490 Localization of extracellular matrix (ECM) on mouse embryos in immunohistochemistry. T. Yoshimura, T. Sawada, T. Kamiya, K. Hattori, T. Aoki, Y. Higuchi, S. Kawakami, M. Fukushima, Department of OB/GYN, Fujita-Gakuen Health University, Toyoake Aichi.

The existence of extracellular matrix (ECM) on the endometrium and embryos has been shown to play a significant role in the process of implantation. The present study was undertaken to determine the localization of fibronectin, collagen type IV, and laminin on mouse embryos using immunohistochemical study. B6C3F1 mice two cell embryos were recovered from oviducts of mated mice at 36 hours after hCG administration. These embryos were cultured in BWW medium supplemented with 10% heat-inactivated women's serum at 5% Co2-95% air up to the hatching blastocyst stage. Localization of fibronectin was not observed in the stage of 2 to 16 cells. Fibronectin was detected immunohistochemically after to morula stage. Further more, the localization was greater in the inner cell mass of blastocyst. Collagen type IV was detected after the 8 cell stage and the localization was greater in the trophoblast of hatching blastocyst. Laminin was detected as early as 2 cell stage embryos, and the localization was greater in embryos after the blastocyst stag than that in early cleavage embryos. Localization of ECM becomes greater relation with embryo development. These date suggest the involvement of ECM in the process of implantation.

491 Negative-charged Lipids in Rabbit Endometrium during the Implantation Period. M.Momoeda, Y.Taketani, M.Mizuno, Dept. Obst. and Gynec., Univ. of Tokyo, Tokyo.

To explore the mechanism of implantation, we examined the change in the amounts of negative-charged lipids in rabbit endometrium. Gangliosides were shown to be present in a very small quantity in rabbit endometrium irrespective of the reproductive stages. Notably, considerable amounts of cholesterol sulfate (CS) was detected both in nonpregnant and pregnant endometrium. The identification of cholesterol sulfate was performed by negative-ion FAB mass spectrometry. The content of CS was relatively low in nonpregnant endometrium. It abruptly increased at day 5 of pregnancy, i.e. at the beginning of implantation, followed by a gradual decline toward day 9. Looking for the distribution of CS in the endometrium, CS levels in the interimplantation sites was about 2-fold as much as those in the implantation sites. CS levels in pseudopregnant endometrium was comparable with those in the interimplantation sites. These results demonstrate that CS is major negative-charged lipid present in periimplantation endometrium in rabbits. We further point to the difference in the concentration of CS between implantation and interimplantation sites, thus suggesting CS as a major participant in the process of implantation.

492 Ultrastructural studies of the interaction between epithelial and stromal cells in the human endometrium. T.Kamiya, A.Kawata, I.Tanaka, T.Nakajima, I.Sawaragi, Dept.Obst.and Gynec., Kansai Medical Univ. Osaka.

In the normal human endometrium, the basement membrane(B.M.) of the epithelial cells(E.C.) is composed of the interaction between the E.C. and the stromal cells(S.C.). In this study, the E.C. were co-cultured with the S.C. in order to reconstruct the B.M. using the collagen gel methods. The E.C. and the S.C. were dissociated from normal endometrium. The S.C. were laid on the basal layer consisting of the Type I collagen gel in the dishes. After 24 hours, the gel mixture with the Type I and IV collagen was coated and the E.C. were overlaid on each dish. And the E.C. were co-cultured with the S.C. in the three dimensional collagen gel culture. The monolayer culture of the E.C. on the Type I collagen gel were performed as the control. The E.C. had the B.M. stained by PAS and gitter stain on the mixed layer with the Type I and IV collagen gel at 5 days in culture. The E.C. were acompanied by the continuous B.M. without no three layers structure by the TEM. However, the B.M. of the E.C. on the Type I collagen gel were not seen at 14 days in culture. The E.C. interacted with the S.C. in the mixed thin layer with the Type I and IV collagen gel, and the B.M. of the E.C. was reconstructed by the Type IV collagen and the S.C.