

Public Abstract

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Title: Morphologic and Histologic Comparisons Between In Vivo and Nuclear Transfer Derived Porcine Embryos

Due to the high failure rate of nuclear transfer (NT) pregnancies very early in gestation we have hypothesized that NT derived embryos have difficulty transitioning through the critical points of elongation and successfully producing a signal for MRP. Because of the high incidence of embryonic loss during a nuclear transfer pregnancy, cloning is considered a relatively inefficient process. However, as the only method currently available for the production of knockout livestock, it remains an invaluable tool.

This study examines abnormalities associated with NT porcine embryos at days 10, 12, and 14. *Methods:* 4 experimental groups were examined: non-pregnant, *in vivo* pregnant, NT recipients, and manipulation control (MC) recipients. Maternal blood samples were collected and assayed for insulin-like growth factor-1 (IGF-1). Embryos were evaluated for embryonic disc diameter, morphology, nucleoli density and mitotic figure index.

Results: NT day 12 ($P \leq 0.03$) and day 14 ($P \leq 0.01$) embryos had increased nucleoli when compared to *in vivo* produced and MC embryos. NT day 14 embryos had an increased ($P \leq 0.03$) mitotic index when compared to *in vivo* produced day 14 embryos. *In vivo* produced day 12 embryos had greater ($P \leq 0.03$) embryonic disk diameters compared to NT and MC embryos, however, *in vivo* produced day 14 embryos had increased ($P \leq 0.01$) embryonic disk diameters only when compared to NT day 14 embryos. *In vivo* produced day 14 embryos were morphologically more advanced ($P \leq 0.01$) than NT and MC day 14 embryos. There were no significant differences between IGF-1 levels.

Conclusions: Nuclear transfer embryos appear to develop at a slower rate than their *in vivo* counterparts as demonstrated by their less advanced morphology and decreased embryonic disk size as gestation progresses. Due to the apparent increase in nucleoli and mitotic figures within the trophoblast cell population of NT embryos, it would seem that the cell cycle is either adversely affected, possibly by incomplete re-programming of that portion of the embryonic genome, or the embryo itself is attempting to compensate for earlier, inadequate growth. The mechanical perturbations caused by the nuclear transfer technique itself also appear to retard development but not to the extent that the actual cloning process does as MC embryos often appear to re-cover some of the developmental ground they lost as they approach day 14. It would appear from these results that increasing the accuracy of the re-programming events would contribute to increasing the survival rate of NT embryos; however the exact means by which to accomplish this are still unknown and require further study.