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A257E Embryology, developmental biology, and physiology of reproduction

Effect of non-esterified fatty acids during *in vitro* oocyte maturation on the development of bovine embryos after transfer

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Metabolic disorders, as in negative energy balance (NEB) dairy cows, are associated with elevated non-esterified fatty acid (NEFA) concentrations, predominantly palmitic acid (PA), in the follicular fluid. These NEFAs are known to jeopardize oocyte *in vitro* maturation and elicit altered blastocyst quality and physiology. Lipotoxic conditions during final oocyte maturation also influence epigenetic reprogramming in the resultant day (D) 7 embryo and may thus affect subsequent development, potentially imprinting lasting marks during later stages of life. Therefore, we hypothesized that exposure of oocytes to high NEFA concentrations during IVM affects post-hatching development of D7 blastocysts after embryo transfer. Bovine oocytes were matured for 24h under 2 conditions: 1) physiological NEFA conditions (28µM stearic acid (SA), 21µM oleic acid (OA), 23µM PA (BAS) and 2) elevated PA concentration as present in follicular fluid during NEB (150µM) with physiological concentrations of SA (28µM) and OA (21µM) (HPA). Matured oocytes were routinely fertilized and cultured in SOF with serum until D7. Cleavage (D2) and blastocyst rate (D7) were compared among treatments using a binary logistic regression model. Eight blastocysts (normal and expanded, equally distributed per treatment and per replicate) were transferred per cow (n=8, 5 replicates). Four cows were attributed to HPA or BAS per replicate and were crossed over for the next replicate. Embryos were recovered at D14 and morphologically assessed (n=46). Glucose, lactate and pyruvate turnover and interferon-tau (IFNT) secretion were measured in extra-embryonic tissue (EXT) after 24h culture (n=62). Morphological, metabolic and IFNT data were tested for normality with a Kolmogorov-Smirnov test and differences between treatment were analysed with a T-test. Data are presented as mean ± SEM. Developmental competence at D7 was not significantly different between treatments (blastocyst rate of 26 vs. 29.6% for HPA and BAS, resp.). Recovery rate at D14 was 30% and 36% for HPA and BAS, resp. ($P>0.05$). HPA during IVM significantly reduced embryo elongation (3.7 ± 1.5 vs. 8.6 ± 1.7 mm, $P=0.001$) but did not affect diameter of embryonic disc compared to BAS. EXT from HPA group consumed similar amount of glucose but tended to produce less lactate compared to EXT from BAS group (1732 ± 211 vs. 2428 ± 355 pmol/mm²/h, $P=0.073$). IFNT secretion was significantly lower in HPA group (0.47 ± 0.71 pg/ml) compared to BAS group (3.79 ± 1.16 pg/ml, $P=0.018$). In conclusion, exposure to elevated PA during *in vitro* oocyte maturation affected post-hatching development at D14. Embryos were less elongated, were metabolically altered and produced less IFNT, a major signal of pregnancy recognition, than their physiological counterparts. This suggests that metabolic stress during oocyte maturation may have long-lasting effects on embryo development that may lead to higher pregnancy loss and reduced fertility in high yielding dairy cows. More research is ongoing to investigate underlying mechanisms through genome wide transcriptome pathway mapping.