

THEMATIC SECTION: 37TH ANNUAL MEETING OF THE BRAZILIAN EMBRYO TECHNOLOGY SOCIETY (SBTE)

OPU-FIV

Use of a sperm separation device as an alternative for sires with low outcomes using gradient selection for *in vitro* embryo production: preliminary results

Marcelo da Cunha Xavier¹, Maurício Antonio Silva Peixer¹, Luiz Gustavo Araújo Oliveira¹, Patricia Furtado Malard², Rodrigo Moura Martins², Rosangela Vieira Andrade², João Henrique Moreira Viana³

¹BIO Reprodução Animal

²Universidade Católica de Brasília (UCB)

³Empresa Brasileira de Pesquisa Agropecuária (EMBRAPA)

E-mail: marcelo@biotecnologiaanimal.com.br

Sperm quality is an important factor affecting *in vitro* embryo production (IVEP) outcomes. The male factor is associated with significant differences in embryo rates, regardless of the oocyte donor used (Peixer *et al.* 2023). The selection of sires to be used in IVEP, however, is mainly based on their genetic merit or importance, rather than on sperm quality. Moreover, currently most IVEP laboratories prepare sperm for *in vitro* fertilization (IVF) using centrifugation in colloid density gradients, such as Percoll. Although a simple and straightforward procedure, this method has been associated with acrosome. The aim of this study was to evaluate the use of a sperm separation device (VetMotl, VetMotl inc., USA) to select viable sperm from sires with known records of low results after IVEP. Frozen-thawed sperm from Nelore sires (n=5) were used. The sperm of each bull was processed either by conventional centrifugation in a percoll gradient (45-90%) or diluted 1:1 in FERT-TALP medium and 850 µL were loaded in a separation chamber, which was then incubated during 30 min in a CO₂ incubator. The supernatant was then recovered from the upper part of the chamber, and evaluated for sperm concentration. Sperm processed by both methods were then co-incubated (aprox. 1x10⁶ sptz/mL) with *cumulus*-oocyte complexes from the same donors for 20h. The presumptive zygotes were cultured in SOF medium, which was adjusted for production of embryos to be transferred as fresh (two sires) or frozen-thawed (four sires) by direct transfer (DT). Cleavage and blastocyst rates were compared by the Chi-squared method. Sperm preparation using the separation chamber resulted in greater cleavage rates (81.8% [63/77] vs. 41.2% [103/250] for the fresh protocol and 51.4% [227/442] vs. 42.3% [135/319] for the DT protocol, respectively; P<0.05) and blastocyst rates (44.2% [34/77] vs. 16.0% [40/250] for the fresh protocol and 17.2% [76/442] vs. 7.5% [24/319] for the DT protocol, respectively, P<0.001), compared with Percoll centrifugation. In all sires, the use of the sperm separation chamber resulted in increase on embryo rates (from +2.4% to +48.1%, on average +26.5 ± 7.1%). In summary, the preliminary results suggests that the use of a sperm separation chamber is an alternative to improve blastocyst rates from sires with previous records of low IVEP outcomes.

Acknowledgements: VetMotl inc. and CNPq INCT 406866/2022-8.