초자화 동결액으로는 ethylene glycol 40% (v/v), Ficoll70 30% (molecular weight 70,000)와 0.5 M sucrose를 포함한 EFS40, 2.75 M의 DMSO와 propylene glycol, 1.0 M sucrose를 포함한 DPS, 그리고 5.5 M ethylene glycol, 1.0 M sucrose의 VS14를 사용하였다.

초급속 동결액은 3.5 M DMSO와 0.25 M sucrose를 포함한 용액을 사용하였다.

각 용액에 대한 독성실험 결과 생존율은 100%, 95.6%, 97.8%, 100% (EFS40, DPS, VS14 and ultrarapid freezing)였으며, 96시간 배양 결과 93.5% (no freezing), 95.6% (EFS40), 93.0% (DPS), 86.4% (VS14), 93.0% (ultrarapid freezing)의 부화율을 보였다.

동결-해빙 후 각 실험군의 생존율은 80.2% (slow freezing), 91.7% (EFS40), 0% (DPS), 69.5% (VS14), 91.8% (ultrarapid freezing)로 나타났다.

또한, 96시간 배양 결과 93.5% (no freezing), 84.1% (slow freezing), 93.9% (EFS40), 47.8% (VS14), 71.2% (ultrarapid freezing)의 부화율을 보여 제 2일째 생쥐 배아를 동결할 때 간편한 방법으로써 초고속 동결법이 유용하다는 것을 알 수 있었다.

P-9 The Cytoskeletal and Chromosomal Constitution of Vitrified Immature Mouse Oocytes

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The objective of this study was to confirm whether the vitrification method using EFS40 has detrimental effect for cytoskeleton or chromosome constitution of the immature mouse oocytes by indirect immunocytochemistry and chromosome analysis. Immature mouse oocytes were vitrified using EFS40 (40% ethylene glycol, 18% ficoll, 0.5 M sucrose) and in vitro maturation was induced during 16 hr after thawing and then matured oocytes which had extruded the first polar body were examined. The results obtained in this experiment were summarized as follows: in vitro survival and in vitro maturation rates of oocytes after vitrification and thawing were 90.3 and 64.7%, respectively, it was similar to those of the exposed group (86.7 and 69.2%, respectively). When the microtubule morphology and microfilament distribution in vitrified oocytes were examined, normal percentage of two cytoskeleton in the vitrified group (93.9 and 100.0%) was not significantly different from that in the control group (100.0 and 100.0%) and the exposed group (94.4 and 100.0%). In addition, the rate of oocytes containing a normal chromosome number in the vitrified group was 65.8%, this result was not significantly different from that in the control group (79.6%) and the exposed group (69.0%). These results indicated that exposure to cryoprotectant or freezing has not effect on the alteration of cytoskeleton morphology and the chromosome constitution of mouse oocytes and that vitrification method using EFS40 was suitable for cryopreservation of immature mouse oocytes.