genomic fragments encoding the mature region of the putative ovine EGF gene, however analysis of the deduced protein sequence indicates that it is a pseudo-gene.

Key Words: Ovine, Epidermal Growth Factor, Genomic

1398 Expression of growth hormone receptor (GHR) 1A, IGF-I, total GHR and cyclophilin (cyclo) mRNA in hepatic tissue of periparturient Holstein cows. R. P. Radcliff*, B. L. McCormack, and M. C. Lucy, *University of Missouri, Columbia MO*.

Growth hormone plays a central role in the change in nutrient metabolism that occurs during the initiation of lactation. Growth hormone#s actions are mediated by GHR whose mRNA is present in three alternatively spliced forms; GHR 1A, 1B and 1C. Liver specific GHR 1AmRNA is transiently decreased around parturition but the exact timing of the decline is not known. Our objective was to characterize GHR 1A, total GHR (1A, 1B and 1C), IGF-I and cyclo mRNA expression in liver of periparturient dairy cattle. Liver biopsies (n = 143) were collected from sixty-six Holstein cows at the University of Missouri Dairy Farm. At least two cows were sampled on each d from 15 d before to 14 d after parturition. Total cellular RNA was isolated and reverse transcribed to cDNA. Target cDNA was measured by quantitative real time polymerase chain reaction using probes and primers specific to each gene. The GHR 1A mRNA declined 2 d before parturition, was lowest 3 to 4 d after parturition and then increased (day; P<0.001). Mean GHR 1A for weekly periods relative to parturition (d #15 to #8, d #7 to #1, d 0 to 7 and d 8 to 14) was 34 \pm 3^a, 20 \pm 4^b, 3 \pm 4^c and 14 $\pm 4^{b}$ fg/ng cDNA respectively (means with different superscripts differ P<0.05). The IGF-I mRNA declined 1 d after parturition, was lowest 2 to 5 d after parturition and then increased (day; P<0.01). Mean IGF-I for weekly periods relative to parturition was 4.2 ± 0.4^a , 2.7 ± 0.5^{bc} , $1.4\,\pm\,0.5^{c}$ and $3.1\,\pm\,0.5^{ab}$ fg/ng cDNA. Total GHR mRNA was not affected by day (P>0.1). Cyclophilin was not a valid housekeeping gene because its mRNA increased from 2 wk before parturition (72 \pm 8^a, 99 $\pm 9^{b}$, 116 $\pm 10^{b}$ and 102 $\pm 10^{b}$ fg/ng cDNA; day, P<0.01). A detailed profile of GHR 1A, IGF-I, total GHR and cyclo mRNA expression during the periparturient period was provided. The decreases in GHR 1A and IGF-I during the transition period are rapid and occur immediately before (GHR 1A) or shortly after (IGF-I) parturition. Changes in IGF-I are correlated with changes in GHR 1A but not total GHR.

Key Words: GHR 1A, liver, cattle

1399 Inhibition of nitric oxide synthase increases glucose uptake and lipolysis in ovine hind-limb by a mechanism independant of insulin. J.J. Cottrell*^{1,2}, M.B. Mc Donagh², R.D. Warner^{1,2}, and F.R. Dunshea², ¹Victoria University, Werribee, Victoria, Australia, ²Natural Resources and Environment, Werribee, Victoria, Australia.

The jugular vein, abdominal aorta and lateral saphenous veins were catheterised in eight Border Leicester x Merino cross lambs (50-55 kg) to determine the effect of nitric oxide synthase (NOS) inhibition on hind-limb metabolism. Lambs were housed in metabolism crates with $ad\ libitum$ access to food and water. Two bleeds were conducted 3 days apart, starting on the 3rd day post-surgery. Lambs were infused with either the NOS inhibitor L-arginine methyl ester hydrochloride (L-NAME) 30mg/kg or saline via the jugular catheter in a balanced randomised crossover design. On each infusion day, blood samples were obtained between -60 to 360 minutes relative to the infusion. Post infusion area under the curve for acute and semi-acute (15-120 and 120-360 post-infusion, respectively) responses were calculated relative to mean pre-infusion concentrations of glucose, lactate and non-esterified fatty acids (NEFA). Arterial plasma samples were pooled over pre-infusion, acute and semi-acute phases for analyses of plasma insulin, all data was analysed with ANOVAs. L-NAME acutely increased the glucose arteriovenous difference (AVD) (-6.4 v. 2 mM.min for control and L-NAME treatment, respectively, P=0.049) indicating increased uptake of glucose by the hind limb. L-NAME also increased lipolysis and/or inhibited fatty acid uptake within the hind-limb, as shown by acute (1.8 v. -1.2 mM.min, P=0.06) and semi-acute (4.3 v. -5.2 mM.min, P=0.021) decreases in NEFA AVD. These metabolic changes were independent of insulin concentration, which was unchanged by L-NAME infusion (-42 v. -51 mU/mL.min, P=0.94, and -91 v. 66 mU/mL.min, P=0.35 for acute and semi-acute phases). L-NAME did not appear to alter hind limb glycogenolysis as indicated by unchanged lactate AVD ($1.4~\mathrm{v}$. 0.6 mM.min, P=0.82 and 4.5 v. -10.1 mM.min, P=0.26 for acute and semi-acute phases). In conclusion, NO regulates carbohydrate and fat metabolism in the hind limb of the lamb by a mechanism independent of insulin. Supported in part by Meat and Livestock Australia.

1400 Glucose and hormonal profiles of Large White and Genex-Meishan gilts in early and late gestation. C. Farmer*1 and J.R. Cosgrove2, 1 Agriculture and Agri-Food Canada, Dairy and Swine R & D Centre, Lennoxville, QC, Canada, 2 Genex Swine Group, Regina, SK, Canada.

This project was done to determine the effect of genotype, namely Large White (LW) vs Genex-Meishan (GM; containing 50% Meishan genetics) on circulating concentrations of glucose, insulin, IGF-I and cortisol in early and late pregnancy. Jugular canulae were inserted non-surgically in 8 LW and 9 GM gilts on days 37 and 106 of gestation to obtain serial blood samples on days 38 and 107. Three preprandial samples (the mean of which represented the baseline) were obtained at 0740, 0750 and 0800 as well as postprandial samples every 20 min thereafter until 1100. Glucose and insulin concentrations were measured in all samples while values for cortisol and IGF-I were determined on all preprandial samples and hourly on postprandial samples. All gilts were weighed and their backfat measured on days 38 and 107 of gestation. Average ages of gilts at mating were 240.6 and 245.3 days for LW and GM, respectively. GM gilts were lighter on days 38 (138.3 vs 151.4 kg, SEM = 3.4, P = 0.02) and 107 (173.4 vs 183.7 kg, SEM = 3.7, P = 0.07) of gestation and their backfat was greater on both days 38 (23.2 vs 17.3 mm, SEM = 1.3, P = 0.004) and 107 (24.3 vs 16.8 mm, SEM = 1.4, P = 0.002). There was a breed x stage of gestation effect on baseline IGF-I concentrations (P=0.019) with values being greater (P=0.003) in GM on day 38 only. Baseline concentrations for glucose (P < 0.001) and insulin (P =0.02) were lower in GM than in LW gilts on both days of gestation and baseline insulin was greater (P < 0.0001) on day 38 compared to day 107 of gestation in both breeds. There was a breed x stage of gestation interaction on the area under the curve (AUC) for IGF-I (P = 0.028). The AUC for postprandial values of IGF-I was greater (P = 0.004) in GM than LW on day 38 while that of glucose was lower (P < 0.001) in GM on both days; AUC for insulin and cortisol were unaffected by breed (P > 0.10). Glucose AUC was also altered by stage of gestation, being greater (P < 0.001) on day 107 than on day 38. Meishan-derived and LW gilts therefore differ in their metabolism and this varies with stage of gestation (Thanks to Genex Swine Group for the animals and to Shur-Gain for supplying the feed).

 $\textbf{Key Words:} \ \mathrm{Swine}, \ \mathrm{Gestation}, \ \mathrm{Meishan}$

1401 Effect of time and day of injection on plasma β -hydroxybutyrate, NEFA, and urea N during 14-day subcutaneous injections of several dosages of glucagon in dairy cows. G. Bobe*1, B. N. Ametaj², R. N. Sonon¹, D. C. Beitz¹, and J. W. Young¹, ¹lowa State University, Ames, IA, ²Purdue University, West Lafayette, IN.

We showed previously that single injections of 2.5 and 5 mg glucagon decreased plasma NEFA concentrations in dairy cows in midlactation. Furthermore, we showed that plasma β -hydroxybutyrate (BHBA) and NEFA concentrations are decreased only in cows with elevated plasma BHBA and NEFA concentrations, respectively, during 14-day continuous intravenous infusions of glucagon beginning at d 21 postpartum, whereas plasma urea N (PUN) concentrations were nor affected. We tested whether time and day of injection changes the plasma BHBA, NEFA, and PUN response during 14-day subcutaneous injections of several dosages of glucagon. Multiparous Holstein cows (n=9) were assigned randomly to 3 groups and received beginning d 8 postpartum 0 (Saline), 2.5 (7.5 mg/d Glucagon), or 5 mg (15 mg/d Glucagon) in 60 ml saline (pH 10.25) by subcutaneous injections of glucagon every 8 h for 14 d. Plasma BHBA, NEFA, and PUN responses were measured by taking 18 serial samples during the 8 h after the first (D1-2PM), second (D1-10PM), middle (D7-2PM), and third last (D13-2PM) injection. Glucagon injections decreased plasma BHBA and NEFA concentrations only in cows with elevated plasma BHBA (>8 mg/dl) and NEFA (>0.5 mM) concentrations, respectively (P < 0.1). Glucagon decreased primarily plasma BHBA concentrations 6-8 hrs after injections at d 7 and d 13 of the injection period and plasma NEFA concentrations for 1-4 hrs at the first injection day, with responses in greater magnitude and at earlier injection times in "15 mg/d Glucagon" cows ($P \leq 0.1$). Glucagon injections did not affect PUN concentrations (P > 0.1). We conclude that 5 mg subcutaneous glucagon injections every 8 h for 14 d beginning at d 8 postpartum decrease elevated plasma BHBA and NEFA concentrations more effectively than the 2.5 mg dosage in dairy cows. The effects of time and day of glucagon injections on plasma BHBA and NEFA concentrations depend on glucagon dosage, time postpartum, and plasma BHBA and NEFA concentrations. (Partly supported under CREES-USDA agreement 99-35005-8576).

Key Words: Fatty Liver, Glucagon, NEFA

1402 Effect of time and day of injection on plasma glucose and insulin during 14-day subcutaneous injections of several dosages of glucagon in dairy cows. G. Bobe*1, B. N. Ametaj², R. N. Sonon¹, D. C. Beitz¹, and J. W. Young¹, ¹lowa State University, Ames, IA, ²Purdue University, West Lafayette, IN.

We showed previously that single injections of 2.5 and 5 mg glucagon increased plasma glucose and insulin concentrations in midlactation dairy cows for 5 and 2 h, respectively. Furthermore, we showed that plasma glucose and insulin response changed during 14-day continuous intravenous infusions of glucagon beginning at d 21 postpartum. We tested whether time and day of injection changes the plasma glucose and insulin response during 14-day subcutaneous injections of several dosages of glucagon. Multiparous Holstein cows (n=9) were assigned randomly to 3 groups and received beginning d 8 postpartum 0 (Saline), 2.5 (7.5 mg/d Glucagon), or 5 mg (15 mg/d Glucagon) in 60 ml saline (pH 10.25) by subcutaneous injections of glucagon every 8 h for 14 d. Plasma glucose and insulin response were measured by taking 18 serial samples during the 8 h after the first (D1-2PM), second (D1-10PM), middle (D7-2PM), and third last (D13-2PM) injection. Glucagon injections of 2.5 and 5 mg increased plasma glucose concentrations for 3 and 4hrs, respectively, and plasma insulin concentrations for 3 and 5 hrs, respectively, at 2 PM but not at 10 PM ($P \leq 0.1$). The time effect of glucagon was associated with increased plasma glucose and insulin concentrations prior to injection at 10 PM ($P \leq$ 0.1). During the glucagon injection period, the length of the plasma glucose and insulin response increased and the peak response decreased but increased in significance, with responses in greater magnitude and at earlier injection times in "15mg/d Glucagon" cows ($P \leq 0.1$). Elevated liver TAG concentrations (>15 mg TAG/g wet weight) diminished plasma glucose and insulin peak responses to glucagon, in particular in "7.5 mg/d Glucagon" cows ($P \leq 0.1$). We conclude that cows in the early postpartal period and cows with fatty liver have a decreased response to glucagon injections that can be overcome by increased glucagon dosages. Time and day of glucagon injections affect plasma glucose and insulin responses to glucagon injections with greater increases in plasma glucose and insulin concentrations with higher glucagon dosages, progressing lactation, and lower liver TAG, plasma glucose and insulin concentrations. (Partly supported under CREES-USDA agreement 99-35005-8576).

Key Words: Fatty Liver, Glucagon, Insulin

1403 Effect of oxytocin injection before milking, attachment delay of milking teat cup and milking frequency on performance of Holstein cows. G. Ghorbani*1 and A. jafari¹, ¹ isfahan University of Technology.

In order to determine the effect of oxytocin injection before milking. attachment delay of milking teat cup and milking frequency, six primiparous and six multiparous Holstein cow were selected. Levels of first factors were oxytocin injection before milking, attachment delay of teat cup and rapid attachment of milking teat cup and levels of second factor were two times milking per day and three times milking per day. Collected data were analyzed in a 6 by 6 latin square design in a 3 by 2 factorial experiment. Oxytocin injection compared to attachment delay of teat cup significantly increased milk production and 3.2 percent FCM in primiparous cows, multiparous cows and total cows. Also fat yield in primiparous cows, and fat, protein and lactose yield in multiparous cows and total cows, were increased significantly, but decreased percentage of fat and total solid in multiparous cows and total cows. Attachment delay of teat cup compared to rapid attachment of teat cup significantly decreased milk production and 3.2 percent FCM in primiparous cows, multiparous cows and total cows. Also fat, and lactose yield in primiparous cows; fat, protein and lactose yield and total solid percentage in multiporous cows and dry matter intake and fat yield in total cows were decreased significantly, but fat percentage increased in multiparous cows and total cows. Three times milking per day compared to two times milking per day significantly increased milk production. 3.2 percent FCM, fat and lactose yield in primiparous cows; 3.2 percent FCM in multiparous cows and milk production, 3.2 percent FCM, protein yield and lactose percentage in total cows, but decreased percentage of protein, lactose and total solid in primiparous cows. The results of this experiment indicated that oxytocin injection before milking, rapid attachment of teat cup and three times milking per day have the best effect on the cows performance in the middle of lactation.

Key Words: oxytocin, Attachment of teat cup, milking frequency

1404 Endocrine responses to isoglucogenic infusions of whey protein and propionic acid in dairy cows. L. Misciattelli*, M. Vestergaard, and T. Hvelplund, *Danish Institute of Agricultural Sciences*.

Endocrine responses to isoglucogenic infusions of a whey protein isolate into the duodenum and propionic acid into the rumen was assessed in order to investigate the mechanisms that regulate milk production in response to changes in protein supply. Four mid-lactation dairy cows with rumen, duodenal and ileal cannulas were used in 4×4 Latin square design, with 14 d periods. The 4 treatments provided the cows with equal quantities of glucogenic substrates, but differing in the proportion coming from propionate or amino acids. The cows were fed 12.7 kg DM/d of a TMR low in intestinally absorbable protein. The ration composition was (% DM): clover grass silage (55), peas (23), rumen protected fat (7), beet molasses (14) and minerals (1). The data was analysed with PROC GLM for standard Latin square design, with one missing observation (treatment 200:200). Plasma obtained by venepuncture at 13:00 on d 11, 12 and 13 was analysed for BHBA, urea, NEFA, glucose (GLUC), tri-acyl glycerides (TG), fructose amine (FCA), IGF-I and triiodothyronine (T3). Catheters were placed in a jugular vein on d 14, and blood samples were taken at 3 h intervals for a 24 h period and analysed for: insulin (INS), glucagon (GLG) and growth hormone (GH). Milk yield, milk protein %, plasma BHBA, urea and NEFA increased significantly with increasing proportion of protein infused, while plasma GH was negatively correlated with protein infusion level. The results suggest that the stimulatory effect of extra absorbable protein on milk yield mainly was due to an increased supply of amino acids to the mammary gland rather than an endocrine mediated effect.

Whey protein, g/d Propionate, g/d	600 0	400 100	200 200	0 300	SEM	Р
Milk yield, kg/d	20.2	19.7	18.0	16.3	0.6	*
Milk protein %	3.27	3.27	3.23	3.08	0.03	*
BHBA, mmol/L	2.9	2.5	1.7	1.7	0.2	*
UREA, mmol/L	8.0	7.7	7.6	6.3	0.2	*
NEFA, meq/L	0.135	0.132	0.122	0.115	0.003	*
GLUC, mmol/L	3.54	3.72	3.96	3.75	0.08	T
TG, mmol/L	0.15	0.15	0.18	0.14	0.02	NS
FCA, $\mu \text{mol/L}$	216	210	252	206	9.6	NS
INS, pmol/L	92	101	104	96	6.7	NS
GLG, ng/L	139	132	127	129	7.8	NS
$GH, \mu g/L$	3.2	3.1	3.2	5.9	0.9	*
IGF-I, $\mu g/L$	100	97	95	92	5.4	NS
T3, nmol/L	1.42	1.55	1.50	1.51	0.07	NS

NS: P>=0.1, T: P<0.1, *: P<0.05

Key Words: Protein, Milk production, Hormonal regulation

1405 Glucose metabolism and insulin sensitivity in Gulf Coast Native and Suffolk ewes during late gestation and early lactation. C.C. Williams*1, K.J. Calmes¹, J.M. Fernandez¹, C.C. Stanley¹, J.C. Lovejoy², H.G. Bateman¹, L.R. Gentry¹, D.T. Gantt¹, and G.D. Harding¹, ¹Louisiana State University Agricultural Center, Baton Rouge, LA 70803, ²Pennington Biomedical Research Center, Baton Rouge, LA 70808.

Two experiments were conducted to evaluate possible differences in glucose metabolism and insulin responsiveness in Gulf Coast Native (GCN) and Suffolk (SFK) ewes. In experiment 1, 30 GCN and 41 SFK ewes in late gestation were used to identify hyperglycemia and hyperinsulinemia. Blood samples were obtained via jugular venipuncture and analyzed for plasma insulin, glucose, thyroxine, albumin, and urea N (PUN) concentrations. In experiment 2, at 2 wk postpartum, 19 GCN and 19 SFK ewes were administered frequently sampled intravenous glucose tolerance tests (FSIVGTT) consisting of a pulse dose of glucose (0.5 mg/kg BW) followed by an infusion of insulin (0.03 IU/kg BW). Blood samples were collected via jugular catheter at timed intervals over a 3hr period after glucose infusion. Samples from the FSIVGTT were analyzed for insulin and glucose concentrations, and the minimal model computer program was then used to assess glucose effectiveness (S_G) , an estimate of insulin-independent glucose disappearance; insulin sensitivity (S_I) , an estimate of insulin-dependent glucose disappearance; and acute insulin response (AIR $_G$), insulin secretion relative to glucose administration. Concentrations of albumin, thyroxine, and PUN were measured in samples collected prior to glucose infusion. In experiment 1, insulin concentrations were greater (P < 0.01) in SFK ewes while glucose concentrations were not different (P > 0.05) between breeds. Concentrations of albumin (P < 0.01), thyroxine (P < 0.05), and PUN (P < 0.05) were greater in SFK ewes. In experiment 2, S_G and S_I did not differ between breeds, while AIR_G tended to be greater (P = 0.06) in SFK ewes. There were no differences (P > 0.05) in albumin, thyroxine, or PUN concentrations. These data indicate slight differences in insulin sensitivity between GCN and SFK ewes during late gestation. However, there were no apparent differences in glucose metabolism between these breeds during the early lactation period.

Key Words: Glucose metabolism, Insulin sensitivity, Sheep

1406 Influence of zinc deficiency on the mRNA expression of zinc transporters in adult rats. MW Pfaffl*1 and W Windisch¹, ¹Department of Animal Physiology, Center of Life and Food Sciences, Techn. Univ. Munich.

The accumulation of zinc (Zn) in the cell is a sum of influx and efflux processes via transporter proteins, like the four Zn transporters (ZnT1-4), the divalent cation transporter 1 (DCT1) and of storage processes mainly bound to metallothionein (MT). To study the effect of Zn deficiency on mRNA expression levels adult rats were used as an animal model. Rats were fed an almost Zn free diet for 29 d, which induced Zn mobilization from body Zn stores. Tissues representing Zn absorption (jejunum, colon), Zn storage and utilization (muscle, liver), and Zn excretion (kidney) were retrieved. Real-time reverse transcription

(RT) polymerase chain reaction (PCR) assays were developed and a relative quantification on the basis of GAPDH was applied. Assays allowed a relative and accurate quantification of mRNA molecules with a sufficiently high sensitivity and repeatability. All known Zn transporter subtypes were found in the tissues. Expression patterns and reactions to Zn deficiency were specific for the tissue analyzed. The table shows the n-fold expression levels up (+) or down (-) regulated by Zn deficiency. Expression results imply that some transporters are expressed constitutively, whereas others are highly regulated (*) in tissues responsible for Zn homeostasis. The most distinct changes of expression levels were shown in colon which can therefore be postulated as a highly Zn sensitive tissue. MT was down-regulated in all tissues, in parallel with intracellular Zn status, and is therefore a potent candidate gene for Zn deficiency. This study provides the first comparative view of regulation of gene expression and fully quantitative expression analysis of all known Zn transporters in a non growing adult rat model.

	MT	DCT1	ZnT1	ZnT2	ZnT3	ZnT4
jejeunum	-1.39	+1.27	-3.79	-5.56	+8.04	+1.24
colon	-6.74***		+10.06*	+1.39*	+9.29	+2.09*
liver	-154.7***	-1.88	+1.04	-1.71	-1.03	+1.13
muscularity	-1.28	-1.26	-1.55	-3.04	-9.98	-1.20
kidney	-1.27	+2.56	-4.57	+1.07	+4.49*	-1.18

Key Words: Zinc deficiency, Zinc transporters 1-4, Adult rat model

1407 Metabolic effects of zinc deficiency on the somatotropic axis in non-growing rats as a new animal model to adult individuals. MW Pfaffl¹, RM Bruckmaier*¹, and W Windisch¹, ¹Department of Animal Physiology, Center of Life and Food Sciences, Techn. Univ. Munich.

Model studies on zinc (Zn) deficiency are usually performed with fast growing rats. But the intensive anabolic situation produces severe interactions between Zn deficiency per se and the metabolism in toto. Respective results may thus not fully reflect the situation in adults. To overcome this methodological disadvantage, we developed an animal model to study Zn deficiency in adult non-growing rats. Feed intake was restricted to 8 g/d containing 2 g Zn/g fortified with pure phytate in Zn deficiency rats and 58 g Zn/g in controls (n=7). At day 1, 2, 4, 7, 11, 16, 22, and 29 of Zn deficiency, 3 animals each were euthanized. Zn deficiency was evident from reduced plasma Zn, plasma alkaline phosphatase activity and severe mobilization of Zn from tissue stores (mainly skeleton), while feed intake and body weight remained unaffected. Metabolic effects on key players of the somatotropic axis were analyzed on mRNA and protein level. A relative quantification in real-time RT-PCR was applied in liver RNA. Expression levels of insulinlike growth factor 1 (IGF-1), IGF-1 receptor (, growth hormone receptor, and three IGF binding proteins (IGF-BP1-3) were quantified. All transcripts were expressed in liver and each factor exhibited a specific pattern. IGF-BP2 mRNA declined slightly (1.8-fold down-regulation, P<0.05). All other transcripts remained unchanged over 29 d Zn deficiency. Growth hormone and IGF-1 plasma concentrations, analyzed by radioimmunoassay, remained constant during Zn depletion (3314 ng/ml and 27546 ng/ml, respectively). Furthermore non-esterified fatty acids (NEFA) and glucose plasma levels were measured enzymatically. Concentrations of NEFA (0.580.16 $\mathrm{ng/ml}$) and glucose (25434 $\mathrm{ng/ml}$) did not change with time. In conclusion, Zn deficiency did not affect mRNA expression and protein levels on key players of the somatotropic axis, except for IGF-BP2, as well as key metabolites.

Key Words: Zinc deficiency, Somatotropic axis, Adult rat model

1408 Effect of drinking diluted seawater on some physiological aspects of camels. H. Abdel Rahman*¹, M. El Sherif², S.S. Omar¹, M.A. ElSayed¹, and N.M. Ibrahim², ¹Minufiya University, Faculty of Agriculture, ²Desert Research Institute, Egypt.

Four she camels Camelus dromedaries, aged 12 years and weighed 558.6 kg on average, were kept in 15 x 11 m open yard and fed ad libitum on fresh acacia and clover hay. Tap water (680 ppm TDS) was available ad libitum for 2 weeks in each summer and winter as control periods (CP), followed by consecutive 20 days treatment periods (TP), when they drank diluted sea water (SW) mixed at the rate 1/1.5 sea/tap water containing TDS 14539 ppm. Blood samples were collected on the day before and last day of treatment with saline water. Diurnal changes in

respiration rate (RT), rectal (RT), skin (ST), coat CT), ambient temperatures and relative humidity were recorded. Activity of glutamic pyruvic (GPT), glutamic oxalic (GPT) transaminases and cocentration of Na, K, Mg, T3 and T4 in blood plasma were determined. Drinking SW decreased RT (P<0.01), increased CT (P<0.05) and insignificantly RR specially in summer. Significant diurnal changes were detected in RT, ST, CT (P<0.01) and RR (P<0.05) in the same order of AT changes. Drinking SW also significantly increased plasma concentration of Na, K, hemoglobin, MCH and MCHC indices, activity of GPT and decreased T3 and T4. Generally, camels can tolerate drinking diluted seawater containing enormously high total disolved salts up to 14540 ppm.

Key Words: Drinking seawater, physiological aspect, camels

1409 Effect of different levels of passive immunity on response to intravenous immunoglobulin in calves. C. J. Hammer *1 , J. D. Quigley 2 , and H. D. Tyler 1 , 1 lowa State University, 2 APC Company, Inc.

Adequate concentrations of IgG are imperative for the health and survival of neonatal calves. Oral and intravenous IgG supplements have been developed to supplement or replace maternal colostrum (MC) when it is unavailable or when the calf has failure of passive transfer. The objective of this study was to determine the effect of different levels of passive immunity on calf response to administration of intravenous immunoglobulin (IVIG). The IVIG was concentrated from bovine abattoir blood to a final concentration of approximately 35 g IgG/L. Dairy breed bull calves (n = 32) were removed from their dams immediately after birth and assigned to one of four treatment groups. Calves in the high group (H) received 2 L of pooled MC at 1 h and 12 h after birth. Calves in the low group (L) received 1 L of MC mixed with 1 L of milk replacer at 1 h and 12 h after birth. Calves in the deprived group (D) received 2 L of milk replacer at 1 h and 12 h after birth. Calves in the control group (C) received 2 L of pooled MC at 1 h and 12 h after birth. At 3 d of age, calves in the H, L, and D group all received 500 ml of IVIG administered via jugular catheter. Calves in the control group received 500 ml of 0.9% NaCl. Blood was collected by jugular venipuncture prior to infusion, and again at 1, 3, 7, 10, 14, 21, and 28 d post infusion for determination of plasma IgG by turbidimetric immunoassay. Mean plasma IgG concentrations at 3 d of age were different between the H, L, and D group of calves, but not between H and C. Mean plasma IgG at 3 d of age were 12.1, 6.3, 0.0, and 11.8 g/L for calves in H, L, D, and C, respectively. Calves in all treatment groups had a greater rise in mean plasma IgG compared to C calves at 1 d post infusion. Mean increase in plasma IgG at 24 h post infusion were 1.8, 2.4, 3.0, and #0.1 g/L for calves in H, L, D, and C, respectively. These data indicate that IVIG can increase plasma IgG levels in calves regardless of the level of passive immunity present at infusion.

Key Words: Calf, Immunoglobulin, Colostrum

1410 Characterization of *Staphylococcus* species in bulk tank milk. N. V. Hegde*, R. Butchko, and B. M. Jayarao, *The Pennsylvania State University, University Park, PA, USA*.

A total of 126 dairy herds from 14 counties in Pennsylvania were examined for number and type of Staphylococcus species. Staphylococcus aureus (SA) was detected in 49 of 126(40.2 %) of bulk tank milk (BTM) samples. Counts ranged from 0 - 275 cfu/ml (mean, 45 cfu/ml). Coagulase negative Staphylococci (CNS) were observed in 117 of 126 (92.8%) of BTM samples. Counts ranged from 0 -15,175 cfu/ml (mean, 1246 cfu/ml). A total of 434 isolates from 122 BTM samples were examined to species level; 434 isolates belonged to 18 different species. Staphylococcus aureus, S. xylosus, S. chromogenes, and S. hyicus were detected in 40.1, 30.3, 25.4, and 25.4% of bulk tank milk samples, respectively. High CNS counts (>1,000 cfu/ml) were associated with high bulk tank somatic cell counts (mean, 414,109 cells/ml) and/or high standard plate counts (mean, 12,289 cfu/ml). A critical review of farm management practices using a self administered questionnaire followed by consultations with dairy producers strongly indicated that; 1) Dairy producers who used sand as bedding had low (<500 cfu/ml) CNS counts in bulk tank milk, and 2) Most of the dairy producers (94%) who practiced forestripping, and pre-and post-dipping had low (<500 cfu/ml) CNS counts.

The findings of the study suggest that proper management practices related to bedding and milking practices can influence the number and type of CNS in bulk tank milk.

 $\mbox{\sc Key Words:}$ bulk tank milk, Staphylococcus~aureus, coagulase negative staphylococci

1411 Leptin attenuates the central effects of neuropeptide-Y on somatotropin but not gonadotropin secretion in cows. M. R. Garcia*1,2, M Amstalden1,2, D. H. Keisler³, N Raver⁴, A Gertler⁴, and G. L. Williams¹,2, ¹ Texas A&M University Agricultural Research Station, Beeville, TX/USA, ² Texas A&M University, College Station, TX/USA, ³ University of Missouri, Columbia, MO/USA, ⁴ The Hebrew University of Jerusalem, Rehovot/Isreal.

Objectives were to determine whether the action of centrallyadministered neuropeptide-Y (NPY) on secretion of LH and GH in ovariectomized cows could be attenuated by recombinant ovine leptin (oleptin). A secondary interest was to examine the interactive effects of NPY and leptin on FSH secretion. Six ovariectomized cows, each surgically-fitted with third-ventricle guide cannulas, were assigned randomly to each of three treatment groups in a Latin Square arrangement: 1) Saline-Saline-Saline (SSS); s.c. and i.v. saline at 0 and 70 min, respectively; intracerebroventricular (ICV) infusion of phosphate buffered saline (PBS)-0.3% bovine serum albumen (BSA) at 90 min, 2) Saline-Saline-NPY (SSN); same as group 1 except 500 μ g pNPY in PBS-0.3% BSA at 90 min, and 3) Leptin-Leptin-NPY (LLN); s.c. and i.v. oleptin (30 μg/kg) in saline at 0 and 70 min, respectively; 500 μg pNPY ICV in PBS-0.3% BSA at 90 min. Plasma concentrations of leptin increased (P < 0.01) 4-fold $(39 \pm 9 \text{ ng/ml})$ after leptin treatment and remained elevated, relative to controls (6 \pm 2 ng/ml), throughout the remainder of the sampling period (200 min). Secretory patterns of LH, FSH, and GH did not change (P > 0.1) after leptin treatment. Variation (P <0.05) in baseline and mean serum concentrations of LH, FSH, and GH were detected between replicates, days, and animals. Variation (P < 0.01) in pulse amplitude and frequency of GH and FSH were detected between replicates. Plasma concentrations of LH began to decline immediately after the infusion of NPY in SSN and LLN treatments, and were markedly lower (P < 0.01) than SSS within 2 h after infusion. Plasma concentrations of GH were greater (P < 0.01) during the first h following NPY treatment in SSN, but this increase was attenuated in LLN. Serum GH did not change in the SSS group, and FSH was similar among all treatments. Results confirm suppressive and stimulatory effects of NPY on LH and GH, respectively, and that leptin can attenuate the action of NPY on GH but not LH secretion in cows.

Key Words: Leptin, NPY, GH

1412 Effects of three post-weaning management regimes on protein-abundance of lipogenic enzymes and adipogenic activities in adipose tissues of beef cattle. E. Okine*1, M.A. Price¹, L. Goonewardene², P. Mir³, Z. Mir³, J.A. Basarab², V. Baron⁴, and J.J. Kennelly¹, ¹AFNS, University of Alberta, Edmonton, AB T6G 2P5, ²Livestock Industry Division, AAFRD, Edmonton, AB. T6H 5T6, ³Agriculture and Agri-Food Canada, Lethbridge, AB T1J 4B1, ⁴Agriculture and Agri-Food Canada, Lacombe, T4L 1W1.

The objectives of this study were to determine the effects of three postweaning management regimes on the activities and protein-abundance (PA) of acetyl CoA carboxylase (ACC) and fatty acid synthetase (FAS) in subcutaneous adipose tissue of 54 steers. Incorporation of [I -¹⁴C|palmitate into triglycerides and adipogenic activities (using cultured mouse 3T3-L1 preadipocytes) in these steers were also determined. The regimes were feedlot (F), background-feedlot (BF) and backgroundpasture-feedlot (BPF). ACC activity in the F group was 75 and 65% (P <0.05) lower than BF and BPF groups, respectively. PA of ACC ranged from 10 to 17 OD $\rm mm^{-2}$ (P $<\!0.05)$ in F, BF and BPF groups. Similar results were obtained for FAS activity and abundance in these steers. Esterification of palmitate was (P<0.05) higher for BF and BPF steers vs F steers. Cellular-GPDH activity (a marker for adipocyte differentiation) and retinol concentrations were higher (P<0.05) in the F group and not different for BF and BPF groups. The high enzyme activities and esterification rates in BF and BPF steers indicate that fatty acid synthesis was still active in these groups compared to F steers. These results were reflected in the marbling grades for the F, BF and BPF groups which were: A and AA (83.3, 70.8 and 100)%), AAA (16.6, 25

and 0%), B1 (0, 4.2 and 0%), respectively. We conclude that adipogenic and anti-adipogenic components interact to determine the activities and protein abundance of ACC and FAS in adipose tissue. As cattle near finish, the effects of anti-adipogenic components may outweigh the influence of adipogenic substances leading to low PA and activities of ACC and FAS and low esterification rates.

Key Words: Cattle, Lipogenesis, Adipogenecity, Acetyl-CoA carboxylase, Adipose tissue, Fatty acids synthesis

1413 Effects of diets high in linoleic acid on carcass fat and CLA content, serum leptin, and age at puberty in beef heifers. M. R. Garcia*1,2, M Amstalden1,2, C. D. Morrison3, D. H. Keisler3, and G. L. Williams1,2, 1 Texas A&M University Agricultural Research Station, Beeville, TX, USA, 2 Texas A&M University, College Station, TX, USA, 3 University of Missouri, Columbia, MO, USA.

Objectives were to determine the effects of feeding a diet high in linoleic acid on total carcass fat content and fatty acid composition, circulating metabolic hormones, and age at puberty in developing crossbred (Angus or Red Angus x Hereford x Brahman) beef heifers. Heifers were weaned and blocked by BW (Heavy, n=10; Light, n=10) and allocated randomly within block to receive isocaloric and isonitrogenous diets formulated with either added fat (HF, n=10) or no added fat (C, n=10) 10) from 4 mos of age until post-pubertal slaughter. Whole sunflower seed (55 % oil content) was utilized in HF diets to provide 5 % added fat from the start of the study until heifers weighed 250 \pm 8 kg, at which time added fat was increased to 7 % of DM until slaughter. Puberty was confirmed based on serum concentrations of progesterone ≥ 1 ng/ml for 2 consecutive samples and visual confirmation of corpora lutea (CL) by transrectal ultrasonagraphy. Heifers were slaughtered at 325 \pm 10 d of age. Total carcass composition was estimated from longissimus muscle, with CLA composition determined in internal and s.c. fat. HF-Heavy heifers tended (P < 0.10) to reach puberty later than all other heifers, and one HF-Light heifer did not reach puberty during the study. Linoleic acid and cis-9, trans-11 CLA tissue content was higher (P <0.03) in the HF group, but neither total carcass fat nor percent DM differed due to diet, although the percent protein tended (P < 0.10) to be lower in HF heifers. Serum leptin did not differ due to diet; however, leptin increased ($P \leq 0.01$) linearly throughout the study. Serum GH and IGF-I increased or remained relatively constant during the first 2-10 weeks of feeding, then began a slow decline (P < 0.05) until the onset of puberty, with serum IGF-I lower ($P \leq 0.01$) in HF heifers. Serum insulin and total cholesterol increased (P < 0.01) throughout the study in both groups, but only total cholesterol was affected by the HF diet (P < 0.05). Growing diets high in linoleic acid appear to have little or no effect on total carcass fat, serum leptin, or age at puberty in beef heifers despite increased CLA tissue content.

Key Words: Puberty, CLA, Leptin

1414 The role of ghrelin and GHS receptor on proliferation and differentiation of ovine preadipocytes. SG Roh*, KC Choi, Y Shrestha, C Yoon¹, and S Sasaki, *Lab of Animal Molecular Physiology, Faculty of Agriculture, Shinshu University, JAPAN*, ¹Dept of Animal Science, Iksan College, Iksan, KOREA.

Ghrelin is a novel endogenous natural ligand for the growth hormone secretagogue receptor (GHS-R or ghrelin receptor) that has been recently isolated from the rat stomach. These 28 amino acids constituting peptide has n-ocatnoylation in its serine-3 residue, which is essential for the ghrelin activation. Ghrelin has been demonstrated to regulate pituitary GH secretion via GHS-R when administered into the central nervous system or peripherally along with GHRH and somatostatin. GHS-Rs have been identified in many other tissues, other than the hypothalamic neurons and in the brainstem, like heart, lung, pancreas, intestine, uterus and adipose tissue, but their functions are unknown on these tissues. The mechanism by which ghrelin and GHS-R affect the proliferation and differentiation of ovine preadipocytes has remained largely unknown. This study was conducted to examine the role of ghrelin and GHS-R on the proliferation and differentiation of ovine primary preadipocytes in culture. The preadipocytes, which were obtained from sheep subcutaneous adipose tissues, were proliferated to confluence and then differentiated to adipocytes in differentiation medium for 10 days. The confluent preadipocytes and differentiated adipocytes at days 3, 7 and 10 were harvested for total RNA extraction and RT-PCR of GHS-R mRNA. Ghrelin decreased the proliferation of preadipocytes. The level of GHS-R mRNA was significantly increased during the differentiation period, although this was not detected in the confluent preadipocytes. Furthermore, ghrelin stimulated the differentiation of preadipocytes and increased the level of GHS-R during the differentiation. In conclusion, our results demonstrate that ghrelin and GHS-R have an important role on the process of adipogenesis of ovine preadipocytes.

Key Words: Ghrelin, Adipocyte, Sheep

1415 Slow-release somatotropin reduces plasma leptin in lactating dairy cows. F. Rosi*1 and L. Pinotti², ¹Ist. Zootecnia Generale, Facoltà di Agraria , ²Dept. VSA, Facoltà di Medicina Veterinaria- Università di Milano I-20133 Milan Italy.

Somatotropin has dramatic effects on adipose tissue and lipid metabolism. Leptin, produced and released primarily by adipose cells, exerts a regulatory control on energy homeostasis. The aims of this study were to determine the effects of bST administration on milk production, plasma leptin and selected plasma metabolites in lactating dairy cows. Forty Holstein cows(90±33)were randomly divided into 2 groups: Control and bST. The bST group received 640mg/4wk of slowrelease bST(Posilac) for two cycles. Milk yield and composition were measured at 7 days post-injection of each cycle. Blood samples were collected on the same day before feeding, and analyzed for leptin, NEFA, total protein, α -amino nitrogen, and urea nitrogen. Both milk yield and milk fat percentage were increased (29.9 vs. 35.3 kg/d; P \leq .01; 3.59 vs. 3.92%; P \leq .05) by bST administration, while milk protein content was unaffected by treatment (3.19 vs. 3.13%). At 7 days post-injection, bST decreased (P<.01) plasma leptin by 33% (4.26 vs. 2.86 μ g/l) whereas plasma NEFA was drastically increased (225 vs. 875 μ mol/l; P \leq .01). Plasma total protein was increased by bST (78.9 vs. 81.4 g/l; P≤.05), while both α -amino nitrogen and urea in plasma of treated cows were reduced (P<.01) by 20% (2.54 vs. 2.03 mmol/l; 6.55 vs. 5.27 mmol/l, respectively). These data confirmed a galactopoetic effect of bST, which imposed, on peak of response, a higher demand of nutrients sustained by an enhanced lipolysis in adipose tissue. Lower plasma leptin observed in present study could be due to reduced body fat mass as consequence of lipolysis induced by bST. This is in line with higher plasma NEFA concentration observed in bST group. Plasma nitrogen metabolites indicate a higher efficiency in protein metabolism in treated cows. This study show that plasma leptin is linked with the nutritional status of cows, even though other hormones and metabolites are also involved in the signaling and control of body energy store.

Key Words: bST, Leptin

1416 The influence pre-calving Somatotropin treatment on the quantity and quality of colostrum in beef cattle. N. Macewko, G.A. Angliss, E.F. Jones, K.E. Govoni, M.F. Loughlin, D. Cissel, S.A. Zinn, D. Schreiber, and T.A. Hoagland, University of Connecticut, Storrs, Connecticut.

Fourteen Hereford beef cows (3-8 years old) were randomly assigned to receive no Posilac (Monsanto) or Posilac (500mg) every two weeks for either four (three injections) or eight (five injections) weeks before calving. At calving, all cows were milked with a portable vacuum machine for 15 minutes. The weight and colostrometer reading of the milk was recorded. Samples of milk were obtained for determination of IgM, IgG1, and IgG2 concentrations. Percent fat, protein, and total solids of the milk were determined. The weight of milk was similar for the control, four, and eight week Posilac treated cows (1.5, 1.3, and 1.3 kg respectively; SEM=0.3). Colostrometer readings and the concentrations of IgM and IgG1 were not significantly influenced by the Posilac treatments. There was a linear effect (P≤0.05) of Posilac treatment on milk IgG2 concentrations for the control, four and eight week Posilac treated cows (5370, 5864, and 8208 mg/dl respectively; SEM=1452). The percent fat, protein, and total solids were similar in the milk across treatments; however, the percent protein and total solids were greater (p=0.21) in the Posilac treated groups. The average daily gain from birth to weaning (0.9, 1.1, and 1.2 kg/day: SEM=0.05 respectively for the control, four, and eight week Posilac treated cows) was significantly (P≤0.05) greater for the calves suckling the Posilac treated cows. In conclusion, Posilac treatment pre-calving influenced the quality of colostrum by increasing the concentration of IgG2 and increased the average daily gains of the calves suckling the treated cows.

Key Words: colostrum, beef cows, immunoglobulins

1417 Protocols for detection of EPSP synthase gene in sheep fed diets containing Roundup Ready® canola. R. Sharma*1, T.W. Alexander^{1,2}, D. Damgaard¹, R.J. Forster¹, and T.A. McAllister¹, ¹Agriculture and Agri-Food Canada, Lethbridge, AB, ²University of Alberta, Edmonton.

Standardized protocols were developed for detecting the gene encoding 5-enolpyruvylshikimate-3-phosphate synthase (EPSPS) in samples collected from sheep fed barley-based diets containing 6.5% (DM basis) Roundup Ready® canola (RRC). Glyphosate tolerance in RRC is conferred by the coding region of EPSPS derived from Agrobacteriumtumefaciens CP4. Genomic DNA was extracted from diets and rumen digesta using a modified CTAB extraction procedure, but was found unsuitable for reproducible PCR amplification of EPSPS gene fragments from complete diets. Passing the genomic DNA from the CTAB extraction through a DNeasy $^{\rm TM}$ plant mini-kit column (Qiagen), however, did produce a good yield of PCR-quality DNA. A Wizard $^{\rm TM}$ genomic DNA purification kit (Promega) was used for extraction of DNA from blood samples. Primer sets were designed and PCR reaction conditions standardized that allowed amplification for of eight different regions (144to 527-bp fragments) of the 1.3-kb EPSPS coding region. Positive PCR $\,$ controls for both plant and bacterial DNA were included in screening diet and digesta samples. PCR controls for animal DNA with ovine tissues, and all three controls with fecal samples. Screening samples for the presence of the transgene entailed PCR assays for the eight fragments at two genomic DNA concentrations (ng and pg range). Using positive controls (spiked samples), the assay was confirmed sensitive enough to detect pg quantities of transgene in diet and blood, and ng quantities in digesta samples. Results of PCR assays were confirmed with Southern blot hybridizations. This protocol has been optimized for DNA extraction and sensitivity of detection, and has proven highly reliable. Transgene fragments were not detected in blood samples collected from sheep 2 to 3 h after feeding the RRC diet. Rubisco small subunit-specific (~500 bp) and chloroplast DNA-specific fragments (653 bp) could be detected in digesta and fecal samples, whereas no fragments of the EPSPS coding region were found.

Key Words: EPSP Synthase, Roundup Ready®, PCR

1418 Influence of nutrition and body condition score on plasma concentrations of IGF-I and thyroxine (T4) in gestating beef cows. C. A. Lents*1, R. P. Wettemann¹, J. M. Bolanos², F. J. White¹, I. Rubio¹, N. H. Ciccioli¹, and L. J. Spicer¹, ¹Department of Animal Science, Oklahoma Agricultural Experiment Station, Stillwater, 74078, ²Ministry of Agriculture, San Jose, Costa Rica.

Pregnant Angus x Hereford cows (n = 73) were used to determine the effects of nutrient intake and body condition score (BCS: 1 = emaciatedand 9 = obese) on concentrations of IGF-I and T4 in plasma. At 2 to 4 mo of gestation, cows were blocked by BCS and assigned to one of four nutritional treatments: high (ad libitum access to a 50% concentrate diet in the drylot), or adequate native grass pastures and one of three amounts of a 40% CP supplement each day (moderate, 1.6 kg; low, 1.1 kg; or very low, 0.5 kg). After 115 d of treatment, all cows grazed dormant native grass pasture and received 1.6 kg/d of a 40% CP supplement. At 70 and 125 d of treatment, cows were gathered and plasma samples were collected by tail venipuncture (fed sample). After 18 h without feed and water, a second plasma sample was collected (fasted sample). Concentrations of IGF-I and T4 were determined by RIA. BCS was similar for all groups (4.6 ± 0.1) at the initiation of treatment. After 70 d, BCS was greatest (P < 0.01) for high cows and similar for moderate, low, and very low cows. High cows had greater (P < 0.05) concentrations of IGF-I in fasted samples than all other groups, but IGF-I was similar in fed samples for all treatments. Treatment and access to feed did not influence plasma concentrations of T4. BCS at 70 d was correlated with plasma IGF-I in fasted samples (r = 0.43: P < 0.001) but not in fed samples. After 125 d of treatment, BCS was greatest (P < 0.01) for very high cows (6.4 ± 0.1) , similar for moderate and low cows (4.8 \pm 0.1), and least (P < 0.01) for very low cows (4.5 \pm 0.1). Plasma concentrations of IGF-I at 125 d in fasted and fed samples were not influenced by previous treatments and were greater (P <0.001) in fasted than fed cows. Body condition at 125 d was correlated with IGF-I in plasma in fasted samples (r = 0.33; P < 0.01) and in fed samples (r = 0.41; P < 0.01). We conclude that concentrations of IGF-I in plasma of cows are correlated with BCS in late gestation.

Key Words: BCS, IGF-I, Thyroxine

Ruminant Nutrition Feed Additives, Fiber, and Minerals

1419 Preliminary report on chemical composition and ruminal degradation of *Aloe vera*. J. A. Vergara, M. A. Cuauro, and O. E. Araujo Febres*, *The University of Zulia, Maracaibo, Venezuela*.

Ruminal degradability of dry matter (DMS) and organic matter (DMO) of Aloe vera(AV) and Brachiaria humidicola(BH) as a reference were evaluated. Ten grams samples of AV and BH milled to 3 mm were incubated in nylon bags at 0, 6, 12, 24, 48 and 72 h in two crossbred steers with permanent rumen canulae (350 kg LW). Non-linear regression was used to calculate the parameters: a, b and c, while a + b was the potential degradability (PD); c, ruminal degradation rate (DR); a, instant degradability (ID). A completely randomized design with 4 replicates was used; means of degradation at 48 and 72 h and equation parameters were compared by LS Means. Chemical composition of AV was: DM: 90%; OM: 86.9%; crude protein: 7.5%; ADF: 39.6%; and NDF: 38.5%. DMS of AV was stabilized among 48 and 72 h (89.90%) higher than (P<0.05) values for BH at 48 h. Same performance had DMO. PD of MS and MO of AV (99.87% and 99.84%) were higher (P < 0.05) than BH values (71.44% and 68.67%). DR of MS in the rumen was similar among by products (0.022 vs. 0.029, respectively); while DR of MO was 0.031and 0.022 for AV and BH, respectively. The higher value (P<0.05) of DI of AV (40.9%) increased PD. AV degradability and Venezuelan production potential of this specie determine a high importance for animal feeding in arid zones.

 $\mbox{\sc Key Words:}$ Aloe vera, Ruminal Degradability, Arid Zone

1420 Influence of addition of fibrolytic enzymes on enzyme activities and fermentation patterns of pure substrates *in vitro*. D. Colombatto*, D. P. Morgavi, and K. A. Beauchemin, *Research Center*, *Lethbridge*, *Alberta*, *Canada*.

A completely randomized study was carried out to investigate possible modes of action of an enzyme mixture (Liquicell 2500, Specialty Enzymes and Biochemicals, CA) with potential to be used in ruminant diets. The enzyme contained mainly xylanase and cellulase activities, with residual amylase and pectinase. Microcrystalline cellulose (CE), oat spelt xylan (XYL) and a mixture (1:1 v/v, CEXYL) were incubated in Hungate tubes (100 mg/tube, eight replicates), untreated or treated with Liquicell 2500 applied at 0.51 and 2.55 l g DM $^{-1}$ (L1 and L2, respectively). Interaction time was 20 h at 24C. Rumen fluid was collected 5 h post-feeding from a steer fed alfalfa hay ad libitum, and incubated at 39C with anaerobic buffer (1:4 v/v). At 1, 6, 18 and 48 h post incubation, samples from the liquid fraction were analyzed for xylanase, endoglucanase (CMCase), β -glucosidase and β -xylosidase activities (39C and pH 6.0). Volatile fatty acids (VFA) were quantified at 6, 18 and 48 h. Samples from 6 and 18 h of incubation were processed to obtain a feed-particle associated bacterial fraction (FPA), which was analyzed for enzymic activities as previously described. Addition of Liquicell 2500 at L2 increased (P<0.05) the initial (up to 6 h) xylanase, CMCase and β -glucosidase activities in the liquid fraction by an average of 85%, indicating that the exogenous mixture supplied extra enzymes and that these enzymes were resistant to the proteolytic action of rumen fluid. Across substrates, xylanase, and CMCase activities in the FPA fraction after 18 h were increased (P<0.05) with L2 by an average of 32%, suggesting an increase in the fibrolytic activity of rumen microbes. Total VFA were numerically (P>0.05) increased by L2 com-