

Calves were born between 2001 and 2004. Carcass traits collected were finished weight, hot carcass weight, marbling score, and longissimus muscle area. Microsatellite markers on chromosomes 5 and 6 were selected based on their relative position. Markers used on chromosome 5 were BM6026, RM103, BM321, RM084, BMS1216, BM315, and BM597. Markers used on chromosome 6 were ILSTS093, ILSTS090, BM1329, BMS518, ILSTS035, BM8124, and BMC4203. Individual marker analysis was conducted because homozygosity of the bulls for some markers hindered interval mapping. Family 1 exhibited allelic effects for finished weight, hot carcass weight, and marbling score on chromosome 5. Markers RM103 and BM321 were associated with finished ( $P < 0.01$ ) and carcass ( $P < 0.05$ ) weights. An association with marbling score was identified with BM6026 ( $P < 0.05$ ), RM103 ( $P < 0.01$ ), and BM321 ( $P < 0.01$ ). On chromosome 6, BMC4203 was associated with longissimus muscle area in family 1 ( $P < 0.05$ ) and family 2 ( $P < 0.001$ ). No association was detected ( $P > 0.05$ ) on family 3. Association of these markers with carcass traits on chromosomes 5 and 6 are consistent with findings from independent studies.

**Key Words:** Markers, Bovine, Carcass

### 627 Associations between single nucleotide polymorphism in the Dgat2 gene and several economic traits in commercial feedlot steers.

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DGAT2 was found to be a new diacylglycerol acyltransferase gene family in 2001. It plays more important role in mammalian triglyceride metabolism than DGAT1 does, but associations between DGAT2 and beef cattle economic merits have not been reported. The objective of this study was to try to find single nucleotide polymorphisms (SNP) with PCR-SSCP method in bovine DGAT2 gene and evaluate the

association of that with carcass and meat quality traits from 157 commercial feedlot steers distributing to 3 Chinese local breeds and 4 cross breeds. In this study, 15 SNP have been discovered in the Dgat2 Intron3 (GenBank Accession No. AY589091) at positions 65, 128, 178, 210, 241, 255, 270, 312, 328, 334, 365, 366, 371, 415 and 437 (Named as their position in PCR amplified fragments). Only 7 of them were analyzed (128, 178, 241, 270, 312, 328 and 371), because three groups (65-128-255, 178-210-365 and 241-334-366) were in complete linkage disequilibrium within group, SNP at position 415bp is a mutation of deletion and that at position 437bp is a null mutation. These data were analyzed using two different models, 1) genotype, and 2) allele substitution. Contemporary groups were fit as a fixed effect and formed from feed group in feedlot plus breed type. Significant statistics are listed in table 1. An initial conclusion was that associations exist between DGAT2 gene and several beef cattle economic traits that concern with carcass fat. For the small sample size of this study, proposal is further effort should be required to validate these findings in larger populations.

**Table 1. List of significant statistics**

Model	Trait <sup>1</sup>	SNP	LSM or regression coefficient estimates	F	Pr>F		
Genotype	REA	128	AA: 62.9 ± 6.7 <sup>a</sup>	GG: 77.9 ± 0.9 <sup>b</sup>	5.4	0.028	
Genotype	KPH	270	CC: 40.1 ± 3.2 <sup>a</sup>	CT: 42.5 ± 2.2 <sup>a</sup>	TT: 47.8 ± 1.0 <sup>b</sup>	4.0	0.019
Allele substitution	NMP	270	270-C	0.81 ± 0.40	4.0	0.046	
Allele substitution	REA	128	128-A	-7.48 ± 3.37	4.9	0.028	
Allele substitution	KPH	270	270-C	-4.27 ± 1.53	7.8	0.006	
Allele substitution	KPH	312	312-A	3.36 ± 2.43	4.9	0.029	

<sup>1</sup>IMF: intramuscular fat percentage (%); REA: rib eye area (cm<sup>2</sup>); KPH: kidney, pelvic and heart fat weight (kg); NMP: net meat percentage (Retail cuts weight / Live body weight, %). <sup>a,b</sup>Within a row, least square means with different superscript letter differ, P<0.05.

**Key Words:** DGAT2 Gene, SNP, Carcass traits

## FASS Environment, Waste Management and Ecosystems: Transforming Forages to Improve Nitrogen Use by Dairy Cows and Decrease Nitrogen Emissions

**628 Source, amount and fate of nitrogen on US dairy farms.** R. A. Kohn<sup>\*1</sup> and M. Wattiaux<sup>2</sup>, <sup>1</sup>University of Maryland, College Park, <sup>2</sup>University of Wisconsin, Madison.

The purpose of this review is to identify factors related to forage crop production that affect the efficiency of nitrogen (N) utilization on dairy farms, and may decrease N losses to the environment. Nitrogen enters most dairy farms in the form of fertilizer, N fixed by leguminous crops, atmospheric deposition, and imported feeds or manures. Typically less than half of the N entering the farm can be accounted for in milk, crop, cattle, or manure outputs, indicating considerable leakage of various chemical forms of N to the environment. Typically, less than 25% of the manure N produced by a dairy herd is recycled into home-grown feed N. Considering the entire agricultural system in support of milk production, all N in milk protein ultimately derives from atmospheric N. Atmospheric N becomes biologically active in one of two ways:

chemically by the Haber-Bosch process or biologically by the rhizobial bacteria associated with the roots of legumes. Total N inputs to agriculture for milk production are 3 to 10 times greater than the outputs as N in milk and meat. Most N is lost to ground and surface water through leaching and runoff, or lost to air via volatilization as ammonia, nitric oxide or nitrous oxide. These losses are harmful to the environment and human health in various ways. Some N is also denitrified back to the atmosphere as harmless N<sub>2</sub> gas. Whereas chemical fixation of N is highly dependent on non-renewable energy sources, legume crops fix N near crop roots for efficient utilization. However, high concentrations of legumes in rations oversupply ruminally-degraded protein, which increases N excretion, in particular in urinary N, which is highly vulnerable to volatilization. Although minimizing N losses to the environment requires that crops use N efficiently, it is equally important for animals to utilize feed N efficiently. Thus, the ideal cropping system would fix its own N,

or recycle manure N very efficiently, but at the same time, produce adequate carbohydrate and a protein source with ruminally undegraded protein and the ideal amino acid profile for milk production.

**Key Words:** Nitrogen emissions and runoff, Protein requirements, Forage crop improvement

**629 Importance of forages on dairy farms, beyond their use as feed.** M. P. Russelle\*<sup>1</sup>, N. P. Martin<sup>2</sup>, and D. H. Putnam<sup>3</sup>, <sup>1</sup>*USDA-Agricultural Research Service, St. Paul, MN*, <sup>2</sup>*USDA-ARS-US Dairy Forage Research Center, Madison, WI*, <sup>3</sup>*University of California, Davis*.

The decline in use of perennial forages in US dairy herd rations is due in part to higher yield, greater energy content, and more uniform forage quality of corn silage. Although conversion of land from rotations of perennial with annual crops to continuous annual cropping has logistical advantages, it increases the need for fertilizer nitrogen (N), pest control, and energy, and raises the risk of soil erosion, nutrient runoff, nitrate leaching, and impaired soil, water, and air quality. Perennial forages help replace soil organic matter, which improves soil aeration, water holding capacity, and nutrient supply. Thus, these forages help offset organic matter declines due to increased soybean production and replacement of solid manure with manure slurry. Additional benefits to wildlife and aesthetics may play roles on some farms, such as those near suburban development. Alfalfa offers a combination of advantages available in no other perennial forage crop. It has the capacity for high N fixation, which provides a source of free N to the farm. Fixation of new N decreases when soil N supply is high, however, alfalfa helps buffer swings in N supply. Recent research results on a regional scale and at a feedlot remediation site demonstrate the importance of this buffering. Alfalfa's deep roots and large N requirement help reduce nitrate leaching far better than shallow rooted legumes, and some grass forages offer the same benefit. Expanding the acreage planted to perennial forages likely will require new markets or external support. The Conservation Security Program has offered funding in targeted watersheds for practices that reduce degradation of public resources. Examples of new markets include the potential for biomass energy from alfalfa stems, with leaves as a valuable by-product, and a new facility in southern Minnesota that will be extracting four products from alfalfa and returning the residue to dairies as feed. Several improvements in perennial forages would enhance their benefits, including increased yield potential, reduced winterkill, adaptation to less frequent harvests, greater utilizable protein, greater phosphorus uptake, and reduced potassium accumulation.

**Key Words:** Alfalfa, Nitrogen, Environment

**630 Preservation of protein during harvest and storage.** L. Kung, Jr.\*<sup>1</sup> and R. E. Muck<sup>2</sup>, <sup>1</sup>*University of Delaware, Newark*, <sup>2</sup>*USDA-ARS, Madison, WI*.

Prior to harvest, 80 to 95% of the crude protein (CP) in perennial grass and legume forages is true protein. The remainder is primarily peptides, free amino acids, nitrate and ammonia. The integrity of plant cells is gradually lost through physical damage from the mower-conditioner or reduction in turgor from drying in the field. This loss of cell integrity releases plant endopeptidases and exopeptidases that cleave proteins to peptides and free amino acids. Dependent upon temperature and the rate and length of drying, the soluble nonprotein nitrogen (NPN) in a forage when baled or chopped for silage may be 15 to 40% of total CP. Further reductions in true protein during storage are generally small in hay. However during ensiling, more proteolysis occurs that may result

in upwards of 60 to 70% of the total CP being NPN. Microbial activity during ensiling may change the composition of the NPN. Clostridia, enterobacteria and some lactic acid bacteria may use amino acids as substrates producing primarily ammonia. Proteolysis during harvest and storage is affected by forage species. Generally more proteolysis is expected in legumes than grasses, but perennial ryegrass species are as susceptible to proteolysis during ensiling as alfalfa. Within legume species, proteolysis is negatively correlated with tannin content. However, tannin content in some species may be high enough to adversely affect nitrogen utilization by the cow. Red clover has a polyphenol oxidase that reduces proteolysis. Currently, producers have few options to reduce proteolysis. Silage inoculants have little effect on the loss of true protein but do reduce adverse microbial activity, minimizing ammonia and preserving more free amino acids and peptides. Acid additives, such as formic acid, are common in northern Europe and can reduce proteolysis. Cost and safety issues have kept these products from adoption in North America. Ideally, modifying forages to contain either polyphenol oxidase and its substrates or low levels of tannins would be beneficial for preserving forage protein during harvest and storage.

**Key Words:** Protein, Proteolysis, Hay

**631 Challenges in utilization of high protein forages by lactating dairy cows.** P. Huhtanen\*<sup>1</sup>, G. A. Broderick<sup>2</sup>, and J. B. Russel<sup>3</sup>, <sup>1</sup>*MTT Agrifood Finland, Jokioinen, Finland*, <sup>2</sup>*USDA-ARS, Madison, WI*, <sup>3</sup>*USDA-ARS, Ithaca, NY*.

Forages are good for the environment and cow health. However, forages of sufficient quality often have elevated levels of rumen-degraded protein (RDP), much of which is converted to ammonia by ruminal microorganisms and excreted as urea. As a result, efficiency of N utilization by lactating cows fed high-forage rations can be low, leading to an increased risk of N leaving the farm through ammonia volatilization from manure or via nitrate losses in water. Despite high CP concentration in high quality grass and legume silages, protein supplementation has resulted in economical milk protein yield responses. The rumen operates as a continuous culture system and microbial growth is dependent on appropriate sources of both N and energy; this has led to suggestions that lack of synchronization of N and energy release is one reason for the low efficiency of the capture of N by ruminal microbes. However, there is little experimental evidence to support improved N utilization in response to timing energy and N release. For grass silage based diets, the incremental N from earlier harvest is better utilized than that from increased N fertilization due to improved fermentable energy supply. Feeding red clover silages has improved N utilization compared with alfalfa silages but, compared with grass silages, N utilization has been lower. Red clover has decreased proteolysis in the silo compared with both alfalfa and grass silages. This may account for the greater ruminal out-flow of non-ammonia N, mainly as feed N. However, this has not translated into improved milk protein yield in cows fed red clover silages. Certain bacteria isolated from the rumen have very high activity for producing ammonia from free amino acids. Suppressing these organisms in the rumen may aid in improving the supply of metabolizable amino acids in dairy cows fed these hay-crop silages. Strategies for improving N utilization in cows fed diets based on high quality forages, such as supplementary feeding of energy, protein and amino acids, will be addressed. Different aspects of modification of forage plants in relation to the potential to improve N utilization also will be discussed.

**Key Words:** N utilisation, Forage, Dairy cow

**632 Manure nitrogen transformations in air, soil and crops on dairy farms.** J. M. Powell<sup>\*1</sup>, K. F. Knowlton<sup>2</sup>, M. P. Russelle<sup>3</sup>, and M. D. Hanigan<sup>2</sup>, <sup>1</sup>USDA-ARS Dairy Forage Resh. Center, Madison, WI, <sup>2</sup>Virginia Tech University, Blacksburg, <sup>3</sup>USDA-ARS Dairy Forage Resh. Center, St. Paul, MN.

Only 25 to 35 % of the crude protein (CP) consumed by dairy cows is converted into milk. Such poor use of dietary CP may be due to inefficiencies associated with forage nitrogen (N) capture and metabolism. Manure N excreted in feces and urine, and the transformation of manure N in air, soil and crops are highly influenced by what dairy cows consume. For example, reducing dietary CP resulted in less total manure N, especially urine N excretion. Ammonia loss from manure from a low CP diet (13.6%) was lower than from a high CP diet (19.4%), representing 9 and 25% of applied manure N, respectively. Increasing condensed tannin content of dietary forage legumes also reduced urine N excretion. Ammonia emissions from barn floors were greater from manure derived from alfalfa silage (AS)-based diets than either birdsfoot trefoil with low or high tannin levels. After application to soil, feces from cows AS-based diets generally lead to higher soil inorganic N (IN) levels than soils amended with feces from corn silage-based diets; feces from AS-based diets increased plant yield and N uptake; feces from high CP diets resulted in greater soil IN levels than feces from low CP diets; and feces from low CP diets did not increase soil IN but decreased plant yield and N uptake. Only a small increase in N efficiency is necessary to make substantial reductions in the dairy industry's contribution to the environmental N load. There appears to be a range of dietary options that satisfy the nutritional requirements of high-producing dairy cows, yet produce manure that has differential effects on post-excretion transformations and environmental losses. Dairy production efficiencies may be gained and manure N losses reduced by incorporating moderate levels of tannins or other protein protection compounds into forages to enhance CP use and reduce dietary CP concentrations, and by developing perennial forages that tolerate manure applications, have improved ammonia absorption and assimilation potential, and are able to assimilate excess soil nitrates.

**Key Words:** Forages, Manure, Nitrogen cycling

**633 Transforming forage plants to increase nitrogen utilization in dairy systems: What are the possibilities?** R. Hatfield<sup>\*1</sup>, J. Grabber<sup>1</sup>, M. Sullivan<sup>1</sup>, G. Waghorn<sup>2</sup>, and M. McCaslin<sup>3</sup>, <sup>1</sup>USDA-ARS, Madison, WI, <sup>2</sup>Dexcel Limited, New Zealand, <sup>3</sup>Forage Genetics, St. Paul, MN.

Forages can supply adequate protein to meet the nutritional needs of high producing dairy cows, at least as the crop stands in the field. However proteins are one of the most labile nutritional components in most forages, often being excessively degraded during ensiling and ruminal digestion, leading to depressed amino acid absorption and excessive urea excretion by cattle. Even when forages are grazed, protein-use efficiencies are often low due to rapid plant cytoplasmic protein degradation in the rumen. To maintain high production, dairy diets are frequently supplemented with a protein source to compensate for poor forage protein use. Traditional breeding and molecular approaches can be used to modify forages for improved protein-use by cattle. For example, redesigning alfalfa to produce polyphenol oxidase and *o*-diphenols or condensed tannins would lead to decreased protein degradation during ensiling and ruminal digestion with a likely increase in amino acid absorption by cattle. Production and feeding of such a forage would reduce urea excretion and possibly slow nitrogen release from feces and crop residues, thereby reducing nitrogen losses from farms. Altering specific gene expression in the lignin pathway may allow decreased lignification and increased fiber digestion for improved nitrogen utilization. Genetic selection or molecular alteration of forages to produce greater quantities of rapidly fermented carbohydrates should enhance conversion of non-protein nitrogen to ruminal protein for utilization by cattle. Increasing total biomass production that has good quality remains a challenge for forage production. Exploiting the genetic potential for total biomass production in forages is just now being explored. Redesigning forages to function more efficiently as effective nitrogen sources for dairy cows is not impossible; it could decrease the need for protein supplements, and ultimately decrease nitrogen losses to the environment.

**Key Words:** Protein, Plant-modification, Nitrogen waste

## Physiology and Endocrinology: Endocrinology

**634 An erythropoietin receptor (EPOR) gene polymorphism (SNP) alters EPOR mRNA in fetal liver of swine during early gestation.** J. L. Vallet<sup>\*</sup> and B. A. Freking, USDA, ARS, U.S. Meat Animal Research Center, Clay Center, NE.

We previously reported that an EPOR gene C/T SNP was associated with litter size. The T allele created a putative GATA-1 site, which was predicted to increase EPOR gene expression. This experiment determined whether the SNP was associated with: (1) EPOR gene expression by the fetal liver or (2) maturation of the fetal blood supply during early gestation. CC and CT gilts were unilaterally hysterectomized-ovariectomized at 160 d of age, mated to boars of like genotype and slaughtered on d 25 (n = 13 CC, 13 CT), 30 (19, 25) and 40 (14, 15) of gestation. Numbers of corpora lutea (CL) and live fetuses were recorded. For CT gilts only, a blood smear was prepared for each fetus, the fetus was weighed and fetal liver and other tissues were collected. Percentage of nucleated blood cells was assessed on d 30 and 40 (all blood cells were nucleated on d 25). DNA was prepared from fetal tissues to determine EPOR genotype. Total RNA

was prepared from fetal liver of one fetus of each genotype for each litter (d 25 and 30), and EPOR mRNA was measured using real time RT-PCR. Number of fetuses decreased ( $P < 0.01$ ) between d 30 ( $11.7 \pm 0.4$ ) and 40 ( $8.5 \pm 0.5$ ) but did not differ between gilt genotypes. Percent nucleated cells decreased significantly between d 30 and 40 but were not affected by fetal genotype. Fetal liver EPOR gene expression was greater ( $P < 0.01$ ) on d 30 compared to d 25 of gestation and a significant additive effect of genotype ( $P < 0.01$ ) was observed (d 25,  $3.8 \pm 0.7$ ,  $4.6 \pm 0.7$ ,  $5.4 \pm 0.7$ ; d 30,  $9.8 \pm 0.5$ ,  $10.3 \pm 0.5$ ,  $11.3 \pm 0.5$  relative units; CC, CT, and TT, respectively). Although these results do not indicate an effect of the SNP on litter size, uterine capacity affects litter size on d 40 or later, and the number of gilts on d 40 were likely inadequate. The SNP also did not affect maturation of the fetal blood supply. However, the T allele was associated with increased EPOR gene expression during early pregnancy as predicted and, thus, could influence erythropoiesis and fetal survival.

**Key Words:** Blood, Erythropoiesis, Fetus