**520** Wool quality and growth traits of Tasmanian pasture-fed crossbred lambs and relationships with plasma metabolites. A. E. O. Malau-Aduli\*, C. F. Ranson, and C. W. Bignell, *University of Tasmania, Hobart, Tasmania 7001, Australia.* 

Wool quality, growth and plasma metabolite traits of 500  $F_1$  progeny from Merino dams sired by 5 ram breeds were investigated to study the influences of sire breed, sex and their interactions with plasma metabolites aimed at dual-purpose crossbreeding options. Coopworth, Texel and White Suffolk sired progeny had significantly (P<0.05) heavier weaning weights (WWT) and average daily gains (ADG) than those sired by Dorset or East-Friesian rams. Coopworth-sired sheep had the highest WWT (31.3±1.7Kg) and East-Friesian sired sheep the lowest (22.9±3.1Kg) with ADG ranging from 0.15kg/day in East-Friesian to 0.23 Kg/day in Texel and White Suffolk sire breeds. Highly significant (P<0.01) sex by sire breed interaction were evident; Coopworth-sired ewe lambs had the highest WWT and ADG (34Kg, 0.27 Kg/day) and Dorset-sired ewe lambs the least (22kg, 0.15 Kg/day). Greasy fleece weight ranged from a minimum of 964g to a maximum of 1303g in Dorset and Coopworth-sired lambs respectively, with Coopworth and Texel sire breeds having significantly heavier (P<0.05) fleece weights than either Dorset, White Suffolk or East-Friesian. Texel-sired sheep had significantly larger (P<0.05) micron fibre diameter (23.4 $\mu$ m) than the 21µm recorded in White Suffolks and East-Friesians. There were also highly significant differences (P<0.01) between sire breeds in staple length (range 50-68mm) and staple strength (range 39-52Nktex), with males having finer fibre diameter (21 vs 23µm) and shorter staple length (55 vs 60mm). Regardless of sire breed or gender, blood plasma metabolites were well within the normal range. A strong, positive and significant phenotypic correlation of 0.72 existed between marking and weaning weights. There were no significant correlations between the wool quality and growth traits, essentially implying that producers can select for finer wool without compromising growth. Coopworth x Merino first cross was the overall best performing sheep breed studied because of its heavier liveweight, faster daily gain, heavy fleece weight and a comparatively lower micron fibre diameter than the other crossbreds.

Key Words: Tasmanian crossbreds, wool quality, plasma metabolites

**521** Bayesian estimation of genetic parameters for body weight traits and litter size of Moghani sheep using Gibbs sampling. N. Ghavi Hossein-Zadeh\*<sup>1,2</sup>, <sup>1</sup>University of Tehran, Karaj, Iran, <sup>2</sup>University of Guilan, Rasht, Iran.

The objective of the present study was to estimate genetic parameters for body weights at different ages and litter size in Moghani sheep. Traits were included birth weight (BW), 3 months weight (3MW), 6 months weight (6MW), 9 months weight (9MW), yearling weight (YW) and litter size (LS). Data and pedigree information used in this research were collected at Breeding Station of Moghani sheep (Ardebil, Iran) during 1987-2005. Linear and threshold animal models with additive genetic, maternal genetic, maternal permanent environmental and residual effects were implemented by Gibbs sampling methodology. A single Gibbs sampling with 100,000 rounds was generated by the MTGSAM program. The posterior means of genetic parameters were estimated based on the 900 samples that were left after elimination of 10,000 rounds in the burn-in period and 100 rounds of each thinning interval. Posterior means of direct heritability estimates for BW, 3MW, 6MW, 9MW, YW and LS were 0.29, 0.13, 0.14, 0.10, 0.31 and 0.10, respectively. Posterior mean estimates of maternal heritabilities were 0.29 for BW, 0.08 for 3MW, 0.11 for 6MW, 0.06 for 9MW, 0.10 for YW and 0.17 for LS. All the posterior mean of phenotypic correlation estimates among body weight traits at different ages were positive and changed from 0.08 to 0.68. But, the estimates of phenotypic correlations between litter size and body weights were negative and ranged from -0.69 to -0.08. A moderate negative direct genetic correlation has been estimated for 9MW-YW, but the estimates of direct genetic correlation between other body weight traits were positive and ranged from 0.08 to 0.88. But, there were negative medium to high direct genetic correlations between body weights at different ages and litter size, ranging from -0.92 to -0.28. Thus, selection for increased growth or LS may have a negative genetic effect on the other trait. The medium to high negative estimates of direct-maternal correlations for body weight traits or litter size suggest that it would be difficult to jointly improve direct and maternal growth ability for Moghani sheep.

Key Words: Bayesian inference, Moghani sheep, body weight

## Dairy Foods: Dairy Foods/Microbiology

**522** Molecular and technological characterization of lactic acid bacteria isolated from the Egyptian white pickled cheese. M. El Soda\*, M. Mohammed, S. Anwar, and S. Awad, *Department of Dairy Science, Faculty of Agriculture, Alexandriau University, Alexandria, Egypt.* 

Egyptian white pickled cheese samples were collected from different areas in Egypt. One hundred isolates obtained from the cheese samples were identified using repetitive genomic element-PCR (Rep-PCR) fingerprinting. The identified isolates were tested for efficiency of biomass production and separation, acidifying activity, autolytic, aminopeptidase and antagonistic activities and exopolysaccharide production. The obtained results revealed that Enterococcus faecium, Enterococcus faecalis, Lactobacillus paracasei subsp. paracasei, Lactobacillus plantarum and Lactobacillus delbrueckii subsp. lactis were the predominant species in Egyptian white pickled cheese. Fifteen percent of Lactobacillus and 2% of Enterococcus isolates showed fast acidifying activity. Aminopeptidase and autolytic properties were generally higher for Lactobacillus strains when compared to the enterococci. Among the lactobacilli, Lactobacillus paracasei subsp. paracasei was the highest in aminopeptidase activity and autolytic properties. Antagonistic activity was detected in 70% of Lactobacillus and 30% of Enterococcus

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isolates. Two strains of *Lactobacillus paracasei* subsp. *paracasei* and one of *Lactobacillus plantarum* were capable of producing exopolysaccharides in milk.

Key Words: Rep-PCR, Egyptian white pickled cheese, lactic acid bacteria

**523** Physiological and transcriptional response of *Lactobacillus casei* ATCC **334** to acid stress. R. Thompson\*<sup>1</sup>, V. Deibel<sup>2,3</sup>, J. Steele<sup>2</sup>, and J. Broadbent<sup>1</sup>, <sup>1</sup>Utah State University, Logan, <sup>2</sup>University of Wisconsin, Madison, <sup>3</sup>TracMicro, Madison, WI.

*Lactobacillus casei* is used as a starter culture in fermented foods, as a probiotic, and in the industrial production of lactic acid. *Lb. casei* produces lactic acid as a major end product of carbohydrate fermentation, which acidifies the environment. Cell survival in acidic environments is critical to industrial application of *Lb. casei*, so a fundamental knowledge of cellular physiology during acid stress may reveal strategies to enhance its industrial performance. Here, we investigated the effect of acid adaptation of *Lb. casei* ATCC 334 on viability during acid chal-

lenge at pH 2.0, cytoplasmic membrane fatty acid (CMFA) composition, and cellular transcriptome using microarray technology. Experiments investigating the effect of acid adaptation at several pH values (range=pH 3.0-5.0) on viability of Lb. casei at pH 2.0 indicated that some degree of acid tolerance was induced over a broad range in pH, but the highest survival was noted in cells that were acid adapted for 10 min at pH 4.5. Fatty acid (FA) methyl esters of CMFAs were prepared, separated and quantified by gas chromatography. The ratio of saturated:unsaturated FAs increased from 0.4 in non-adapted control cells to 4.3 and 5.3 after 10 and 20 min of acid adaptation (AA), respectively. The level of cyclopropane FAs increased from 5% in the control to 17 and 18% after 10 and 20 min of AA, respectively. Several differences were noted between the transcriptome of cells grown at pH 6.0 (control) to that from cells that were acid adapted for 5 min (AA1) or 20 min (AA2) at pH 4.5, or acid adapted for 20 min and acid shocked at pH 2.0 (AA-AS). For example, AA1 significantly (P<0.05) affected the expression of 15 genes, of which one (7%) was up-regulated. AA2 significantly affected 322 genes (of which 105 [33%] were up-regulated), while AA-AS significantly affected the expression of 66 genes (12 [19%] up-regulated). Real-time quantitative PCR (RT-PCR), an independent assay to confirm microarray data, was performed on ten selected genes. In general, the microarray results were validated by the RT -PCR data, and there was a positive correlation (r = 0.89) between the two methods.

Key Words: lactobacillus, acid shock, stress response

**524** Genotyping for strain-level differentiation of *Bifidobacterium animalis* ssp. *lactis.* J. R. Loquasto<sup>\*1</sup>, E. P. Briczinski<sup>2</sup>, A. M. Roberts<sup>1</sup>, E. G. Dudley<sup>1</sup>, R. Barrangou<sup>3</sup>, and R. F. Roberts<sup>1</sup>, <sup>1</sup>Pennsylvania State University, State College, <sup>2</sup>University of Wisconsin, Madison, <sup>3</sup>Danisco USA Inc., Madison, WI.

While numerous health benefits are attributed to probiotic organisms, these benefits are generally considered to be strain-specific. Bifidobacterium animalis ssp. lactis strains are widely used in dietary supplements and fermented and non-fermented dairy foods for their probiotic function. However, the high level of genetic homogeneity reported among B. animalis ssp. lactis used in commercial dairy products presents a challenge to differentiation using common phenotypic or DNA-based techniques. In an effort to differentiate *B. animalis* ssp. *lactis* isolates, 22 strains obtained from national culture collections (2) and commercial culture suppliers (20) were characterized using a variety of phenotypic and DNA-based methods (PFGE and RAPD-PCR). The only phenotypic difference observed was the inability of 10 strains to ferment glucose and only one strain (ATCC 27536) could be differentiated by PFGE after restriction with XbaI and SpeI. In an effort to locate genetic targets for strain-level differentiation, the genomes of *B. animalis* ssp. *lactis* DSMZ 10140 (the Type strain) and Bl-04 (a commercial strain) were sequenced and compared. Comparative analyses of the genomes revealed four insertion/deletion sites (for a total of 443 bp) and 47 single nucleotide polymorphisms (SNPs), confirming a high level of genetic relatedness. PCR primers designed to amplify the INDELs and regions containing each identified SNP were used to determine the allele in each of the 22 strains under study. To date, evaluation of two INDELs, one containing a portion of the CRISPR locus, and 15 SNP sites has identified 9 unique allelic types. To our knowledge, this represents the first SNP-based typing scheme for differentiating strains of B. animalis ssp. lactis and perhaps the first practical strain-level typing scheme for B. animalis ssp. lactis. This typing scheme will be useful to starter manufacturers, dairy products processors and clinical researchers who are interested in verifying strain identity for this subspecies.

Key Words: Bifidobacterium animalis ssp. lactis, SNP, probiotic

**525** CpG oligodeoxynucleotide from *Streptococcus thermophilus* regulates anti-inflammatory responses. T. Shimosato<sup>\*1</sup>, M. Tohno<sup>2</sup>, T. Sato<sup>3</sup>, and H. Kitazawa<sup>2</sup>, <sup>1</sup>Shinshu University, Kamiina, Nagano, Japan, <sup>2</sup>Tohoku University, Sendai, Miyagi, Japan, <sup>3</sup>Yokohama City University, Yokohama, Kanagawa, Japan.

Immunostimulatory sequence of oligodeoxynucleotides (ISS-ODNs) from lactic acid bacteria, such as CpG and AT ODNs, are recently found as potent stimulators of innate immunity (1-3). In this study, we identified a strong immunostimulatory CpG ODN, which we named MsST, from the lacZ gene of Streptococcus thermophilus ATCC19258, and we precisely evaluated its immune functions. In vitro studies revealed that MsST had a similar ability as the murine prototype CpG ODN 1555 to induce inflammatory cytokine production and lymphocyte mitogenicity. In mouse splenocytes, MsST increased the number of CD80<sup>+</sup> CD11c<sup>+</sup> and CD86<sup>+</sup>CD11c<sup>+</sup> dendritic cells and CD4<sup>+</sup>CD25<sup>+</sup> regulatory T cells. We also analyzed the effects of MsST on the expression of various cytokines by real-time quantitative PCR. MsST was more potent at inducing anti-inflammatory cytokines such as interleukin (IL)-10 and IL-33 expression after 48 h of stimulation, whereas IL-6 was down regulated. Together with recent findings, these results indicate that MsST may be a potential therapeutic ODN for inflammatory disease by secreting IL-10 and IL-33. In addition, Streptococcus thermophilus, which contains a strong ISS-ODN, may also be a good as a candidate starter culture for the development of new physiologically functional foods. 1. Shimosato T., et al., Anim Sci J. in press. 2. Shimosato T., et al., Cellular Microbiol.8(3):485-95.2006. 3. Shimosato T., et al., Biochem Biophys Res Commun. 326(4):782-7.2005.

Key Words: CpG ODN, S. thermophilus, IL-33

**526** Survival of probiotic adjunct cultures added to low-fat, reducedfat, and full fat cheddar cheese. C. J. Oberg<sup>\*1</sup>, L. Moyes<sup>1</sup>, C. Brothersen<sup>2</sup>, and D. J. McMahon<sup>2</sup>, <sup>1</sup>Microbiology Department, Weber State University, Ogden, UT, <sup>2</sup>Western Dairy Center, Utah State University, Logan.

A variety of delivery systems are used to deliver probiotic bacteria such as yogurt and milk. Cheese may have benefits over other dairy delivery systems including greater in vivo survival, longer shelf life, and being a more common component in the average diet. Cheese was made at three different fat levels (33, 16, 6%) using Lactococcus lactis ssp. lactis. Commercial strains of Lactobacillus acidophilus, Lactobacillus casei, Lactobacillus paracasei, and Bifidobacterium were added as adjuncts. Selective media, designed to promote growth of certain lactic acid bacteria (LAB) over others, were used to detect individual LAB types during cheese storage. Microbial flora of the cheese was analyzed at 1, 30, 60, 90, 120, 180, and 270 d. Between 90 and 120 d of storage, bacterial counts changed on selective media for Bifidobacterium indicating NSLABs may be appearing. Differences in cheese chemistry, particularly the salt-in-moisture levels related to cheese fat levels appears to play a role in probiotic adjunct counts since NSLABs appear on Lb. casei selective media (MRS + vancomycin) after only 30 d in low fat control cheeses. In low fat cheese, one Lb. acidophilus adjunct was observable to 60 d while Lb. casei and Lb. paracasei maintained high counts through the entire storage period. In some cheeses, one Lb. casei adjunct had the ability to grow on both Lb. casei and Bifidobacterium selective agar. Since no Bifidobacterium had been added to these cheeses Bifidobacterium counts could be discarded. In all cheeses with added Bifidobacterium, counts decreased after 90 d of storage. In full fat cheese, Lb. acidophilus counts remained high to 120 d. For many cheeses, nonstarter LABs seemed to appear on a number

of the selective media during aging. Probiotic adjunct cultures can be detected in cheddar cheese out to 90-120 d when growth of nonstarter LABs obfuscates results on selective media. Some adjunct cultures can be detected beyond this time, particularly *Lb. casei* and *Lb. paracasei*. Results indicate selected probiotic adjuncts can survive for at least 90 d in low-fat, reduced-fat, and full fat Cheddar cheese.

Key Words: probiotic, cheese, lactic acid bacteria

**527** Intrinsic resistance and stress responses to hydrogen peroxide in bifidobacteria. T. S. Oberg<sup>\*1</sup>, S. C. Ingham<sup>2</sup>, J. L. Steele<sup>2</sup>, and J. R. Broadbent<sup>2</sup>, <sup>1</sup>Utah State University, Logan, <sup>2</sup>University of Wisconsin, Madison.

The interest and use of bifidobacteria as a probiotic in function foods has increased dramatically in recent years. Due to the anaerobic nature of bifidobacteria, however, oxidative stress can pose a major challenge to the viability of bifidobacteria during storage in functional foods. To better understand oxidative stress resistance in two industrially important species of bifidobacteria, we examined the response of three strains of Bifidobacterium longum and three strains of Bifidobacterium animalis subsp. lactis to hydrogen peroxide. Each strain was exposed to a range of hydrogen peroxide concentrations (0.1mM to 10mM) to evaluate and compare intrinsic resistance to H<sub>2</sub>O<sub>2</sub>. Next, strains were tested for the presence of an inducible oxidative stress response by 20 or 60 min exposure to a sublethal level of H2O2 followed by challenge at a lethal H<sub>2</sub>O<sub>2</sub> concentration. Results showed B. longum subsp. infantis ATCC 15697 and B. lactis D2908 had the highest level of intrinsic H<sub>2</sub>O<sub>2</sub> resistance among tested strains of B. longum and B. lactis, respectively. Inducible H<sub>2</sub>O<sub>2</sub> resistance was only detected with two strains; B. longum NCC2705 showed a 17-fold increased survival in 5.25mM H<sub>2</sub>O<sub>2</sub> after a 60 min induction in 1.25mM H<sub>2</sub>O<sub>2</sub>, and survival of B. lactis D2908 increased 2.5-fold in 5.25mM H<sub>2</sub>O<sub>2</sub> after 20 min induction in 1.25mM H<sub>2</sub>O<sub>2</sub>. Other strains showed either no difference or increased sensitivity to H<sub>2</sub>O<sub>2</sub> after induction treatments. These data indicate that intrinsic and inducible resistance to hydrogen peroxide is strain specific in B. longum and B. lactis and suggest that for some strains, sublethal H2O2 treatments could help increase cell resistance to oxidative damage in production and storage of probiotic foods.

Key Words: bifidobacteria, probiotic, oxidative stress

**528** Cholesterol removing ability and bile tolerance of lactic acid bacteria isolated from fermented yak milk. Y. Jiao<sup>1</sup>, L. Zhang\*<sup>2</sup>, and H. Yi<sup>2</sup>, <sup>1</sup>*Heilongjiang University of Chinese Medicine, Harbin, China*, <sup>2</sup>*College of Food science and engineering, Harbin Institute of Technology, Harbin, China*.

Cholesterol assimilating ability and bile tolerance of 23 strains of lactic acid bacteria isolated from fermented yak milk of Gansu province were

examined. All strains have varying capabilities to remove cholesterol in vitro. The ability of cholesterol assimilation of most of the strains in the media with oxgall was better than without. The heat killed strains assimilated less cholesterol than the normal ones. All types of bile salts could inhibit the growth of strains, the most powerful of which was sodium glycocholate, bile acid took the second place, oxgall was the weakest. There was no relationship between bile salts tolerance and cholesterol assimilation. Finally, it was found that the strains of H1, I10,and W2 acted better, indicating that these strains may be promising candidates for use as a dietary adjunct to lower serum cholesterol in vivo.

Key Words: fermented yak milk, lactobacillus, cholesterol removal

**529** Factors affecting the total bacteria count of raw milk preserved with azidiol (liquid or tablet) and bronopol. M. O. Leite<sup>\*1,2</sup>, N. J. Andrade<sup>3</sup>, M. M. O. P. Cerqueira<sup>1,2</sup>, L. M. Fonseca<sup>1,2</sup>, and R. Rodrigues<sup>1,2</sup>, <sup>1</sup>Federal University of Minas Gerais (UFMG), School of Veterinary Medicine, Department of Food Technology and Inspection, Belo Horizonte, MG, Brazil, <sup>2</sup>Laboratory of Milk Quality Analysis, UFMG, Belo Horizonte, MG, Brazil, <sup>3</sup>Federal University of Viçosa, Viçosa, MG, Brazil.

The objective of the present work was to evaluate the influence of several parameters on the milk quality results of electronic analyses for total bacteria count (TBC), somatic cell count (SCC) and composition. The parameters were: storage time and temperature, azidiol (liquid and tablet), and bronopol. Six samples were collected, subdivided, added preservative and stored at room temperature for up to eight days, and incubated under three different temperatures (4, 7, and 10°C) for until ten days. An electronic equipment, Bentley CombiSystem 2300<sup>®</sup> was used for the composition and SCC analyses. The total bacterial counting (TBC) was analyzed using an electronic equipment Bactocount IBC (Bentley®) and standard plate counting for mesophilic aerobic microrganisms. The design was split-plot, and the results were evaluated by Analysis of Variance with analysis of minimal significant difference by Duncan Test. The results showed that samples kept at 30°C can be analyzed, for composition and SCC until the fourth and fifth day, respectively, after collection. Cooled samples can be analyzed for composition, SCC and TBC until 10 days after collection. However the samples can not be stored at room temperature for TBC. There was a significant statistical difference on the levels of lactose and SCC for samples preserved with azidiol instead of bronopol. Therefore azidiol is not suitable for sample preservation in the electronic analysis for composition and SCC. Sample preserved with bronopol is not indicated for TBC because it underestimates the bacterial population. The results indicated that the tablet of azidiol can be used to replace the liquid presentation of this preservative in raw milk samples. Acknowledgements: FUNDEP/UFMG; FAPEMIG; CNPq; CAPES.

Key Words: azidiol tablet, milk quality, bronopol

## **Extension Education**

**530** A diagnostic tool to assess calf welfare and management onfarm. E. Vasseur\*<sup>1</sup>, J. Rushen<sup>2</sup>, A. M. de Passillé<sup>2</sup>, D. Lefebvre<sup>3</sup>, G. Fecteau<sup>4</sup>, and D. Pellerin<sup>1</sup>, <sup>1</sup>Université Lavalé, Quebec city, Quebec, Canada, <sup>2</sup>Pacific Agri-Food Research Centre, Agriculture and Agri-Food Canada, Agassiz, British Columbia, Canada, <sup>3</sup>Valacta, Dairy Production Centre of Expertise Quebec-Atlantic, Sainte-Anne-deBellevue, Quebec, Canada, <sup>4</sup>Veterinary Faculty, Université de Montréal, Sainte-Hyacinte, Quebec, Canada.

Unweaned calf morbidity remains high, which is a costly animal welfare concern. A previous survey, of 115 Quebec dairy farmers found mean perinatal calf mortality of 8.8%, which was underestimated by 20 to 50% by producers with 94% believing calf morbidity was not a problem.