cellular and transepithelial water movement. The luminal fluid microenvironment of the uterus is important for sperm capaciation and preimplantation embryo development. Steroids regulate water imbibition in the uterine endometrium. The aim of this study was to determine the expression of AQPs mRNA gene and localization of protein regulated by estrogen and progesterone in mouse uterus.

Method: Ovariectomized (OVX) ICR mice were treated with injection of 17β-estradiol (E2, 0.3 μg/mouse) and progesterone (P4, 1 μg/mouse). Mice were killed 6, 12, and 24hr after 17β-estradiol or progesterone injection by cervical dislocation. Another group of mice was given progesterone following 17β-estradiol treatment for 24 hr, and was killed 6, 12, and 24 hr after progesterone injection. The levels of AQPs mRNA were examined by RT-PCR, and laser capture microdissection (LCM). To determine whether these mRNAs were translated, cellular distribution of these proteins were investigated by immunohistochemistry.

Results: AQP 4, 5, and 8 were highly expressed in E2 or E2 + P4 treated group than P4 treated group. Immunohistochemical results showed that AQP 4, 5 and 8 protein were expressed in the myometrium, luminal and grandular epithelial cell. AQP 4, 5, and 8 protein were highly expressed in the luminal epithelial cell in E2 or E2 + P4 treated group than P4 treated group.

Conclusions: These results suggest that the expression of AQP 4, 5, and 8 is up-regulated by estrogen. Thus AQP 4, 5, and 8 may regulate water imbibition and luminal fluid production in the mouse uterus by estrogen-dependent manner.

P-40 Effects on in vitro Development of Mouse Preimplantation Embryos by Co-culture System with vero Cells Monolayer in Media with Different Composition of Glucose and Pyruvate

JH Kim[1], C Won[2], HB Song[3], JG Rhee[1], KH Jung[1], BH Kang[1], KC Kang[1], KH Lee[1]

Department of Obstetrics & Gynecology, Chungnam National University Hospital[1], Moa Obstetrics & Gynecology Clinic[2], Department of Life Resources, Taegu University[3]

Background & Objective: The purpose of this study was conducted to examine the effects on in vitro development of early preimplantation mouse embryos by co-culture system with vero cells monolayer in culture media with different composition of glucose and pyruvate.

Methods: Two-cell embryos were collected from 4~5 weeks old ICR mice. A total 382 embryos were co-cultured with vero cells monolayer in four different media that were manufactured by mixture ratio using DMEM with (DMEM-GGP) or without (DMEM-G) glucose and pyruvate. In control group, DMEM-G medium which is currently using for human embryo culture in our infertility clinic was used. Group I was cultured in medium which was mixed three volume of DMEM-G and one volume of DMEM-GGP, and group II was cultured in medium which was mixed same volume of DMEM-G and DMEM-GGP, and group III was cultured in DMEM-GGP. All media were supplemented with 20% hFF. Results between different groups were analyzed using a Chi-square test, and considered statistically significant when p value

was less than 0.05.

Result: After 24 hrs, the developmental rate into ≥ 3-cell was significantly higher ($p<0.05$) in group I (97.0%) than in group II (88.8%) and III (86.6%). The developmental rate into ≥ 8-cell in group I (44.0%) was significantly ($p<0.05$) higher than in group III (26.8%), however, group I was no significant differences compared with control (32.2%) or group II (31.6%). At 48 hrs, the developmental rate into ≥ morula was significantly higher in group I (89.0%) than in control (74.7%) and group III (71.1%). However, in developmental rate into blastocyst, control (14.9%) was shown to the significant differences compare with group III (4.1%). At 72 hrs, the developmental rate into ≥ expanded blastocyst was significantly higher in group III (15.5%) than in control (5.7%), and developmental rate in total blastocyst was no significant differences between all experimental groups, but group I (55.0%) was highest, and group II was lowest (42.9%). At 96 hrs, the developmental rate into ≥ hatching blastocyst was significantly higher in group II (30.6%) and III (33.0%) than in control (17.2%), but the developmental rate in total blastocyst was no significant differences between all experimental groups involved control.

Conclusions: Development of early preimplantation mouse embryos during 48 hrs in culture was effective in group I, however, development into ≥ expanded blastocyst at 72 hrs and into ≥ hatching blastocyst at 96 hrs were more effective in group III. These results were likely that it was caused by consumption on glucose and pyruvate of vero cells in culture media. Therefore, energy sources have to be supported in culture media by culture time or cleavage stages not only in conventional culture systems, but also in co-culture systems.

P-41 Insulin, Insulin-like Growth Factor-I과 Tumor Necrosis Factor Alpha 상호작용에 의한 생쥐 초기배아에서 발생과 사멸 조절 및 신호전달

한양대학교 생명과학과, 울산의대 산부인과[1]

한현주 · 강병문[1] · 계명찬

Background & Objectives: 당뇨성 모체의 자궁환경은 배아의 발달 저해 및 손상을 유발하며 tumor necrosis factor alpha (TNF-α)는 배아의 손상과 밀접히 관련되어 있다. 수정으로부터 착상까지 초기 배아 발달과정은 배아자체적인 발생 프로그램에 의해 지배되지만 배아 주변의 다양한 peptide 호르몬, 성장인자 및 사이토카인들은 배아 발생 프로그램의 정교한 조절을 담당한다. 특히 당뇨성 모체환경하에서 배아의 손상 기작을 이해하기 위해서는 이들에 의한 배아 내 신호전달 수준의 조절 기작을 이해하는 것이 중요하다. 본 연구는 insulin, insulin-like growth factor-I (IGF-I), TNF-α에 의한 초기 배아 형태적 발생 및 사멸의 조절기작을 규명하고자 이들 ligand 수용체 하위에서 mitogen activated protein kinase (MAPK) cascade 수준의 신호 교류 및 MAPK 활성의 변화를 조사하였다.

Method: Insulin, insulin-like growth factor-I (IGF-I), TNF-α에 의한 초기 배아 형태적 발생 및 사멸의 변화를 확인하고 그 조절기작을 규명하고자 이들 ligand 수용체 하위에서 mitogen activated protein kinase (MAPK) cascade 수준의 신호 교류 및 MAPK 활성의 변화를 조사하였다. 생쥐의 상실배 및 포배