



A245 Embryology, developmental biology, and physiology of reproduction

### **Does lipopolysaccharide affect the rate of cleavage and embryonic development *in vitro*?**

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Cows with postpartum uterine infection reduce the growth of dominant follicles and the production of estradiol, which results in postpartum delay in ovulation (Sheldon, *Biology Reproduction*, 81, 1025, 2009). Gram-negative bacteria, such as *Escherichia coli*, contain on its outer membrane lipopolysaccharide (LPS) that produces an acute inflammatory response at systemic level, which causes an increase in body temperature and serum levels of pro-inflammatory cytokines (Carroll, *Innate Immunity*, 15, 81, 2009). Thus, the objective of this work was to evaluate the effect of the supplementation of MIV medium with LPS (0.1, 1.0 and 5.0  $\mu\text{g} / \text{mL}$ ) in the cleavage rate and initial embryo development *in vitro* in cattle. *In vitro* embryo production was performed in a 5% CO<sub>2</sub> incubator at 39°C using commercial media (Progest - Biotechnology in animal reproduction, Botucatu, SP). Oocytes (COCs) were obtained from slaughterhouse, washed and selected by morphology. COCs of grade I, II and III were randomly distributed into four groups (n = 50 COCs / group) according to the addition of LPS in the IVM medium (G1: 0  $\mu\text{g} / \text{mL}$ , G2: 0.1  $\mu\text{g} / \text{mL}$ , G3: 1.0  $\mu\text{g} / \text{mL}$  and G4: 5.0  $\mu\text{g} / \text{mL}$  LPS, SIGMA-ALDRICH®, St. Louis, MO, USA). IVM occurred for 22 hours. IVF was performed with a concentration of 1x10<sup>6</sup> sperm/mL during 20 hours. After this period, the probable zygotes were cultured in CIV medium covered with mineral oil for 7 days. At day 3, the cleavage rate (cleaved / inseminated) was assessed and on the same day, and also at day 5, 70% of the culture medium was renewed. On day 7 the embryonic development rate (blastocysts / inseminated) was evaluated. Thus, 4 replicates were performed with approximately 150 oocytes / group. The effect of LPS on the cleavage and embryo development rate was analyzed by repeated measurements of ANOVA after data transformation and the Tukey post-hoc test. There was no difference for cleavage rate, G1: 21.5  $\pm$  3.3 (58.9%); G2: 22.5  $\pm$  8.1 (55.6%); G3: 23.7  $\pm$  5.3 (60.1%); G4: 30.2  $\pm$  10.6 (72.9%) (P > 0.05); or embryonic development: G1: 6.7  $\pm$  2.1 (13.7%); G2: 8.0  $\pm$  3.5 (15%); G3: 9.3  $\pm$  5.1 (17.7%); G4: 10.7  $\pm$  8.1 (19.3%) (P > 0.05). These data suggest that exposure of oocytes to LPS does not affect the initial embryonic development *in vitro* in cattle.