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Differential Expression of Aquaporins During Murine Preimplantation DevelopmentHyeonsang Shin^P, Ji Won Lee¹, Su Man Kang¹, Sung Eun Lee¹, Han Seung Kang¹, Myung Chan Gye¹, Jin Hyun Jun², Moon Kyoo Kim^C*^{PC1}Department of Life Science, College of Natural Sciences, Hanyang University, Seoul 133-791; ²Laboratory of Reproductive Biology and Infertility, Sungkyunkwan University, School of Medicine, Seoul 110-745*

Aquaporins (AQPs) are transmembrane channel proteins, and their function as molecular water channel allows water to flow across plasma membranes in the direction of osmotic gradients. Blastocyst formation is mediated by a trans-trophectoderm ion gradient(s) established, in part, by Na⁺/K⁺-ATPase, which drives the movement of water through AQPs across the epithelium into the extracellular space of the blastocyst to form the fluid-filled blastocoel. The purpose of this study was to evaluate the temporal pattern of the expression of AQP-8 and -9 during preimplantation development of mouse embryos. AQP-8 mRNAs were not present in early cleavage stages but were detected in morula and blastocyst stage embryos. On the contrary, AQP-9 mRNAs were expressed in mouse oocytes and embryos up to the blastocyst stage. Real time RT-PCR revealed 2-fold increase during blastocyst development, suggesting these AQPs support expansion of blastocyst. Now we are investigating the effect of cAMP agonist on the expression of AQP-8 and -9 in the blastocysts. In the confocal microscopy, AQP-8 and -9 were localized in membranes of the trophectoderm, suggesting that AQPs may regulate the trophectoderm fluid transport during blastocyst formation.

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Effect of Steroid Hormones on Aquaporin Gene Expression in the Ovariectomized Mouse UterusSu Man Kang^P, Hyeonsang Shin¹, Ji Won Lee¹, Sung Eun Lee¹, Han Seung Kang¹, Myung Chan Gye¹, Moon Kyoo Kim^C*Department of Life Science, College of Natural Sciences, Hanyang University, Seoul 133-791*

The aquaporins(AQPs) are a family of homologous water channels expressed in many epithelial and endothelial cells involved in fluid transport. To date, 11 AQPs, numbered 0-10, have been cloned in mammals. Estrogen and progesterone are steroids that play important roles in the regulation of mammalian reproduction. One primary action of these hormones is to regulate the development and function of the uterus. The luminal fluid microenvironment of the uterus is important for sperm capacitation and preimplantation embryo development. So, the aim of this study was to determine the expression of aqp genes regulated by these steroids in ovariectomized(OVX) mouse uterus. To evaluate the effects of estrogen and progesterone on aqp genes expression, adult virgin OVX females were allowed to recover (2wks) and treated with 17 estradiol(300ng/head, sc), progesterone (1ug/head, sc), or a combination of these. One group of OVX females received 17 estradiol was killed 6, 12, and 24hr later. A second group of mice given progesterone was killed 6, 12, and 24hr later. A third group of mice was given progesterone following 17 estradiol treatment for 24hr, and was killed 6, 12, and 24hr after progesterone injection. The levels of aqp mRNA were analyzed by RT-PCR and real-time PCR. The aqp4, 5, and 8 mRNA were up-regulated in the OVX uterus after 17 estradiol injection. Progesterone, however, did not alter aqp genes expression. The present results suggest that estrogen up-regulates the expression of aqp genes in the uterus during preimplantation period and thus regulate water imbibition and luminal fluid production in the mice.

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Changes in Aquaporin Gene Expression During Estrous Cycle in Mouse UterusJi Won Lee^P, Hyeonsang Shin¹, Su Man Lee¹, Sung Eun Lee¹, Han Seoung Kang¹, Myung Chan Gye¹, Moon Kyoo Kim^C*Department of Life Science, College of Natural Sciences, Hanyang University, Seoul 133-791*

Mouse estrous cycle divided into four phases consisting the diestrus, proestrus, estrus and metestrus. Estrus period is marked by edematous changes in the uterus in response to hormonal stimuli. In mice, increased uterine vascular permeability occurs in response to estrogen and certain vasoactive mediators, but the mechanism regulating the fluid transport during estrous cycle has been not fully understood. In mammals 11 aquaporins (AQPs) which facilitate bulk water transport have been identified. To verify possible involvement of AQPs in the edematous change in uterus, we examined expression of AQP1, 3, 4, 5, 8, 9 in mouse uterus at different stages of estrous cycles. Results show that AQP1 and 3 showed constitutive expression throughout the estrous cycle, AQP4 is highly expressed at estrus stage, AQP5 expression is peaked at proestrus but gradually decreased until diestrus, AQP8 expression was higher during proestrus and estrus stage than other stages, and AQP9 expression increased through diestrus to metestrus. These results suggest that AQP 4, 5, 8, is highly expressed during follicular phase. Taken together, these results suggest that expression of aquaporins in mouse uterus might be regulated by estrogen.

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Aquaporin Gene Expression in Mouse OvarySung Eun Lee^P, Hyeonsang Shin¹, Ji Won Lee¹, Su Man Kang¹, Myung Chan Gye¹, Han Seung Kang¹, Moon Kyoo Kim^C*Department of Life Science, College of Natural Science, Hanyang University, Seoul 133-791*

Follicle development has two major steps in mammalian ovary. The initial stage of folliculogenesis occurs independently of gonadotrophic hormones. But antrum development is dependent on follicle stimulating hormones(FSH). In response to FSH, the antrum formation is dramatically accelerated, and which requires rapid and massive transport of water. Aquaporins (AQPs) support a large volume of water flow through transcellular pathways. To date, 11 AQPs have been identified in mammals. However, little is known about AQP expression during folliculogenesis. To investigate the possible regulation of AQPs expression in the ovarian follicles by gonadotropin injected mice, we examined the effect of PMSG and hCG on the expression of AQPs in the ovary of immature female mice. Aquaglyceroporins (AQP 3,7,8) increased following stimulation by PMSG and hCG until the ovulation and luteal development. After ovulation, AQPs expression decreased below the basal level. Taken together, this suggests that AQP3,7,8 are important for fluid accumulation during antral development mouse ovary.