P4086 Identification of uterine microRNAs in pigs

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MicroRNAs (miRNAs) are a class of single-stranded small (18-25 nt) non coding RNAs that have regulatory roles on gene expression at the post-trascriptional level controlling a wide range of biological processes. Several studies have shown the involvement of miRNAs during embryogenesis but little is known about the porcine miRNAs expressed in uterus during this period. An F_2 intercross was created from 3 Iberian (Ib) boars and 18 Meishan (Me) sows in order to study prolificacy traits in pigs. Fourteen F_2 IbxMe sows, classified into two groups regarding the number of embryos (NE) at 30-32 days of gestation as high (NE≥14; n=5) or low (NE≤11; n=9) prolificacy, were used to elaborate small RNA libraries from uterus. High-throughput sequencing with the 454 Genome Sequencer FLX Titanium (Roche®) was used to determine the level of miRNAs expression in sow uterus as well as to determine if differentially expressed miRNAs could be associated with prolificacy performance. Overall, 249 miRNAs were found when our deep-sequence dataset was aligned with miRBase v.15.0. The most abundant miRNAs in pig uterus at 30-32d of gestation were miR-125b, miR-200b, miR-200c, miR-23a and miR-23b. Several miRNAs showed differences in frequency between high and low prolificacy sows suggesting that miRNAs could be involved in embryo survivability during gestation in pigs. Quantitative PCR analyses are now being performed in order to validate in a more accurate way the results obtained with the deep sequencing approach.

P4087 Sequencing and Polymorphisms in the Porcine Glucocorticoid Receptor

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Both animal and human studies have shown that during early childhood and old age, the brain is extremely sensitive to stress. High levels of stress lead to release of glucocorticoids, which bind to the glucocorticoid receptor forming a complex that acts as a transcription factor. This regulates gene expression for development, metabolism, and the immune response. Data from the rat suggests that the GR gene is epigenetically altered in the offspring of stressed mothers. The importance of this observation to other mammalian species needs further investigation. The pig is the model organism chosen for this study as its size and physiology as well as the size and structure of its chromosomes are highly similar to that of humans. The glucocorticoid receptor gene sequence in pigs has recently been completed, however many of its structural variants (SNPs and CNVs) remain unknown. We have identified and sequenced the coding and promoter regions of the porcine GR gene via a comparison of the porcine genome to the published human sequences. The sequences and polymorphisms of the 9 porcine exons are currently being compared between different individuals. Preliminary results indicate that Duroc pigs have more polymorphisms. The sequence information produced in this study will hopefully allow those regions of the promoter already known, to undergo epigenetic modification in other species to be interrogated in pig.

P4088 Transcriptome alterations due to physiological normoxic (2% O_2) culture of embryonic stem cells

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A study carried out in ARK-Genomics looked to identify consensus transcriptional changes across three well characterised embryonic stem cell lines (ESCs) in response to being cultured in 2% oxygen (physiological normoxia) compared to 21% oxygen has been reanalysed. Previous studies of culturing ESCs at 2% have reported reductions in spontaneous differentiation, reduced spontaneous chromosomal aberrations, enhanced clonality and smaller, less complex cells. We took this data, previously analysed externally, and reanalysed using our standard laboratory software. ARK-Genomics, as part of its support for gene expression, has access to a wide variety of tools, the most commonly used within the laboratory being Partek Genomics Suite and Ingenuity Pathway Analysis (IPA). While mostly in agreement with the previous analysis, the use of the GO ANOVA tool in Partek followed by subsequent analysis in IPA generated previously unidentified changes in gene expression.

P4089 An INDEL polymorphism in DGAT1 3'UTR correlates with milk production parameters in Brazilian Guzerat dairy breed

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One of the strongest QTLs influencing milk production yet identified, DGAT1 gene provides a good example of the kind of work required to transfer a QTL from taurine to indicine breeds. In *Bos taurus* breeds, the SNP DGAT1 K232A, explains 2-50% of the variance in such parameters. DGAT1 232A allele has been correlated with lower saturated fat. We found low frequencies of DGAT1 232A in Brazilian Zebu breeds (0-2%). However, an allele being absent or rare, and therefore barely detectable in association studies, does not mean that the gene itself is not a QTL. Searching for breed specific polymorphisms, we sequenced DGAT1 3' UTR in 8 Guzerat individuals. We identified a new INDEL polymorphism. We screened the INDEL in a sample of 97 Guzerat cows evaluated for: total milk production (kg), lactation length (days), content of protein, fat, lactose and solids (kg), expected progeny difference (DEP), and breeding values (BV). Significant associations ($p = 0.05$) of INDEL polymorphism were found for BVs in 305 days, total fat and protein. Comparing to +/+, genotype +/- had a loss of 297 kg in production and genotype -/- a reduction of 113 kg (comparing to +/-). The heterozygote genotype is associated with a 9 kg reduction on BV for total fat, while genotype -/- causes an additional loss of 11.5 kg. A similar trend was observed for proteins, with losses of 12.4 kg (genotype +/-) and 16 kg (genotype -/-). These results confer additional support for DGAT1 as a QTL.

Support: FAPEMIG, CNPq, PRONEX/FAPEMIG.

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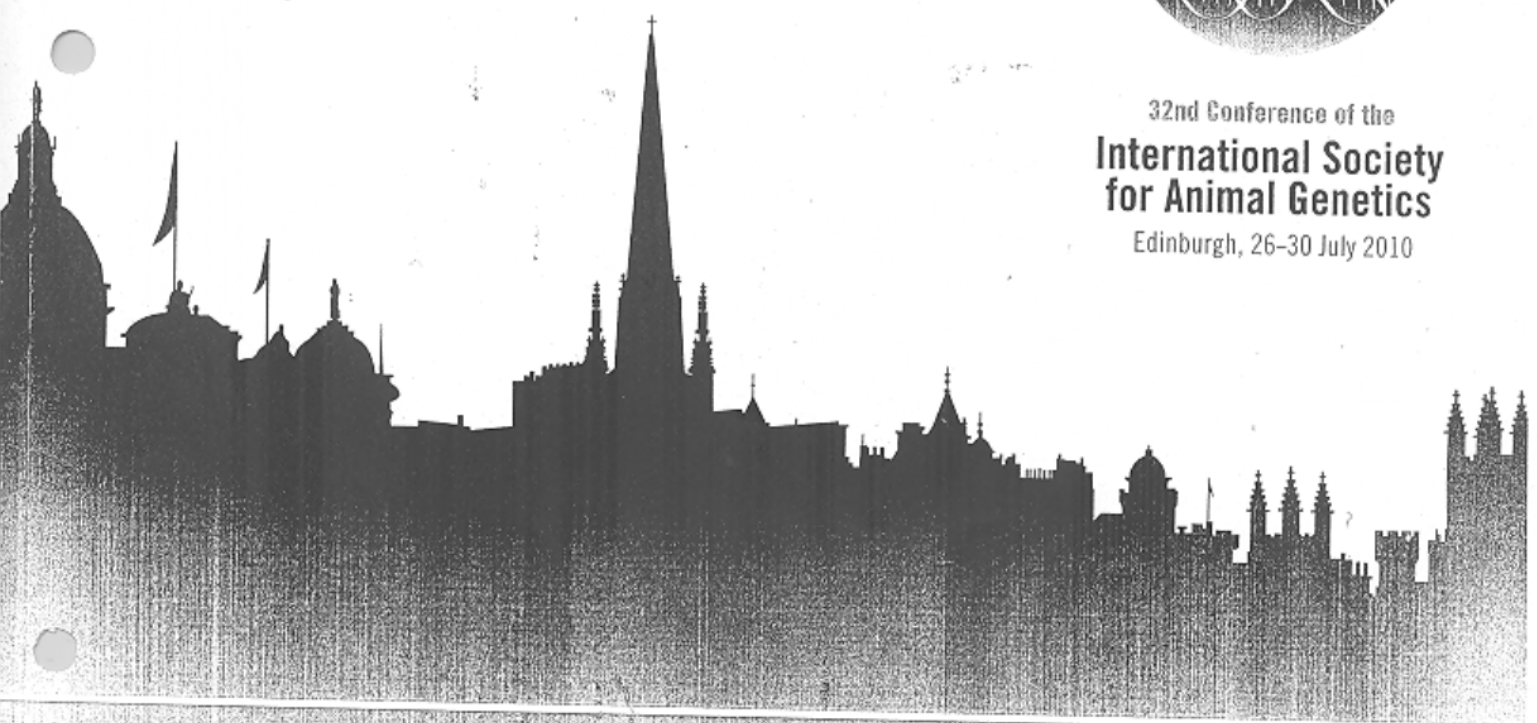

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