

Embryos and culture cells: A model for studying the effect of progesterone

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Abstract

A positive association between P4 concentration and initial bovine embryo survival has been reported. The objective of this study was to establish two coculture systems as a model to study the influence of progesterone on the initial bovine embryo development. Granulosa cells (GC) or bovine oviduct epithelial cells (BOECs) were used at the base of embryo culture medium microdroplets (TCM199 and 10% of superovulated oestrus cow serum, (SOCS)) supplemented or not with progesterone (P4, 33.4 ng mL⁻¹) and/or a progesterone receptor antagonist (onapristone, OP, 2.2 × 10⁻⁵ M). Presumptive zygotes were transferred to monolayers after *in vitro* maturation and fertilization of bovine oocytes with thawed *swim-up* selected sperm. Embryo development was carried out according to the following groups: experiment 1, BOEC (*n* = 378) and BOEC plus OP (*n* = 325); experiment 2, GC (*n* = 514); GC plus OP (*n* = 509); BOEC (*n* = 490); BOEC plus P4 (*n* = 500); BOEC plus P4 and OP (*n* = 502). Embryos were checked for cleavage at day 2 and for stage development between days 8 and 12 of culture. In experiment 1, no differences (*P* > 0.05) were identified between BOEC and BOECOP groups for embryo rates of development, quality or developmental stages. Also in experiment 2, no differences were found in embryo rates of development, quality or developmental stages between embryos cultured under the two coculture systems when no supplementation was added. Embryo development rates were not affected by OP presence in GCOP group. However, P4 negatively affected Day 8 (D8) embryo development rates in BOEC system (BOECP4 = 16.8 ± 2.6% vs. BOEC = 23.7 ± 1.7%, *P* = 0.02). This negative effect was abolished when P4 antagonist (OP) was added to the culture medium. BOEC supplementation with P4 also induced a delay on embryo development at D8 as confirmed by a lower development score (BOECP4 = 3.0 ± 1.4 vs. GC = 3.4 ± 0.1, GCOP = 3.5 ± 0.1, BOEC = 3.4 ± 0.1 and BOECP4OP = 3.5 ± 0.1; *P* < 0.05).

These results demonstrate that OP supplementation had no harmful effect on embryo development either in granulosa, where P4 is naturally synthesised, or in BOEC coculture systems. Also we can not confirm a

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