MORPHOLOGY AND CASPASE-3 EXPRESSION ON GOAT OOCYTE AFTER WARMING WITH INSULIN TRANSFERRIN SELENIUM (ITS) AND HEAT SHOCK PROTEIN 70 (HSP70) SUPLEMENTATION

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ABSTRACT

This research aims to know the potential of the combination Insulin Transferrin Selenium (ITS) and Heat Shock Protein 70 (HSP70) as vitrification medium suplementation. Ovarium was collected from slaughter house and oocyte was collected by aspirated using 18 G needle form follicle. The classification of oocyte quality was perform, then oocytes with compact cumulus and the cytoplasm homogenized was maturated in vitro. As many as 20 oocyte were divided into four treatment groups. Group 1 as control (PBS+30%EG+Sucrose 1M). Group 2 (PBS+30% EG+Sucrose 1M +ITS 5 μg/ml). Group 3 (PBS+ 30% EG+ Sucrose 1M+ HSP70 0.5 μg/ml). Group 4 (PBS + 30% EG+ Sucrose 1M+ ITS 5 μg/ml+ HSP70 0.5 μg/ml). Vitrification used modified method. A pasteur pipette were pulled in a flame and cut in half to get a suitable internal diameter about 125 μm. Oocytes entered to the vitrification medium, then oocytes were loaded onto hemistraw. Furthermore hemistraw dipped in liquid nitrogen and put in a largestraw. Then the largestraw fixed on each tips and inserted into the cassette straw. Furthermore cassette inserted in container goblet of liquid nitrogen. For warming, oocytes were transferred to diluent solution with the gradual concentration of sucrose, oocytes dropped in to PBS + 1M sucrose for 2 minutes, and then transferred to PBS + 0.5M sucrose for 2 minutes, then transferred to PBS medium for 2 minutes.

The results showed that suplementation using Insulin Transferrin Selenium (ITS) and combination of Insulin Transferrin Selenium (ITS) and Heat Shock Protein 70 (HSP70) able to maintain oocyte morphology and suppress the expresion of caspase-3 after vitrification compared with controls. ITS suplementation shows the expression of caspase-3 the least.

Keywords: Oocyte, Vitrification, Insulin Transferrin Selenium, Heat Shock Protein, Caspase-3