P-24 Effect of Ethylene Glycol (EG) and 1,2-Propanediol (PROH) on Mouse and Human Embryos during Slow Freezing Protocol

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Objective: There were controversial reports on result of cryopreservation using the EG or PROH as a cryoprotectant. Recently, EG has been introduced in mammals as a cryoprotectant for freezing of oocytes and embryos because of low toxicity and rapid permeation into the cells. Therefore, this study was conducted to compare the effects of EG and PROH on cryopreservation of mouse and human embryos.

Design: In experiment 1, mouse intact 6, 8-cell embryos were collected from ICR mouse and divided into two groups: group 1; EG, $n = 347$, group 2; PROH, $n = 319$. In experiment 2, human cleavage embryos derived from fertilized eggs showing 3 pronuclei were obtained from patients undergoing IVF and divided into two groups: group 1; freezing with EG, $n = 106$, group 2; freezing with PROH, $n = 113$. In experiment 3, mouse intact 6, 8-cell embryos were collected from ICR mouse and divided into three groups: group 1; control of exposure time during freezing step, $n = 465$, group 2; control of exposure time during thawing step, $n = 331$, group 3; control of exposure time during freezing and thawing step, $n = 585$.

Materials and Methods: Mouse and human embryos were cryopreserved with 1.5 mol/l EG + 0.2 mol/l sucrose and 1.5 mol/l PROH + 0.2 mol/l sucrose using the slow freezing method. The embryos survived after thawing in all groups were cultured in Preimplantion-1 (P-1) medium with 10% SSS. Survival and development up to blastocyst of embryos was examined under a stereomicroscope after culture. Statistical analysis was carried out by use of a χ^2 test.

Results: In experiment 1, the rates of survival were 80.4% (279/347) in group 1 and 79.3% (253/319) in group 2. The developmental rates to blastocyst were 44.1% (123/279) in group 1 and 45.1% (114/253) in group 2, respectively. In experiment 2, the rates of survival were 96.2% (102/106) in group 1 and 96.2 (106/113) in group 2. In experiment 3, reducing the time of exposure in 1.5 mol/l EG with 0.5 mol/l sucrose dose not seem to influence the development of mouse embryo when using a slow freezing protocol.

Conclusion: Cryopreservation of mouse and human embryos at cleavage stage by using EG or PROH had no statistical difference in the survival rate and developmental rate to blastocyst. It might be preferable to expose the embryo to the cryoprotectant for short period to avoid its toxic effects because of a high permeation of EG. Further study is requested for correlation between the penetration time and the concentration of cryoprotectant.