Animal Reproduction

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CRYOTOLERANCE OF IN VITRO PRODUCED BOVINE EMBRYOS ORIGINATED FROM OOCYTES MATURED IN MEDIUM SUPPLEMENTED WITH RECOMBINANT HUMAN FSH

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Resumo

The recombinant human FSH (rhFSH) has been used during in vitro maturation (IVM) of cattle oocytes as an alternative to the FSH obtained from porcine pituitary (pFSH). The rhFSH show less variation on biological activity, and lacks the sanitary risks associated with the use of protein extracts obtained from other species. However, few studies have compared the efficiency of rhFSH and pFSH considering not only the results of IVM, but also subsequent embryo quality. The aim of this study was to evaluate the cryotolerance of embryos produced in vitro after IVM with rhFSH. Cumulus-oocyte complexes (COC, n=2,040) recovered from slaughterhouse ovaries and morphologically classified as grades I or II were used. The COC were randomly allocated into three groups, which were IVM in TCM199: 1) without FSH (-FSH, n=680); 2) with 0.5 µg/mL pFSH (pFSH, Folltropin-V, Vetoquinol, n=680); or 3) with 0.1 UI/mL rhFSH (rhFSH, Gonal, Merck, n=680), all groups in the same culture conditions (38.5°C, 5% CO2). Cumulus expansion was evaluated at 22 h of IVM and subjectively classified as poor, intermediate, or good. Sperm from a single sire with known fertility was used for in vitro fertilization. The presumptive zygotes were cultured under low oxygen concentration (5% CO2, 5% O2, and 90% N2, at 38.5°C). The cleavage, blastocyst and hatching rates were addressed at days 3, 7, and 10 of culture, respectively. At the day 7, a subset of grade I expanded blastocysts (n=54, 69, and 71 embryos from groups -FSH, pFSH and rhFSH) were cryopreserved by vitrification, stored in liquid nitrogen, and then thawed and in vitro cultured under low oxygen concentrations. Hatching rates were evaluated after 72h. Data were evaluated by the Chi-squared method using the SAS software (SAS Institute). Cleavage and blastocyst rates, were higher in the rhFSH, compared with -FSH and pFSH groups (80.4% and 46.3% vs 71.5% and 34.6%, and 70.0% and 38.0%, respectively, P=0.0069 and P=0.0207). However, there was no difference in hatching rates (74.5%, 79.0%, 70.1% for -FSH, pFSH and rhFSH groups, respectively, P>0.05). There was no difference in hatching rates after vitrification among groups (75.0%, 72.5% and 73.2%, P=0.8875, for groups -FSH, pFSH and rhFSH, respectively). In summary, there is no evidence that the presence or the source of FSH (porcine or recombinant human) during IVM affect subsequent embryo cryotolerance.

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