CHIMERA PRODUCTION IN BOVINE AND ITS SPERM CAPACITY

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ABSTRACT

In vitro produced bovine embryos were used to produce chimeras by combining the 8-cell stage embryos. Aggregated embryos were embedded in either 0, 1.0 and 1.2% agar and cultured in vitro. The aggregation rate of the embryos cultured without agar embedding was lower (P<.05) than with agar embedding (62%, 92% and 94% for 0, 1.0 and 1.2% agar, respectively). Five aggregated embryos were transferred non-surgically, resulting in the birth of 2 chimeric calves. Base on chromosome analysis, the chimera bull had apparently normal chromosomes (29 acrosentric pairs, one large submetacentric X chromosome and one small submetacentric Y chromosome). Capacity of the Chimera (C) sperm were used for IVF to compare with sperm of the Japanese Black (JB), Limousin (L), Japanese Red (JR) and Holstein (H) bulls. Fertilization rates by using Chimera sperm were higher (P<.05) than JR and H sperm, but did not differ from JB and L sperm (81.8%, 80.0%, 69.4%, 44.2% and 18.2% for C, JB, L, JR and H, respectively). The blastocyst rates oocytes inseminated with C sperm were higher (P<.05) than with L, JR and H sperm, but not differ from JB sperm (38.1%, 39%, 25.0%, 23.3% and 17.8% for C, JB, L, JR and H sperm, respectively). These findings suggested that chimera in bovine could be able to produce by aggregation method resulted in the normal calves. The sperm collected from chimeric bull could be used for producing bovine IVF embryos.

Key words: Chimeric bull, Sperm, IVF

INTRODUCTION

Chimeras have been successfully produced in laboratory animals either by the aggregation of precompaction-stage embryos (Mintz, 1971; Tarkowski and Wroblewska, 1967) or by the microinjection of the inner cell mass (ICM) into the blastocyst (Gardner, 1968; Rorie *et al.*, 1989). Attempts to produce chimeras in domestic animals derived from surgically flushed embryos have been reported in sheep (Fehilly *et al.*, 1984; Polzin *et al.*, 1987; Rorie *et al.*, 1989) and cattle (Brem *et al.*, 1984; Picard *et al.*, 1990; Summers *et al.*, 1983). The blastocyst injection technique (Picard *et al.*, 1990; Summers *et al.*, 1983) or aggregation of bisected embryos with the zone pellucida intact (Brem *et al.*, 1984) was used in cattle to produce chimeric calves.

Sperm from individual bulls differ in their ability to fertilize matured oocytes in vitro and support embryo development to the pre-implantation stage (Iritani *et al.*, 1986; Leibfried-Rutledge *et al.*, 1987). Reproduction of interspecific chimeric between sheep and goat (MacLaren *et al.*, 1993) have been reported. However, the ability of tetraparental bovine chimeric sperm to fertilized matured oocytes and support further embryo development in vitro has not been reported.

The primary objective of this study was to evaluate the developmental capacity of the aggregated embryos. In a series experiments conducted, we investigated (1) the effect of agar embedding for protection of aggregated embryos from disaggregation during culture in vitro, (2) the chromosome composition of chimeric calf (3) the ability of sperm from a tetraparental chimera to fertilize matured oocytes and developmental capacity of in vitro produced embryos.

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