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OMEGA-3 FATTY ACIDS ENHANCE DEVELOPMENTAL COMPETENCE OF BOVINE OOCYTES UNDER METABOLIC STRESS CONDITIONS *IN VITRO*

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Metabolic stress conditions such as negative energy balance in dairy cows are associated with fat mobilization and elevated saturated (stearic; SA, palmetic; PA) and monounsaturated (oleic; OA) fatty acids (FAs) in serum and follicular fluid. We have shown that these FAs have direct detrimental effects on oocyte quality (Van Hoeck et al., ARS, 149:19-29, 2014). In contrast, we demonstrated that polyunsaturated α-linolenic acid (*n-3* 18:3; ALA) can enhance oocyte competence (Marei et al., BOR, 81:1064-1072, 2009). Here, we examined the effects of ALA supplementation (at physiological follicular fluid concentration; 50 µM) during *in vitro* oocyte maturation on subsequent embryo development in the presence of high follicular fluid concentrations of SA, PA and OA (HNEFA, 425µM). Cumulus cell expansion was scored at the end of oocyte maturation (0-3: 0; not expanded, 3; fully expanded). The proportions of cleaved and fragmented embryos were recorded on day 2 post-fertilization. Blastocyst rates were recorded on day 7 and 8. Day 8 blastocysts were categorized as Normal (not expanded), Expanded, or Hatched, and were fixed and immunostained with anti-cleaved-caspase-3 antibody and Hoechst. Total cell counts and apoptotic cell indices were calculated. Data were obtained from 5 independent repeats using 1529 oocytes derived from slaughter house material. A total of 179 blastocysts were stained. Categorical data were analyzed by binary logistic regression using SPSS, and numerical data were analyzed using ANOVA. Pairwise comparisons were performed using Bonferroni correction. *P* values <0.05 were considered significant. Compared with FA-free solvent controls, supplementation with HNEFA resulted in: inhibition of cumulus cell expansion (score: 1.7±0.2 vs. 2.8±0.04, *P*<0.05); higher fragmentation rates (16.8% vs. 9.5%, *P*<0.05); and lower blastocyst rates on day 7 (*P*<0.05), either expressed as a proportion from the total number of fertilized oocytes (15.6% vs. 22.8%) or from the total number of cleaved embryos (20.4% vs. 30.6%). Hatched and expanded blastocysts produced from HNEFA-exposed oocytes had higher apoptotic cell indices. In contrast, these negative effects were alleviated by ALA supplementation. In the HNEFA+ALA group, cumulus expansion score (2.4±0.16), fragmentation (6.9%), blastocyst rate on day 7 (21.4% from total fertilized oocytes and 28.7% from cleaved embryos), and apoptotic cell index were similar to the controls. In addition, HNEFA+ALA group had significantly higher total cell numbers in expanded and normal blastocysts compared with those from HNEFA group. In conclusion, ALA supplementation enhanced oocyte developmental capacity during maturation under metabolic stress conditions. The underlying mechanisms of action are currently under investigation. These results may have clinical implications to improve fertility through dietary interventions in animals and humans suffering from metabolic disorders associated with lipolysis.