

Artificial Oocyte Growth in Domestic Species

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Fully-grown immature oocytes collected from the ovaries of commercially slaughtered cows can be used for *in vitro* fertilization (IVF) following their *in vitro* maturation (IVM). Ovaries also contain a huge number of growing and non-growing oocytes. Once non-growing oocytes (mouse: 15–20 μ m, pig and cow: 30 μ m) in primordial follicles enter the growth phase, they grow to their final size (mouse: 75 μ m, pig and cow: 125 μ m) taking a long period of time. When fully-grown oocytes receive the gonadotropic stimulation, they resume the meiosis, mature to the second metaphase and are ovulated. However, the population of the oocytes that grow to the final size and mature in the ovaries is quite small. Artificial growing-up of small oocytes collected from ovaries could provide a new source of mature eggs for livestock production and assisted reproduction in humans.

Baby mice have been produced by *in vitro* growth (IVG) culture of oocytes in primordial follicles (Eppig & O'Brien, 1996). In large domestic species, baby calves have been produced from IVG-cultured oocytes (Yamamoto *et al.*, 1999; Hirao *et al.*, 2004). However, the oocytes used in the experiments were growing ones at the mid-growth phase (90–99 μ m in diameter) from early antral follicles of 0.5 mm in diameter. IVG systems have not been established for non-growing oocytes of large domestic species.

Xenotransplantation of small oocytes to nude mice or SCID (severe combined immune deficiency) mice is a substitute for an effective long-term IVG culture. We xenotransplanted bovine primordial follicles (40 μ m in diameter, oocyte: 30 μ m) and secondary follicles (150–200 μ m in

diameter, oocyte: 55 μ m) separately into SCID mice. After 4–6 weeks, secondary follicles developed to the antral stage, with oocytes reaching their final size, although primordial follicles survived but did not grow in the grafts (Senbon *et al.*, 2003). We extended the experiment to porcine primordial follicles. Primordial follicles from adult pigs did not grow in xenografts. On the other hand, primordial follicles from piglets developed to the antral stage with oocytes reaching their final size after 2 months. These findings show that secondary follicles and their oocytes have already entered their growth phase and are able to develop to the final stage in appropriate conditions, and that primordial follicles/oocytes especially from adult animals require some factor(s) to enter the growth phase. Improved IVG and xenotransplantation systems enhance our knowledge about the initiation of oocyte growth as well as follicular selection in mammalian ovaries.

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