

Improving the efficiency of nuclear transfer using stem cells

H. Zhu, J. Craig, P.W. Dyce, Nicole Sunnen and J. Li
Department of Animal and Poultry Science, University of Guelph
519-824-4120 ext. 52713, jli@uoguelph.ca

Background

The efficiency of nuclear transfer (NT) in pigs is extremely low. One of the current hypotheses is that the type of donor cell plays a major role in the success of NT. Stem cells are proposed to be ideal donor cells due to less differentiated state than adult cells. To test the suitability of using stem cells isolated from porcine fetuses as nuclear transfer donor cells, we have compared the pre-implantation development competency of embryos cloned from stem cells and fibroblast cells.

Objectives

1. To determine the suitability of using porcine stem cells as nuclear transfer donor cells, by evaluating pre-implantation development.
2. To investigate nuclear remodelling events that occurs after nuclear transfer.
3. To evaluate chromosomal abnormalities of fibroblast and stem cells in culture.

Results

- Stem cells show significantly less abnormal chromosomal abnormalities than fibroblast cells after long term *in vitro* culture (more than a month; Table 1).
- A higher proportion of stem cell NT embryos had the expected nuclear configuration (1 pronuclei) following NT than fibroblast NT embryos (Table 1)
- A higher percentage of embryos derived from stem cells develop to the two cell, blastocyst stage than those from fibroblast cells, and show significant increases in the cell number per blastocyst.

Table 1: Comparison of cloning efficiency of using stem cells and fetal fibroblasts as donor

Donor cell	Abnormal karyotype (%)	1 Pronuclei (%)	Cleavage	Blastocyst	Cell number / blastocyst
Stem cell	$1.4 \pm 0.18^{**}$	$79.6 \pm 2.2^*$	69.6 ± 3.6	$26.5 \pm 1.7^*$	$29.6 \pm 1.3^*$
Fetal Fibroblasts	4.7 ± 0.19	65.6 ± 1.5	71.2 ± 3.1	20.9 ± 1.4	25.1 ± 1.3

* $p < 0.05$, ** $p < 0.01$ compared to the group cloned using fibroblasts as donor.

Conclusions

Embryos reconstructed with stem cells derived from porcine fetuses show better pre-implantation development than those from fibroblast cells. The results suggest that porcine stem cells may be better donor cells for nuclear transfer than fibroblast cells in the pig.

Acknowledgments

This work was supported by NSERC, Agriculture and Agri-Food Canada, Ontario Ministry of Agriculture and Food, Ontario Pork, and the Food System Biotechnology Center at the University of Guelph.