## D105 The Binding of Progesterone to the Plasma Membrane of *Xenopus* Oocytes: Characteristics of Binding and Hormonal and Developmental Control.

Jung Won Ju*, Wook-Bin Im, Hyuk-Bang Kwon and Hueng-Sik Choi
Hormone Research Center, Department of Biology, Chonnam National University

Progesterone induces maturation of the amphibian oocyte through its action on the plasma membrane. Action of progesterone was transduced through the unidentified progesterone membrane receptor on the surface of oocytes, and the presence of a specific progesterone receptor on the surface of oocytes was demonstrated by photoaffinity labeling with synthetic progestin and by radioreceptor binding assays using total plasma membrane fraction in *Xenopus* and in *Rana pipiens*. In this study, the binding activity of progesterone was characterized in devitelline oocytes, which vitelline membrane was removed, in *Xenopus laevis*. The binding affinity of progesterone to devitelline oocytes was examined by using immobilized progesterone. Fluorescein isothiocyanate-labeled progesterone 3-0-carboxymethyloxime-BSA(P-BSA-FITC) was localized on the outside surface of the devitelline oocytes. The binding affinty of P-BSA-FITC to devitelline oocyte is higher than that of steroid-BSA-FITC(estrogen, testosterone). The binding affinity of P-BSA-FITC appeared to be high at pH 7.0-7.5, but low at more acidic(pH 6.0-6.5) or alkali(pH 8.0-9.0) conditions. The binding affinity reached maximum levels by two-hours of incubation with P-BSA-FITC, and the affinity was weaker in Stage IV oocytes than that observed in Stage VI oocytes. P-BSA induced oocyte maturation of denuded oocytes but E-BSA failed to induce oocyte maturation. This result was similar with that observed with *Rana dybowskii*. These results strongly suggest that a specific receptor for progesterone exist in plasma membranes of *Xenopus* oocytes, and progesterone acts initially on this receptors at the oocyte surface where it triggers generation of membrane-mediated second messengers during oocyte maturation in amphibians.

## D106 Effect of Heat Shock on P4-induced Oocyte Maturation in *Rana dybowskii*.

Jung Won Ju*, Wook-Bin Im, Hyuk-Bang Kwon and Hueng-Sik Choi
Hormone Research Center, Department of Biology, Chonnam National University

In amphibians, full-grown oocytes are arrested in prophase I and reenter cell cycle by the stimulation of progesterone. When progesterone induces the maturation of amphibian oocyte through its action on the plasma membrane, various kinases in the signal transduction pathway are activated by phophorylation/dephosphorylation and followed by the translation of maternal mRNAs which encoded proteins involved in oocyte maturation. Using *Rana dybowskii*, we investigated the mechanism of P4-induced oocyte maturation by heat shock(35℃). When oocytes were exposed to heat shock, the rate of GVBD was significantly decreased. This inhibition effect of heat shock on GVBD was observed in oocytes exposed to heat shock for 60 and 90min, but was not observed in oocytes which exposed for 30min. When the activity of MAPK and cdc2 kinase, which play essential roles in oocyte maturation, was measured during the maturation of oocytes exposed to heat shock, the activity of these kinases was not inhibited by heat shock for 30min. However, when oocytes were exposed to heat shock for 60 and 90min, the activity of MAPK was inhibited completely, and that of cdc2 kinase was decreased significantly.

Taken together, these results demonstrate that P4-induced oocyte maturation is delayed by heat shock and the activity of MAPK and cdc2 kinase are inhibited by heat shock. Thus, it is evident that MAPK and cdc2 kinase play essential roles in the completion of oocyte maturation in *Rana dybowskii*.