

# THE ROLE OF CYTOPLASMIC POLYADENYLATION ELEMENT SEQUENCE ON MRNA ABUNDANCE IN PROCINE EMBRYOGENESIS

Kyle Dobbs

Dr. Randall Prather, Thesis Supervisor

## ABSTRACT

Development of a porcine germinal vesicle oocyte (GVO) to a 4-cell stage embryo occurs during a transcriptionally silent period when the oocyte/embryo relies on maternally derived mRNA to encode proteins required for development. Regulation of translation and degradation of maternal mRNA is thought to be partially dependent upon cytoplasmic polyadenylation elements (CPEs) within the 3' untranslated region of the mRNA. The goal of this study was to determine how CPE sites affect the abundance of mRNA during embryogenesis and parthenogenetic development, and how cordycepin, a 3'-deoxyadenosine (3'-dA) which inhibits poly (A) tail formation, affects polyadenylation and transcript abundance. Expressed sequence tags (ESTs) from oocytes and 4-cell stage embryos were scanned for the presence of five consensus CPEs. Nineteen different transcripts containing one to three putative CPEs were selected and transcript abundance was determined in GVO, metaphase II, 2-cell and 4-cell stage embryos via real-time PCR; and the length of the poly (A) tail was determined by using a Poly (A) tail PCR (PAT) assay. Real time PCR was performed on three biological and two technical replicates for each stage. There was no direct correlation between poly (A) tail length, transcript abundance and the CPE. Cordycepin prevented polyadenylation of transcripts that normally undergo noticeable polyadenylation. Thus CPEs may not be the only factors that regulate message stability, and parthenogenetic activation does not result in changes in transcript abundance that mimic in vitro fertilization.