Public Abstract
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Graduation Term:SP 2010
Department:Animal Sciences
Degree:MS
Title:Transcriptional Profiling by Deep Sequencing Identifies Differences in mRNA Transcript Abundance in In Vivo Derived Vs. In Vitro Cultured Porcine Blastocyst Stage Embryos

In vitro embryo culture systems promote development at rates lower than in vivo. The goal of this project was to discover transcripts that may be responsible for a decrease of embryo competency in blastocyst stage embryos cultured in vitro. Gilts were artificially inseminated on the first day of estrus and on Day 2 one oviduct and the tip of a uterine horn were flushed and the recovered embryos were cultured in PZM3 for four days. On Day 6 the gilts were euthanized and the contra-lateral horn was flushed to obtain in vivo derived embryos. Total RNA was extracted from 3 pools of 10 blastocysts from each treatment. First and second strand cDNA was synthesized and then sequenced using Illumina sequencing. The reads generated were aligned to a custom-built database designed to represent the known porcine â€œtranscriptomeâ€• A total of 1,170 database members were different between the two groups (P<0.05), and 588 of those were at least 2-fold different. Eleven transcripts were subjected to real-time PCR and each validated the sequencing. There was an overall decrease in both inner cell mass and trophectodermal cell numbers in embryos cultured in vitro, however, no difference in ICM:TE ratio was found. Interestingly, the transcript (SLC7A1) was higher in the in vitro cultured group. This difference disappeared after addition of arginine (0.36 mM) to the 4 day culture. In conclusion, Illumina sequencing and alignment to a custom â€œtranscriptomeâ€ successfully identified a large number of genes that yield clues to the derivation of culture media.