

LABORATORY OF CELL SOCIOLOGY

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Animal organs consist of several types of cells. They are organized in an ordered fashion wherein the proportion of each cell type is constantly maintained. The ordered cell arrangement and proportion are built up during organogenesis by cell-cell interactions. Since *Notch* has been postulated to play a role in cell fate decision by mediating cell-cell interactions, we are trying to find out cellular and molecular mechanisms working in organogenesis by studying the function of *Notch*.

Organogenesis of the mouse placenta occurs during early pregnancy, embryonic days 7-9, before the establishment of molecular transport mechanisms in the definitive placenta takes place. Trophoblasts not adjacent to the inner cell mass differentiate into trophoblast giant cells and lie at the outside, forming interface with maternal deciduas. The polar trophoblast gives rise to cells of the chorion, and ectoplacental cone. They produce labyrinthine and spongiotrophoblast layer, respectively. While maternal red blood cells begin to perfuse into trophoblast cell layers and reach at labyrinthine layer by E9.5, invasion of embryonic allantoic mesenchyme into labyrinthine layer and differentiation of fetal red blood and endothelial cells which line fetal capillary takes place around E9.5.

The mutation in the ankyrin repeats of mouse *Notch2* results in embryonic lethality by embryonic day 11.5 due to a poor maternal vascular beds formation. The mutant placenta showed an early invasion of angiogenic allantoic mesenchyme followed by premature formation of fetal blood vessels in the mutant placentas as early as E9.0. However, specification of trophoblast subtypes appeared not to be drastically disturbed. Thus, in the developing mouse placenta, *Notch2* is unlikely involved in cell fate decision, but rather participates in a formation of circulatory systems in the labyrinth layer where the expression of *Notch2* was detected (Figure 1). Although inadequate formation of maternal vascular beds was partially restored by aggregating mutant diploid embryos with wild type tetraploid embryos (Figure 2), networks of the mutant fetal vasculatures and maternal blood spaces appeared still comprised in the 4N chimeric placenta. These results indicate that *Notch2* works promotive in vasculogenesis. Thus, *Notch2* is not cell autonomously required for the early cell fate determination of subtypes of trophoblast cells, but plays an indispensable role in coordinated maternal and fetal vasculogenesis in the developing mouse placenta.

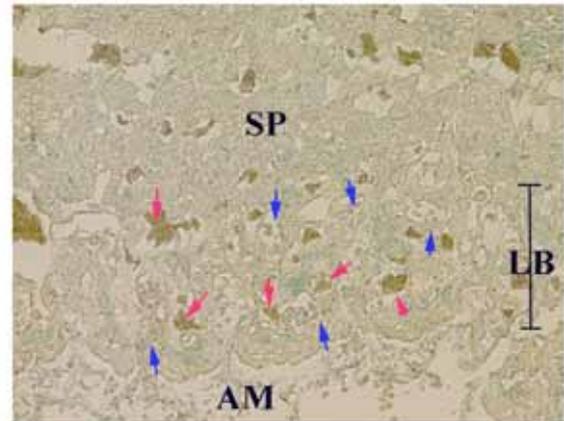


Figure 1. Expression of *Notch2* gene in developing mouse placenta. A cryo-section of *Notch2*^{+/LoxZ} placenta at E10.0 was incubated in X-gal staining solution. Trophoblast in maternal vascular bed side expresses the gene, but not in allantoic mesenchyme (AM) side in labyrinth layer (LB). The expression also occurs in the mesenchyme. Enucleated maternal red blood cells are indicated by red arrow heads. Nucleated fetal red blood cells are by blue arrow heads.

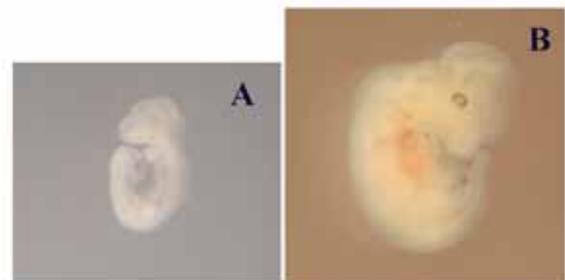


Figure 2. Survival of *Notch2* mutant embryo in a chimera with tetraploid wild type embryo. The mutant obtained in the intercrosses by the heterozygotic mice shows developmental retardation at E10.5 (A) and die before E11.5. The mutant is able to survive in a chimera with wild type tetraploid embryo until E13.5 (B).

Publication List:

Original papers

- Gotoh, N., Manova, K., Tanaka, S., Murohashi, M., Hadari, Y., Lee, A., Hamada, Y., Hiroe, T., Ito, M., Kurihara, T., Nakazato, H., Shibuya, M., Lax, I., Lacy, E., and Schlessinger, J. (2005). The docking protein FRS2alpha is an essential component of multiple fibroblast growth factor responses during early mouse development. *Mol. Cell. Biol.* 25, 4105-4116.
- Nakamura, Y., Hamada, Y., Fujiwara, T., Enomoto, H., Hiroe, T., Tanaka, S., Nose, M., Nakahara, M., Yoshida, N., Takenawa, T., and Fukami, K. (2005). Phospholipase C-δ1 and -δ3 are essential in trophoblast for placental development. *Mol. Cell. Biol.* 25, 10979-10988.