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STEM CELLS INJECTION IN THE OVARY INCREASED THE OOCYTE AND EMBRYO PRODUCTION IN BOVINE FEMALES.

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Abstract.Abstract1:

OBJECTIVE: The aim of this study was to evaluate the follicular population, oocytes recovery rate per OPU and embryo production after the injection of allogeneic mesenchymal stem cells (MSCs) in the ovarian cortical layer.

DESIGN: 27 Nelore (*Bos indicus*) cows were synchronized for follicular growth wave [D -65: insertion of an intravaginal P4 device (1.0 g) and administrations of EB (2.0 mg im) and PGF2α (0.53 mg im,)]. On D -60 the P4 device was removed and the animals submitted to ultrasonographic evaluations and ovum pick up (OPU) followed by *in vitro* embryo production (IVEP) at 30-day intervals (D -30 and D0). On D1, cows were distributed to one of three experimental groups: CONT (no stem cells application; n=7), MSC1 (MSCs application in one ovary - $5x10^6$ cells per ovary - n=10) and MSC2 (MSCs application in two ovaries - $5x10^6$ cells per ovary - n=10).

MATERIALS AND METHODS: Allogeneic MSCs from adipogenic origin were isolated and cultivated in IMDM culture medium with 20% FBS and 1% P/S, at 37 °C in 5% of CO_2 for cellular expansion from third passage. MSCs were characterized by Flow Cytometry, with the CD44 and CD29 as positive markers, presenting 93.2% and 98.4% positivity and the CD18 and CD45 negative markers, that showed 96% and 99.4%. After MSCs application, the cows of all groups were again submitted to follicular synchronizations, ultrasonographic evaluations and OPU-IVEP procedures at 30, 60, 90, 120, 150 and 180 days. The data were analyzed as time-repeated measures using the GLIMMIX procedure of SAS.

RESULTS: After the MSCs treatment was observed interaction between treatment and time for the numeric variables: total follicles aspirated (P=0.001) total oocytes retrieved (P=0.01), viable (P=0.03) and cleaved (P=0.02), total embryos per OPU session (P=0.01) and hatched blastocysts (P=0.05). In cows receiving MSCs, the production curve of numeric variables presented less reduction in time in relation to CONT group. Interaction between treatment and time was not observed for the rates evaluated. It was

verified that cows receiving MSCs had an increase in the oocytes recovery rate (CONT=62.6%^{b}, MSC1=62.8%^{b} and MSC2=67.8%^{a}; P=0.01), viable oocytes rate (CONT=63.0%^{b}, MSC1=64.3%ab and MSC2=67.8%^{a}; P=0.01), cleaved rate (CONT=63.8%^{b}, MSC1=67.9%^{a} and MSC2=64.5%ab; P=0.03), blastocyst rate (CONT=34.7%^{b}, MSC1=39.0%^{a} and MSC2=33.3^{b}%; P=0.001) and hatched blastocysts rate (CONT=26.8%^{b}, MSC1=31.5%^{a} and MSC2=26.2%^{b}; P=0.0003).

CONCLUSIONS: We concluded that MSCs application increased the OPU-IVEP efficiency in bovine female models.

Abstract.Abstract2:

References: None.

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