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Chimeric Blastocysts by Aggregation between Parthenogenetic and Fertilized Bovine Embryos

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Abstract: Chimeric blastocysts were produced by aggregation of parthenogenetic and fertilized bovine embryos. To induce parthenogenetic activation, mature oocytes were treated with 7% ethanol and 5 mg/ml cytochalasin D and then cultured at 38.5℃ under 5% CO_2 in air. Normal fertilized embryos were obtained by *in vitro* maturation, fertilization and culture. Aggregated embryos were prepared by four methods as follows: [1] injection of 2 blastyomeres of a parthenogenetic embryo (16-cell stage) into a fertilized embryo (4-cell stage), [2] injection of a parthenogenetic demi-embryo (8-cell stage) into a fertilized demi-embryo (8-cell stage), [3] injection of a fertilized demi-embryo (8-cell stage) into a parthenogenetic demi-embryo (8-cell stage) and [4] aggregation of whole parthenogenetic and fertilized embryos (8-cell stage). The developmental rate *in vitro* of aggregated embryos produced by aggregation of the whole blastomeres (method [4]) tended to be higher than those by other methods (method [1] to [3]). Karyotyping thirteen aggregated embryos revealed 8 of them to be chimeric chromosome composition of XX and XY. These results verified that the parthenogenetic cells can be contributed to the development of the aggregated embryo.

Key words: parthenogenetic embryo, fertilized embryo, aggregated embryo, chimeric blastocyst

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Introduction

Parthenogenetic development of embryos to give live offspring occurs naturally in many non-mammalian species[1], and the phenomenon can also be induced experimentally in non-mammalian vertebrates and invertebrates[2]. Although the parthenogenetically activated diploid mammalian embryos develop normally through preimplantaion, they rarely reach to the forelimb-bud stage[3] in mouse.

There are distinct differences between the paternal and maternal contribution to the embryonic development. The paternal genome appears to be more important for the proliferation of the extra embryonic tissues and the maternal genome plays a key role in preimplantation and early postimplantation development [4].

Vital functional differences between the parental genomes of mammals have been demonstrated by embryonic lethality of parthenogenetic, gynogenetic and androgenetic uniparental genotypes[5]. Cells derived from uniparental embryos were rescued by integration with normally fertilized embryos, which results in a chimeric organism[6-8]. These phenomena may relieve the problem of mammalian parthenogenesis.

In the bovine oocytes, there are several effective ways to induce diploid parthenogenesis, such as exposure to ionophore A23187[9] or ethanol, electric