

**535** Culture of trophoblast cells by growing chorionic villus explants and effect of the culture supernatants of trophoblast cells on lymphocytes proliferation. T.Fujino, Y.Nagata, Dept.Obst.and Gynec.,Fac.Med.Kagoshima Univ.,Kagoshima.

To obtain long-term trophoblast single cell culture, chorionic villus explants were grown without enzyme digestion at the primary culture. Chorionic villus tissues were obtained from term placentas, first-trimester placentas and hydatidiform moles. The tissues were minced into small pieces, and then cultured in flasks. In about a week cells were observed budding from the outer surface of the villus tissues. At first the culture was composed of more than one type of cells: round cells, spindle shaped cells, and so on. But, they gradually became multiangular and multinucleated, and in part they fused each other. For passages 0.25% trypsin was used. After 2 passages hCG production was 150mIU/ml/ $3 \times 10^4$  cells/36hrs. So far culture was possible up to 4 passages and cells survived for 4 months. The cultured cells were confirmed as trophoblast by immunohistochemistry. Effect of culture supernatants of trophoblast cells on lymphocytes proliferation was tested by measuring <sup>3</sup>H-thymidine uptake. Culture supernatants of multiangular, fused cell-dominant trophoblast cell culture had a stimulatory effect on lymphocytes proliferation.

**536** Accumulation of CD16<sup>-</sup>CD56<sup>+</sup>natural killer cells with high affinity interleukin 2 receptors in human early pregnancy decidua. S.Saito, Y.Kato, M.Enomoto, M.Ichijo, Dept.Obst.and Gynec.,Nara Med.Univ.,Nara.

Most human peripheral blood NK cells express the phenotype CD16<sup>+</sup> CD56<sup>+</sup>. However, a very minor subset of NK cells express CD16<sup>-</sup> CD56<sup>+</sup>, and these NK cells bear both IL-2R $\alpha$  (p55) and IL-2R $\beta$  (p75) (high affinity IL-2 receptors). In this report, we demonstrate that in human early pregnancy decidua, an interface between maternal immunocompetent cells and fetus (placenta), abundant (~83%) CD16<sup>-</sup> CD56<sup>+</sup> NK cells with high affinity IL-2 receptors were present, and these cells responded to low amounts of IL-2 (4.5pM). These CD16<sup>-</sup> CD56<sup>+</sup> NK cells significantly expressed an early activation antigen, CD69, in vivo, whereas peripheral blood CD16<sup>-</sup> CD56<sup>+</sup> NK cells did not express CD69. These findings suggest that CD16<sup>-</sup> CD56<sup>+</sup> NK cells in early pregnancy decidua may be activated in vivo, and may play an important role in immunoregulation during early pregnancy. Also, decidual lymphocytes may be useful materials to study the mechanism of MHC-unrestricted cytotoxicity of this type of NK cells.

**537** Expression of functional high affinity interleukin-2 receptor on decidual CD56 positive lymphocytes. Y.Kurahayashi, S.Uehara, S.Tanigawara, T.Takabayashi, K.Okamura, A.Yajima, Dept. Obst. and Gynec., Tohoku Univ. Sch. Med., Miyagi.

High affinity interleukin-2 receptor (IL-2R) is composed of at least two subunits, p55 and p75. We studied expression and function of IL-2R of decidual lymphocytes. Two-colour flow cytometric analyses showed that 62.7% of decidual lymphocytes were CD56 strong positive, and expressed both p55 and p75. In contrast, the ratio of CD56 positive lymphocytes in peripheral blood of normal pregnant women (PBL) or cord blood were 18.9%, 8.5%, and these cells expressed only p75. Decidual CD56 positive cells were so characteristic that the reactivity to IL-2 and the magnitude of proliferation were higher than those of PBL. These responses were blocked by p55 or p75 antibody. On the other hand, cytotoxic activity of decidual lymphocyte was weaker than those of PBL.