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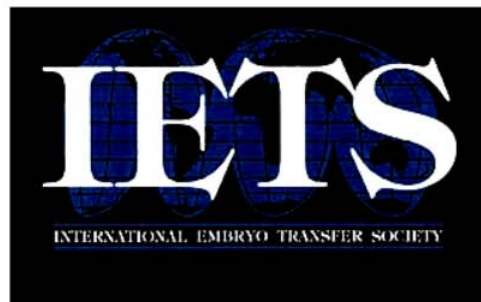
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*“ADVANCES AND NEW CONCEPTS
IN THE UNDERSTANDING OF...”*



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73 EFFECT OF THE BUFFER SYSTEM, CRYOPROTECTANT, AND ANTIOXIDANT ON SPERM FROM ENDANGERED CATALONIAN SHEEP XISQUETA AND ARANESA AND GOAT BLANCA DE RASQUERA BREEDS DURING PRESERVATION

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Abstract

In an attempt to increase the possibilities of a better sperm cryopreservation from these endangered small ruminant catalonian breeds, we are studying different strategies. One of them is to study the effect of the buffer system Test (TEST) compared with the Tris and citric acid (TRIS) buffer system, testing simultaneously both systems in a 1% (soybean lecithin or in a 15% (v/v) powdered egg yolk-based media supplemented both with 5% glycerol and to as also the inclusion of 5 mM of butylated hydroxytoluene (BHT) as an antioxidant on the cryopreservation media. The main objective of the present study was to test first the effect of the different media when the sperm were cultured at 5°C. Briefly, fresh ejaculates from 6 young bucks of Blanca de Rasquera breed (1 year old) were collected by an artificial vagina in favourable reproductive period and immediately mixed in equal quantities. Pooled ejaculates were into 2 equal aliquots and washed by centrifugation (twice for 10 min at 600g) in TRIS- or TEST-based media without cryoprotectants and antioxidant. Afterward each pellet was split into 4 equal aliquots, re-suspended in TRIS or TEST depending on the experimental group, soybean lecithin, or powdered egg yolk-based media, and supplemented or with BHT and kept for 4 h at 5°C. Likewise, fresh ejaculates from 8 young rams (4 rams of Xisqueta and 4 rams of Aranesa breed, 1 year old) were collected and processed as buck semen samples. Sperm survival before cryopreservation was determined by eosine-nigrosine stain, and sperm motion parameters were analysed by a computer-assisted sperm analysis system (ISAS[®]). Six replications were performed in both species, and General Linear Model (SAS[®], Cary, NC, USA) was used for the statistical analysis. The highest sperm viability percentage (mean ± SE) sperm cultured 4 h at 5°C was observed in the extender with TRIS buffer system in powdered egg yolk-based media supplemented with BHT (81.1 ± 2.8), not showing significant differences with the other extenders, except with the viability of the samples in the extenders with TEST buffer system in soybean lecithin-based media supplemented (56.5 ± 2.9; $P < 0.001$) or not with BHT (60.1 ± 5.1; $P < 0.001$). On the other hand, no significant differences in sperm viability were observed on ram sperm between treatments. Nevertheless, the sperm quality motion characteristics (not shown) were quite different between all the treatments in both species. Considering that the present results are preliminary, we suggest that more analysis should be made to explain how the different composition of the extenders affect sperm quality during the cryopreservation process.

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