Animal Reproduction

THEMATIC SECTION: 37TH ANNUAL MEETING OF THE BRAZILIAN EMBRYO TECHNOLOGY SOCIETY (SBTE)

OPU-FIV

Effect of the quantity of COCs and the injection quality on embryo recovery after Intrafollicular Transfer of Immature Oocytes (IFIOT)

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The Intrafollicular Transfer of Immature Oocytes (IFIOT) has emerged as an alternative for bovine embryo production. However, the technique still yields unsatisfactory results for commercial-scale implementation. This study aimed to evaluate whether the quantity of cumulus-oocyte complexes (COCs) injected into the preovulatory follicle, along with injection quality, impacts the final embryo recovery. Immature oocytes were obtained from slaughterhouse ovaries. Subsequently, twenty-two Nelore cows synchronized as previously described (Faria *et al*., Reproduction, Fertility and Development 33(5) 372-380, 2021) were subjected to IFIOT using a 27-gauge needle (Unisis®, Fukuoka – Japan) for the injection of either 25 (T25 treatment , n= 9) or 50 COCs (T50, n= 8), loaded in 10 µl of PBS medium. The injection quality was classified into grades 1 (n= 12) (injection into the center of the follicle and visualization of the entry of all structures) or 2 (n= 5) (injection into the periphery of the follicle and entry of the structures too fast (vortex) or too slow (almost imperceptible), more than one perforation in the follicle and no visualization of the entrance of the structures and/or reduction of the follicle after removing the needle). Immediately after IFIOT, ovulators females were artificially inseminated with a dose of conventional semen from a Nelore bull with known fertility data. Simultaneously, groups of 25 to 30 COCs were allocated for IVP. Nine days after IFIOT, uterine flushing of ovulators females was performed. The structures recovered were classified as zona pellucida, unfertilized oocyte, degenerated embryo, morula, early blastocyst, blastocyst or expanded blastocyst. The recovery rate of structures and the rate of embryos per treatment and injection quality grade were evaluated. Expanded blastocysts produced by IFIOT and IVP were evaluated for diameter and total number of cells. Data were analyzed by analysis of variance with mixed models (SAS, 9.4 Version). The recovery rate of structures after IFIOT was not affected (P>0.05) by the quantity of injected COCs (T25 - 21.69%; T50 - 19.83%) or the injection quality (grade 1 - 24.66%; grade 2 - 16.86%). Similarly, the recovery rate of embryos was also not affected by the quantity of COCs (T25 - 3.77%; T50 - 3.56%) or the injection quality (grade 1 - 6.83%; grade 2 - 0.50%). Furthermore, there was no interaction between the quantity of COCs and the injection quality for the analyzed parameters (P >0.05). From 178 COCs, 43 (24.2%) blastocysts were obtained in the IVP group. IFIOT and IVP embryos showed similar (P > 0.05) diameter (IFIOT - 175,0 µm; IVP – 179,1 µm) and number of cells (IFIOT – 124,2; IVP – 141,3). It can be concluded that IFIOT embryo recovery is not affected by injection quality or the quantity of injected COCs. Additionally, IFIOT and IVP embryos showed similar quality, based on the evaluated parameters. Financial Support: FAP-DF, Embrapa, CAPES.