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Methyl beta cyclodextrin use as agent of sperm capacitation on the *in vitro* production embryos buffaloes (preliminary results)

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Cyclodextrins are cyclic oligosaccharides which have the ability of lipids such as cholesterol incorporation and can be used to alter the cell membrane cholesterol content (Visconti et al., 1999). Studies show their use in cholesterol removal from seminal membranes and in the induction of bovine sperm capacitation (Purdy et al, 2004, BiolReprod, 71, 522-527; Kato et al, 2010, Zygote, 19, 21. 30). Thus, the aim of this work is to evaluate the use of cyclodextrin as sperm capacitation agent, analyzing their influence on IVPE in buffalo. Buffalo ovaries were collected from slaughterhouse and the cumulus-oocyte complexes (COCs) were matured *in vitro* in TCM-199 medium supplemented with 10% FBS, FSH and LH, for 22 hours at 38.5 ° C in 5% CO₂. COCs were fertilized in TALP - FERT medium supplemented with penicillamine, hypotaurine and epinephrine, modified according to the experimental groups: Negative Control - NC (without BSA, heparin or cyclodextrin), Positive Control - PC (with BSA and heparin as capacitation agent) and groups with different Methyl Beta Cyclodextrin (MBCD) concentrations (Sigma, St Louis, USA) (MBCD-0.5mM, MBCD-0.75mM and -MBCD-1.5mM) and incubated under the same conditions mentioned for IVM. 24 hours after fertilization, the zygotes were placed on SOF medium drops supplemented with BSA (6 mg /mL), 10% FBS, aminoacid, pyruvate, gentamicin and antioxidant. The cleavage and blastocyst rates were evaluated on the 2nd, 6th and 7th cultivation day, respectively, and the results were analyzed by ANOVA and Tukey post-test, adopting the significance level of 5%. The cleavage rate there was no significant difference between PC and MBCD-0.5 mM (0 vs 39.23 ± 4.06 and 32.15 ± 17.25), but NC differed MBCD-0.75mM and MBCD-1.5mM (0 vs 46.94 ± 17.00 and 46.45 ± 17.70, respectively), but it had no significant difference between the groups with MBCD. In the production of blastocysts at 6th day of culture, the PC groups (22.25 ± 5.75), MBCD- 0.75mM (18.18 ± 8.27) and MBCD-1, 5mM (17.77 ± 6.74) did not differ significantly (p>0.05), however they differ from NC groups and MBCD 0.5mM (0 vs. 10.74 ± 6.24, respectively) (p<0.05). And at 7th day of culture PC groups (33.97 ± 3.38) and MBCD- 1.5mM (25.90 ± 10.68) are not different, but differed from NC (0), MBCD-5mM (11.36 ± 10.54) and MBCD-0.75mM (18.18 ± 8.27). Thus, more repetitions are needed in order to confirm that the use of buffaloes MBCD in concentration of 1.5mM shows similar results to the positive control, thus demonstrating that MBCD can be used as a heparin substitute.