

- 57 Immunohistochemical analysis of progesterone receptor in human Fallopian tube. A. Ikegami, J. Kato, Dept. Obst. and Gynec., Yamanashi Med. College., Yamanashi.

In human Fallopian tube, the surge of progesterone after ovulation causes deciliation and loss of secretory activity in the epithelial cells. The concentrations of progesterone receptor in a particular cell are partly regulated by the plasma levels of estradiol and progesterone. We have examined the variations in progesterone receptor (PR) of human Fallopian tube during the menstrual cycle using immunohistochemical techniques. Three segments (isthmus, ampulla, and infundibulum) of fallopian tube were removed from the surgical specimens of 13 women of reproductive age who had hysterectomies. Level of PR were evaluated by determining the distribution and intensity of staining. Epithelial, stromal, and smooth muscle cell nuclei of fallopian tubes had nuclear localization of PR. The intensity of staining for PR was strong in fallopian tubes that were removed from the proliferative phase of the menstrual cycle.

- 58 Detection of PRmRNA in human uterus using RT-PCR. T. Osada, S. Hirata, K. Hagihara, M. Hirai, J. Kato. Dept. of Obst. and Gynec., Yamanashi Med. School, Yamanashi.

The existence and distribution of progesterone receptor (PR) in human endometrium (EM) and myometrium (MM) have been investigated extensively, but few reports on PRmRNA in those tissues have been available so far. In the present study, we have examined the level of PRmRNA in human EM and MM by the use of reverse transcription-polymerase chain reaction (RT-PCR).

Total RNA was extracted from EM and MM by the guanidium thiocyanate-cesium chloride technique. The RNA was reverse transcribed, followed by PCR using two oligonucleotide primers specific for a part (320bp) of progesterone binding domain of the human PRcDNA. It was confirmed that the amplified fragments corresponded to the part of the human PRcDNA by nucleotide sequencing. Subsequently, Southern blot analysis was carried out for detection and quantification of PRmRNA.

In the present study, amplified genes were detected in human EM and MM by Southern blot analysis. The level of amplified genes in EM was greater than that in MM. From these results, it seems that PR in EM and MM may be regulated by the level of PRmRNA.

[Supported by grants from the Ministry of Education, No. 01440069 to J.K.]

- 59 Evaluation of oocyte maturation in terms of intracellular calcium oscillation at fertilization. T. Fujiwara, O. Tsutsumi, T. Ayabe, T. Yano, N. Mitsuhashi, M. Mizuno and S. Miyazaki*, Dept. Obst. and Gynec., Univ. of Tokyo, Tokyo, *Dept. Physiology, Tokyo Women's Medical College, Tokyo.

Repetitive calcium (Ca) release from intracellular Ca store is observed in mature hamster eggs at fertilization, which can be induced by inositol 1,4,5-trisphosphate (IP_3). We have determined intracellular Ca concentration ($[Ca^{2+}]_i$) using Fura-2 loaded into eggs. Peak levels of $[Ca^{2+}]_i$ were 500-600nM in mature eggs, while those of immature eggs were about 200nM. We also studied the changes of $[Ca^{2+}]_i$ after iontophoretic microinjection of IP_3 into hamster eggs during oocyte maturation. It revealed that the response to IP_3 in mature eggs was all-or-none type Ca rise although it was not in immature eggs. All-or-none type Ca response was observed 12h after hCG administration in animals. It was also observed when cumulus oocyte complex was cultured 10h in BWW medium. Peak levels of $[Ca^{2+}]_i$ induced by IP_3 were 500-600nM in mature eggs and 300nM in immature ones. These results suggest that intracellular Ca oscillation develops both in nature (all-or-none) and its sensitivity, which may serve as a useful marker of maturation.