ABSTRACT

GENETIC AND MOLECULAR CONTROL OF OOCYTE FUNCTION

By

Ashley L. Severance

A high-quality oocyte will successfully accomplish three critical tasks: segregate chromosomes, reprogram its own genome and that of the incoming sperm to an embryonic state, and support the metabolism and early development of the embryo. This dissertation focuses on two critical determinants of oocyte quality, maintenance of the meiotic spindle and mRNA regulation during meiotic maturation. Chapter 3 of this dissertation focuses on the role of the translational repressor, EIF4EBP1, at the meiotic spindles. I show that blocking EIF4EBP1 phosphorylation, which is normally permissive of translation, disrupts the first meiotic spindle, and that inhibiting Polo-like kinase 1 (PLK1) affects EIF4EBP1 phosphorylation and spindle formation at both meiotic divisions. This exciting result suggests PLK1 as an important regulatory focus for controlling EIF4EBP1 and mRNA translation locally at the spindle, to enable

the supply of essential proteins during meiotic maturation. Chapter 4 of this dissertation builds on the theme of understanding maternal mRNA regulation by characterizing the dynamic changes in the pool of mRNAs during oocyte maturation and addresses how this varies with oocyte quality. By comparing maturation changes in the transcriptome between two inbred strains (C57BL/6J and DBA/2J) and F1 hybrids between the two strains (BDF1), I discover differences in maternal mRNA regulation associated with superior BDF1 oocyte characteristics (i.e., oocyte hybrid vigor). This includes differences in regulating mRNAs related to mitochondrial physiology and histone production. I also show that many of the differences

between the three genotypes arise during maturation and thus in the absence of transcription, indicating differences in regulating mRNA degradation. Overall, a combination of processes underlies the differential regulation of maternal mRNA in BDF1 oocytes compared to parental

strains. A small amount of transgressive gene expression is seen, but the most prominent mechanisms responsible for differential mRNA regulation in BDF1 oocytes include “blending” (F1 expression levels intermediate between parental expression levels) and additive dominance. Interestingly, up to 25% of the mRNAs differ significantly between genotypes at the MII stage. This includes many proposed markers of oocyte quality, for which mRNAs differential

expression between strains exceeded what was reported for differences associated with oocyte quality. Because all three genotypes are fertile, this suggests that single markers may not be reliable indicators of oocyte quality. Overall, these results clearly show that the oocyte utilizes complex regulatory mechanisms at both the meiotic spindles and throughout the ooplasm. This dissertation opens the door for many potential future directions to probe further into understanding these oocyte-specific adaptations. In the future, this research along with other studies could be useful to develop therapies to improve fertility outcomes in both agricultural species and in humans.

For the hero, Hercules.

()() (‘.’) ( )