

( $P < 0.05$ ) by LAB2 but not by LAB1, LY1 and LY2. The expressions of MCP-1, MCP-2, MUC-1 and IL-13 were unaffected by LAB1 and LAB2 at all time points, whereas they were ( $P < 0.05$ ) restricted by LY1 and LY2 as incubation time increased. These results indicate that mode of action on expression of cytokines vary with probiotic strains, and the immune status of BKE cells can be modulated by either enhancing or restricting certain cytokines' expression *in vitro*.

**Key Words:** probiotic, lactic acid bacteria, live yeast, immune modulation, Bovine kidney epithelial cells, *in vitro*

**PSVII-25** **Grazing-induced transcriptomic changes in bovine biceps femoris muscle, subcutaneous fat, and liver mRNAs and plasma exosome microRNAs.** S. Muroya<sup>1</sup>, H. Ogasawara<sup>2</sup>, K. Nohara<sup>2</sup>, M. Oe<sup>1</sup>, K. Ojima<sup>1</sup>, M. Hojito<sup>2</sup>, <sup>1</sup>*Institute of Livestock and Grassland Science, NARO, Tsukuba, Ibaraki, Japan*, <sup>2</sup>*Kitasato University, Hokkaido, Japan*

Grazing alters skeletal muscle physiological properties in cattle. We hypothesized that changes in circulating microRNA (c-miRNA) levels are attributed to release and/or uptake of exosomal miRNA at adipose and liver tissues. In this study, to determine grazing-induced mRNA and miRNA transcriptomic changes at those tissues, we conducted microarray analyses in biceps femoris muscle (BF), subcutaneous fat (ScF), and liver tissues of Japanese Shorthorn steers. Three steers (aged 20–22 mo) were allocated to each of grazing and housed groups; grazing steers were fed on pasture for 3 months from middle of May, when the other steers were housed and fed in a free-stall barn with grass harvested every morning. The samples were collected at 3 mo of grazing period, with surgical biopsy in case of solid tissues. Plasma exosomes (pEX) were prepared from the blood samples, using ultracentrifugation. Total RNAs were prepared from the tissues and pooled within each tissue, and finally applied to both mRNA and miRNA expression microarray analyses. The number of changed genes was the highest in BF (52 increased and 1088 decreased genes). On the other hand, the number of genes showing more than 2-fold increase (INC) and decrease (DEC) compared to the housed was the highest in ScF and the lowest in liver. Especially in ScF, we observed 169 INC and 249 DEC genes, indicating dominance of DEC genes over INC genes in ScF of the grazing steers. Using the list of INC and DEC genes, we conducted gene ontology analyses and extracted the term 'extracellular exosome' from the

DEC genes in ScF, as a term associated with changes in ScF of the grazing steers. This was coincident with greater number of the decreased miRNAs than the increased in pEX. These results suggest an association of grazing-induced changes in c-miRNAs with ScF transcriptome in the steers.

**Key Words:** Grazing cattle, Subcutaneous fat, microRNA

**PSVII-30** **Intracellular toll-like receptors correlates positively with interferon-stimulated genes during the peri-implantational period in beef cattle.** A. Sanz<sup>1</sup>, B. Serrano-Pérez<sup>2</sup>, E. Molina<sup>2</sup>, A. Noya<sup>1</sup>, I. López-Helguera<sup>2</sup>, I. Casasús<sup>1</sup>, D. Villalba<sup>2</sup>, <sup>1</sup>*CITA de Aragón-IA2, Zaragoza, Spain*, <sup>2</sup>*Universitat de Lleida (UdL), Lleida, Spain*

The objective of this study was to assess the possible interrelations between expression patterns of intracellular toll-like receptors (TLR) and interferon-stimulated genes (ISG) during the peri-implantational period in peripheral blood mononuclear cells of two beef cattle breeds. Thirty Parada de Montaña and 19 Pirenaica multiparous cows were synchronized to estrus and artificially inseminated (AI). Pregnancy diagnosis was performed by ultrasonography on day 37 and confirmed 91 days post-AI. Blood samples were drawn on days 18 and 21 after AI. Gene expression of TLR7, TLR8, ISG15, OAS1 and MX2 was analysed by real time PCR. Pearson correlation coefficient ( $r$ ) test was used to identify possible relationships between gene expression levels of TLR and ISG with JMP statistical software (SAS Institute Inc., Cary, NC). On day 37, pregnancy was confirmed in 35 dams. In pregnant dams, TLR7 expression was positively correlated with ISG15 expression on day 18 ( $r = 0.34$ ;  $P < 0.05$ ), and TLR8 expression was positively correlated with ISG15 ( $r = 0.65$  and  $r = 0.70$ ;  $P < 0.05$ ) and MX2 expression ( $r = 0.42$  and  $r = 0.69$ ;  $P < 0.05$ ) on days 18 and 21, respectively. In non-pregnant dams, positive association was also observed between TLR8 and ISG15 on day 18 ( $r = 0.55$ ;  $P < 0.05$ ). These results confirm that embryo signalling during the recognition period may lead to activation of a wide range of immune signals in local and peripheral tissues. Positive association observed between TLR8 and ISG15 in non-pregnant dams could be consequence of embryo signalling in dams that suffered embryo loss afterwards. Our results indicate that intracellular TLR7 and TLR8 are positively associated to ISG15 and MX2 during pregnancy establishment in beef cattle.

**Key Words:** Interferon tau, pregnancy establishment