

IDENTIFICATION AND QUANTIFICATION OF DIFFERENTIALLY REPRESENTED TRANSCRIPTS IN PREIMPLANTATION BOVINE EMBRYOS

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Abstract

Identification of transcripts present at key development stages in preimplantation embryos is critical for a better understanding of early embryogenesis. To that end, we characterized the relative abundance of multiple transcripts during several developmental stages, including metaphase II-stage oocytes, and 2-cell-stage, precompact morula, and *in vitro*-produced blastocyst-stage embryos, along with differences in the relative abundance of transcripts present in *in vivo*-, *in vitro*-, and nuclear transfer-produced blastocysts. Our thesis was that the identification of differentially represented transcripts from these stages would reveal not only developmentally important genes, but also genes that might be aberrantly expressed due to embryo production techniques. Individual clusters from a large bovine expressed sequence tag (EST) project were compared using Fisher's Exact Test. Of the 3,144 transcripts that were present during embryogenesis, 125 were found to be differentially represented ($P < 0.01$) in at least one pairwise comparison. Fifteen of these transcripts were selected for further examination using quantitative real-time PCR, which confirmed that nine of the 15 transcripts were significantly differentially represented in at least one pairwise comparison, while three more of the transcripts exhibited a strong trend ($P < 0.05$) of different abundance levels in at least one pairwise comparison. Further investigation of these results may not only help us to better understand the developmental implications of embryo manipulation, but may also lead to the improvement of current assisted reproductive technologies.