quality traits are considered, EF are a good alternative to CF for production of heavy pigs destined to the dry-cured industry.

Key Words: Iberian Pigs, Gilt Castration, Carcass Quality

396 Effects of pump rate and cooked temperature on pork loin instrumental, sensory descriptive and consumer-rated characteristics. R. T. Baublits*, J.-F. Meullenet, J. T. Sawyer, J. M. Mehaffey, and A. Saha, *University of Arkansas*, *Fayetteville*.

Fresh pork loins (n = 15; muscle sections, n = 30; arranged in an incomplete block design) were utilized to evaluate the effects of untreated muscles (0% pump rate), or muscles enhanced with a solution comprising 0.4% sodium tripolyphosphate and 1.0 % sodium chloride at either a 6% or 12% pump rate, and cooked to a 71°C or 82°C end-point internal temperature on meat quality, instrumental texture characteristics, sensory profiles, and consumer acceptance. Loins enhanced at a 12% pump rate had a higher ($P < 0.05$) pH than untreated loins. While there were no differences in Warner-Bratzler shear force due to cooked temperature, chops enhanced at a 12% pump rate had lower ($P < 0.05$) shear force values than untreated chops. Additionally, chops enhanced at 6% or 12% pump rates had lower ($P < 0.05$) razor shear force values than untreated chops. Descriptive sensory analyses revealed chops cooked to 71°C had a more intense ($P < 0.05$) blood serum flavor than chops cooked to 82°C. Untreated chops had less intense ($P < 0.05$) pork fat flavor, and more intense ($P < 0.05$) blood serum, livery, and cardboard or oxidized flavor characteristics than chops enhanced at 6% or 12% pump rates. Additionally, there were no differences ($P > 0.05$) in metallic intensity between enhanced and untreated chops. Sensory panelists reported chops enhanced at 6% or 12% pump rates to generally be more tender than untreated chops. Consumers reported a higher ($P < 0.05$) overall acceptability for chops enhanced at 6% or 12% pump rates. Overall acceptance scores were in average 2 points higher for the enhanced chops (mean =7.03) than for the untreated chops (mean =4.94) on the 9-point hedonic scale. Furthermore, both sensory panelists and consumers reported chops enhanced at 6% or 12% pump rates to be similar ($P > 0.05$) in juiciness characteristics, regardless of end-point temperature. However, untreated chops cooked to 82°C were less juicy ($P < 0.05$) than untreated chops cooked to 71°C, suggesting retained palatability when enhanced chops are cooked to more abusive temperatures.

Key Words: Pork, Enhancement, Cooked Temperature

Milk Protein and Enzymes: Milk Protein Interactions

397 Casein micelles and whey proteins: Physical interactions and functional properties. S. G. Anema*, *Fonterra Research Centre*, *Palmerston North, New Zealand*.

The functional properties of milk products are inextricably linked to the denaturation of the whey proteins and their interactions with other milk protein components. As the level of whey protein denaturation is easily measured, milk products are often classified by their level of whey protein denaturation. We examined the denaturation of the major whey proteins in milk, the factors affecting this denaturation, and the relationship between the level of denaturation and the functional performance of the milk in a simple acid gel system. From these studies, models to predict the degree of denaturation of the whey proteins at a range of temperature, heating time, and composition combinations were generated. However, the relationship between the level of denaturation and the functional performance of the milk in acid gels was rather poor.

An understanding of the denaturation reactions of the whey proteins provides information on only the initial steps of a complex series of aggregation reactions that occur when milk products are heated. Analysis and interpretation of these aggregation reactions are often difficult. In our further studies, we examined one specific aggregation reaction considered to be of importance to functionality: the interaction between the denatured whey proteins and the casein micelles. The effects of numerous factors (pH, concentration, composition, heating regime, etc.) on these interactions, and the relationship between these interactions and the functional performance of the milk in our simple acid gel system, were studied. We found that it was possible to substantially manipulate the degree of interaction between the denatured whey proteins and the casein micelles by altering the conditions of the milk at heating. These changes in interaction behavior had a marked effect on the physical properties of the milk and the functional performance in our acid gel system, allowing significant modification of the textural properties. These results exemplify the importance of the aggregation reactions of the denatured whey proteins in determining the properties of heated milk systems.

398 Process-induced intermolecular bonds in milk protein gels and their impact on rheological properties. J. Hinrichs*, *University of Hohenheim*, *Germany*.

The various texture properties in milk products are mainly determined by the protein content, the interacting protein fractions, the milieu conditions and the process technology applied. Furthermore, the kind of technology applied may force or enhance the formation of intermolecular interactions stabilizing the nano- and microstructure which on their part may correlate with the functional properties or texture of milk protein gels. An extraction test applying different buffering agents was established in order to quantify the stabilizing covalent and non-covalent bonds in milk gels. The texture properties were characterized by dynamic rheological measurements and penetration tests simultaneously. This enables us to study the process–structure–function relationship in pressure-induced, heat-induced, rennet-induced and acid-induced milk gels.

The combined characterization of the gel structure demonstrated that heat-induced and pressure-induced whey protein gels - predominantly stabilized by covalent disulphide bonds - appeared elastic, which is expressed by the low loss angle. In high pressure-induced whey protein gels, the loss angle declined when the amount of stabilizing disulphide bonds was increased by technological means. Contrary to that, the loss angle remained constant while the storage modulus increased when the amount of calcium bridges respectively the amount of hydrophobic interactions increased in rennet-induced respectively acid-induced gels via technological means. The gels appeared firmer but the viscoelastic properties remained unchanged.

In summary, the kind and quantity of the intermolecular interactions stabilizing milk protein gels can be determined by applying various buffer systems. It seems likely that analysis of process-induced covalent and non-covalent bonds will become an additional tool for texture analysis in scientific issues and for solving technological problems.

399 The 500 Myr story of the evolution of phosphoproteins that made milk possible. C. Holt* and R. A. Clegg, *Hannah Research Institute*, *Ayr, UK*.

Because of recent genome sequencing projects, casein genes are now recognised to have evolved as members of a paralogous group of secretory calcium (phosphate) binding phosphoproteins (SCPPs). The group's origins lie in ancient homologues of a protein called SPARC which gave rise to a SPARC-like protein (SPARCL1) and an osteopontin-like protein (OPN) at the end of the Cambrian period, about 500 Myr ago [1,2]. The earliest mineralised tissues (denticles) employing calcium phosphate emerged at this time, to be followed by external bony plates and scales and later endoskeletons. In extant mammals, members of the SCPP family are found in the biological fluids saliva and milk and in the mineralised tissues of bone, dentine and enamel. OPN, however has

a wider distribution in soft and hard tissues and many other biological fluids.

In previous work we have shown that a number of casein phosphopeptides can sequester amorphous calcium phosphate [3,4] to form a thermodynamically stable [5] nanocluster of radius 4nm. Calcium phosphate nanoclusters occur naturally as substructures in the colloidal casein particles of milk, known as casein micelles [6]. They allow a high concentration of an otherwise highly insoluble calcium salt to be achieved without danger of precipitation of a solid phase. Accordingly, we have suggested that the casein micelle is a solution to the problem of pathological calcification in the mammary gland.

We will consider the structural and thermodynamic requirements for the formation of nanoclusters and show that one other non-casein member of the group of SCPPs is able to sequester calcium phosphate and form nanoclusters. These findings suggest that calcium phosphate sequestration by phosphoproteins may be part of a general solution to the problem of the control of biocalcification.

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400 HAMLET, an alpha-lactalbumin folding variant that induces tumor cell apoptosis. C. Svanborg*, *University of Lund*, *Sweden*.

HAMLET (**H**uman **a**-lactalbumin **M**ade **LE**thal to **T**umour cells) is a structurally defined protein-fatty acid complex derived from human milk. The complex kills cancer cells but leaves healthy differentiated cells intact. In the laboratory, HAMLET kills >40 different cancer cell lines, with leukemic cells being the most sensitive. The cells die by an apotosis like mechanism which is paradoxical as most tumour cells carry mutations that prolong their life span by allowing them to avoid apoptotic cell death. Thus, HAMLET appears to identify a death mechanism that is conserved in tumor cells, but lost as cells differentiate and mature. As the protein and lipid that form HAMLET occur naturally in human milk it may be speculated that HAMLET might contribute to the lowered cancer incidence in breast-fed children.

Since the discovery of HAMLET in 1995, we have studied

1. The **molecular characteristics of the compound**

2. **The mode of action on tumour cells.**

3. **The protective potential of HAMLET in tumor models** and human patients

HAMLET was used for brain tumour treatment in a human rat xenograft model. (Ref: ***Cancer Res** 64:2105, 2003*.) Infusion of HAMLET into the tumor was shown to delay tumor growth and prolong survival of immmunodeficient rats, carrying a human glioblastoma tumor. Apoptotic cells were detected in the tumor but not in surrounding healthy brain tissue.

HAMLET was also used for human skin papilloma treatment (Ref: ***N Engl J Med** 350:2663, 2004*.). A placebo controlled trial of topical HAMLET treatment was carried out and completed with a two-year follow-up. HAMLET was shown to reduce the volume of skin papillomas and to promote the resolution of the lesions.

The molecular, functional and therapeutic aspects will be discussed

Nonruminant Nutrition: Stable Isotope Tracer Techniques for Nonruminant Nutrition Research and Their Practical Applications

401 Mass isotopomer distribution analysis (MIDA) for studying intermediary nutrient metabolism. B. J. Bequette*, *University of Maryland*, *College Park*.

Functional genomic and proteomic investigations are beginning to characterize the metabolic controls and relationships between gene function and nutritional physiology. Still missing in this metabolic roadmap is characterization of the activities or fluxes through these integrated pathways that ultimately determines nutrient utilization. In the past 10 years, stable isotope (^{13}C, ^{15}N, ^{2}H) labeling (mass isotopomer) with mass spectrometric analysis has allowed fluxes through metabolic pathways to be measured in vivo and in vitro. MIDA refers to the measurement of the mass distributions in a molecule or molecular fragment that are characteristic of the unique biochemical pathway(s) of the nutrient's metabolism. This review will discuss the use of MIDA and how it can be used to dissect the integrative pathways of macronutrient (protein, carbohydrates, fat) metabolism. For example, in studies with laying hens, fish and chicks, MIDA has been applied to ascertain the nutritional essentiality, and thus metabolic inability for synthesis, of nucleic acids and some "non-essential" amino acids. Another application of MIDA has been the determination of the pathways and cycles of essential amino acid metabolism with regards to their metabolic roles other than for protein synthesis. When applied to the measurements of new gluconeogenesis and tissue utilization of glucose, MIDA has exposed the importance of amino acids as glucogenic substrates, and also highlighted the importance of the interconnectivity of the pathways of metabolism of carbohydrates, volatile fatty acids, amino acids and fatty acids to maintain anaplerosis and cataplerosis (metabolic balance) via the Krebs cycle. As gene and protein expression profiles begin to build the global roadmap of nutrient utilization, it will be necessary to determine the functional and quantifiable significance of these metabolic pathways that make up the roadmap. Here, the use of MIDA, when applied to the study of macronutrient metabolism, can provide the details of the biochemical networks of nutrient utilization.

Key Words: Stable Isotope, Amino Acid, Glucose

402 Measuring splanchnic amino acid metabolism by using stable isotope tracers. B. Stoll* and D. Burrin, *USDA-ARS Children's Nutrition Research Center, Baylor College of Medicine*, *Houston, TX*.

The splanchnic bed is comprised of the liver and the portal drained viscera (PDV). The PDV, which include the stomach, intestines, pancreas, and spleen, represent 4-6% of body weight, yet they account for 20-35% of whole-body protein turnover and energy expenditure. The high nutrient needs of the gut are met first as a result of first-pass metabolism. Consequently, the first-pass metabolism of dietary nutrients by the gut, especially amino acids, has a critical influence on their availability to peripheral tissues and whole body requirements. Moreover, the systemic availability of dietary amino acids is key determinant of lean body growth rate. A complicating factor in the measurement of gut nutrient utilization is that the intestinal mucosa receives nutrients from two sources, the diet and the arterial circulation. However, combining measurements of the net portal balance with enteral and intravenous infusions of stable isoto-