Glucose and Lactate Concentrations Affect the Metabolism of

In Vitro Matured Porcine Oocytes

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It is important to meet the specific metabolic needs of the oocyte for complete cytoplasmic maturation. We hypothesized that maturation media containing substrate concentrations similar to follicular fluid would result in mature oocytes with metabolic activity similar to that of in vivo matured oocytes, and improved blastocyst development.

Cumulus oocyte complexes and follicular fluid were collected from abattoir ovaries. Follicular fluid was analyzed by ultramicrofluorimetry for concentrations (mM) of glucose, lactate, and pyruvate. Oocytes were matured in PPM mat.2 in a 3 x 2 factorial design with either 2, 5 or 10 mM glucose (G) and 6 or 8 mM L-lactate (L). Forty two hours later, oocytes were denuded with hyaluronidase and either assessed for glycolytic and pentose phosphate pathway activity, or fertilized in PPM fert.3 and cultured in PPM 1.2/2.2. Development was assessed at 144 hpi. All data were analyzed using GLM ANOVA, and statistical differences were analyzed using the Bonferroni Multiple Comparison Test.

Follicular fluid concentrations of glucose were higher (P < 0.05) in large (7-9 mm) follicles $(1.81 \pm 0.17 \text{ mM})$ than in small (3-5 mm) follicles $(1.03 \pm 0.14 \text{ mM})$. Lactate and pyruvate concentrations were not different between large and small follicles $(6.12 \pm 0.19 \text{ mM} \text{ and } 5.76 \pm 0.09 \text{ mM}; 0.01 \pm 0.01 \text{ mM}$ and $0.02 \pm 0.01 \text{ mM}$, respectively). Glycolytic activity was greater (P < 0.05) in oocytes matured in 2G6L than 2G8L, 5G8L, 10G6L and 10G8L. Pentose phosphate pathway (PPP) activity was higher (P < 0.05) in oocytes matured in 2G6L compared to 10G6L and 10G8L. Interestingly, higher concentrations of glucose and lactate decreased glucose metabolism. No differences in cleavage or blastocyst development were observed between treatment groups. Low blastocyst development may have resulted from remaining deficiencies during in vitro maturation or inadequacies in either the fertilization or culture media. These results indicate that physiological concentrations of energy substrates (2 mM glucose, 6 mM lactate) during maturation increase glycolytic and PPP activity in porcine oocytes closer to that observed in vivo, suggesting increased oocyte viability and more complete cytoplasmic maturation during the first step of in vitro embryo production in the pig.