P-1 In Vitro Development and Remodeling of Porcine Embryos Following Nuclear Transfer using Porcine and Mouse Fibroblasts

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The objective of this study was that the transfer of nuclei from on cell to another provided a powerful tool for studying the interactions between the cytoplasm of one cell and the nucleus of another. Thus, we determined the developmental capability of porcine fibroblast (PF; from a 35day-old male fetus) and mouse fibroblast (MF; from a 3 week-old male) after transferring into enucleated porcine oocytes. Nuclear transfer of PF and MF into the enucleated porcine oocytes was accomplished by membrane to membrane method. Reconstructed porcine eggs were cultured in 50 µl of NCSU 23 containing 0.5% BSA for 4 days and then cultured in 50 µl of NCSU 23 containing 10% fetal bovine serum for 3 days. Nuclei of porcine eggs nuclear transferred by PF and MF were developed to the mitotic division and 2 cell stages at 24 hr. Porcine 2-cell embryo reconstructed by MF showed the normal 20XY by chromosome analysis. However, the development capacity of porcine embryos reconstucted by donor cells from two different species showed significantly difference (p<0.05) between PFs and MFs. The developmental rate to the morula to hatching blastocyst stages (37.4% of cleaved embryos) of reconstructed eggs using PFs was significantly higher than that of reconstructed eggs using MFs (7.5% of cleaved embryos). Therefore, our finding suggests that mechanisms regulating early embryonic development may be conserved among mammalian species. In addition, porcine oocyte cytoplasm can successfully reprogram the somatic nuclei of different species and develop up to the blastocyst stage in our culture system.

P-2 Integration of EPO Gene in Bovine Embryos Following Nuclear Transfer using Bovine Fetal Fibroblasts Transfected by Retrovirus Vector

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The present study demonstrated the successful integration of erythropoietin (EPO) and neyomycin resistant (Neo^R) genes in bovine embryos following nuclear transfer using transfected bovine fetal fibroblasts with these genes by retrovirus-mediated infection. First, transfected bovine fetal fibroblasts were selected by G418 of 800 µg/ml. Then matured bovine oocytes were enucleated by aspirating the first polar body and adjacent cytoplasm (approximately 30% of ooplasm)