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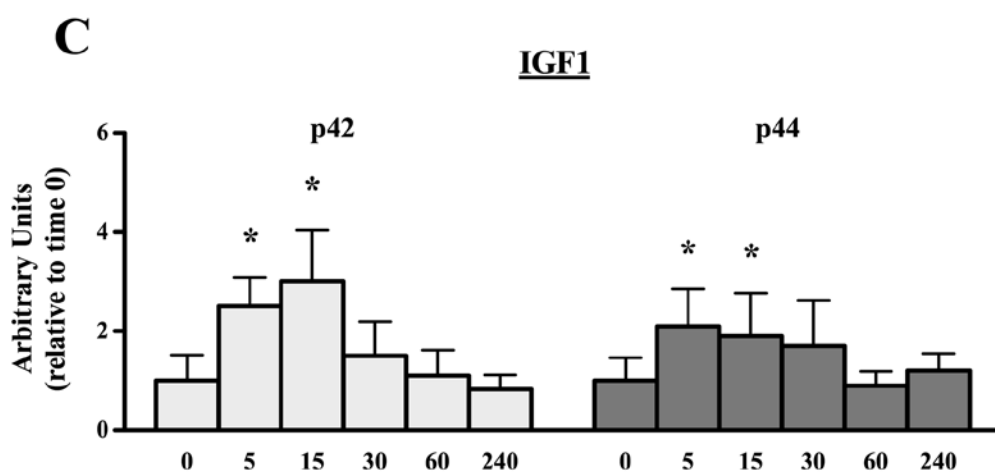
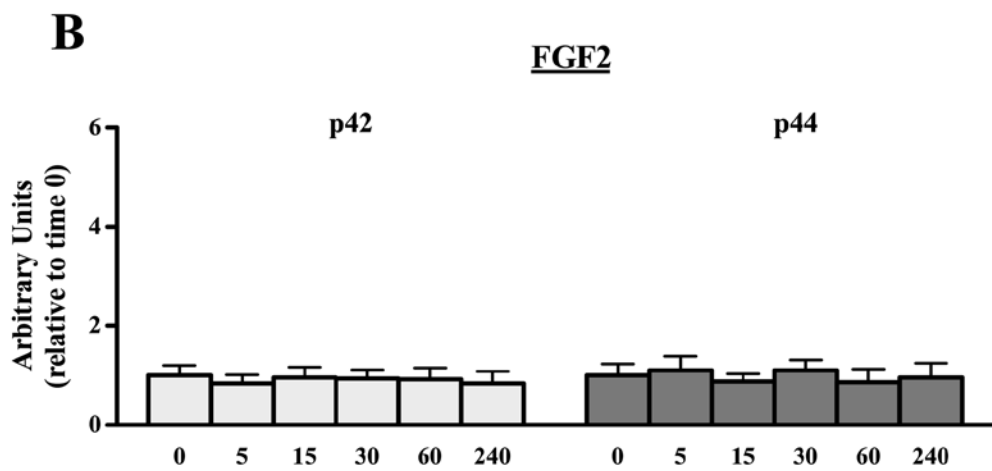
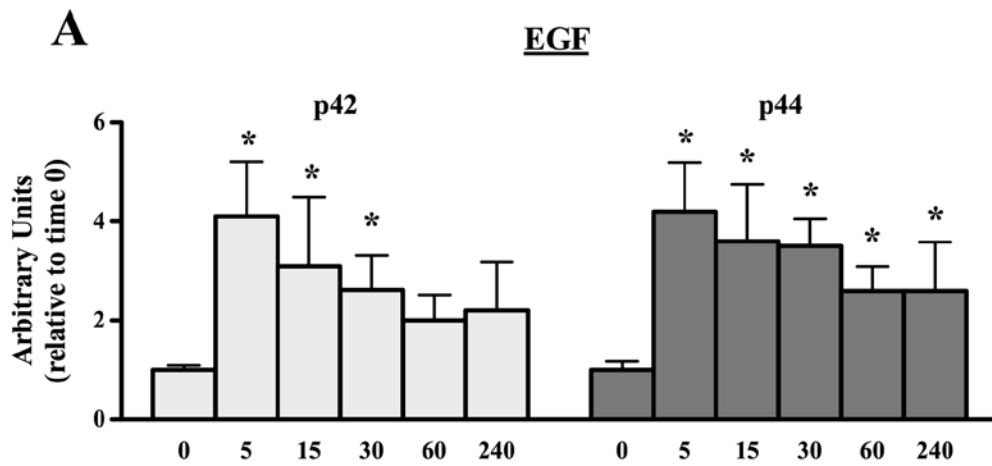
Supplementary Material

Combinatorial effects of epidermal growth factor, fibroblast growth factor 2 and insulin-like growth factor 1 on trophoblast cell proliferation and embryogenesis in cattle

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Time
(Relative to Supplementation, min)

Fig. S1. Densitometric analysis of EGF-, FGF2-, and IGF1-dependent changes in MAPK3/1 phosphorylation status. CT1 cells lysates were collected either immediately before (time 0) or at specific periods after treatment with 10 ng/ml rhEGF (Panel A), 10 ng/ml rbFGF2 (Panel B), or 50ng/ml rhIGF1 (panel C). Lysates were electrophoresed in polyacrylamide gels and transferred to nylon membranes. Thereafter they were immunoblotted with antibodies recognizing total or phospho-specific (p) MAPK3/1. Three independent studies were completed. Densitometric scanning was completed on the p42 and p44 bands. The phosphorylation status relative to total p42 or p44 is presented. The asterisk (*) indicates time-points with increased ($P < 0.05$) phosphorylation when compared with time 0.

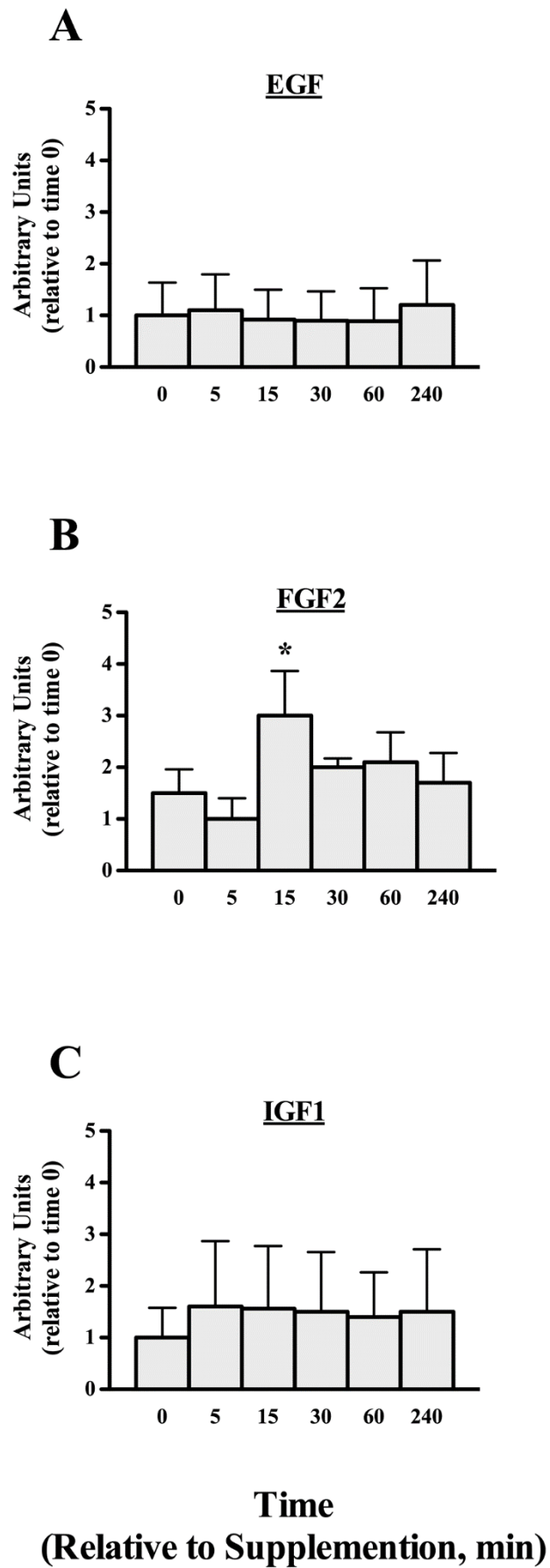


Fig. S2. Densitometric analysis of EGF-, FGF2-, and IGF1-dependent changes in AKT phosphorylation status. CT1 cells lysates were collected either immediately before (time 0) or at specific periods after treatment with 10 ng/ml rhEGF (Panel A), 10 ng/ml rbFGF2 (Panel B), or 50ng/ml rhIGF1 (panel C). Lysates were electrophoresed in polyacrylamide gels and transferred to nylon membranes. Thereafter they were immune-blotted with antibodies recognizing total or phospho-specific (p) AKT. Three independent studies were completed. Densitometric scanning was completed. The phosphorylation status relative to total AKT is presented. The asterisk (*) indicates time-points with increased ($P < 0.05$) phosphorylation when compared with time 0.