Identification of a novel *Skp1* isoform that encodes a novel SKP1 protein in mouse oocytes

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GV oocytes are transcriptionally quiescent and rely on stored mRNA that is recruited for translation. Translation of stored mRNA is essential for maintaining oocyte quality, and sustaining early embryonic development. The complexity of the oocyte proteome is driven by the diverse pool of stored mRNA that have undergone alternative splicing. However, the impact of alternative splicing on proteome diversity within the oocyte remains poorly understood. To better understand this relationship we utilized direct RNA-sequencing of cumulus oocyte complexes (COCs) to directly measure transcriptome complexity at a single, full-length molecule resolution, coupled with mass spectrometry. Direct RNA-sequencing was performed using total RNA from 3,751 COCs, generating an average of 467,843 (SEM ± 33,510.67) full-length reads across three separate sequencing runs, with mean molecule length of 1.08 Kb (SEM ± 0.012). We identified 45,086 unique transcripts mapping to the mouse genome, of which 85% were single transcript genes and 15% were multi-transcript genes. Within multi-transcript genes, we identified 15,571 alternative splicing events, with alternative exon usage the most predominant (46% of all events). Next we assessed the protein coding potential for the 45,086 unique transcripts. Using the Ensemble database, 82.6% of the transcripts were unannotated, while 13.0% were listed as protein coding. We used *in silico* translation to estimate protein coding potential and observed that 40.1% of transcripts identified to be protein coding. Next we performed mass spectroscopy using a group of 40 GV oocytes. Identified peptides were searched against a Mouse Swiss Prot database and a custom protein database generated from *in silico* translation of the direct RNA-sequencing data. We identified a novel *Skp1* mRNA isoform that encodes a novel protein. Analysis of the novel protein revealed a loss of the key N-terminal region essential for interaction with the ubiquitin ligase Cullin-1. Skp1 is a key component of the SCF complex and is essential for maintaining synapses during meiosis. Our studies reveal that alternative splicing within GV oocytes can result in the production of novel proteins that could impact normal oocyte function. Future studies will assess the role of these novel proteins in oocyte physiology.