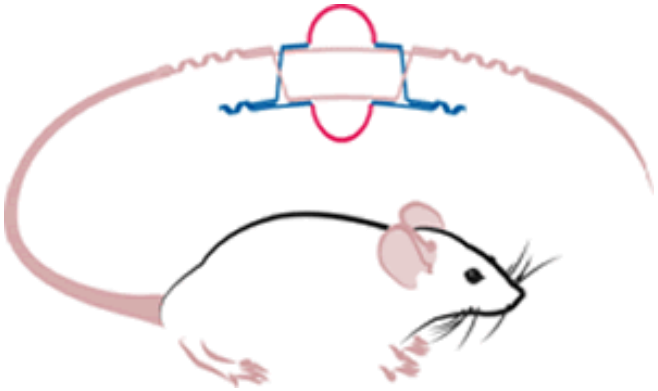
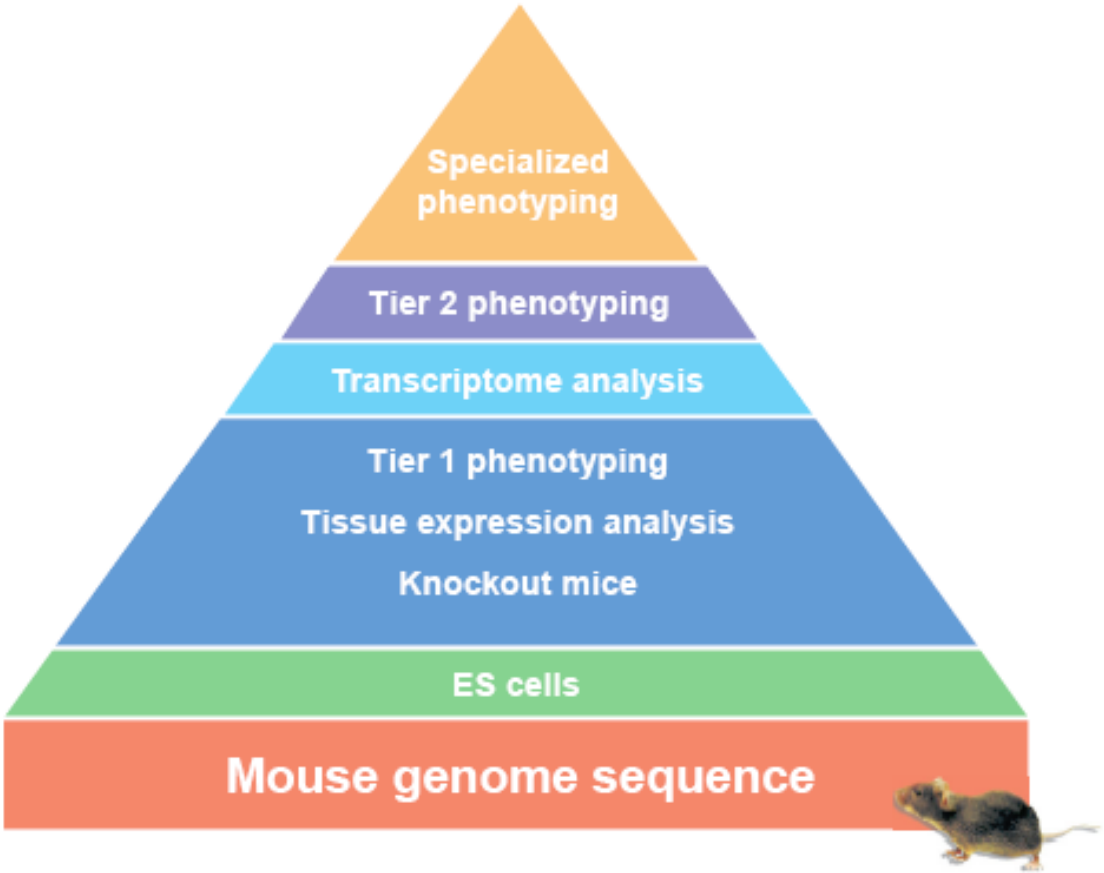


Knockout Mouse Project (KOMP) and Knockout Mouse Production and Phenotyping (KOMP^2)

Colin Fletcher, Ph.D.
Common Fund
Apr. 9, 2013



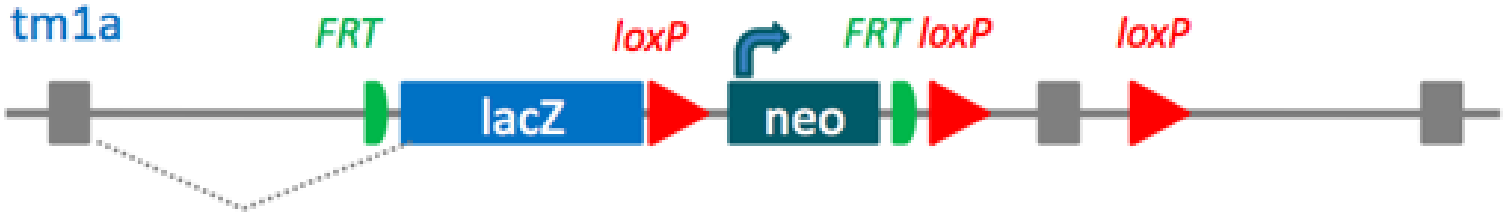
The vision for KOMP was articulated in a meeting at the Banbury Center, Cold Spring Harbor in 2003, calling for high throughput production of gene knockouts, and phenotyping, for every gene in the mouse genome.

KOMP

- *"…a high-throughput international effort to produce…knockouts for all mouse genes, and place these resources into the public domain."*
- The KOMP was launched in 2006 by NIH
  - $56.6 million over 5 years from the ICs
  - a goal of creating 8,500 ES cell lines
  - alleles are nulls or conditional-ready, contain reporter
- The KOMP Research Network
  - Two production centers
  - A repository
  - A Data Coordination Center
  - Three ES cell development projects
- KOMP and EUCOMM along with other international efforts formed the International Knockout Mouse Consortium (IKMC) and have jointly produced > 17,000 mutant ES cell lines and made them available from public repositories.

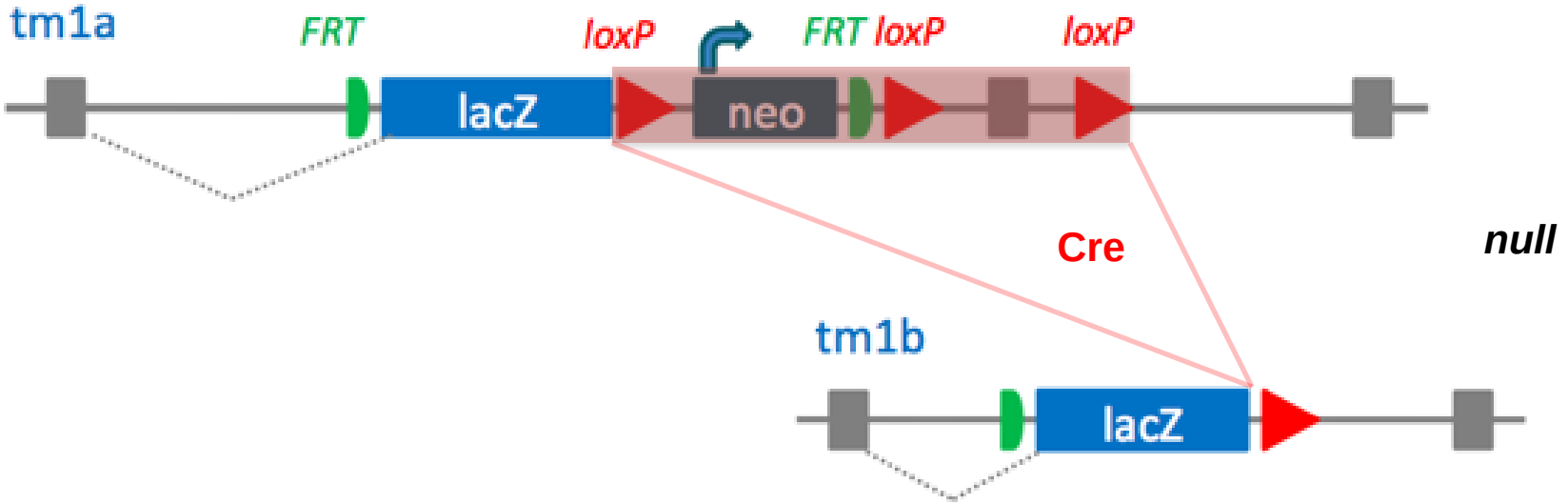
KOMP Allele Design

CSD



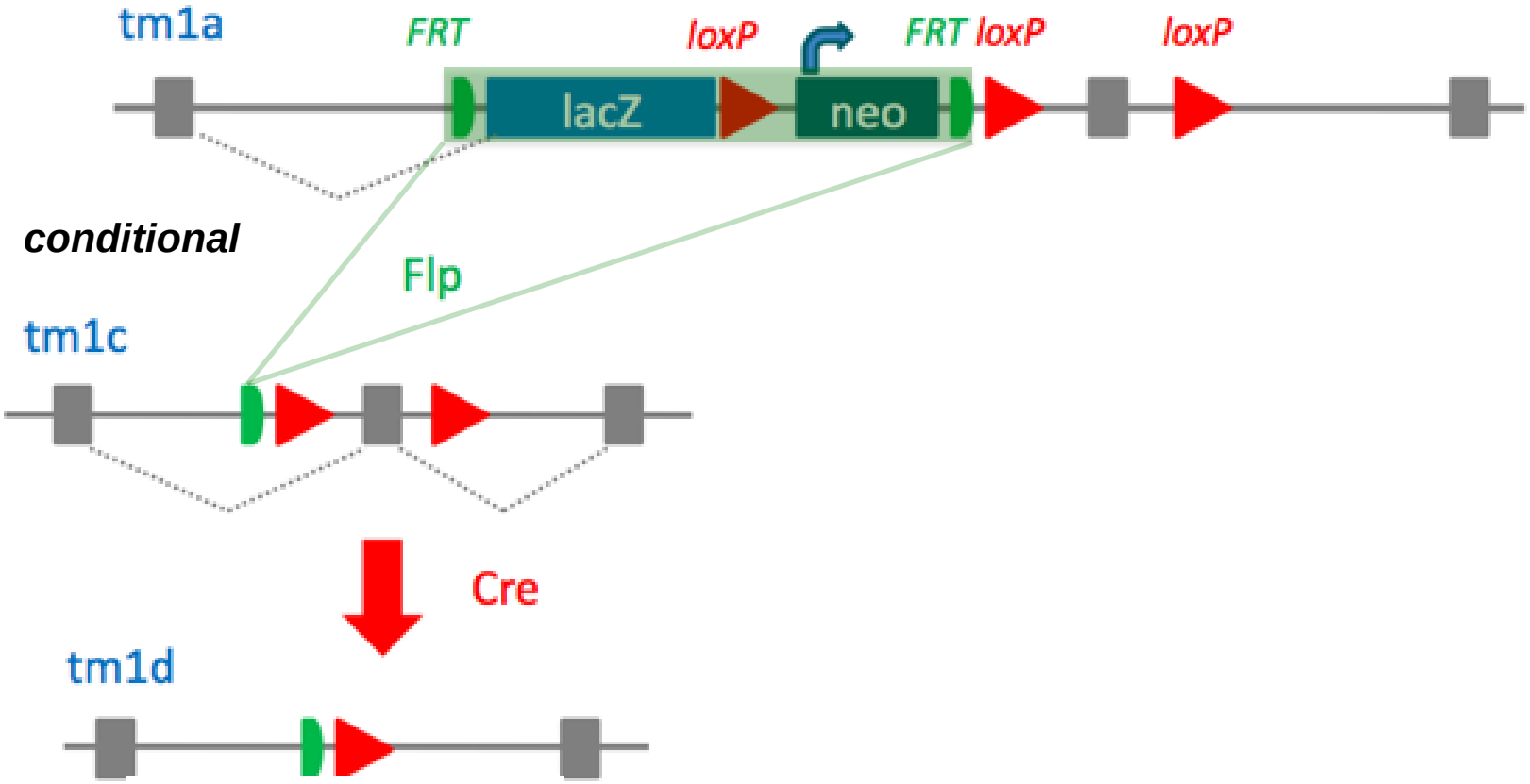
KOMP Allele Design

CSD



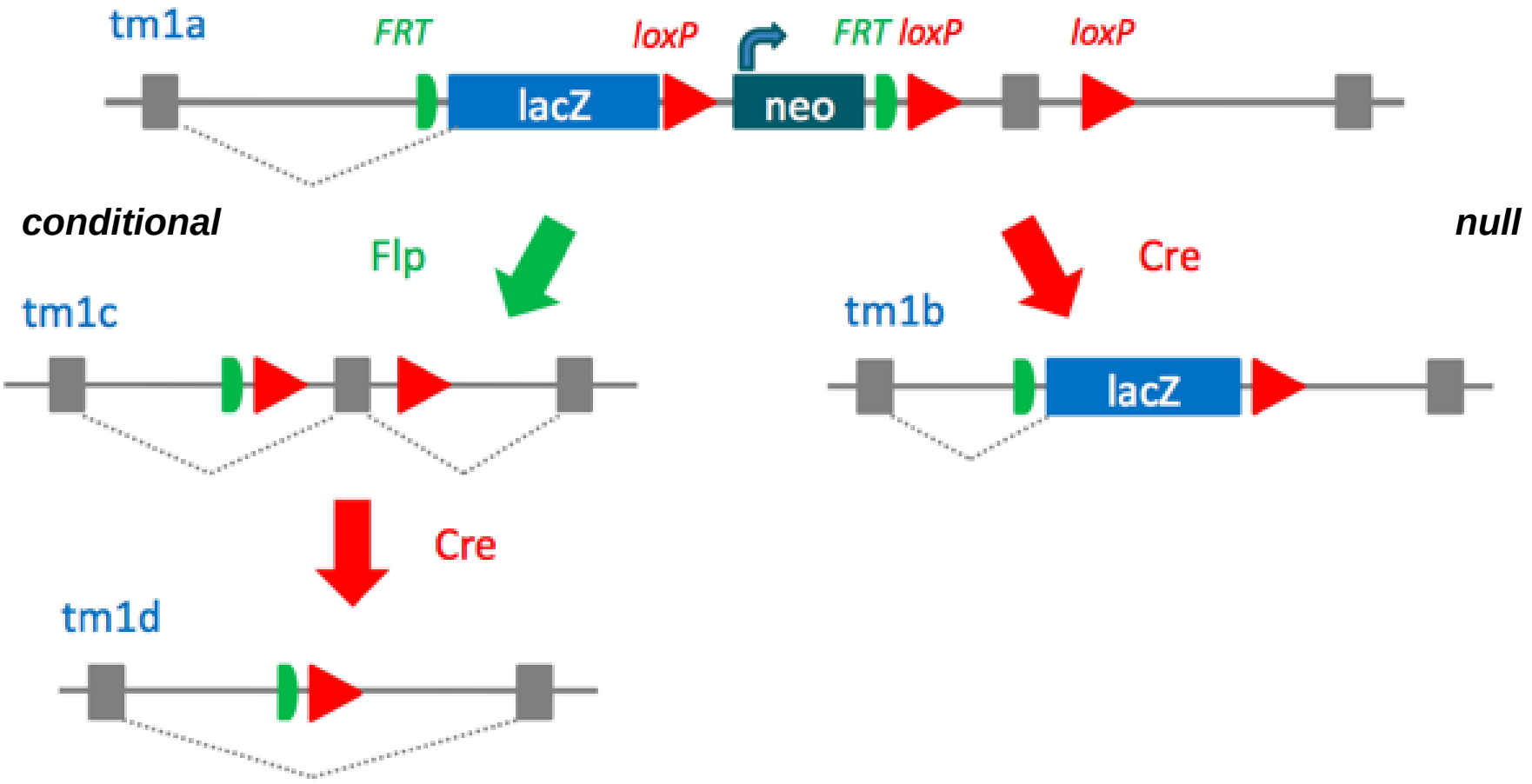
KOMP Allele Design

CSD

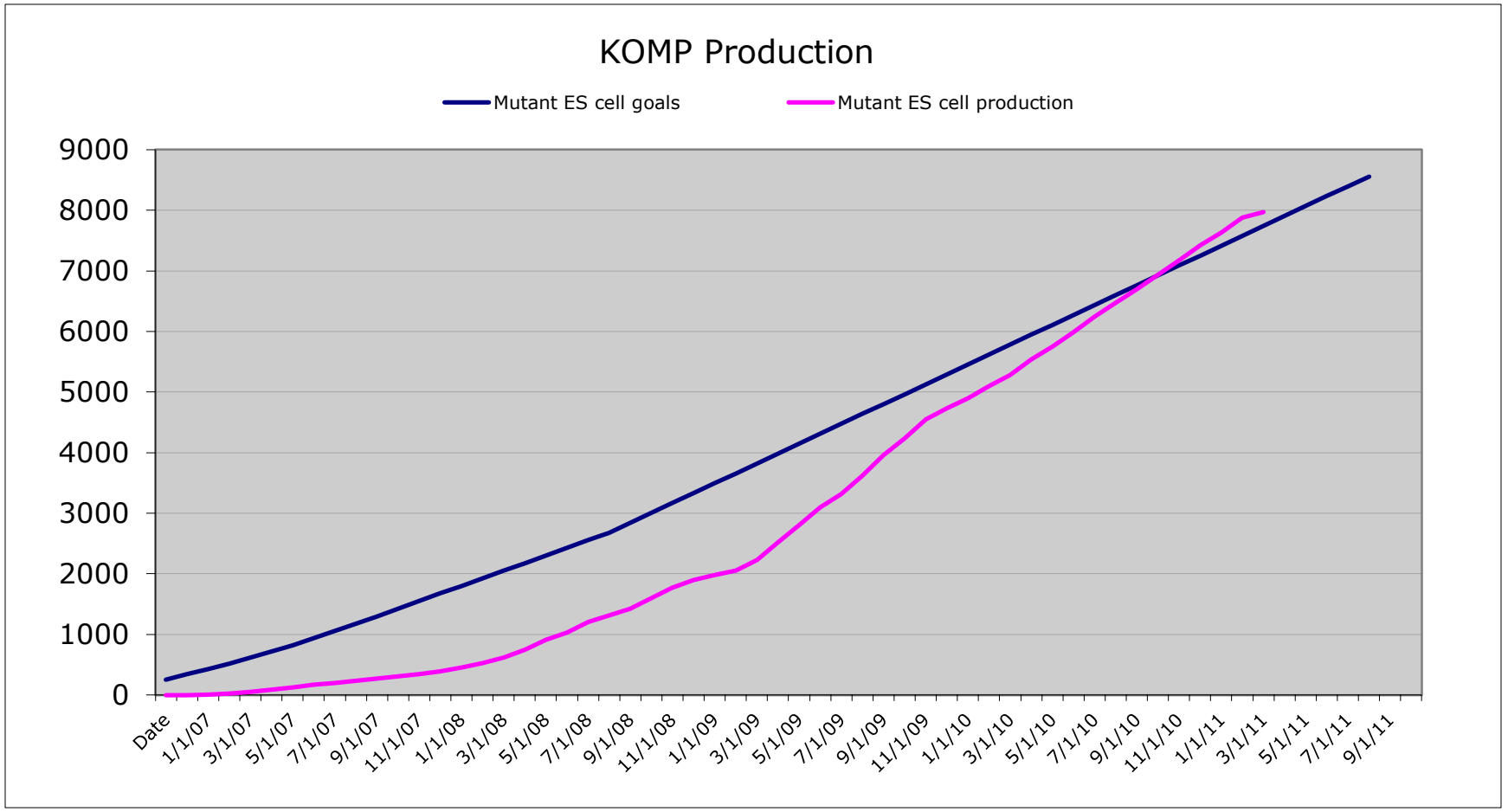


KOMP Allele Design

CSD



Goals and Progress 2006-2011



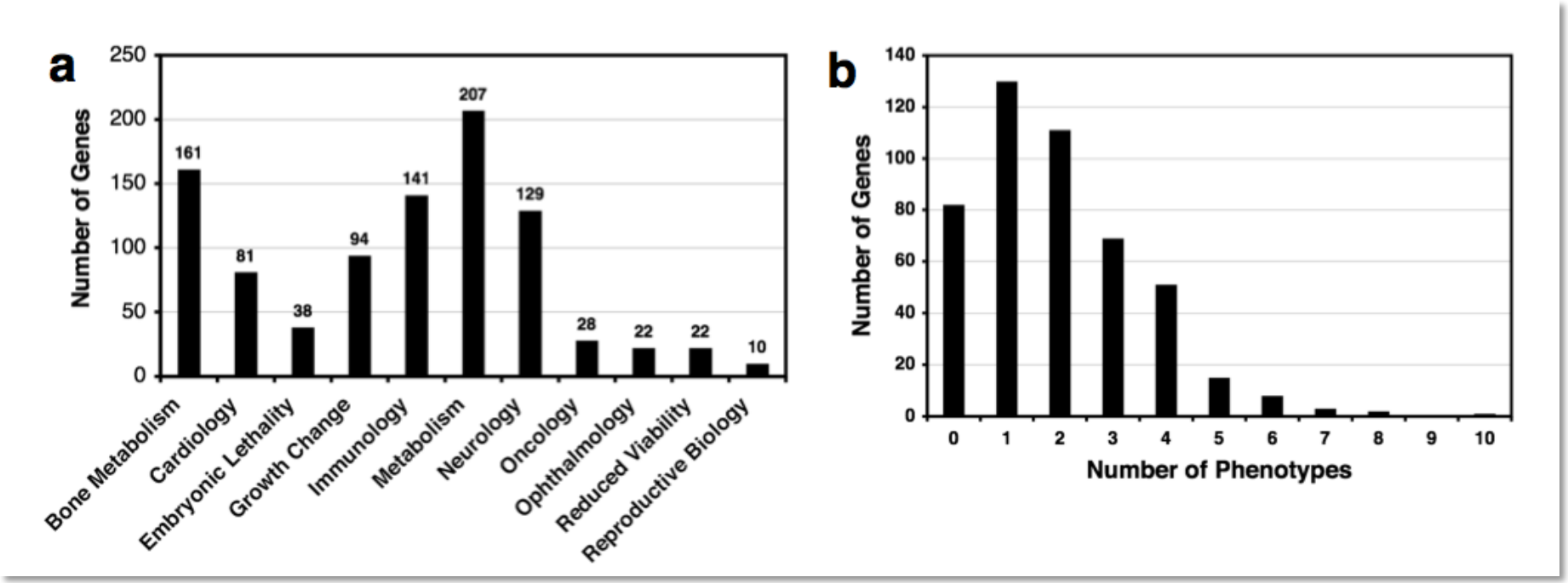
Rationale for a Large-scale KOMP²

Supporting a broad phenotyping effort would provide the following advantages:

- Eliminate the redundancy and waste inherent in the “cottage industry” approach
- Each mutant mouse will be characterized for a broad set of phenotypes to allow direct comparisons & result in a thorough description of gene function.
- Novel genes will be brought to light that would otherwise be ignored
- Quality standards will be established and maintained, so the data will be of the highest reliability.
- The risk of not finding a phenotype will be greatly reduced.
- Important, but unpublishable, negative results will be captured.
- Potential for breakthrough discoveries

Genentech/Lexicon Mouse Phenotype Project

472 Mouse knockouts were broadly phenotyped



130 (27%) strains had 1 phenotype
245 (52%) strains had 2-5 phenotypes

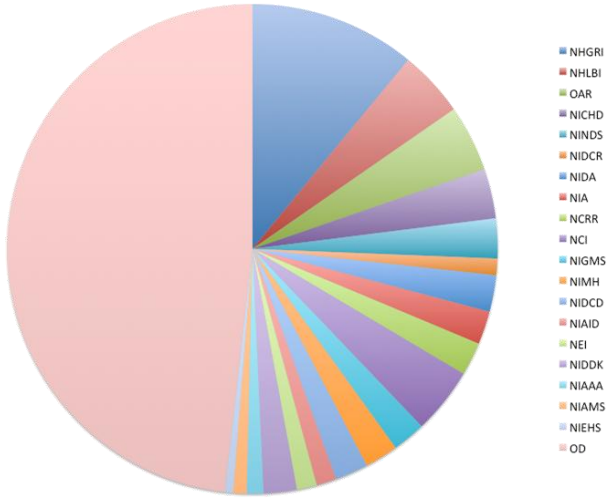
Broad phenotyping shows that knockouts have pleiotropic effects

Andy Peterson, Genentech

Funding, Awards, and Coordination

Funding:

- NIH Common Fund – 50%
- Participating NIH ICs – 50%
- $110M over five years



Common Fund Website:

- http://commonfund.nih.gov/KOMP2/

Production and Phenotyping Centers:

- UCDavis (with Sick Kids and Charles River)
- Baylor (with MRC Harwell, Wellcome Trust Sanger)
- The Jackson Laboratories

Data Coordination Center and Data Base:

- European Bioinformatics Institute (with MRC Harwell)

Repository:

- The KOMP Repository, UCDavis

Coordinating with The International Mouse Phenotyping Consortium

KOMP² Tests Multiple Systems

Neurological/ Behaviour
- Open Field
- Modified SHIRPA/Dysmorphology
- Grip Strength
- Acoustic Startle/PPI
- Pain Test

Metabolism
- Weight
- Calorimetry
- Intraperitoneal Glucose Tolerance Test
- Body Composition (DEXA)
- Clinical Blood Chemistry
- Insulin Blood Level

Cardiovascular
- ECG / Echo
- Heart Weight

Pulmonary
- Challenge Whole Body Plethysmography

Reproduction
- Fertility

Sensory
- Auditory Brain Stem Response (2+2)
- Slit Lamp
- Opthalmoscope

Musculo-skeletal
- Grip Strength
- Body Composition (DEXA)
- X ray (5 + 5)

Immune
- Hematology
- FACS analysis blood/spleen

General
- Modified SHIRPA/Dysmorphology
- Gross Pathology & Tissue Collection (2+2)
- Tissue embedding &Block Banking (2+2)
- Histopathology (2+2) - from blocks where required

Tests in development or under consideration

Mandatory tests

Non-mandatory tests

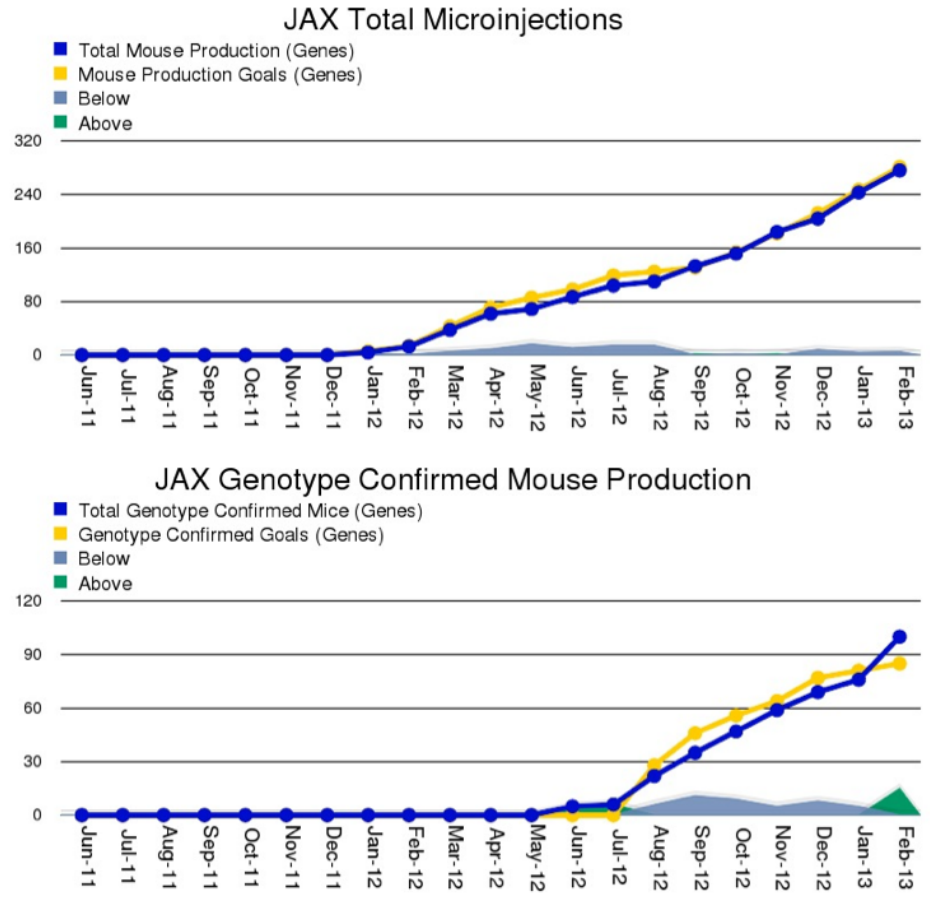
Coordinating with The International Mouse Phenotyping Consortium

Progress Tracking by DCC

JAX

Download as CSV Download MI image Download GC image

Status	Current Total (up to and inc. February)	Last Complete Month (February)
All genes	734	16
ES Cell QC	319	20
ES QC Confirmed	270	28
ES QC Failed	17	4
Microinjections	276	33
Genotype Confirmed Mice	100	24
Intent to Phenotype	78	13
Cre Excision Complete	27	7
Phenotyping Complete	0	0



https://lims.mousebiology.org/dashboards/remote

(New!) Embryonic Lethal Pipeline

HET × HET

Genotype pups (P14-21)

0% HOM

HET × HET

Embryo viability checkpoint

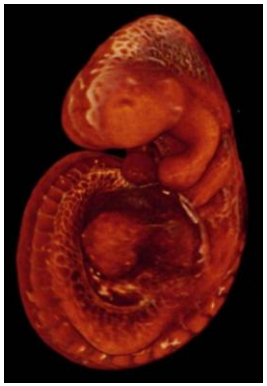
HET

Adult HET Phenotyping

E12.5

Embryo Assessment

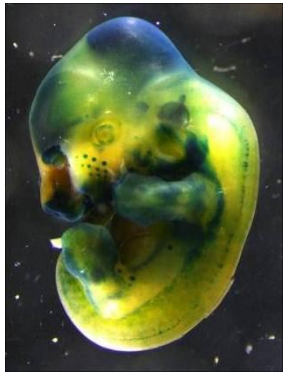
OPT



E9.5 HOM

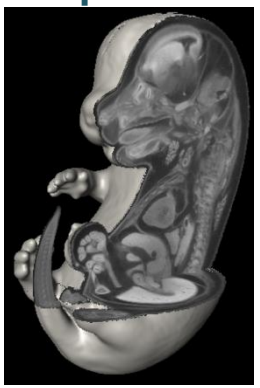
+ Targeted Histopathology
Embryo & Placenta

LacZ



TCF/Lef-LacZ reporter

µCT

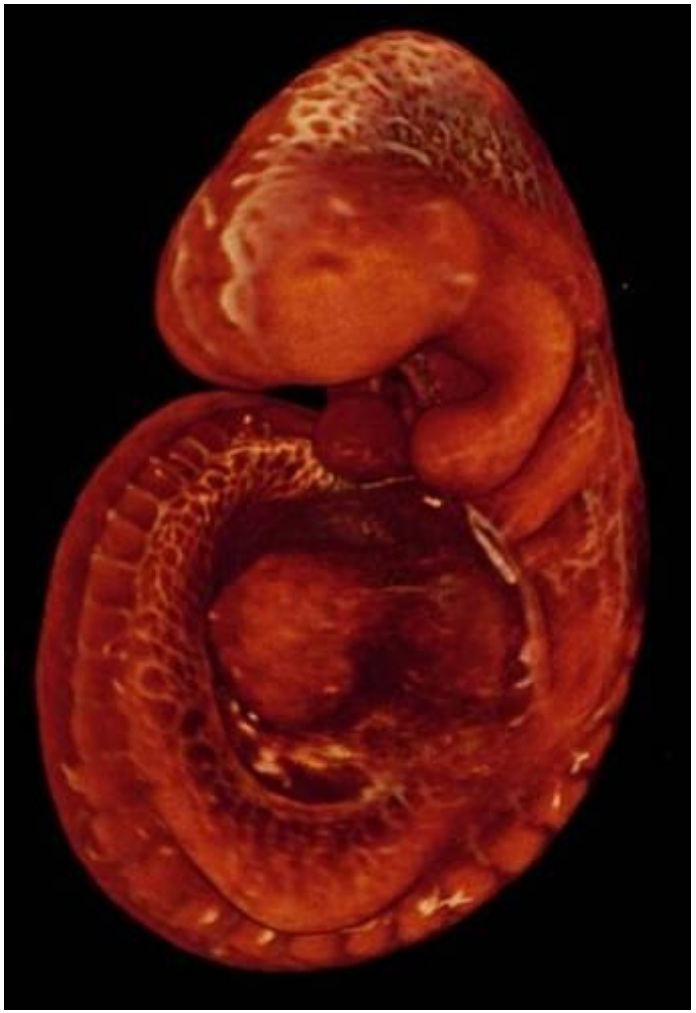


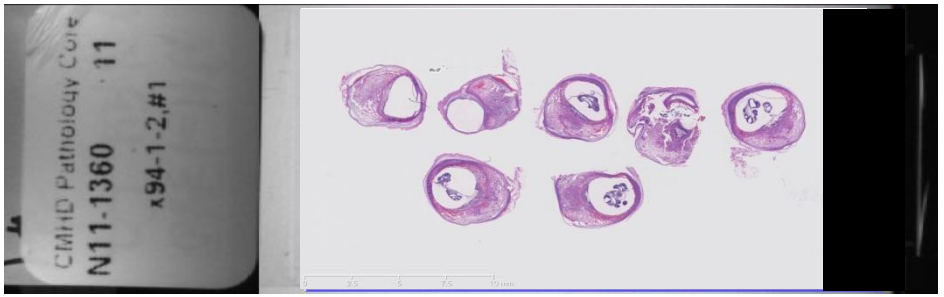
E15.5 HOM

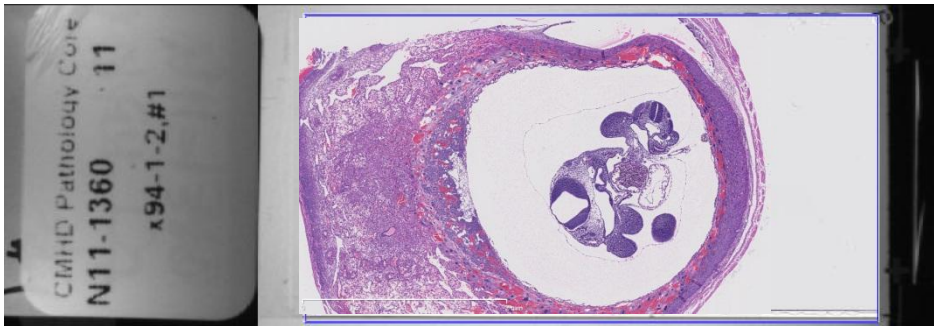
+ Targeted Histopathology
Embryo & Placenta

Imaging Mouse Embryos

OPT and Histopathology



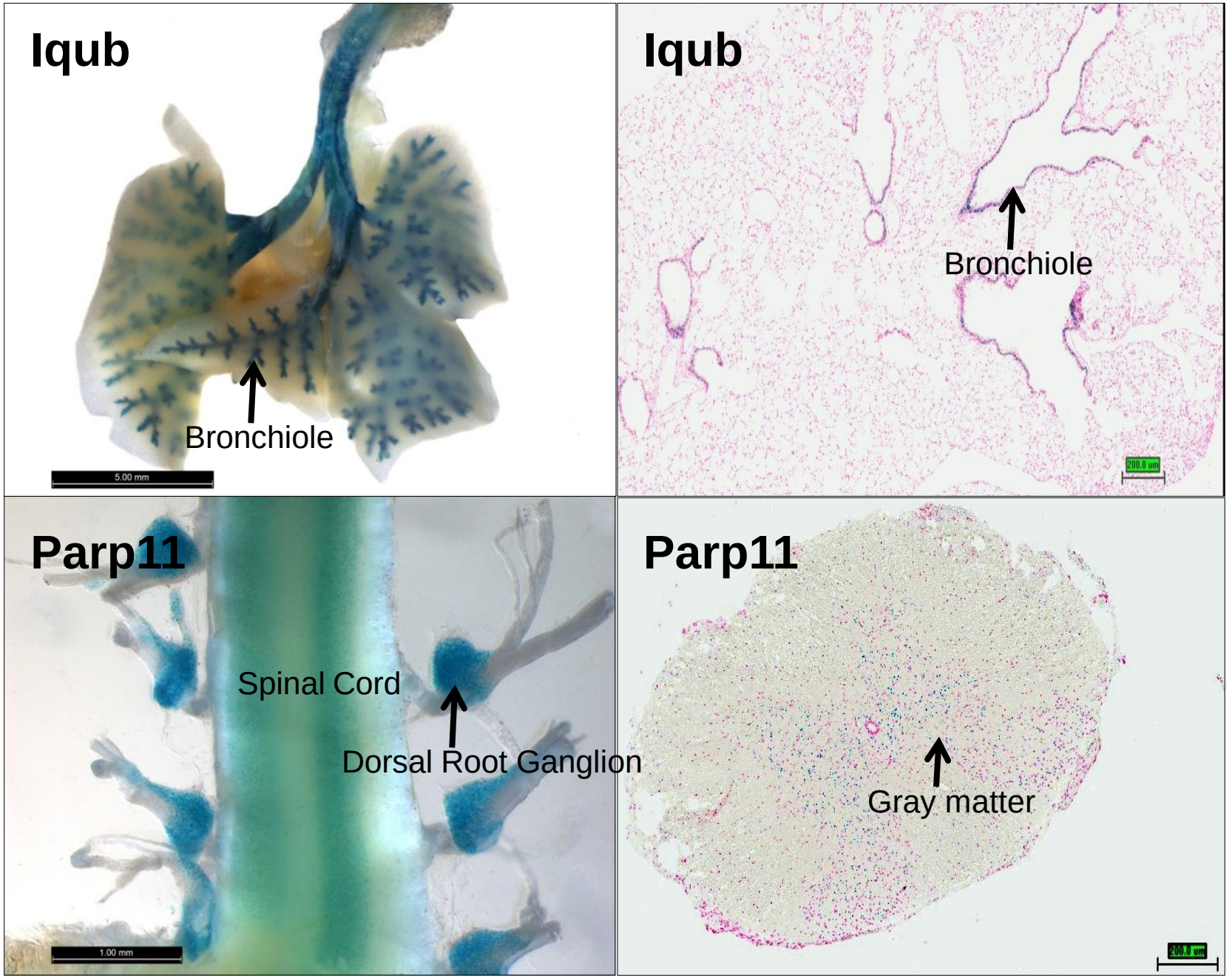




Automated analysis using image registration

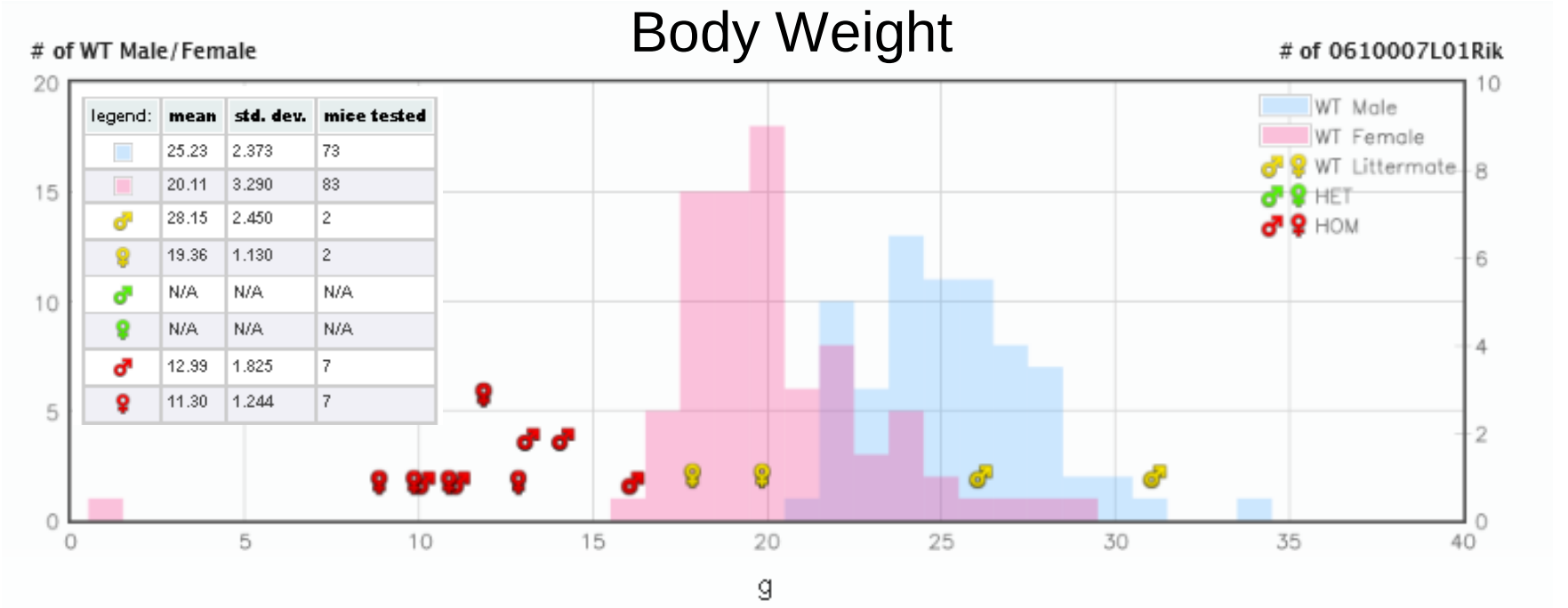
Histological analysis of embryo and placenta

Representation of Phenotype Data?


Iqub
Bronchiole
5.00 mm
Iqub
Bronchiole
200.0 um
Parp11
Spinal Cord
Dorsal Root Ganglion
1.00 mm
Parp11
Gray matter
200.0 um

K312 Phenotype Screen: 0610007L01Rik

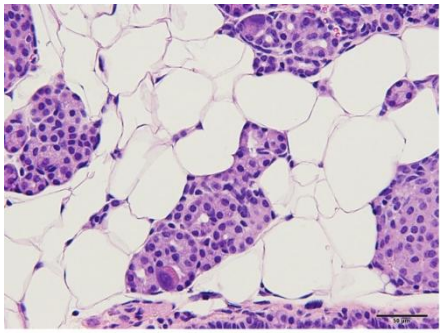
Body Weight



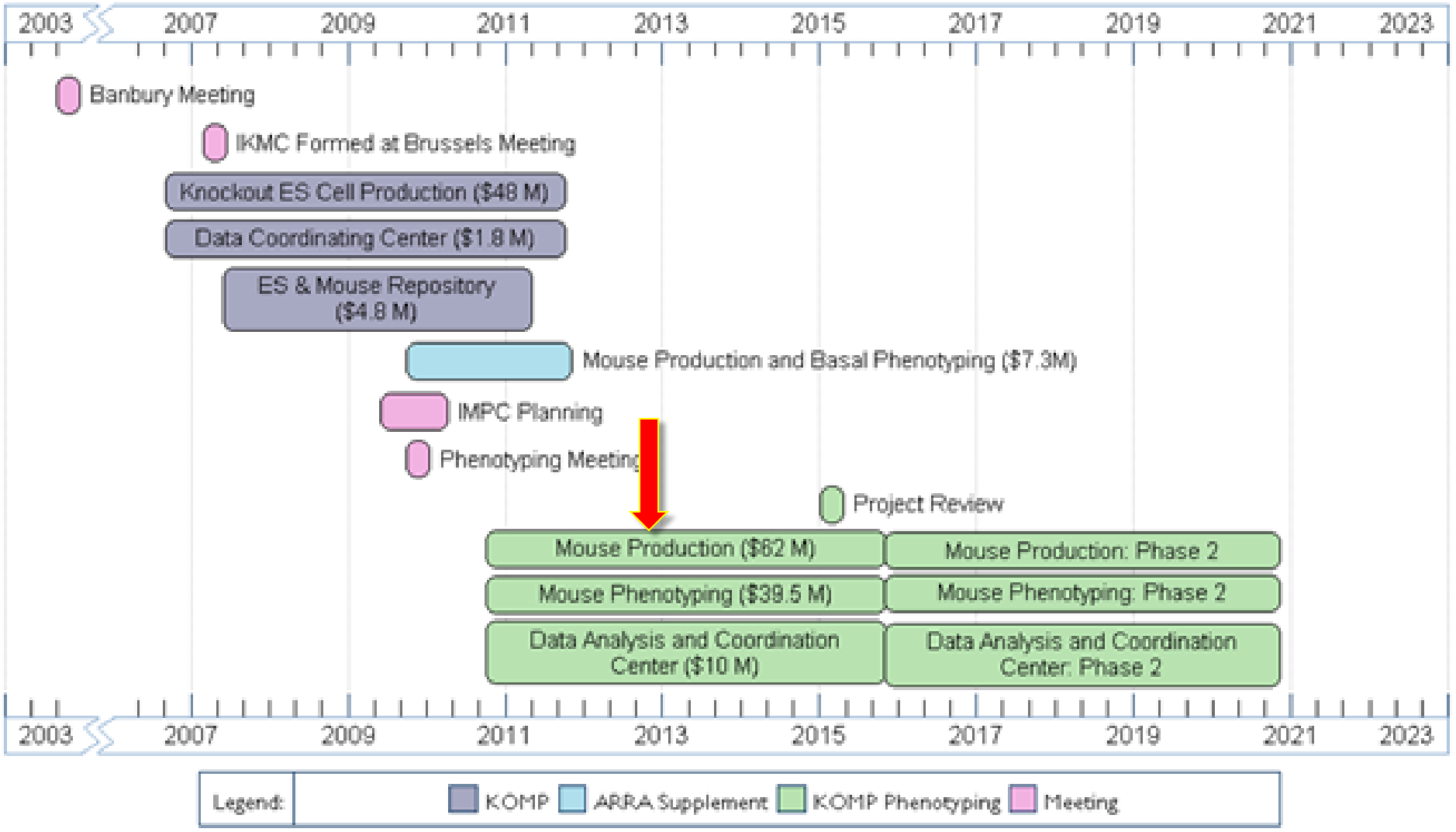
Clinical Features 0610007LO1Rik:

- Stunted
- Little adipose tissue
- Distended cecum and colon
- Atrophied pancreas

Thymus has a decreased cortical:medullary distinction and medulla is slightly decreased in size. Pancreas is small with marked fatty infiltration. The pancreas consists predominantly of ducts and islets with only few individual acinar cells which are enlarged, some are binucleated, and distended with zymogen granules..



KOMP/KOMP² Timeline



Thank You!