Efficient production of transgenic piglets using ICSI-SMGT in combination with RecA protein

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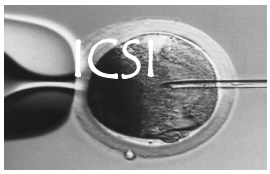
Objective: investigate whether integration and expression of exogenous DNA into the pig genome is improved by RecA in a SMGT-ICSI system

Material and Methods

Embryo transfer

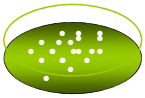
IVM

RecA:ssDNA:sperm



386 oocytes

TALP

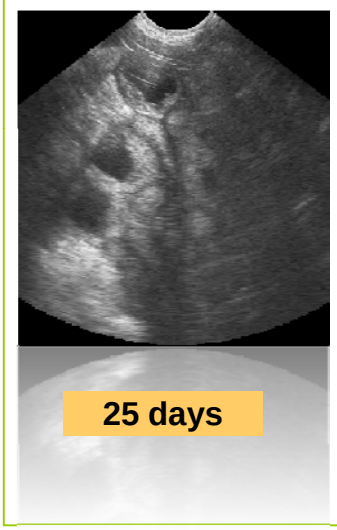


3 prepuberal gilts

Embryo transfer

1h after ICSI

Pregnancy diagnosis



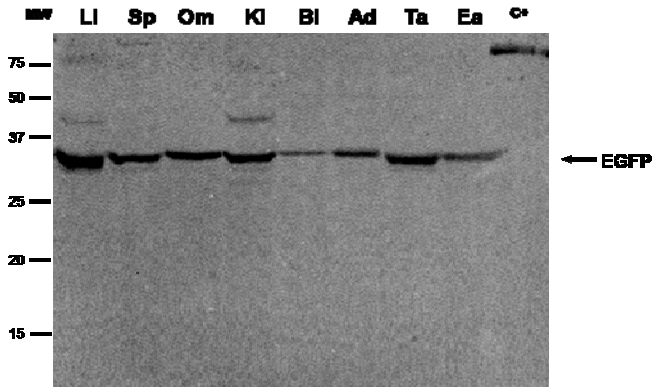
Piglets analysis

- Protein expression: Western Blot
- Integration: PCR

Results

In 6 animals (4 lives and 2 stillborn) were positive for *PCR* technique. All analysed tissues (liver, spleen, greater omentum, kidney, blood, adipose, tail and ear) were positive in live piglets and only skin sample was positive in stillborn animals. In samples from positive animals, *Western Blot* analysis was used to detect the expression of EGFP protein, and was detected in all of tissues analyzed (100%)

	Embryos transferred	Pregnancy	Litter size
Sow 1	119	+	3
Sow 2	125	+	4
Sow 3	142	Return to oestrus after > 21 days	-
Total	128.6 11.9	66.67%	7



Conclusion: This method of "active transgenesis" using recombinases improve transgenic rate animals (86%) and reduce the mosaicism (66.7%) in comparison to passive transgenesis methods

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