A308 Support biotechnologies: Cryopreservation and cryobiology, diagnosis through imaging, molecular biology, and "omics"

Effect of sex on survival of bovine *in vitro* produced embryos vitrified by Cryotop

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Several studies have shown that male and female embryos are different not only in speed of development, but also for metabolism, gene expression, epigenetic patterns and stress response. This study aimed to evaluate if the cryopreservation effects caused on embryos may vary between gender. Oocytes obtained from slaughterhouse ovaries underwent IVM for 24 hours, were inseminated with 1 x 10^6 spermatozoids/mL, co-incubated in IVF medium for 16-18 hours, and possible zygotes were cultured *in vitro* (IVC) for 8 days. Cleavage at D2 and blastocyst rates at D6, D7 and D8 were evaluated. At D7, grade I embryos at expanded blastocysts stage, according to IETS manual, were removed from IVC and divided in two treatments: control (C) and vitrified (V) by *Cryotop*. After warming process, embryos returned for additional 24 hours in ICV conditions, for survival (not degenerated embryos) and evolution rates evaluation. Afterwards, embryos from both treatments (C: n=129; V: n=165) were individually stored in DM-PBS with lysis buffer, at – 20 °C, for sex determination, that was assessed by polymerase chain reaction and confirmed in 1.5% agarose gel. Data were analyzed by Mann Whitney test ($P < 0.05$). Male (n=57 [44%] e n=89 [53.9%]) and female (n=72 [55.8%] e n=76 [46%]) embryos percentage were similar for both control group and vitrified one ($P > 0.05$), respectively. For the vitrified embryos difference between male and female data was not seen for survival rate (n=87 [55.1%] e n=71 [44.9%], respectively), evolution form expanded to hatched blastocyst rate (n=61 [52.4%] e n=50 [47.6%], respectively) and degeneration rate (n=2 [28.6%] e n=5 [71.4%], respectively). These results suggest that male and female embryos have the same vitrification tolerance for *Cryotop* method.