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Effectiveness of *in vitro* maturation strategies to reduce the lipid accumulation in buffalo embryos

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In vitro embryo production (IVEP) is a procedure that can promote genetic improvement in a short time frame. However, the success rates obtained with this biotechnology in buffaloes are still inconsistent, which can be associated with the high concentration of lipids in the cytoplasm of oocytes and embryos. Considering the potentially positive impacts of strategies to replace/reduce the supplementation of fetal bovine serum (FBS) during IVEP, the objective of this study was to evaluate the effects of reducing the concentration of FBS and/or use of 5 mM L-carnitine (LC) during *in vitro* maturation on the development and lipid accumulation of buffalo embryos. In the first experiment, we aimed to determine the lowest concentration of FBS in the IVM medium able to maintain the embryo development rate obtained by the control group (10% FBS). Buffalo oocytes were placed in IVM in bovine serum albumin (BSA) medium supplemented with 0%, 2.5%, 5% or 10% FBS for 22 h, and then fertilized in Talp-IVF medium for 24 h, and *in vitro* cultured in modified SOF medium supplemented with 1.5% FBS at 38.5 °C and 5% CO_2 atmosphere in air for 7 days. Blastocyst rates were evaluated and the data analyzed using the analysis of variance (ANOVA) and Tukey test. After defining the lowest effective concentration of FBS as 5% [27/79; 34.18%^{a}, similar to 10% - 52/105; 34.67%^{a} and superior to 0% (11/104 - 10.58%^{b}) and 2,5% (16/83 - 19.28%^{b}) groups], we performed a second experiment in which the 0%, 5% and 10% FBS groups were also evaluated regarding the addition of 5 mM of L-carnitine in the IVM medium. The blastocysts produced in this experiment were submitted to lipid quantification tests, involving staining followed by observation by optical (OilRed O) and confocal (BODIPY 493/503) microscopy. The lipid quantification data were evaluated by the nonparametric Kruskal-Wallis test. All the statistical analyses were performed with the SPSS version 22.0.0.0 software, except for the lipid data, which were evaluated with GraphPad Prism 7 version 7.03. No difference was observed between the 5% (60/184 - 32.61%^{a}) and 10% FBS (82/227 - 36.12%^{a}) groups in blastocyst rate, which were superior to 0% (34/270 - 12.59%^{c}) and groups supplemented with L-carnitine (5% FBS-LC: 32/144 - 22.22%^{b} and 10% FBS-LC: 38/153 - 24.84%^{b}). There was no difference regarding embryo lipid accumulation. The results indicate that it is possible to reduce the FBS concentration to 5% in IVM media for buffalo embryo production and the supplementation of the maturation medium with L-carnitine at a concentration of 5 mM did not cause an increase in the embryo production of this species. Furthermore, alterations in the lipid accumulation during the IVEP were not found, with or without the presence of FBS and addition of L-carnitine during the IVM, indicating the need for further research, mainly involving the *in vitro* culture step of buffalo embryos.