6 Developmental Capacity of Bovine Follicular Oocytes after Ultra-Rapid Freezing by Electron Microscope Grid

II. Cryopreservation of *In Vitro* Matured Bovine Oocytes

Eun Young Kim, Nam Hyung Kim*, Myo Kyung Kim, Bong Kyung Yi, Hyeon Sook Lee, San Hyun Yoon, Sepill Park, Kil Saeng Chung* and Jin Ho Lim****

*Maria Infertility Medical Institute (마리아기초의학연구소), *College of Animal Husbandry, Kon-Kuk University, **Maria OB/GYN, Seoul*

Mature bovine oocytes were frozen ultra-rapidly with the electron microscope grid and cryoprotectant solution consisting of 30% ethylene glycol, 0.5M sucrose, 18% ficoll and 10% FBS added D-PBS (EFS30) or exposed to the freezing solution. Assessments of morphological survival, fertilization and embryonic development were recorded. Also, cytoskeletal alteration of bovine oocytes according to exposure to the freezing solution or after freezing/thawing was examined. The results obtained in this experiment were summarized as follows; After ultra-rapid freezing and thawing, a high mean percentage of oocytes (67.4%) was survived. When fertilization was identified at 18 h after insemination, the fertilization rate in control and freezing group was evaluated as total penetration (96.7, 90.0%), normal 2 pronuclei formation (74.6, 68.9%) and mean number of sperm/oocyte (1.50, 1.44). The rate of two-cell formation to freezing group was 67.2%, and was not significant different when compared to control (71.4%)and exposure (78.6%). In addition, development to the blastocyst on day 8 after fertilization of freezing group was 32.8% from the cleaved oocytes. Development to the blastocysts of frozen-thawed oocytes was very similar to that of the control (33.3%) and exposure (34.8%). These results demonstrate that developmental capacity of frozen-thawed bovine oocytes can be successfully obtained by ultra-rapid freezing method using electron microscope grid and EFS30.

7 Microtubule and chromatin organization during the first cell cycle following intracytoplasmic injection of round spermatid into porcine oocytes

Jang Won Lee, Nam-Hyung Kim, Hoon Taek Lee and Kil Saeng Chung

Animal Resources Research Center, Kon-Kuk University, Seoul 143-701, Korea

Assessment of normal fertilization and offspring production by injecting spermatid into oocytes could offer exciting prospects not only for understanding gamete physiology during fertilization but for the clinical treatment of male infertility due to defective spermiogenesis. However, little information is available on the role of paternal origin centrosome during spermatid fertilization. The objective of this study was to determine microtubule assembly and chromatin configuration in porcine oocytes during the first cell cycle following round spermatid injection into matured