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Effect of *in vitro* maturation (IVM) method and gas tension during culture upon *in vitro* production of F1 embryos (Nelore x Angus) using Y-sorted semen

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The availability of X-sorted sperm was a turning point for the use of *in vitro* embryo production (IVEP) in dairy breeds in Brazil. Due to a repressed demand for crossbred dairy heifers, IVEP using female sex-sorted semen has boosted up in the past 10 years. On the other hand, less attention was given to the use of Y-sorted semen for IVEP, despite the great potential for large-scale production of F1 male beef calves. Optimization of IVEP in beef, however, must take into consideration some particularities of extensive beef cattle operations, including the usual long distances between the site of oocyte collection and the IVEP laboratory. In this regard, *in vitro* maturation (IVM) during oocyte transportation may represent a critical step for the entire process. The aim of the present study was to evaluate the effects of IVM in tubes versus Petri dishes followed by *in vitro* culture (IVC) under two distinct O_2 tensions, i.e., a randomized complete design with a 4x4 arrangement of treatments: IVM in tube-IVC in high O_2 (n=1043); IVM in tube-IVC in low O_2 (n=1131); IVM in Petri dish-IVC in high O_2 (n=866), and IVM in Petri dish-IVC in low O_2 (n=846). Cumulus-oocyte complexes (COCs) were collected from ovaries of Nelore cows at a slaughterhouse and randomly assigned for IVM in either 5mL tubes (25 COCs per tube in 400mL of medium TCM199) or microdrops on a Petri dish (25 COCs per 90mL drops of TCM199), at 38.5° C, for 22-24h in 5% CO_2. Matured COCs were *in vitro* fertilized (IVF) with commercial Y-sorted semen from a single Angus sire. Sperm preparation included centrifugation in a mini-Percoll gradient followed by co-incubation of gametes for 18-22h. After IVF, putative zygotes were cultured in SOF medium at 38.8°C with two distinct gas tensions: 5.5% CO_2 and 20% O_2 (high O_2) or 5.5% CO_2 and 5% O_2 (low O_2). The endpoints cleavage rate, blastocyst rates at days 7 and 8 (late embryo development), and proportion of grade I embryos were analyzed using the SAS GLM procedure and differences among means compared by the Tukey´s post hoc test. We did not observe an interaction between type of IVM and gas tension during IVC. Nonetheless, IVM in tubes increased cleavage and day 7 blastocyst rates compared to IVM in petri dish (54.2±1.9% *vs* 41.0±2.4% and 19.5±1.5% *vs* 12.7±1.1%, respectively, $P<0.05$), but had no effect ($P>0.05$) on the proportion of grade I embryos nor day 8 blastocysts. O_2 tension did not affect ($P>0.05$) blastocyst rate and proportion of grade I embryos, but low O_2 reduced day 8 blastocyst rate compared to high O_2 (10.5±2.6% *vs* 20.5±3.0%, respectively, $P<0.05$). These results demonstrate that IVM in tubes is a feasible alternative for COC maturation during transport to the laboratory. In addition, IVC performed at high O_2 delayed embryo development, with consequences for the logistics of embryo transfers into recipients. Thus, we conclude that both IVM and IVC still need optimization for large-scale IVEP programs using Y-sorted beef cattle semen.