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Effects of vitrification on morphology and mRNA expression in apoptotic genes in ovine immature cumulus oocytes complexes

Satish Kumar and Suresh Kumar Gahlawat
Chaudhary Devi Lal University, India

Cryopreservation is an important tool for conservation of biological materials. This study reports effects of vitrification on the morphology of ovine immature cumulus oocytes complexes, and mRNA content to observe the expression of key apoptotic genes, BAD, BAK, BAX, BID, BOK, BCL2A1, BCL2, MCL1, P53, and GAPDH on immature oocytes. For morphological evaluation using Zoom stereo-microscope, 853 oocytes were divided into 3 groups for vitrification using 40% Glycerol or 40% DMSO or 20% Glycerol + 20% DMSO. After thawing, a total of 819 oocytes were recovered, resulting in a loss of 34 oocytes during handling. The morphological examination of the oocytes revealed that the percentage of oocytes recovered in a morphologically normal state was minimum $84.2 \pm 2.3\%$ for glycerol 40% and maximum $94.09 \pm 1.1\%$ for 20% Glycerol + 20% DMSO. The proportion of oocytes recovered in a morphologically normal state was significantly lower ($P < 0.01$) for 40% Glycerol ($84.2 \pm 2.3\%$) compared to that for these groups. For scanning electron, microscopic studies 451 oocytes were used in this experiment for vitrification using a combination of 20% Glycerol + 20% DMSO and percentage of oocytes recovered in a morphologically normal state was 95.7 ± 2.07 recorded. The real-time PCR results for the comparative expression of the apoptotic gene using mRNA abundance showed the down-regulation BAD, BCL2A1 and MCL1. The expression of BCL2, BAX and BOK are similar in both the groups. However, BID and BAK were up-regulated. In addition, P53 was also downregulated proves vitrification safe for the preservation of immature oocytes in ovine.

Biography

Satish Kumar has expertise in Assisted Reproduction Technology. He is Assistant Professor at Ambala College of Engineering and Applied Research Ambala, Haryana, India. He is actively engaged in research and teaching in Department of Biotechnology. He has guided eleven M-Tech students in different research areas of Biotechnology and has been member and coordinator of various committees at college and university level. He has MSc degree in Biotechnology and Molecular Biology, Haryana Agricultural University, Hisar and PhD in Animal Biotechnology from Chaudhary Devi Lal University, Sirsa, and Haryana, India. He has authored one book and three book chapters and research articles in Journal of International and National Repute. He also serves as a member of the scientific advisory board of International Journal of Animal Biotechnology, India. He is a popular speaker who delivered lectures on the role of Biotechnology in human welfare and worked with scientists of International repute from top institutions across India. He has delivered many lectures in different institutions in India. He has also delivered a lecture via webinar on the topic "Advances in Biocatalysis and its impact on the early and late development of small molecules" to 12 PhD scientists and over 20 Research Associates to Merck Research Laboratories USA.

satishrana.biotech@gmail.com

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