

Effect of Sphingosine-1-Phosphate on *In Vitro* Maturation of Porcine Oocytes

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Sphingosine-1-phosphate (S1P) is one of the sphingolipid metabolites which affect a variety of cellular processes including the proliferation, differentiation, growth, survival, migration and gene expression. The present study was undertaken to investigate the effect of S1P on nuclear maturation of porcine oocytes. *In vitro* maturation frequency of porcine oocytes were compared in three different media; group I: NCSU23+0.1% PVA, group II: NCSU23+ 10% PFF(porcine follicular fluid), and group III: NCSU23+10% PFF+10 ng/ml EGF+2.5 mM β -mercaptoethanol. Each group containing 0.1 mg/ml cysteine was divided into 4 sub-groups of S1P concentration (0, 50, 500 and 5000 nM). Porcine oocytes were incubated in each maturation medium supplemented with hormones (10 IU/ml PMSG and 10 IU/ml hCG) for 22h and then further cultured in the same medium without the hormones for 22h. After completion of *in vitro* maturation, the oocytes were fixed and stained to examine nuclear maturation by using a rapid stain method. In the group I, the proportions of metaphase II stage among oocytes cultured in 0 nM(control), 50 nM, 500 nM and 5000 nM S1P were 45.4%, 66.7%, 56.6% and 48.7%, respectively. Thus, 50 nM S1P group showed a higher maturation rate than control group($P<0.05$). In group II, maturation rate of porcine oocytes was also higher in 50 nM S1P group compared to control group(83.5% vs 62.7%, $P<0.05$). However, in group III effect of S1P on the nuclear maturation of porcine oocytes was not observed. When sphingosine kinase inhibitor(N,N-dimethylsphingosine), which blocks generation of intracellular S1P, treated at a concentration of 10 nM, 50 nM and 100 nM, maturation rate of the oocytes was 25.0%, 21.8% and 9.8%, respectively, showing a lower maturation rate than control (53.3%, $P<0.05$). These results suggest that S1P plays an important role in the nuclear maturation of porcine oocytes.

Key words) *Sphingosine-1-phosphate, Porcine oocyte, S1P inhibitor, Nuclear maturation.*