



Proceedings of the 29th Annual Meeting of the Brazilian Embryo Technology Society (SBTE); Gramado, RS, Brazil, August 20th to 23rd, 2015, and 31st Meeting of the European Embryo Transfer Association (AETE); Ghent, Belgium, September 11th and 12th, 2015. Abstracts (Abstracts with numbers followed by the letter E are from the AETE).

A255E Embryology, Developmental Biology and Physiology of Reproduction

The effects of hypo- and hyperglycemia during lipolysis-like conditions on bovine oocyte maturation, subsequent embryo developmental and glucose metabolism

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Keywords: glucose, metabolic disorders, NEFA, oocyte.

Elevated follicular NEFA concentrations, commonly present in cattle in NEB or women suffering obesity or type 2 diabetes, are known to disrupt oocyte and embryo development and alter subsequent embryo metabolism. However, NEB cows exhibit systemic hypoglycemia whereas humans suffering metabolic disorders have hyperglycemic insults. Both metabolic features may affect oocyte development. Little is known about whether elevated NEFA concentrations in combination with hyper- or hypoglycemic conditions influences oocyte viability. In this study, we hypothesized that glucose interacts with high NEFA levels during *in vitro* oocyte maturation to affect developmental capacity and metabolism of the resulting blastocysts. Thus, 647 bovine grade I COCs were matured (3 repeats) under 4 conditions: 1) physiological NEFA (72 μ M; palmitic, stearic and oleic acid) and routine IVM glucose (GLUC) concentrations (5.50mM) (CNTRL), 2) pathophysiological NEFA (420 μ M) and routine GLUC (HI NEFA), 3) HI NEFA and high GLUC (10mM) (HI NEFA+HI GLUC) and 4) HI NEFA and low GLUC (2.75mM) (HI NEFA+LO GLUC). Subsequently, matured oocytes were routinely fertilized and cultured for 7 days. At day (D) 7 post insemination (pi) all blastocysts were individually cultured for 24 hours in 4 μ l drops of modified SOF medium under oil after which droplets were analyzed on GLUC concentrations as described by Guerif *et al.* (PLOSone, 8, e67834, 2013). Cleavage (48h pi), blastocyst rates (D8 pi) and the rates of D8 blastocysts from cleaved zygotes were recorded. Developmental competence and GLUC consumption data were compared between 4 treatments using a binary logistic regression model and mixed model ANOVA, respectively. Replicate, treatment and the interaction of both factors were taken into account (IBM SPSS Statistics 20). Significant lower cleavage rates were observed for HI NEFA+LO GLUC (56%) compared with CNTRL (73%; $P=0.006$) and HI NEFA+HI GLUC conditions (70%; $P=0.048$). At D8 pi, blastocyst rates of HI NEFA+LO GLUC exposed oocytes (18%) were significantly lower compared with CNTRL (38%, $P<0.001$), whereas development of HI NEFA+HI GLUC D8 blastocysts (25%) tended to be reduced compared with CNTRL ($P=0.066$). The capacity of cleaved zygotes to develop to blastocyst stage by D8 showed a similar profile: HI NEFA+LO GLUC (32%) significantly reduced and HI NEFA+HI GLUC (35%) tended to reduce development compared with CNTRL (53%; $P=0.024$ and $P=0.066$, respectively). Interestingly, with no significant difference in developmental stage at D7, these HI NEFA+LO GLUC blastocysts consumed significantly less GLUC from D7 to D8 (12.14 \pm 4.10 pmol/embryo/h) compared with CNTRL (25.53 \pm 2.96 pmol/embryo/h; $P=0.020$). In conclusion, low GLUC concentrations seem to be more deleterious than high GLUC concentrations in the presence of elevated NEFAs in terms of embryo development and the lower ability of the surviving D7 embryo to consume GLUC as an energy source for its further development.