

Selection of immature bovine oocytes using Brilliant Cresyl Blue enhances nuclear maturity after vitrification.

ABSTRACT

Beside cooling/warming rates and composition of vitrification solution, developmental stage of immature oocytes may also affect their vitrification outcome. The aim of the present study was to evaluate the selection effect of developmentally competent immature bovine oocytes by Brilliant Cresyl Blue (BCB) on maturity of oocytes after vitrification. Oocytes were obtained from slaughterhouse ovaries. Only oocytes with 4-5 layers of cumulus cells and homogenous cytoplasm were used. After exposure to BCB stain, immature oocytes were divided into colored (BCB+) and colorless (BCB-) cytoplasm groups. Immature oocytes were equilibrated in VS1 (7.5 Ethylene Glycol (EG)+7.5% DMSO) for 10-12 min and then exposed to VS2 (15% EG+ 15% DMSO+0.5M sucrose) for 1 min. Thereafter, oocytes were loaded on Cryotop and directly plunged into liquid nitrogen. After warming, oocytes were examined for presence of polar body and nuclear maturity. Higher number of oocytes in BCB+group extruded first polar body in comparison with other vitrified groups but not significantly ($p>0.05$). Compared to the BCB- oocytes, there was significantly lower percentage of degeneration for BCB+oocytes ($p<0.05$). Within vitrified groups, reaching to the MII stage was significantly higher in BCB+group (51.5%) compared with BCB and vitrified-control groups (27.9 and 40.3%, respectively). These results indicated that selection of potent immature bovine oocytes using brilliant cresyl blue improved the nuclear maturity of immature oocytes after vitrification. In addition, this selection can be a valuable tool to improve the vitrification outcome.

Keyword: Vitrification; Brilliant cresyl blue; Bovine; Immature oocytes.