

91 Effects of high osmotic sucrose medium on embryo growth and on embryo glucose metabolism. T. Saito, H. Saito, M. Hiroi, Dept. Obst. and Gynec., Yamagata Univ. Sch. Med., Yamagata.

[purpose] We examined the effects of high osmotic sucrose medium on embryo glucose metabolism by the enzymic cycling method. [method] Two cell embryos were put into HTF medium containing 0.5mol sucrose for 60 minutes. Then embryos cultured in HTF medium for 40 hours. Treated and non-treated embryos were employed to measure the content and up-take of glucose and the enzymic activity of hexokinase (HK), glucose 6 phosphate dehydrogenase (G6PDH), lactate dehydrogenase (LDH), malate dehydrogenase (MDH). [result] HK, G6PDH, LDH, MDH and Glucose of 2 cell embryos showed no significant difference between control and sucrose treated group. At 40 hours culture, HK and glucose of the control was higher than those of the sucrose group. (HK: 8.0 ± 0.3 VS $6.0 \pm 0.8 \times 10^{-12}$ mol/embryo/hr) (glucose: 1011 ± 53 VS $860 \pm 42 \times 10^{-15}$ mol/embryo) G6PDH and LDH of the control was lower than those of sucrose group. (G6PDH: 3.5 ± 0.2 VS $5.3 \pm 1.3 \times 10^{-12}$ mol/embryo/min) (LDH: 61.2 ± 3.0 VS $75.3 \pm 4.8 \times 10^{-12}$ mol/embryo/min) MDH showed no difference. [conclusion] The enzymes of glucose metabolism in embryos have no damages immediately after sucrose treatment. But 40 hours after sucrose exposure, the enzymatic development of glucose metabolism delayed.

92 Analysis of the uterine environment on mouse embryo development. Y. Goto, Y. Noda, M. Shiotani, T. Nonogaki, Y. Umaoka, K. Narimoto, T. Mori, Dept. Gynec. and Obst., Kyoto Univ. Fac. Med., Kyoto.

Our previous studies have shown that both co-culture with uterine epithelia and addition of uterine fluids stimulated the mouse embryo development. In this study, the effects of uterine fluids from various stages of the oestrous cycle were studied in vitro. Moreover, morulae were transplanted by direct insertion into the pseudopregnant mice, and were recovered 24 hours later. The promoting effect of uterine fluids did not differ by the stage of the cycle. The inhibitory effect was not observed throughout the cycle. When morulae were transplanted into pseudopregnant Day 3 recipient uteri, 66.3% (59/89) of the embryos were recovered and 49 (83.1%) embryos developed to blastocysts. When transplanted into pseudopregnant Day 2, only 1.4% (1/70) of the embryos were recovered and no embryo developed to blastocyst. However, when transplanted into pseudopregnant Day 2 after the ligation of the lower portion of uteri, blastocysts could be recovered (10/26). These results suggest that the uterus may provide the beneficial effect for embryo development throughout the oestrous cycle, and that the excretion of embryos from uteri may cause the low implantation rate in the inappropriate embryo transplantation.

93 Blastocyst formation in vitro of hamster embryos cultured from single cell stage. Y. Umaoka, Y. Noda, S. Natsuyama, T. Mori, Dept. Obst. and Gynec., Faculty of Medicine, Kyoto Univ., Kyoto.

We analyzed the environmental variables influencing the development of hamster embryos in vitro. Female golden hamsters (8-10 weeks old) were injected intraperitoneally PMSG (15 units) to induce superovulation, and housed with males of the same strain 72 hours after PMSG administration. Single cell stage embryos were collected from mated females. The developmental efficacy was evaluated by the rate of eight-cells and blastocysts. Environmental variables examined in this study were (1) culture medium (mTALP or HECM-1) (2) length of time required for embryo collection (5, 10, 15 or 20 minutes) (3) oxygen tension (5% or 20%) (4) supplementation of SOD or (5) thioredoxin to the medium. Better culture efficiency was obtained (1) in HECM-1, (2) with shorter elapsed time for embryo collection (5 minutes), (3) under 5% O₂ (4) with supplementation of SOD or (5) thioredoxin to the medium. Under optimized conditions which satisfy the results above, we have succeeded in developing single cell stage embryos to blastocysts in vitro (20.8%). These results suggest that a change in energy metabolism such as glucose metabolism, not only oxygen radicals, is closely involved in the in vitro developmental blockage of hamster embryos.