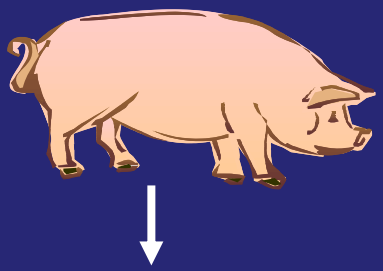
Effect of MAPK Inhibitors on Meiotic Maturation and Embryonic Development in Pigs

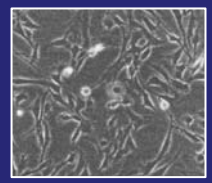
Presented by: Laura Crawford
Supervisor: Dr. V. Bordignon
Department of Animal Science

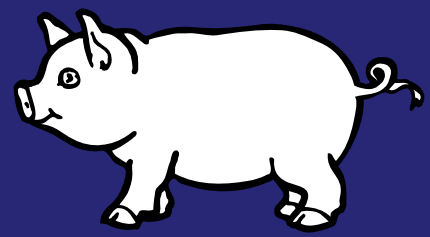
“Reprogramming of somatic cell differentiation by nuclear transplantation”



Oocytes matured in vitro

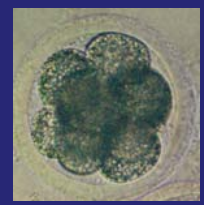
Cell culture

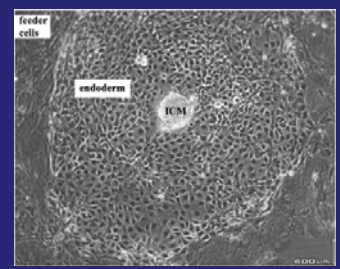




Enucleation and nuclear transfer

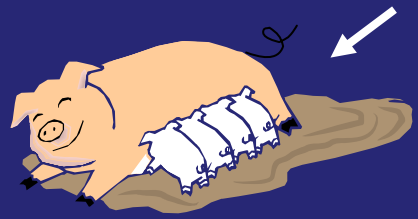
Regenerative medicine





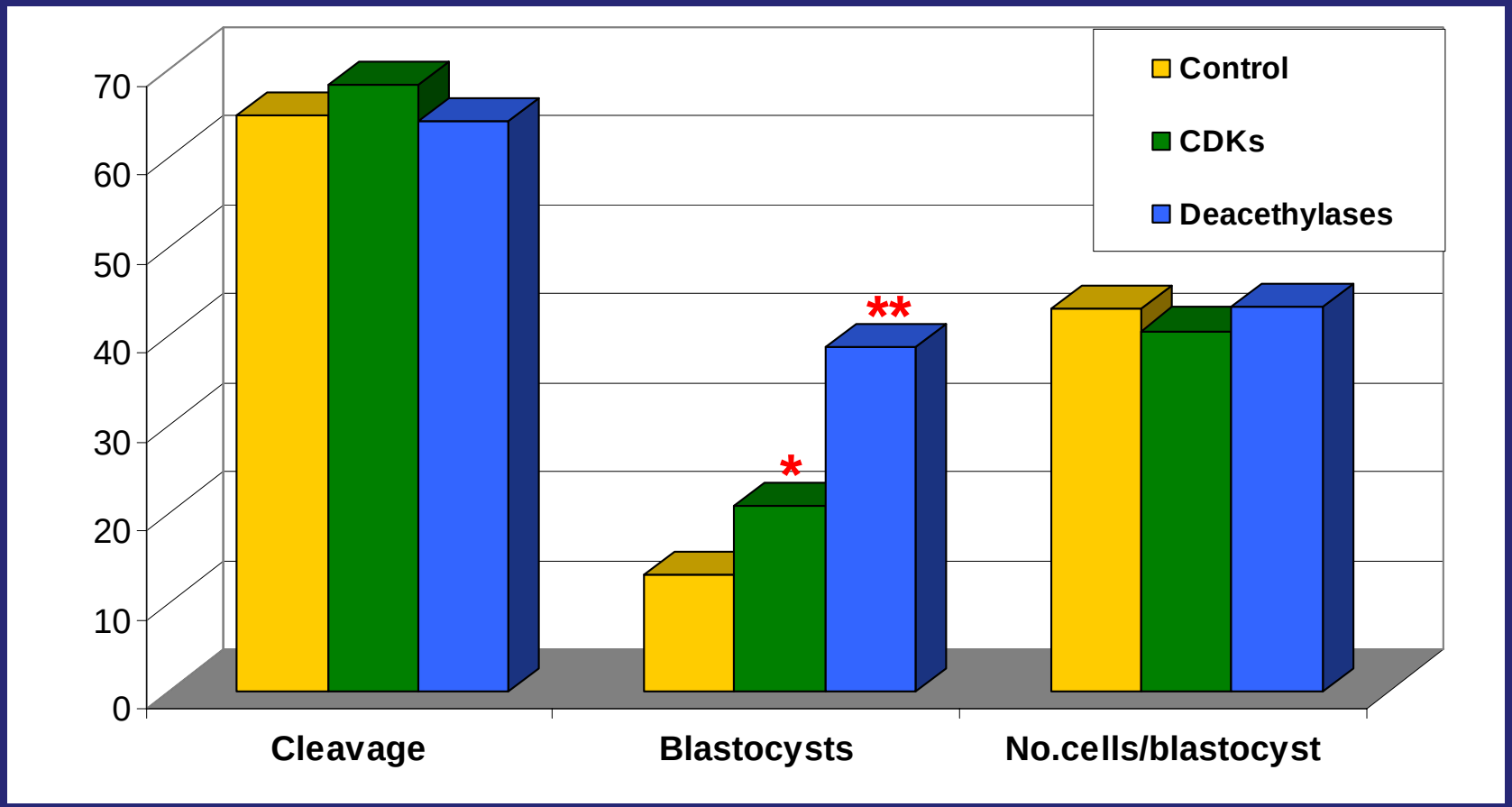
Embryonic stem cell lines

Embryo transfer



Cloned piglets:
-transgenic animals
-disease models
-xenotransplantation

Treatment of nuclear donor cells with modifiers of epigenetic factors increases development of nuclear transfer embryos



(Research being conducted by Limei Che)

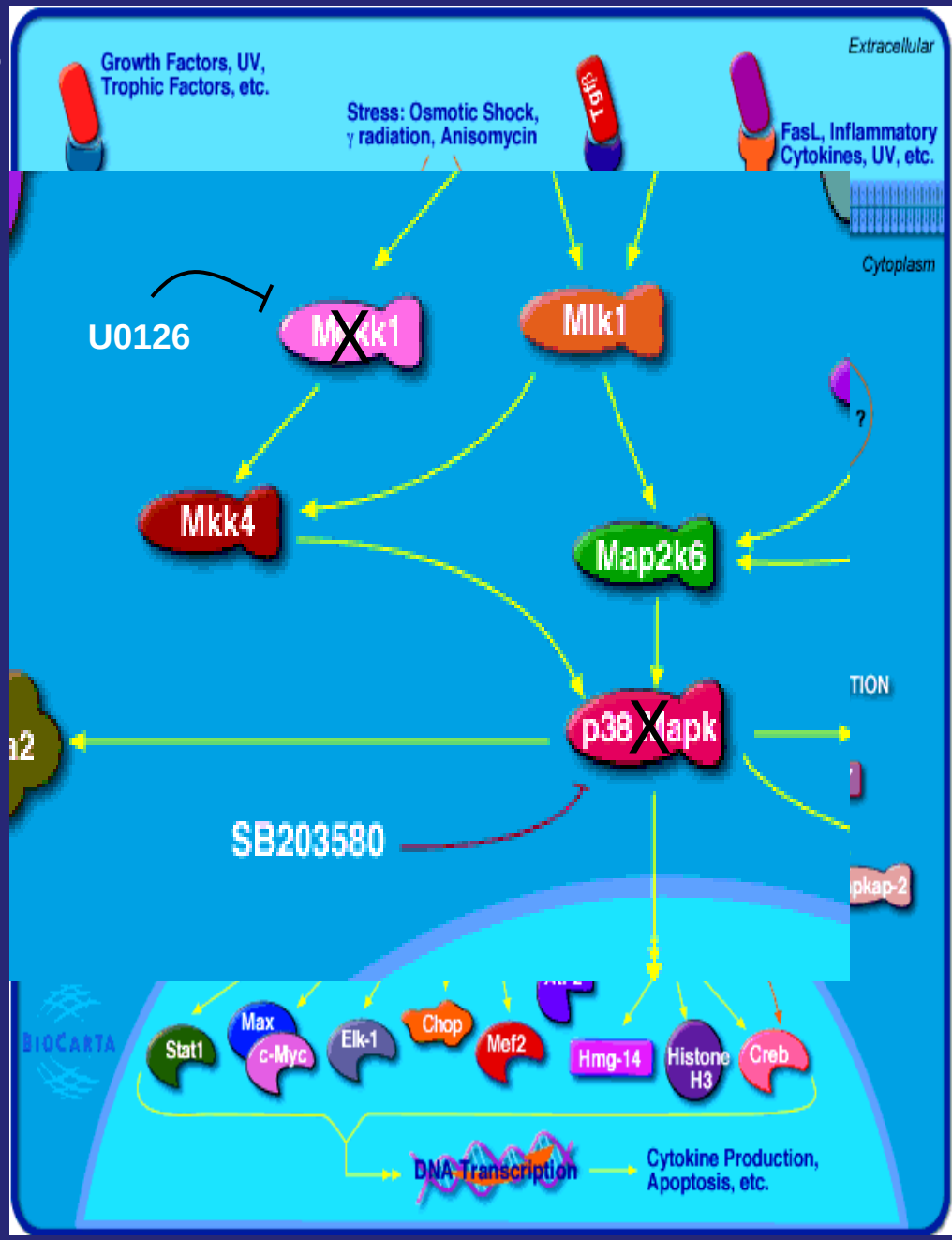
Hypothesis and objectives

Hypothesis:

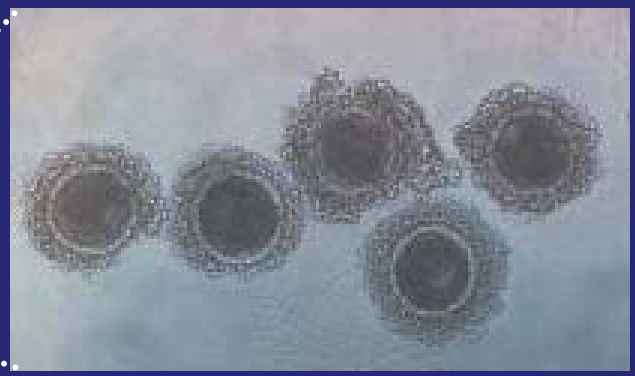
- The blockage of the meiotic resumption for 22 h with inhibitors of MAPK pathway will improve developmental rate of porcine embryos in vitro

Objectives:

- Verify whether inhibitors of MAPK pathway (*U0126 and SB203580*) prevent meiotic resumption of pig oocytes
- Assess the development of embryos produced from oocytes treated with MAPK inhibitors in the presence (or not) of follicular fluid



Experimental protocol



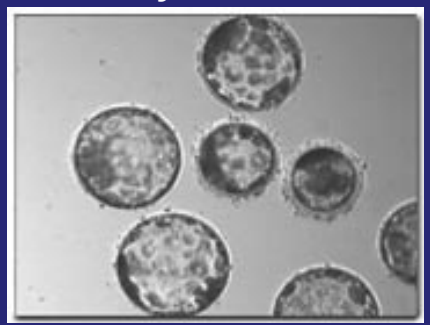
Control U0126 SB203580

Meiotic stage after 22 hrs IVM

Meiotic stage after 48 hrs IVM

Parthenogenetic Activation

Blastocyst stage embryos at D-7

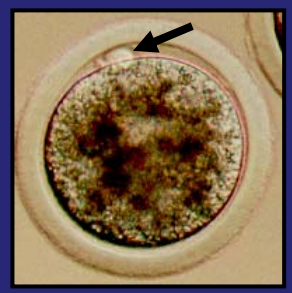


Maturing Oocytes

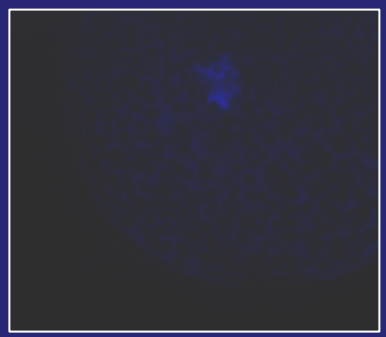
Oocyte-cumulus complex

Expanded OCC

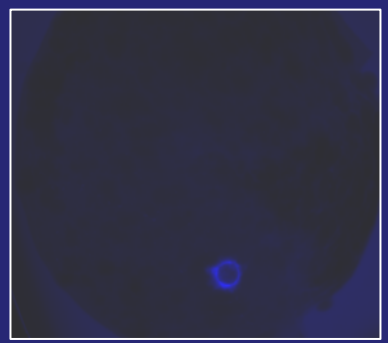
Immature Oocyte



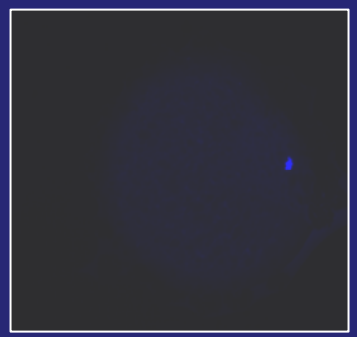
Mature Oocyte w/ Polar Body



GV



GVBD



M1

M2

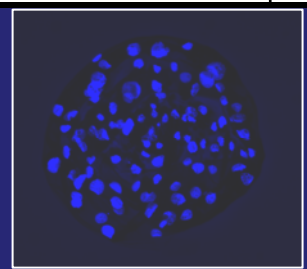
Effect of MAPK Inhibitors on Meiotic Maturation

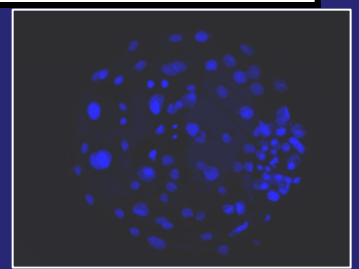
Inhibitor	Meiotically Arested oocytes at 22hrs	Meiotically matured oocytes at 48hrs
Control	31.5%[a]	58.4%
MEK1/2 (*U0126*)	79.0%[b]	57.4%
p38MAPK (*SB203580*)	37.0%[a]	66.7%

* Different letters in the same column denotes statistical significance ($P<0.05$)

In vitro development to blastocyst stage at D-7

Treatment	Follicular fluid (20 %)	# oocytes	Blastocysts (%)	Cell #/embryo
Control	+	61	45.9	38
	-	73	45.2	45.7
U0126 [10µM]	+	58	36.2	35.1
	-	51	43.1	39.6
SB203580 [10µM]	+	46	41.3	32.7
	-	55	49.1	57.9*





Conclusions

- MEK1 (U0126) inhibitor prevents meiotic resumption of porcine oocytes
- Oocytes treated with inhibitors of MEK1 (U0126) and p38MAPK (SB203580) complete meiotic maturation similarly to untreated oocytes
- Development to blastocyst stage was not significantly affected by oocyte treatments

Thanks!

•To NSERC for providing me with this wonderful opportunity!

•To Dr. Bordignon for allowing me to work in his amazing lab and to learn so much!