Id-1 gene was higher expressed than other genes in the OVX/estrogen treatment/6-h/12-h protocols among Id genes. To investigate the differential expression pattern of Id-1 gene, we performed LCM and then identified that Id-1 mRNA was only expressed in the epithelium of the OVX/estrogen treatment/6-h/12-h protocols. Also, Id-1 protein was predominantly localized in luminal and glandular epithelium of the OVX/estrogen treatment/6-h/12-h protocols. Interestingly, the expression of Id-1 mRNA was shown to upregulate at the implantation sites compared to interimplantation sites.

Conclusions: These results revealed that Id-1 was strongly regulated by estrogen in the mouse uterus. Particularly, we suggest that Id-1 may be possessed unidentified function in epithelium of the mouse uterus. The observations presented in the current study suggest that Id-1 gene may have a distinct function in the uterine physiological events, such as the implantation process and the estrus cycle. However, we do not know the molecular mechanisms which regulate the expression of Id-1 genes between the uterus and blastocysts and how steroid hormone might regulate the expression of Id-1 is not clear.

O-18 Ceramidase Inhibition Potentiates Stress-induced Apoptosis of Murine Ovarian Granulosa Cells

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Background & Objectives: (1) To evaluate the effect of ceramide versus ceramine in regulating murine granulosa cell death (2) To examine the mRNA expression of ceramidase in granulosa cells and the effect of inhibition of ceramidase in stress-induced granulosa cell death

Method: In Vivo: Ovaries were collected from C57BL/6 (22~24 days postpartum) female mice prior to and 42 hours after in vivo stimulation with gonadotropin hormone (PMSG; 5 IU) to promote follicle and granulosa cell survival. In addition, corpora lutea were collected from gonadotropin-stimulated (PMSG + hCG) ovaries 84 hours after the start of treatment (approximately 30 hours after ovulation). Northern blot hybridization and in situ hybridization were used to evaluate mRNA expression of acid ceramidase. In Vitro: Female C57BL/6 mice (22~24 days postpartum) were treated with PMSG (5 IU, n=3). Ovaries were collected 42 hours later and healthy antral follicles were punctured to extrude granulosa cells. Granulosa cells were pre-cultured in Waymouth medium with 10% fetal bovine serum for 24 hours. Media from all cultures were replaced with media lacking serum and cultures were continued for 6~24 hours with ceramidase inhibitors; oleylethanolamide (OE) or (1S, 2R)-D-erythro-2-(N-myristoylamino)-1-phenyl-1-propanol (D-MAPP) with ceramide or serum starvation. Cells were fixed with 4% paraformaldehyde and stained with Hoescht for examination of nuclear morphology (apoptosis). Diacylglycerol kinase assay was used to measure ceramide levels.

Results: After 6 hours, C8-ceramide (50 mM) triggered apoptosis in only 28 6% of the cells, whereas C8-ceramine (50 mM) induced apoptosis in all cells (LD_{50} = 1 mM). These data, which suggest that cera-

midase activity is a critical determinant of GC survival, were supported by studies with the ceramidase inhibitors, Both OE and D-MAPP significantly potentiated GC apoptosis induced by either serum starvation or by 50 mM C8-ceramide treatment. By comparison, L-MAPP (an inactive stereoisomer of D-MAPP) had no effect on cell death induced by either serum starvation or C8-ceramide. Acid ceramidase was expressed abundantly in granulosa cells and ovaries and it's expression was significantly increased by gonodotropin in granulosa cells. OE significantly increased ceramide production ($p<0.05$), however, D-MAPP did not increased ceramide production from granulosa cells after 4 hours treatment.

Conclusions: 1) C8-ceramine induced apoptosis in all cells at 50 mM concentration while the same concentration of C8-ceramide caused only 28.6% of cell death. 2) Acid ceramidase was expressed abundantly in granulosa cells and ovaries and its expression was increased by PMSG and, decreased in corpora lutea. 3) Ceramidase inhibition significantly potentiated GC apoptosis induced by C8-ceramide or serum starvation. 4) Ceramide metabolism is a critical determinant of gaunulosa cell fate.

O-19 Expression of Sperm-specific Cation Channel CatSper in Human Spermatozoa

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Background & Objectives: Numerous studies have demonstrated that calcium ion controls sperm motility and capacitation. CatSper, a newly identified cation channel is reported to exist in flagellum of mouse spermatozoa. CatSper is a sperm-specific calcium channel and plays a key role in the sperm motility and the fertility in mouse. In this study, we aimed to elucidate the expression and intracellular localization of CatSper in human spermatozoa.

Method: The sperm samples were isolated from semens of 30 patients. After the CASA, the viability test and the morphological test, samples were divided into two groups (normozoospermia and asthenozoospermia). Using this sperm sample, we performed RT-PCR for mRNA expression and fluorescent immunocytochemistry for protein expression.

Results: In all of the sperm samples, we could found the mRNA expression of CatSper. We compared the expression of CatSper mRNA with sperm motility and progressiveness by semi-quantitative analysis. In comparison of CatSper mRNA expression and sperm motility (not distinguishing dead sperm from whole sperm population), the mean value of CatSper mRNA expression (mean±SD) was 1.5±0.6 in normozoospermia (n=15) and 1.4±0.6 in asthenozoospermia (n=15). When we compared the expression of CatSper mRNA with motility/viability ratio for excluding non-motile dead sperm (group A; >0.5, group B; ≤0.5), group A (n=19) was 1.5±0.6 whereas group B (n=11) was 1.2±0.6. The mean value of group A was 25% higher than that of group B but it was not significantly different. In comparison of CatSper