Chemical Inhibition of IL6ST Compromises the In Vitro Production of Bovine Embryos

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Previous work found that interleukin-6 (IL6) supplementation improves inner cell mass (ICM) cell numbers within in vitro produced bovine embryos. The embryo expresses IL6, and this raises the possibility that embryo-derived IL6 also plays important roles during early embryogenesis. The IL6 receptor complex is comprised of a ligand-specific α subunit (IL6R) and a common IL6-family β subunit (IL6ST). We hypothesized that incubation with the chemical IL6ST inhibitor, SC144, would impair in vitro bovine embryo development. Abattoir-derived cumulus-oocyte complexes were matured and fertilized, and presumptive zygotes were cultured in SOF-BE1 (45-53 embryos/treatment/replicate; 3-4 replicates/study). Development was assessed at days 2, 5, 7 and 8. Embryo cleavage was not affected by supplementing 0.4 μM SC144 from days 1 to 8, but there was a tendency for reduction when 2 μM SC144 was supplemented (P=0.08) and a reduction (P=0.002) when 10 μM SC144 was supplemented. All SC144 concentrations reduced morulae formation on day 5 (P<0.05). There was a tendency for 0.4 μM SC144 to reduce total blastocyst formation on days 7 (P=0.13) and 8 (P=0.08) and for 2 μM to reduce blastocyst development on days 7 (P=0.02) and 8 (P=0.01). No blastocysts were observed in the 10 μM treatment on days 7 and 8. The day 3 to 5 SC144 treatment scheme was used to describe whether IL6ST inhibition influences morula development. Embryos were washed and plated in new SOF-BE1 lacking the inhibitor on day 5 to determine if carryover effects on blastulation were evident. None of the SC144 concentrations affected morulae formation at day 5; however, blastocyst development was reduced at days 7 and 8 in the 0.4μM and at day 7 in the 2μM SC144 treatments (P<0.01). Blastocysts were degenerate on day 8 in the 2 μM treatment group. No blastocysts were observed in the 10 μM SC144 treatment group on either day (P<0.0001). Blastocysts that were present in the 0.4 μM SC144 group contained the same ICM and TE cell numbers and ICM:TE ratios as the control group. A final study examined how SC144 exposure from day 5 to 8 affects blastocyst development. Blastocyst development was reduced at day 7 and 8 at each SC144 concentration (P<0.01), and substantial blastocyst degeneration was observed on day 8 when 2 or 10 μM SC144 was provided. Blastocysts exposed to 0.4 μM SC144 contained fewer ICM (P=0.03) and tended to contain fewer TE (P=0.08) cell numbers, although no changes in the ICM:TE ratio were detected. In summary, chemical inhibition of IL6ST did not compromise the initial cleavage events but did influence morulation and blastulation. Blastocysts contained a greater than normal rate of degeneration, and reduced ICM and TE cell numbers were detected if the inhibitor was present when blastocysts were developing. This project was supported by Agriculture and Food Research Initiative Competitive Grant no. 2021-67015-34485 from the USDA National Institute of Food and Agriculture.