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Cooling and Freezing of Boar Spermatozoa: Supplementation of the Freezing Media With Reduced Glutathione Preserves Sperm Function

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In this study, we evaluated the effects of glutathione (L-7-glutamyl-L-cysteinylglycine; GSH) supplementation of the freezing extender on semen parameters during the cooling (2 hours at 5°C) and freezing phases of the cryopreservation process to compensate for the decrease in GSH content observed during sperm freezing. To fully address these questions, we

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incorporated a new set of functional sperm tests. These included tests of mitochondrial function, inducibility of the acrosome reaction, in vitro penetration (IVP) of oocytes, changes in sulfhydryl group content in membrane proteins, and capacitation status. The main findings emerging from this study were that the addition of GSH to the freezing media resulted in 1) an improvement in percent motility (%MOT) and motion parameters of thawed spermatozoa, as measured by both microscopic analysis and computer-assisted semen analysis (CASA); 2) a higher number of total viable spermatozoa; 3) a higher number of noncapacitated viable spermatozoa; and 4) a decrease in the number of spermatozoa with changes in the sulfhydryl groups in membrane proteins. This protective effect on sperm function was more pronounced with 1 mM of GSH than with 5 mM of GSH.

Key words: Pig spermatozoa, antioxidants, cryopreservation, in vitro fertilization, capacitation status

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