embryos which developed in vitro in the NCSU 23 medium were cultured in the BSA free NCSU 23 medium supplemented with 20% fetal calf serum, the incidence of hatching or hatched out was significantly increased as compared to the control groups. However, addition of amino acids, vitamins or insulin to the NCSU 23 medium did not enhance the early morula developing to the hatched embryos. When either in vivo or IVM/IVF derived 1- to 2- cell stage embryos were cultured 4 days in the NCSU 23 and an additional 4 days in the NCSU 23 supplemented in the fetal calf serum, the percentages of hatched blastocysts were significantly higher than the control groups. These results suggested that dual culture conditions are required to optimize in vitro culture system for the development of the porcine embryos in vitro.

## P-15

## Development of Porcine IVM/IVF Produced Embryos to the Hatching Blastocyst In Vitro as Affected by Amino Acids and Fetal Bovine Serum

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The objective of this study was to test the effect of the amino acids and fetal bovine serum (FBS) to the blastocyst and hatching developments of 2-4 cell embryos produced by porcine in vitro matured-fertilized oocytes. In experiment 1, the development of 2-cell embryos on NCSU medium (0.4% BSA) at 36 hr after in vitro fertilization (IVF) examined according to the time course. The formation of blastocoel

revealed at 96 hr after culture of 2-cell embryos. In experiment 2, the zona thickness (16.3  $\pm$  2.34,  $11.1 \pm 2.89$ ,  $5.2 \pm 2.61$ ), embryo size (155.8  $\pm$ 7.0, 174.9  $\pm$  10.5, 235.7  $\pm$  21.3) and cell number  $(13.1 \pm 3.44, 22.6 \pm 6.37, 57.4 \pm 10.60)$ investigated at various blastocysts (early, middle, expand) at 7 day after IVF. In experiment 3, the blastocyst and hatching development of 2-4 cell embryos at 50 hr after IVF tested on NCSU medium supplemented with the amino acids (2% BME amino acids and 1% MEM non-essential amino acids), FBS (10%) and BSA (0.4%). The embryos cultured in NCSU medium developed to blastocyst when added the amino acids, FBS or BSA. Especially, the hatching development of obtained on the NCSU medium embryos containing the amino acids or FBS. However, the morula and blastocyst of embyos cultured for 3 days on NCSU (0.4% BSA) could be develop to the hatching stage as transfer on NCSU (10% FBS). Therefore, these experiments suggest that the hatching development of porcine in vitro matured-fertilized embryos affects by amino acids and FBS.

## P-16

## Effects of Epidermal Growth Factor (EGF) on Mouse IVF Embryo Development and Their Cell Number

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Control of growth and differentiation during mammalian embryogenesis may be regulated by growth factors. The objective of this study was to determine the effect of EGF on the preimplantation development of mouse IVF embryos and their ICM and TE cell number, and to examine the expression of EGF-R protein on