SP 7554
2017
SP-PP-SP 7554

A099 OPU - IVF and ET

Goat incubator: The doe as a life incubator of bovine oocytes - first step

Ribrio Ivan Tavares Pereira Batista, Luiz Sérgio de Almeida Camargo[2], Joanna Maria Gonçalves de Souza Fabjan[3], Jader Forquim Prates[4], Juliane Teramachi Trevizan[4], Felipe Zandonadi Brandao[3], Jeferson Ferreira da Fonseca[4]

[1]Embrapa Caprinos e Ovinos, Núcleo Regional Sudeste, CEJHB–Embrapa Gado de Leite, Coronel Pacheco, MG; [2]Embrapa Gado de Leite, Juiz de Fora, MG; [3]Faculdade de Medicina Veterinária, Universidade Federal Fluminense, Niterói, RJ; [4]Embrapa Caprinos e Ovinos, Núcleo Regional Sudeste, Coronel Pacheco, MG.

Despite significant improvements in the in vitro production of cattle embryos, the suboptimal in vitro culture environment still limits the embryo quality and production. Techniques that associate the advantages of in vivo and in vitro systems, such as intrafollicular transfer of immature oocytes, have been proposed mainly to increase the embryo quality. In this context, we tried to use a goat as live incubator and associated nonsurgical embryo transfer techniques in small ruminants to perform ex situ (in vivo) maturation of bovine oocytes. For this, immature bovine cumulus-oocyte complexes (COCs) of grade 1 and 2 were randomly distributed into two groups for in vitro (IVM; n = 38) and ex situ (ESM; n = 40) maturation. The IVM was performed for a period of 24 h in TCM-199 medium (Gibco Life Technologies, Inc., Grand Island, NY, USA) supplemented with 20 mg/mL of FSH (Pluset, Calier, Barcelona, Spain), 0.36 mM sodium pyruvate (Sigma Chemical, St. Louis, MO, USA), 10 mM sodium bicarbonate (Sigma Chemical, St. Louis, MO, USA) and 50 mg/mL streptomycin/penicillin (Sigma Chemical, St. Louis, MO, USA) at 38.8 °C in an atmosphere of 5% CO2 in air with maximum humidity. For ESM, a pre-synchronized nulliparous goat (12 months old) received 40 immature COCs in the uterine horn apice by transcervical route (Fonseca et al., 2014 Arq. Bras. Med.vet. Zootec) and 24 h after the procedure the structures were retrieved by the uterine flushing (Fonseca et al., 2013 Small Rumin Res). For analysis of the nuclear maturation rate and lipid quantification, the oocytes were denuded (0.1% hyaluronidase), fixed (4% paraformaldehyde) and stained with 10 µg/mL Hoechst 33342 and 10 µg/mL Nile Red (Molecular Probes, Inc., Eugene, OR, USA) dissolved in physiological saline (0.9% NaCl) with 1mg/mL polyvinylpyrrolidone. Oocytes displaying metaphase II plate were considered matured. The lipid amount was inferred by measuring the fluorescence intensity using the ImageJ program and fluorescence intensity were compared by Student's t-test. Forty-seven percent of the structures were recovered after uterine flushing (19/40). The nuclear maturation rate was 94.5% (18/19) and 81.6% (31/38) for ESM and IVM groups, respectively. In vitro-matured oocytes contained more lipid droplets, expressed as a higher ($p < 0.05$) amount of emitted fluorescence light (858 ± 73 arbitrary fluorescence units) than ex situ-matured oocytes (550 ± 64 arbitrary fluorescence units). This is the first report associating nonsurgical embryo transfer techniques with goat as live incubator for maturation of bovine oocytes. We conclude that transcervical transfer of bovine oocytes to uterine goat may be an alternative to in vitro maturation aiming the reduction of lipids without compromising nuclear maturation. Further studies are required to improve the oocyte recovery rate.

Financial Support: Fapemig (Project CVZ PPM 00042-14) and CNPq (Project 310166/2012-8).

SP 7554