Development of Chimeric Embryos Aggregated with Blastomeres of In Vitro Fertilization (IVF) and of Parthenote Bovine Embryos

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Chimerism has become an important tool for investigating fundamental aspects of early embryonic development and differentiation in mammals for producing transgenic animals. The objective of this study was to evaluate the developmental capacity of chimeric embryos reconstructed with parthenotes and IVF bovine embryos into empty zona pellucida.

The MII oocytes were activated by two treatment groups [Group 1, 5μ M ionomycin, $5 \min$, $+10 \mu$ g/ml cycloheximide (CHX)/5 μ g/ml cytochalasin B (CCB), 3 h; Group 2, 5μ M ionomycin, $5 \min$ + 1.9 mM 6-dimetylaminopurine (6-DMAP), 3 h]. Chimeric embryos were reconstructed by aggregation of each two blastomeres of 4-cell stage parthenotes and IVF embryos. To prepare each blastomeres, the embryos were exposed to a 0.1% pronase solution in order to dissolve their zona pellucidae and isolated the blastomeres in calcium and magnesium free-PBS. Chimeric embryos were than produced by micromanipulation technique.

Blastocyst formation of chimeric embryos aggregated with IVF and parthenote produced by Group 1 treatment were significantly lower than that by Group 2 treatment (P<0.05) or handle control (P<0.01). No difference on blastocyst formation between chimeric embryos produced by Group 2 and handled control. This result shows that parthenotic embryos have potentially possessed the developmental efficiency in chimeric embryos aggregated.

Table 1. Development of chimeric embryos aggregated with blastomeres of IVF and of parthenote embryos.

Treatment	Replicates	No. of embryos	Rate of Development (%)	
			8-cell	Blastocyst
Group 1. IVF(2/4) + CHX · CCB(2/4)	3	13	5 (38)	2 (15) ^a
Group 2. IVF(2/4) + 6-DMAP(2/4)	3	15	6 (40)	4 (29) ^b
Group 3. Handled control(2/4+2/4)	3	14	6 (40)	4 (28) ^b
Group 4. IVF(4-cell stage)	4	27	23 (85)	10 (37) ^c

Different superscripts in the same column indicate a significant difference (a and b, b and c, P < 0.05; a and c, P < 0.01)

Key words) Chimeric embryo, Parthenote

^{*2/4} denotes a pair of blastomeres derived from a 4-cell embryo: Handled controls denotes 4-cell embryos subjected to micromanipulation and reassembled from their original four blastomeres.