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Effect of antioxidants on in vitro nuclear and cytoplasmic maturation of bovine oocytes.

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Resumo

Despite all the advances in IVP, approximately 30 to 40% of oocytes that mature in vitro reach the blastocyst stage. Considering that the quality of the oocyte is the key factor for the success of IVP, inadequate conditions during maturation may be responsible for the low efficiency of the technique. One of the alternatives to improve this step is the use of antioxidants, which regulate the production of reactive oxygen species (ROS) from aerobic metabolism. The objective of this study was to evaluate the effect of ITS and Cysteamine (CYS) supplementation on the nuclear and cytoplasmic maturation of bovine oocytes. COCs were obtained from slaughterhouse ovaries and matured in the presence (+) or not (-) of Cysteamine (CYS) and ITS. Therefore, four treatments were used: T1. Control: -ITS -CIS; T2. -ITS+CIS; T3. +ITS+CIS; T4. +ITS-CIS. The CC expansion during IVM, which was determined by the difference between the mean area of all COCs from each treatment before and after IVM. Nuclear maturation was evaluated by staining the oocytes with lacmoid and determing the stage of meiosis. Subsequently, the COCs of the different treatments were subjected to fertilization and in vitro culture. Embryo were evaluated on D2 for cleavage and D6 and D7 for blastocyst formation. Blastocyst of D7 were differentially stained and total cell number and % trophoblast and inner cell mass (ICM) were determined. The number of embryos produced and the expanded area of COC were evaluated by ANOVA, the kinetic of nuclear maturation by Chi-square and cell count by Kruskall-Wallis. No difference (p>0.05) was observed in the areas (mm2) of COC's expansion among treatments [(mean ± standard deviation (SD)]: T1= 7.501±1.371; T2= 8.043 ± 1.334 ; T3= $6,450\pm1,497$; T4= 6.212 ± 1.167 . Also no effect (p>0.05) of presence of either antioxidant used was detected on nuclear maturation with the majority of oocyte reaching the metaphase II stage: [T1= 89.4% (68/86), T2= 83.5% (71/85); T3= 76.05% (54/71); T4= 67.9% (55/81)]. Similarly, neither cleavage [T1= 72±5% (84/116); T2=74±6% (97/131); T3=72±7% (88/122); $T4=72\pm8\%$ (84/117)] nor blastocyst production at D7 (T1= 35±7% (41/116); T2= 43±7% (56/131); T3= 30±14% (37/122); T4= 35±14% (41/117)] were improved by the treatments. The differential staining showed that total cell number of D7 blastocysts (T1= 179±18; T2= 185±12; T3= 181±11; T4= 186±11) and the percentage of ICM and trophoblast cells of the blastocyst were similar (p>0.05) among all treatments. In conclusion, the use of ITS or CYS does not affect nuclear maturation, cytoplasmic maturation or the quality of in vitro produced embryos.