論文の内容の要旨

獣医学専攻

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論文題目　Roles of Embryonic Hindgut Endoderm in Primordial Germ Cells Migration and Differentiation in Mice

（マウス始原生殖細胞の移動および分化における胚性内胚葉の役割について）

In most animals, primordial germ cells (PGCs) arise very early in development at a site distinct from which gonads are ultimately formed. In mice, PGCs arise around gastrulation between 6.25 and 7 days post coitum (dpc) from a pluripotent population of cells in the proximal epiblast. The germ cell lineage is discriminated from somatic cell lineage during development, and repression of the somatic cell fate is therefore a key event during the germ cell specification. The specification is initiated by signals provided by the extraembryonic ectoderm, and the visceral endoderm that surrounds the epiblast cells and instructs a small population of epiblast cells to become PGCs. Specified PGCs migrate through the primitive streak into the region of the embryonic endoderm that forms the hindgut (7.5~8.5 dpc). Later in development between 8.5~9.5 dpc, the second phase of migration takes place in which PGCs migrate randomly within the hindgut epithelium. Finally, between 9.5 and 10.5 dpc, PGCs emerging from the hindgut are seen in the dorsal mesentery and migrate into two bilateral streams towards the GRs.

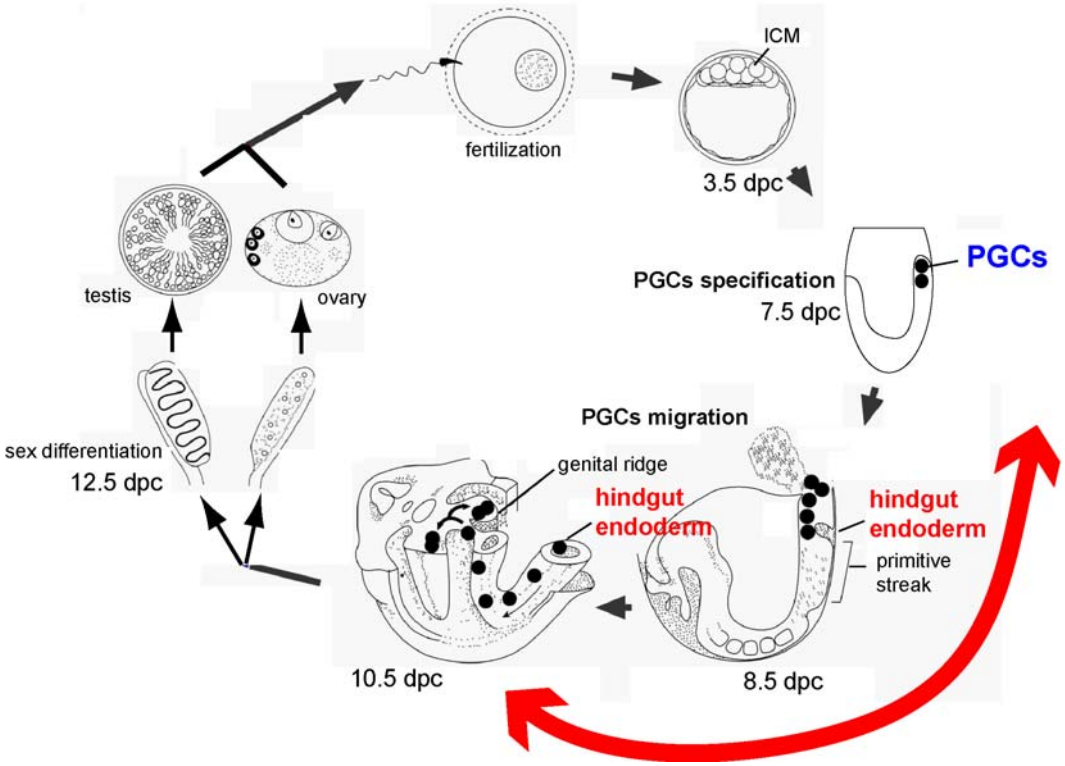

Figure 1. **PGCs migration within the embryonic hindgut endoderm in germ cell development in mouse.** Double-headed arrow indicates the period of PGCs migration within the embryonic hindgut endoderm.

As reviewed above, it requires two days out of the total three-day-migration period for PGCs to migrate through the embryonic hindgut endoderm toward GRs in mouse embryogenesis (double-headed arrow in Fig. 1). The PGCs migration route via the endodermal tissues is widely conserved in various vertebrate and invertebrate species (gut endoderm in nematodes; midgut in flies; endoderm in frogs; hindgut in mice), suggesting a potential role of endodermal tissues in the PGCs migration and differentiation during early migratory stages. Therefore, I hypothesized that the embryonic hindgut endoderm is a key “micro-environment” for PGCs to migrate and differentiate properly during gastrulation. The purpose of this study is to clarify the

possible roles of the embryonic hindgut endoderm in PGCs migration and differentiation during gastrulation in mice.

Here, I provide the direct evidence for crucial roles of the embryonic hindgut endoderm in directing proper PGCs migration, not epigenetic maturation, during late gastrulation to early somite stages. The *Sox17*-null mouse embryos displayed early and specific defects in hindgut expansion during late gastrulation. In these *Sox17*-null embryos, PGCs normally differentiated at the base of allantois, and then only a small number of the front PGCs population could move properly into the presumptive endodermal area. However, possibly due to defective expansion of *Sox17*-null endoderm, these PGCs in endodermal position were blocked to migrate from the primitive streak position into the outer layer of definitive endoderm throughout gastrulation (Fig. 2, center ①). At the same time, a large number of remaining PGCs migrated away from the primitive streak into the ectopic position of the yolk sac visceral endoderm (Fig. 2, center ②), where these ectopic cells properly progressed to epigenetic maturation similar to those of wildtype PGCs in hindgut. Chimera analysis of *Sox17*-null ES cells confirmed that the supply of wildtype endoderm cells can rescue defective migration of the *Sox17*-null PGCs into the proper hindgut position (Fig. 2, right). The present results, therefore, imply that defective embryonic hindgut endoderm expansion promotes ectopic PGCs migration into yolk sac visceral endoderm, highlighting the importance of the continuous supply and expansion of definitive endoderm cells for the selection of the PGCs into the correct path to the GRs. Furthermore, my findings highlight that the regulation of germ-line migration by the gut endoderm during gastrulation is evolutionally conserved among some invertebrates and mammals.

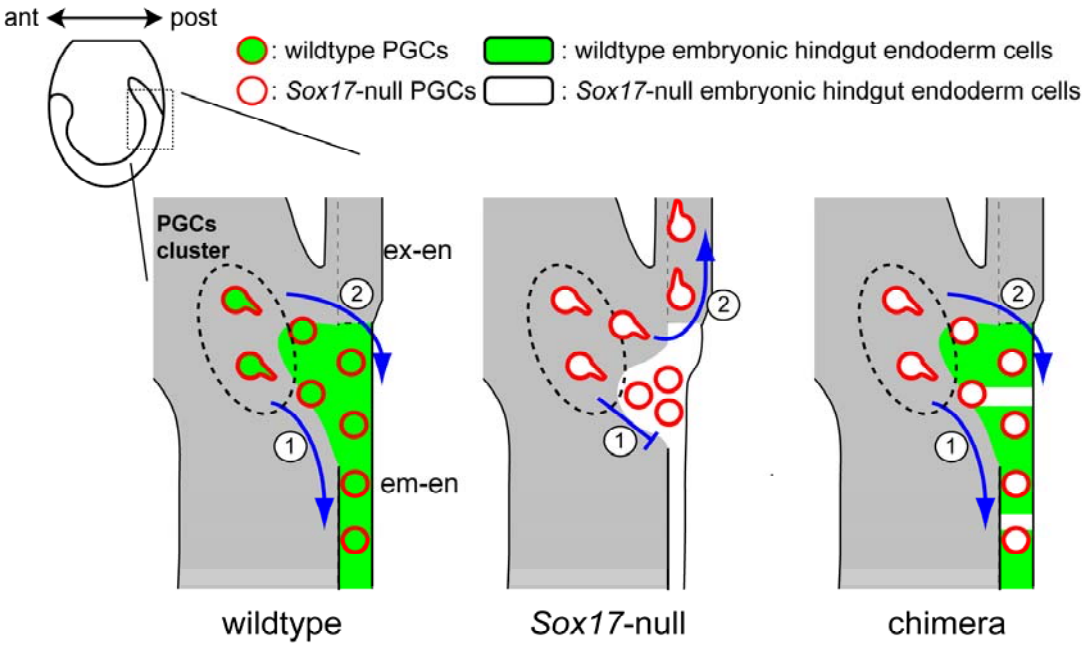

Figure 2. **Comparison of the PGCs migration and the expansion of newly-supplied embryonic hindgut endoderm in wildtype, *Sox17*-null and chimeric embryos.**

In the *Sox17*-null embryos, a front population of the PGCs is immobile within the defective hindgut endoderm (①), but the remaining PGCs ectopically migrate away into the extraembryonic yolk sac endoderm in *Sox17*-null embryos (②). By contrast, supply of wildtype embryonic hindgut endoderm cells can rescue defective migration of the *Sox17*-null PGCs by chimera analysis. These results strongly suggest that the continuous supply and expansion of embryonic endoderm cells is essential for PGCs movement into the correct path to the GRs.

①: Front population of PGCs. ②: The last (remaining) population of PGCs.

Arrows indicate the directions of PGCs migration. ex-en, extraembryonic endoderm; em-en, embryonic endoderm.