

whereas that is 0.6 mM in the mature oocyte. These results demonstrate that a role of intercellular association between granulosa cell and oocyte with microvilli during oocyte maturation.

**D103**

**Involvement of PI3-kinase in the Progesterone-Induced Oocyte Maturation in *Rana dybowskii*.**

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Previously, we have shown that the p70<sup>s6k</sup> play an essential role during the early phase of oocyte maturation in *Rana dybowskii*. To investigate further the early signal transduction components involved in this process, the possible role of phosphatidylinositol-3 kinase (PI3 kinase) during oocyte maturation was examined. Progesterone-induced oocyte maturation was significantly inhibited by wortmannin and LY294002, specific inhibitors of PI3 kinase. Protein synthesis was also significantly suppressed by wortmannin treatment during oocyte maturation. Moreover, PI3 kinase inhibitor suppressed progesterone induced phosphorylation of S6 kinase in a dose dependent manner. Likewise, PI3 kinase inhibitors significantly inhibit the phosphorylation of mitogen activated protein (MAP) kinase which was increased during oocyte maturation. Finally, progesterone induced H1 kinase activity was also inhibited by PI3 kinase inhibitors in a dose dependent manner. Taken together, these results suggest that PI3 kinase play as an initial component of signal transduction pathway which is resided in the upstream of p70<sup>s6k</sup>, MAP kinase, and MPF production during progesterone mediated amphibian oocyte maturation.

**D104**

**Effects of Endocrine Disrupters, Tributyltin and Triphenyltin on Progesterone-Induced Oocyte Maturation in *Rana dybowskii*.**

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The purpose of the present study is to investigate the effect of endocrine disrupters on oocyte maturation in amphibian. We hypothesized that the organotin compounds, tri-n-butyltin (TBT; anti-fouling agent) and tri-phenyltin (TPT), may disrupt progesterone-induced oocyte maturation in Korean brown frog *Rana dybowskii*. Exposure of TBT or TPT was sufficient to inhibit progesterone-induced oocyte maturation in Rana. This inhibitory activity was specific to TBT and TPT; other organotin compounds (Mono-butyltin; MBT, Di-butyltin; DBT) did not show any specific effect on oocyte maturation. When TBT was pretreated for 1hr before P4 treatment, this inhibitory effect of TBT was reached maximum level. The progesterone-induced protein synthesis was significantly decreased by TBT and TPT. Moreover, TBT and TPT also inhibited the activation of MAPK during P4-induced oocyte maturation. However, the inhibition of TBT to oocyte maturation was not observed during spontaneous maturation. We propose that TBT and TPT could inhibit P4-induced oocyte maturation through a step involving inhibition of protein synthesis and MAPK signal pathway.

**D105**

**Gametogenesis and Reproductive Cycle of the Turban Shell, *Lunella coronata coreensis* (Gastropoda:**

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Gonadal development, gametogenesis, reproductive cycle, gonad index, flesh weight rate, and first sexual maturity of the turban shell, *Lunella coronata coreensis* were investigated by histological observation. The materials used were collected monthly from the rocky intertidal zone of Daehang-ri, Buan-gun, Jeollabuk-do, on the west coast of Korea, from July 1998 to June 1999. Sexes of *L. coronata coreensis* were separate. The gonad was widely located in the spirals of the visceral mass buried in the digestive gland. The ovary and testis were composed of a number of oogenic lobules and spermatogenic tubules, respectively. Monthly variations in the gonad index increased from March ( $23.86 \pm 3.73$ ) when the water temperature increased and reached the maximum in July ( $49.76 \pm 6.47$ ). And then, the gonad index sharply decreased in September ( $15.58 \pm 2.33$ ). The flesh weight rate ranged from 25.2% to 32.1%, and its variation showed a similar pattern to the gonad index. Individuals of 5.9 mm and less in shell height could not take part in reproduction in both sexes. Percentages of first sexual maturity of female and male specimens ranging from 7.0~7.9 mm in shell heights were 84.6% and 91.7%, respectively, and 100% in those over 8.0 mm in shell height in both sexes took part in reproduction. By studying the monthly changes of the morphological features and sizes of germ cells during gametogenesis in the gonad, the reproductive cycle of this species could be divided into five successive stages: early active (December to April), late active (January to July), ripe (May to August), spawning (July to September), and recovery (September to March). The spawning period of this species was once a year between July and September, and the

main spawning occurred in July when the seawater temperature reached above 24.8°C. The fully ripe eggs were 150~160 μm in diameter.

**D106****Multiple Signaling Pathways of Bullfrog GnRH Receptors****Da Young Oh<sup>1</sup>, Li Wang, O Im Yun, Hyuk Bang Kwon and Jae Young Seong**

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It has been known that the agonist-bound gonadotropin-releasing hormone receptor (GnRHR) engages several distinct signaling pathways including activation of phospholipase C (PLC), followed by the activation of protein kinase C (PKC), and elevation of intracellular  $Ca^{2+}$  levels. However, it has been questioned whether the GnRHR activation involves cAMP-mediated PKA activation, extracellular signal-related kinase (ERK), and Jun N-terminal kinase (JNK) activation. In the present study we demonstrated bullfrog (bf) GnRHR-mediated multiple signaling cascades, indicating that at least both PKA and PKC pathways play equally important roles in GnRHR-mediated signaling. The activation of bfGnRHRs triggered both inositol phosphate production and cAMP response element (CRE)-mediated luciferase activity in a dose-dependent manner. Treatment with GF109203X, a PKC inhibitor, U73122, a PLC inhibitor, or EGTA an extracellular calcium chelator could partially inhibit the GnRHR-mediated CRE activity, indicating the potential cross-talk between PKA and PKC pathway. Furthermore, we examined the involvement of ERK or JNK signaling by cotransfection of GAL4 response element driving luciferase construct (GAL-Luc) with GAL4-Jun or GAL4-Fos construct. Taken together, this study demonstrates that