than 50% of the investigated plots received a nitrogen input above a deposition level at which the species diversity of the ground vegetation may be at risk.

Key Words: Nitrogen, Critical loads, Animal production

**590** The role of nutrition in reducing nutrient output from ruminants. L.D. Satter<sup>\*1</sup>, T.J. Klopfenstein<sup>2</sup>, and G.E. Erickson<sup>2</sup>, <sup>1</sup>U.S. Dairy Forage Research Center, Madison,WI, <sup>2</sup>University of Nebraska, Lincoln.

Much of the effort expended on nutrient management has focused on the post-excretion product. It is important to keep in mind that management of the diet can have important impacts on quantitative and qualitative aspects of the excreted nutrient. Surveys of nutritionists and extension specialists show that dairy producers are advised to feed .45-.50% phosphorus (P)(DM basis) in their lactating cow diets. This is 20-30% in excess of NRC (2001) requirements. Feeding to requirement would reduce P excretion by 30% or more, and would reduce solubility and potential for runoff of the P that is applied to fields. Nitrogen (N) excretion by dairy cows can also be decreased, but by a lesser amount. Balancing RUP and RDP, and use of protected methionine along with strategic selection of protein supplements that are relatively rich in lysine, may permit a 10-15% reduction in total N excretion, with most of the reduction occurring in urinary N. Urinary urea, following conversion to ammonia, is the N excretion product most vulnerable for loss to the environment. Feedlot cattle routinely consume P in excess of NRC (1996) predicted requirements, and recent research suggests the NRC estimates of the P requirement are high. Decreasing dietary P from the industry average (.35% P) to the NRC predicted requirement (.22-.28%) decreased P input by 33 to 45% and excretion by 40-50% in nutrient balance studies. With grain-based feedlot diets, overfeeding P is inevitable. At minimum, supplemental P sources should be removed from diet formulations. More accurate formulation of feedlot diets for protein provides opportunity for reducing N excretion. Using the NRC model for metabolizable protein, and employing phase-feeding, N inputs may be decreased by 10-20% from the feedlot industry average of 13.5%dietary CP. This translates into a 12-21% reduction in N excretion, and 15-33% reduction in ammonia volatilization in open-dirt feedlot pens. Diet formulation can have important impact on the amount of N and P excreted in both dairy and beef. It is much easier to control potential pollutants by managing their release into the environment than to recover or confine them once they are released.

Key Words: Nitrogen, Phosphorus, Ruminant

**591** Nutritional strategies to reduce environmental emissions from non-ruminants. P.R. Ferket<sup>\*1</sup>, R.C. Angel<sup>2</sup>, E. van Heugten<sup>1</sup>, and T.A. van Kempen<sup>1</sup>, <sup>1</sup>College of Agriculture and Life Sciences, North Carolina State University, Raleigh, NC 27695, <sup>2</sup>Department of Animal Science, University of Maryland, College Park, MD 20742-2311.

The amount of nutrients (i.e. N, P, Zn and Cu) and associated odors emitted from production animals into the environment can be modulated by several different nutritional strategies, but their practical application is dependent upon costs and other biological limitations. In general, nutrient excretion may be reduced by avoiding the feeding of excessive amounts or using nutritional manipulations to enhance nutrient utilization in the animal. Manufacture and handle feed to minimize wastage and improve feed/gain. Develop feeding programs that are specific for sex and strain of animal, increase the number of feed phases, and formulate diets according to the minimum nutrients required to satisfy production goals. Use the ideal protein concept to estimate amino acid requirements and use synthetic amino acids supplements to reduce N emission. Use feed ingredients with high digestibility and nutrient bioavailability, and formulate diets based on nutrient availability instead of total nutrient content. Nutrient digestibility of feedstuffs is dependent upon processing conditions, genetic characteristics of the grains and oilseeds, and the presence of nutritional antagonists in the diet. Avoid feed ingredients that lead to odor production (e.g. fishmeal and some easily fermentable feed ingredients). Use feed additives, such as antibiotics, nonstarch polysaccharides, direct-fed microbials, organic acids, microbial enzymes (i.e. phytase, carbohydrases, and proteases) to increase the digestibility and absorption of nutrients or to modulate microflora. Finally, a cost factor for the control or disposal of nutrients or odor should be considered in the feed formulation to optimize the various nutritional strategies discussed above. Regardless of biological and economic limitations, significant reductions in nutrient and odor emission from non-ruminants can be achieved by appropriate nutritional strategies.

Key Words: nutrition, nutrient and odor emission, non-ruminants

**592** Development of comprehensive nutrient management plans: Practical aspects of getting nutrient management plans implemented . Mary Combs<sup>\*1</sup>, <sup>1</sup>USDA-Natural Resources Conservation Service, Raleigh, NC.

The 1998 Clean Water Action Plan required the EPA and USDA to jointly develop a unified strategy to minimize the environmental and health impacts of the nation's animal feeding operations (AFOs). This Unified National Strategy for Animal Feeding Operations identified a national expectation that all AFOs develop and implement Comprehensive Nutrient Management Plans (CNMPs) by 2009. Focusing on the smaller, non-regulated (federal) AFOs with limited resources, NRCS and its partners may need to assist with an estimated 262,700 CNMPs across the U.S. to meet this expectation.

Significant CNMP development and implementation issues remain: (1) Substantial resources for staff and training are required to provide this accelerated technical assistance for CNMP development. Developing the CNMP is just the first step; considerable follow-up with producers is required to assist with operation, maintenance, and revision of the Plans as producers' needs change. (2) More research is needed in several critical areas to better understand nutrient movement and validate states' phosphorus indexes and models that assess potential nutrient losses. (3) In areas of concentrated AFOs and limited land for application, nutrient management policy may result in no technically or economically feasible solutions for the producer. (4) Both regulators and technical specialists must recognize the economic situation of producers. The cost of waste management systems is site specific, and is not only a function of operation size. The special challenges to limited resource farmers must be considered. (5) Cost sharing and incentives are inadequate to meet the needs. In North Carolina, USDA's Environmental Quality Incentives Program and the N.C. Agricultural Cost-Share Program fund about 1/3 of the existing demand. (6) Ensuring compatibility with state programs, laws, rules, and certification criteria for technical specialists will continue to a significant coordination effort. (7) NRCS's image by its customers continues to evolve. NRCS practice standards, developed to support voluntary USDA programs, are becoming regulatory instruments, as federal or state regulations reference these standards.

Key Words: AFO, CNMP, NRCS

#### **Novel Genes and Gene Products**

**593** Differential display as a tool to identify a steroid-induced gene. Robert Kemppainen\*, *Auburn University, Auburn, Alabama*.

Differential display is one of several methods designed to identify differentially induced or expressed genes and has been used successfully in many studies to identify new genes in various tissues or cells. The basic method involves collection of RNA from target tissues followed by cDNA synthesis using oligo-dT primers designed to make cDNA from subpopulations of the mRNA. These different cDNA's are then used as templates in PCR in conjunction with the original oligo-dT primer and a set of arbitrary upstream primers. Labeled PCR products are loaded onto sequencing gels so that side-by-side comparisons can be made to identify up- or down-regulated genes. We used the technique to identify dexamethasone-induced genes in a pituitary cell line. Since steroid negative feedback requires gene transcription/translation and the identity of steroid-induced genes is unknown, differential display seemed to be an ideal technique for this purpose. Cells were treated with dexamethasone or its vehicle and RNA was collected and used for differential display. The screen performed used 240 primer combinations, surprisingly; only about 20 induced bands were consistently generated. Of the 20, 19 turned out to be false positives and one dexamethasone-induced gene was identified; its full-length cDNA was cloned from a library and sequenced. The cDNA is a novel member of the Ras-superfamily and was named Dexras1 due to its ability to be induced by dexamethasone. Experiments are in progress to characterize the role of Dexras1 in the pituitary and in other steroid-responsive tissues. Overall, differential display was extremely useful in identification of a novel gene, however, it may require considerable effort in terms of testing various PCR primer combinations and the technique may be expected to generate a significant number of false positives.

 ${\sf Key}$  Words: Differential , Display, Dexame thasone

## **594** Genes, Chips and Animal Biology. Nagappan Mathialagan<sup>\*1</sup>, Charles Bolten<sup>1</sup>, Steven Wagner<sup>1</sup>, John Byatt<sup>1</sup>, and Frances Buonomo<sup>1</sup>, <sup>1</sup>Monsanto Animal Agricultural Group.

Genomic technologies have transformed the animal biology research into a new era of discoveries in a similar fashion as the introduction of radioimmunoassay techniques. Genomics has resulted in the identification of thousands of new gene sequences in farm animal species with no real link to functional association. Comparative genomic analysis with completed genomes such as human has been used to discover orthologous genes. However, this approach still leaves out the annotation of genes which are novel to the animal species. Gene expression technologies like Microarrays and Serial Analysis of Gene Expression (SAGE) are used to determine the expression of thousands of genes simultaneously. Species-specific microarrays need to be used to associate a function to these new genes. However, cross-species microarrays may be used in instances where there are no species-specific arrays. We have used Incyte human microarrays for transcript profiling of bovine mammary gland to identify regulated genes associated with milk production. A set of human gene homologues were identified that are regulated during lactation and involution. Genes up-regulated during lactation, identified by heterologous profiling, were selected for confirmation by other methods such as Northern blot analysis, quantitative RT-PCR, subtractive cDNA libraries and nylon arrays. One example of a regulated gene we selected for confirmation was Stearoyl-CoA-Desaturase (SCD), an enzyme involved in the synthesis of conjugated linoleic acid. An increased expression was associated with lactation while a sharp decline in the expression was observed with involution. In addition, our experience with

heterologous arrays showed that genes can be erroneously identified due to sequence identity of bovine genes to unrelated genes in human. This observation emphasizes the preference to use species-specific arrays for gene expression studies.

Key Words: Transcript Profiling, Microarrays, Genomics

### **595** Proteomics in the animal sciences. Lawrence Dangott\*, *Texas A&M University, College Station, TX.*

One of the goals of biologists in the post-genome era will be to characterize all the proteins within an organism, tissue or organelle, in order to describe the pathways and protein interactions that mediate cellular function. Proteomics is the term given to the large-scale analysis of proteins using biochemical, biophysical and chemical techniques of analysis. Although traditionally associated with the two-dimensional display of large numbers of proteins, in the post-genomic era, proteomics is dividing into three main areas; 1) protein identification and microcharacterization; 2) differential expression analysis of proteins in normal and altered tissues; and 3) studying protein-protein interactions. Approaches to achieve these goals require the combination of traditional molecular biological, biochemical and biophysical techniques with the expanding capabilities of high-throughput robotics and high-sensitivity, high-resolution mass spectrometry as well as the development of new technologies. These kinds of approaches are being used in our laboratory and others to explore and explain the functions, interactions and regulation of proteins in animal reproductive biology and environmental toxicology. Proteins involved in embryo implantation are being identified in ovine uteri using 'knock-out' ewes, two-dimensional gels and in gel digestion techniques coupled to automated protein micro-sequencing and MALDI-TOF (matrix-assisted laser desorption ionization time-offlight) mass spectrometry. Similarly, proteins involved in horse sperm differentiation are being identified by applying these techniques to proteins isolated from in vitro cultures of equine seminal tubules. In related experiments, post-translational modifications are being mapped by ion-trap electrospray mass spectrometry and multi-dimensional chromatography coupled with mass spectrometry is being used to identify components of protein complexes.

the nutritional quality of MFGM, a more detailed analysis of the com-

position of buttermilk has suggested the utilization of buttermilk as an

ingredient in the manufacture of foods, for example low fat cheese and yogurt. The behavior of buttermilk as a functional ingredient can be

attributed to the presence of skim milk proteins and the MFGM, how-

ever the role played by the various components and their interactions is

not understood. Processing history and compositional differences also

seem to affect the functionality of MFGM. Understanding the various

components and the functional properties of buttermilk will allow this

Key Words: Proteomics, Mass spectrometry

#### Preharvest and Postharvest Approaches to Modification of Milkfat

# **596** The milk fat globule membrane of buttermilk: a unique ingredient. M. Corredig\*, Department of Food Science and Technology, University of Georgia.

The presence of material derived from the milk fat globule membrane (MFGM) makes buttermilk (the byproduct of buttermaking) distinct from any other dairy product. Studies of MFGM have revealed strong associations of the membrane lipids with various membrane proteins (butyrophilin, xanthine oxidase and some minor proteins). When membrane material is isolated from buttermilk a high ratio of polar lipids is found, in particular phosphatidyl ethanolamine, phosphatidyl choline and sphingomyelin. Phospholipids play an important role in many metabolic processes, and phospholipid-enriched fractions are today marketed as important ingredients in a variety of dietary products. Furthermore, evidence is emerging that sphingomyelin from milk may have anti-cancer properties and other health-related benefits. In addition to

byproduct to become a source of new and unique ingredients. Our discussion will review the current literature in this area and present some thoughts on the further development of commercial products derived from the MFGM.

Key Words: phospholipids, MFGM, buttermilk

#### Role of Extracellular Matrix (ECM) in Growth and Development

**597** The role of the extracellular matrix in growth and development: An introduction. M.W. Orth\*, *Michigan State University*.

Besides providing structural support, the extracellular matrix (ECM) has recently been shown to play a significant role in the regulation of tissue growth and development. As an example, certain ECM molecules can sequester growth factors and release them during tissue remodeling. Also, proteolytic products of ECM molecules can have a unique biological activity via interactions with cell surface receptors. The focus of this symposium will be to examine the role and regulation of the ECM in four tissues of particular interest to the animal scientist. Dr. Sandy

Velleman will describe the architecture of the ECM in skeletal muscle tissue with an emphasis on the function of the proteoglycan component. The predominant focus on myofibrillar proteins traditionally has overshadowed this exciting area of research. Dr. Tom Schmid will discuss the role of the ECM during endochondral bone formation, with an emphasis on ECM proteins found in unique regions of growth plate and articular cartilage that were discovered in his laboratory. He will also discuss the potential of using ECM molecules as biological markers in physiological fluids to monitor the development and health of the skeleton. Dr. Russ Hovey will discuss the development of the mammary gland and its complex architectural structure. Specifically his research interest focuses on understanding the contribution of the mammary stroma dur-