A160 OPU-IVP and ET

Different sperm selection methods used for ovine in vitro embryo production

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The sperm selection method is one step of in vitro embryo production systems and influences the embryo development rates. The aim of this study was to compare the efficiency of different sperm selection methods used in ovine IVP systems in terms of embryo development rates. Methods as Swim up (SW), Mini Optiprep® (MO), Mini Percol® (MP) and Mini Isolate® (MI) were compared in terms of cleavage rates at day 2 and development rates at day 8 (blastocysts/oocytes inseminated). Cumulus oocyte complexes (COCs) were aspirated from ovine ovaries obtained at a local slaughterhouse, selected and matured for 22-24h. The same batch of ovine fresh semen was used in the different sperm selection methods in the experiment. The SW method was performed by layering an aliquot of semen under tris glucose citric acid medium. After 30 min of sperm migration in controlled conditions (39°C), the upper portion was removed and centrifuged at 200G for 5 min. The small volume gradients (MP and MI) were both prepared at 90 and 45%. Semen aliquots were layered over MP and MI and centrifuged at 700G for 5 min. The small volume gradient MO was prepared at 30, 28 and 26% and after semen aliquots were layered it was centrifuged at 900G for 15 min. The sperm pellet was isolated and centrifuged at 700G for 5 min in fertilization media, in all treatments. The *in vitro* matured COCs were randomly distributed in four treatments: SW (n=130), MO (n=152), MP (n=120) and MI (n=110) and inseminated with 1x 10^6 sptz/mL spermatozoa maintained in fertilization media during 18h. Embryo culture was performed during 8 days in SOFaa media with 0.8% BSA in bag system in an atmosphere arrangement of 5% CO_2, 5% O_2, and 90% N_2. Embryo development rates were compared using variance analysis and Duncan test at 5% of significance level (SAS). No differences were observed in the cleavage rates (71%, 76%, 75% and 81%) and development rates at D8 (21%, 24%, 16% and 17%) (P>0,05) for SW, MO, MP and MI respectively. The experiment results showed no influence of the different sperm preparation methods over the embryo development rates found in the ovine *in vitro* embryo production systems.