Genetic Engineering & Model validation

institut Clinique de la Souris

Pipeline for the generation of new mouse models

Design construction

ES cells culture

Microinjection Blastocysts

Breeding

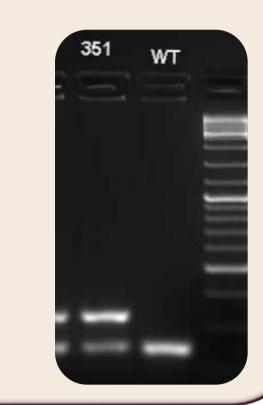**Genotyping**

Gene expression

Genetic Engineering

- Expert advices
- Targeting strategy in accordance with the customer needs
- In-house projects / new tools
- High Scientific/Technical output

- ES cell validation by long range PCR and Southern blot
- QC validation of EUCOMM and KOMP clones

Highly experienced team:
More than 1300 targeted mutagenesis
More than 200 transgenic projects

351 WT

Modification through ES cell

- Knock-out Constitutive, conditional
- Knock- tissues specific and /or inducible
- knock In, genetic targeted insertion
(Point mutation, reporter tag, cell tracking…)
- Humanization

Nuclease genome engineering

- CRISPR/Cas9 *NEW offer * !
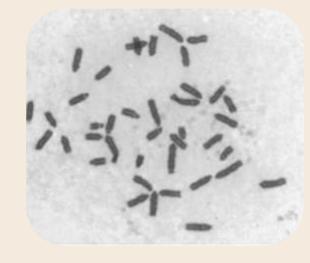
Pronucleus modification

- By injection of fertilized eggs
- Plasmid or BAC

ES cell service

- Derivation of WT and mutant new ES cell lines from different genetic background
- *In vitro* differentiation

In development: Rat ES cells

Male ES cell line

ES cell

- In-house derived C57BL/6N , Sv129/Pas and BALB/cN ES cell line
- Electroporation
- Selection, Amplification, Picking
- Karyotyping

Microinjection

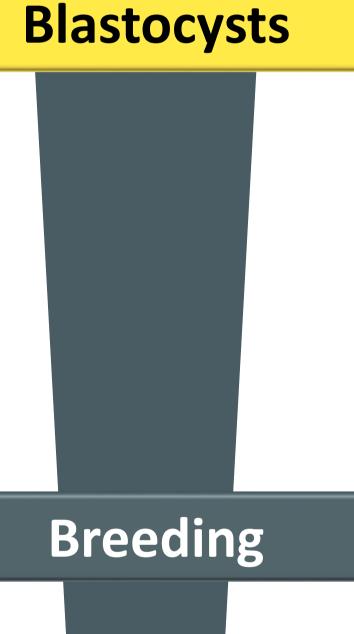- Injection of validated ES cell into blastocysts
- Injection of DNA into fertilized eggs for random insertion of transgenes

Efficient germ line transmission rate:
More than 75 %
GLT for more than 400 IMPC clones

GLT rate

Germ line transmission per injection
(all IKMC & consortia centers)

IKMC pipeline efficiency
ICS metrics PHENOMIN 2012-2013

Animal facility

- Maternity and breeding facility
- Capacity of 45 000 mice
- Improved PEI over the last years
- *In vitro* fertilization running in different genetic background (including C57BL/6N)
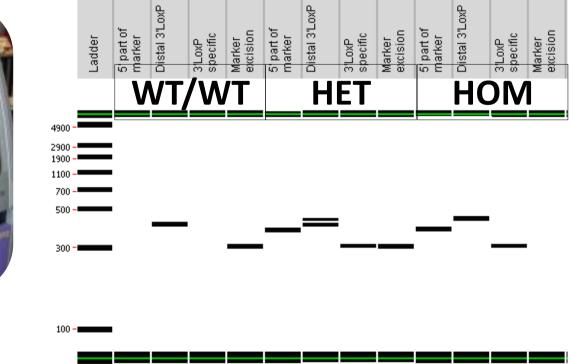

Chimera

Genotyping service

- Fully automated genotyping platform
- Validation of genetic modification
- Confirmation of mutation by sequencing
- Short range PCR or qPCR

Easy-to-use genotyping protocol provided for each generated line

WT/WT   HET   HOM
Typical genotyping screen

Validation of genetic engineered mouse model

Gene expression validation **and** copy number determination	Functional validation
Northern Blot, Southern Blot	Immunohistochemistry
qPCR, RT-qPCR	Western Blot

Knock-out, Knock-in, siRNA model, Cre or CreER^T2, Conditional model. . . .

← Cre-eGFP
← eGFP
GFP antibody