

Fat area and lipid droplet morphology of porcine oocytes during in vitro maturation with trans-10, cis-12 conjugated linoleic acid and forskolin

Lipid droplets (LD) in porcine oocytes form a dark mass reaching almost all cytoplasm. Herein we investigated changes in fat areas, cytoplasmic tone and LD morphology during in vitro maturation (IVM) of porcine oocytes cultured with 100mM trans-10, cis-12 conjugated linoleic acid (t10,c12 CLA) or 10mM forskolin at different time periods. Four groups were constituted: control, excipient, t10,c12 CLA and forskolin, with drugs being supplemented during 44 to 48h and the initial 22 to 24h in Experiments 1 and 2, respectively. In Experiment 3, forskolin was supplemented for the first 2 h. Matured oocytes were inseminated with frozen-thawed boar semen and cleavage rate recorded. Before and during IVM, samples of oocytes were evaluated for LD, total and fat areas and fat gray value or for meiotic progression. Results showed that forskolin supplementation during 44 to 48 h or 22 to 24 h inhibits oocyte maturation (exp. 1: forskolin = $5.1 \pm 8.0\%$, control = $72.6 \pm 5.0\%$; exp. 2: forskolin = $24.3 \pm 7.4\%$, control = $71.6 \pm 5.6\%$) and cleavage (exp. 1: forskolin = $0.0 \pm 0.0\%$, control = $55.4 \pm 4.1\%$; exp. 2: forskolin = $8.3 \pm 3.3\%$, control = $54.5 \pm 3.0\%$). Forskolin also reduced oocyte and fat areas. In Experiment 3, forskolin negative effect on oocyte maturation and cleavage disappeared, although minor ($P < 0.03$) LD and oocyte fat areas were identified at 22 to 24 h of IVM. Oocytes supplemented with t10,c12 CLA during 44 to 48h presented a lighter ($P < 0.04$) colour tone cytoplasm than those of control and forskolin. In conclusion, t10,c12 CLA and forskolin were capable of modifying the distribution and morphology of cytoplasmic LD during porcine oocyte maturation, thus reducing its lipid content in a time-dependent manner.