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Effect of oocyte density on commercial *in vitro* bovine embryo production

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The efficiency of the *in vitro* bovine embryo production (IVEP) is the key to increase the profitability of commercial labs. Some factors may affect embryo development *in vitro*, such as oocyte quality, energy substrate, oxygen tension, and embryo density (Khurana et al., Theriogenology, 54, 741-766). In ovum pick up (OPU) procedures the number of the oocytes recovered can be variable and low and, in commercial labs, oocytes and embryos have to be cultured from each individual donor cow, regardless of the recovered number, becoming necessary to process them in small groups. It has been reported that optimum development is achieved when oocytes are cultured in groups of 20-40, in comparison with groups of 5 and 10 structures (O'Doherty et al., Theriogenology, 48, 161-169). Therefore, the aim of this study was to investigate the effect of oocyte density on IVEP efficiency, which was conducted in a commercial lab of *in vitro* fertilization (IVF). In the lab, cumulus-oocyte complexes (COCs), obtained from random donors, were assigned in six groups: 1-5 COCs per drop (group 1, n=37 drops), 6-10 COCs (group 2, n=91), 11-15 COCs (group 3, n=89), 16-20 COCs (group 4, n=57), 21-25 (group 5, n=45) e 26-30 COCs (group 6, n=27), where n is the number of replicates of each group. Oocyte maturation was performed for 24 hours in drops of 80 μ L. After that period, COCs were fertilized with semen from different bulls, in drops of 70 μ L for 22 hours. After IVF, presumptive zygotes were cultured for seven days, in drops of 60 μ L. The percentage of cleavage (third day of culture) and blastocysts (seventh day of culture) were calculated on the number of viable oocytes. All the steps of embryo production were performed with the same conditions in the six groups, including volume of media and the incubation at 38,8 °C in a 5% CO₂ in air with high humidity. Data were analyzed by the method of least squares using analysis of variance by proc GLM. Differences between means were compared by Tukey test with 5% significance. Differences in cleavage rate between groups were not observed (groups 1-6, respectively: 84.9%±32.6%, 82.5%±26.1%, 85.3±19.5%, 80.3%±22.4%, 77.6%±22.5% and 82.1%±21%, P>0.05). Embryo production rate also did not differ between groups (groups 1-6, respectively: 35.4%±42.9%, 35.5%±37.8%, 40.5%±31%, 28.7%±26.3%, 30.5%±25.2% and 39.4%±28.6%, P>0.05). In conclusion, there was no influence of oocyte density on the IVEP, which confirms the efficiency observed in commercial scale, in which the number of oocytes is variable. Besides, other factors - as donors and bulls - which were not the focus of analysis in this study, can be involved.