UK Medical Research Council

IMPC Production, Phenotyping, Engagement

Tom Weaver
Director, Mary Lyon Centre, MRC Harwell

International Mouse Phenotyping Consortium Launch!
28/29th September 2011
NIH, Washington DC, USA

Overview

MRC Harwell & the MRC IMPC

Engagement with Community

Operations – Management & Immediate Plans

Future Developments (if have time)

UK Medical Research Council

Established in 1911

Mission: Improve human health through world-class medical research

$1B Budget (1/30th size of NIH)

Over 4,000 people in 50 Institutes, Units and Centres

Rich Scientific History

- 29 Nobel Laureates => DNA Structure, Targeted Mutagenesis in Mice

Commitment to Basic & Translational Research

MRC Strategic Plan 2009-2014:
MRC Specific Committment to Mouse Genetics

Focus Funding on Mouse Models To:

- Understand mechanism of action of disease genes and pathways
- Identify new drug targets
- Develop better animal models of human disease

- ✓ **Work with partners to comprehensively phenotype mutants and share data**
  - $15M Support towards IMPC (Proportionate to $450M)
- Promote Animal Welfare => National Centre 3Rs (education, guidelines, r&d funding)

MRC Harwell: MGU and MLC Overview

Centre of Excellence in Mouse Genetics Since 1950

- Impact of Radiation on Genomes,
- X Chromosome Inactivation,
- Imprinting,
- Systematic Mutagenesis & Phenotyping,
- Mouse Models of Disease

Mammalian Genetics Unit ("MGU") – Science Lead

- Genetics and functional genomics research into a wide variety of disease models
- **10 Research Programs: Imprinting, Deafness, Neurobehavior and degeneration, Diabetes, Liver, Bone, Development,…**
- Steve Brown, Director

Mary Lyon Centre ("MLC") – Operational Lead

- National Infrastructure for Mouse Genetics
- SPF Vivarium (15k cages) & Dedicated Service Labs
- Service Provision to over 120 labs around the UK
- $1.5M/year "external" funding via services
- **High Throughput Production and Phenotyping**
- **National Biorepository & Distribution Centre**
- Tom Weaver, Director

MRC Harwell

Mary Lyon Centre

Operational Engine
Comprehensive Service Infrastructure

Production, Archiving, Distribution

- Live mice; frozen embryos/sperm, >1500 strains & 10K ENU archive; 300 EMMA strains distributed to 26 countries.

Mutagenesis Programs

- Transgenesis; Chemical/Sensitized Screens

Adult and Embryonic Phenotyping

- Range of Platforms and Testing Pipelines For behavior, metabolism, sensory, development, ….

Molecular Analysis

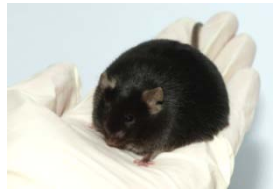
- Genotyping; Mapping; FACS; Analytical Chemistry

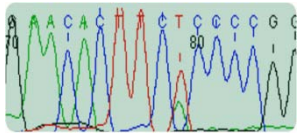
PharmaBiotech Contract Research

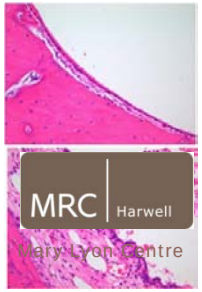
- Rederivation; Breeding & Holding; Therapeutic Efficacy Testing

MouseBook™

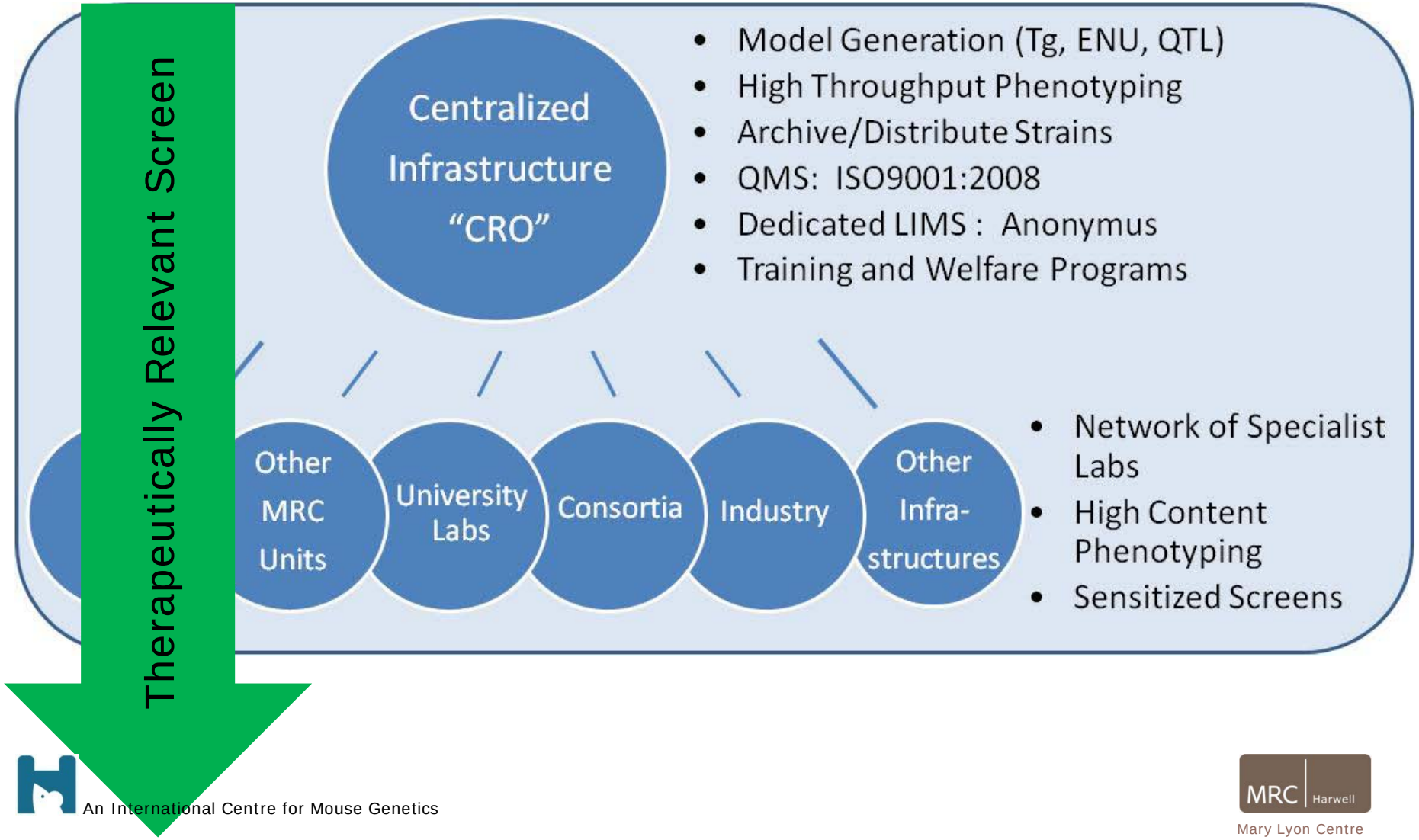
EMMA







Harwell Phenotyping Pipelines & UK Network of Clinical Researchers & Consortia



Obesity and Metabolism

- Metabolism
- Cardiovascular
- Bone & Cartiledge
- Sensory
- Neurobehavior & Motor function
- Respiratory & Immune System
- Development, Reproductive Disorders

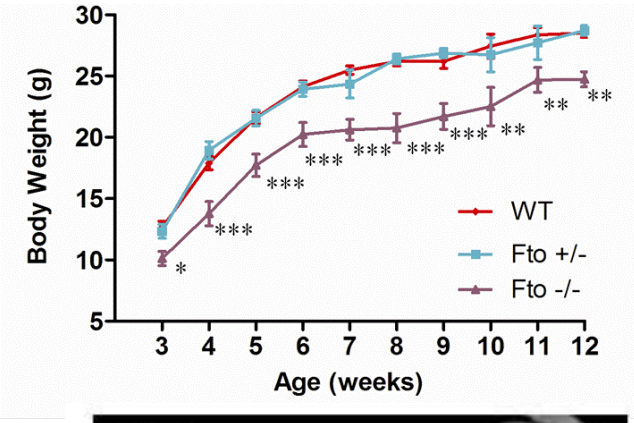
Intra Peritoneal Glucose Tolerance test (IPGTT)

Body Weight, Composition

Morphology

Serum Chemistry

Calorimetry



Cardiovascular

- Metabolism
- **Cardiovascular**
- Bone & Cartiledge
- Sensory
- Neurobehavior & Motor function
- Respiratory & Immune System
- Development, Reproductive Disorders

ECG

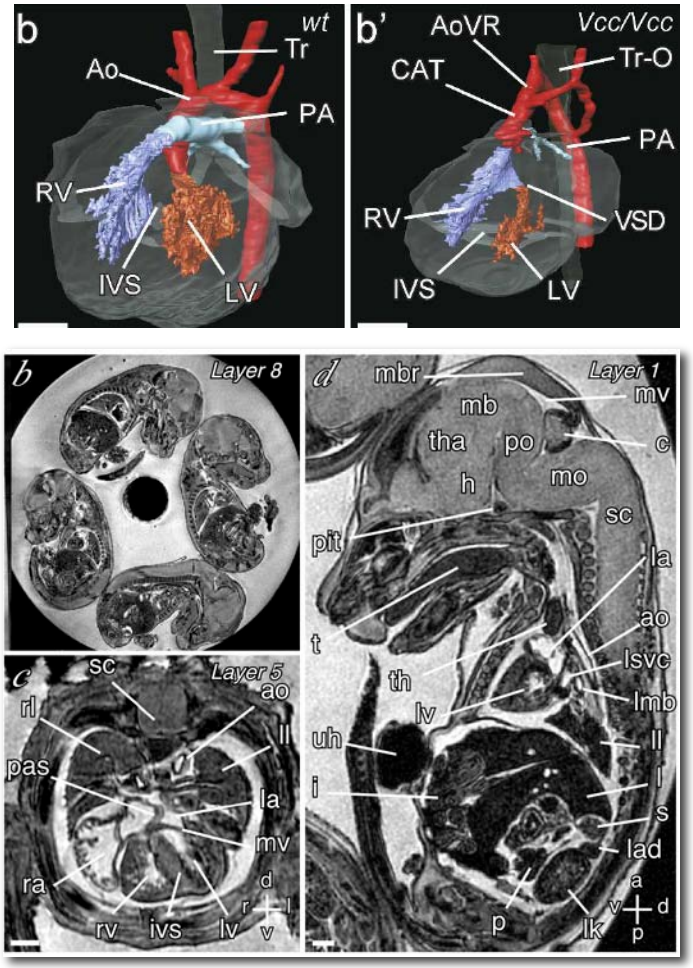
Echo-cardiography

Non-Invasive Blood Pressure

Heart Weight

Calorimetry

MRI



Bone and Cartelidge

- Metabolism
- Cardiovascular
- **Bone & Cartilage**
- Sensory
- Neurobehavior & Motor function
- Respiratory & Immune System
- Development, Reproductive Disorders

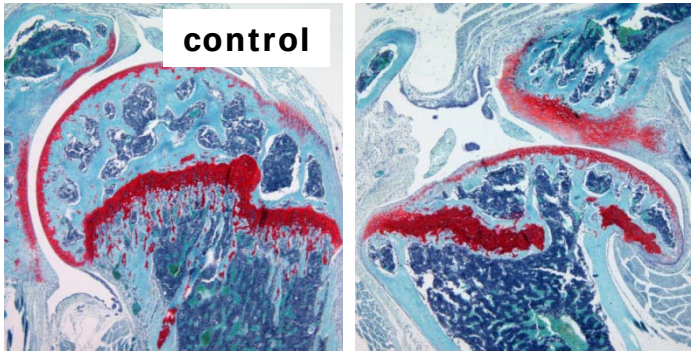
Dismorphology (X-Ray)

Bone and Tissue Composition (Dexa)

Serum Chemistry (Clinical Analyzer)

Histology

Echo-MRI

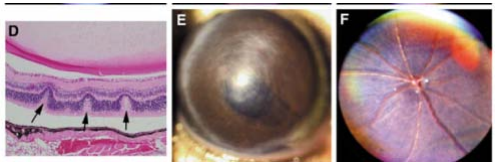


Sensory

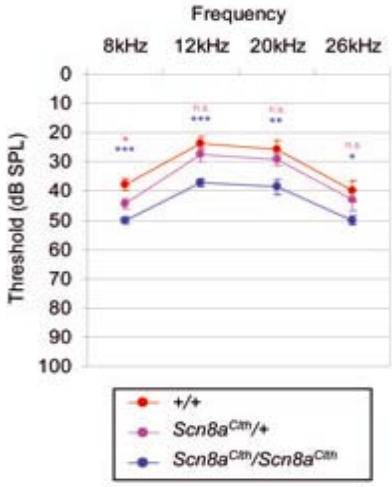
- Metabolism
- Cardiovascular
- Bone & Cartiledge
- **Sensory**
- Neurobehavior & Motor function
- Respiratory & Immune System
- Development, Reproductive Disorders

Eye morphology and retinal degeneration
- Slit lamp
- Opthalmascope

Visual Acuity
- Optokinetic drum



Auditory response
- Acoustic startle (click box)
- Pre-pulse inhibition (PPI)
- Auditory brainstem response (ABR)



Neurobehavior & Motor function

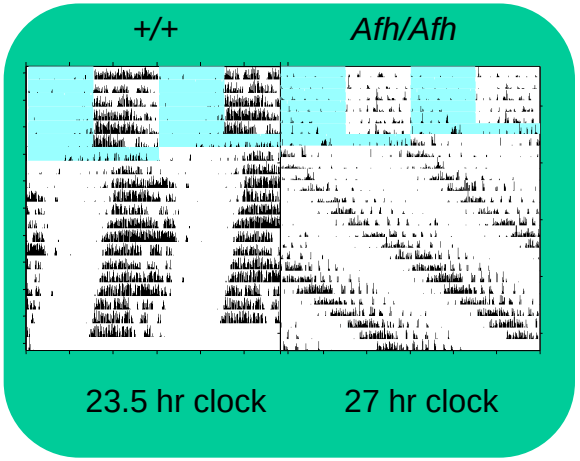
- Metabolism
- Cardiovascular
- Bone & Cartiledge
- Sensory
- **Neurobehavior & Motor function**
- Respiratory & Immune System
- Development, Reproductive Disorders

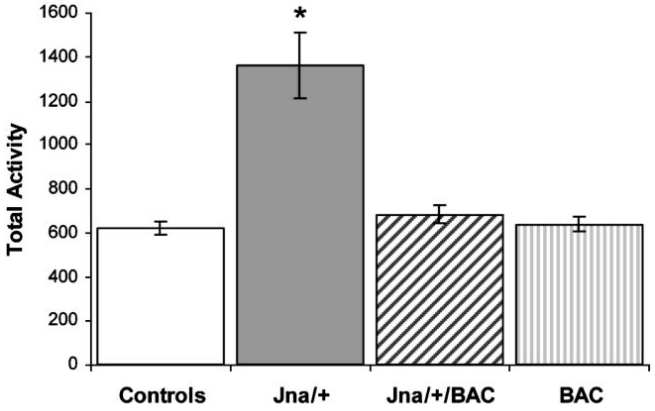
Observation
- SHIRPA Test
- Grip Strength
- Rotarod

Circadian Rhythm
- Wheel Running
- Sleep Boxes

Anxiety
- Open Field Test
- PPI
- Water Maze

Cognition
- Nose Poke



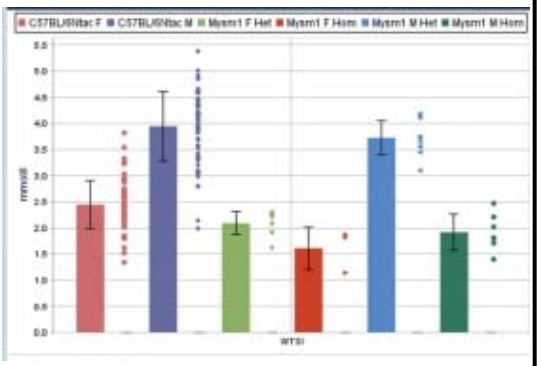


Respiratory & Immune Systems

- Metabolism
- Cardiovascular
- Bone & Cartiledge
- Sensory
- Neurobehavior & Motor function
- **Respiratory & Immune System**
- Development, Reproductive Disorders

Respiratory Challenge

- Plethysmography

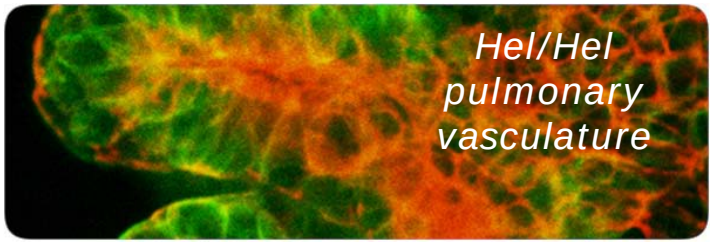


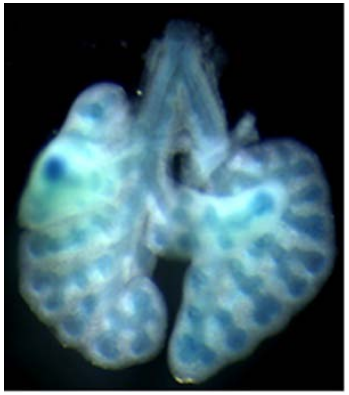
Immune Cell Count

- FACS

Immunoglobin Concentration

- ELISA
- OmniPlex

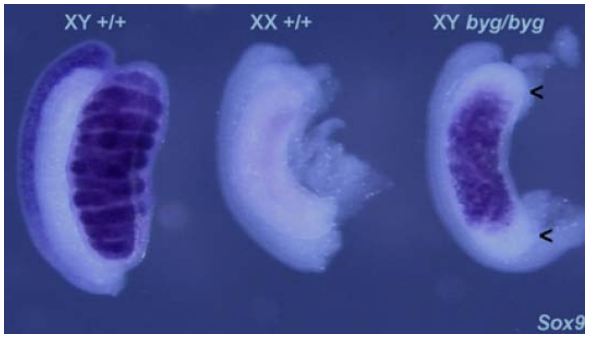




Development

- Metabolism
- Cardiovascular
- Bone & Cartiledge
- Sensory
- Neurobehavior & Motor function
- Respiratory & Immune System
- Development, Reproductive Disorders

Dysmorphology
- Optical Projection Tomography
- Confocal
- MRI

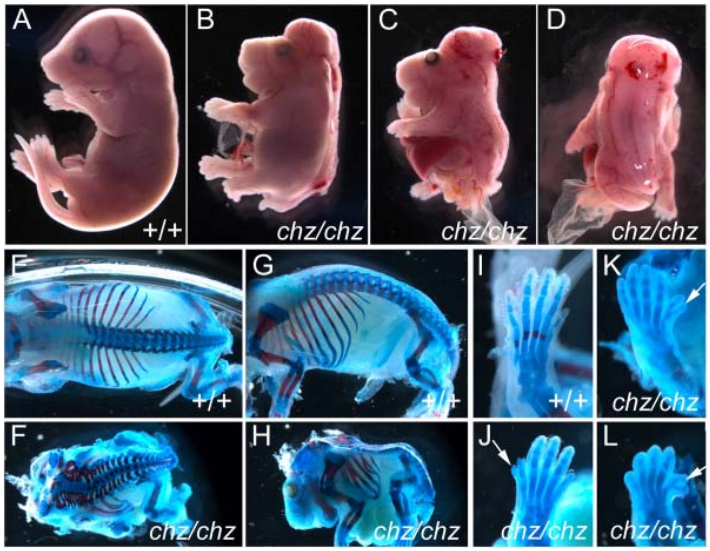


Lethality Test

Fecundity Test

Histology

Pathology



Quality Assurance Systems
(Mark Gardiner)

ISO Certified

Quality Plan

KPI Linked Objectives

SOPs & Training Register

Auditing

Non-Conformity Register

Customer Focused

CERTIFICATE OF APPROVAL

This is to certify that the Quality Management System of:

Medical Research Council
Didcot, Oxfordshire
United Kingdom

has been approved by Lloyd's Register Quality Assurance to the following Quality Management System Standards:

ISO 9001:2008

The Quality Management System is applicable to:

The breeding of mouse models for research into human diseases.

Approval
Certificate No: LRQ 4005566

Original Approval: 20 July 2010
Current Certificate: 20 July 2010
Certificate Expiry: 19 July 2013

Issued by: Lloyd's Register Quality Assurance Limited

UKAS MANAGEMENT SYSTEMS 001

AnonyMus (Alison Walling)
A custom built Laboratory Information Management System (LIMS)

Reports, Phenotyping Data Capture and Export

- **Sample Workflow Management**
- Reporting, with job scheduling of automated excel output
- Mouse husbandry and breeding
- Genotyping
- Cage locations & histories
- Cryo-preservation
- Tissue storage
- Phenotyping
- User account management etc
- Procedural information
- Home office Licence Administration, PPLs, PILs, reporting, tracking

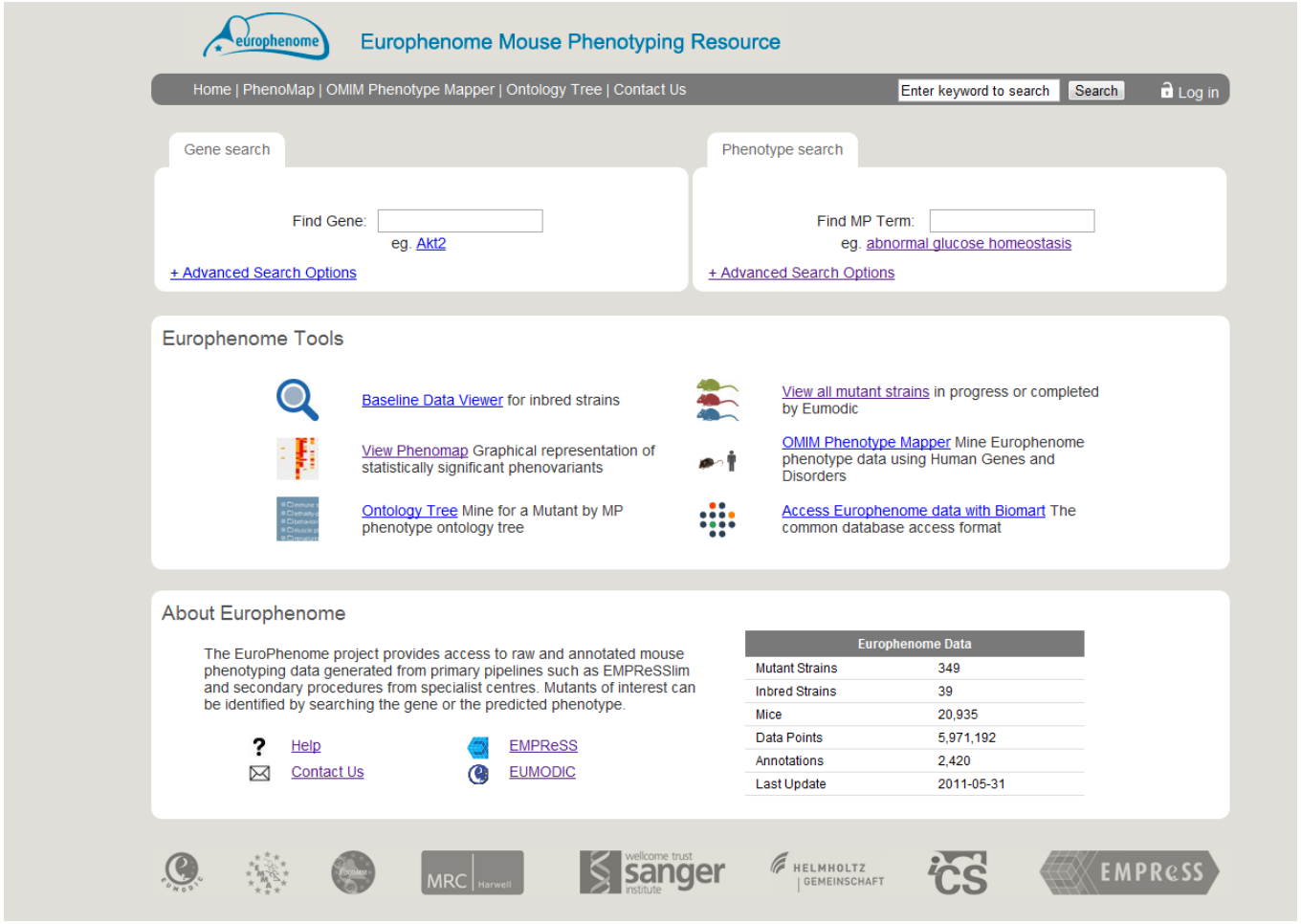
AnonyMus

Software Developed By An In House Team

Modular Architecture for Bespoke Solutions

- Allows for new requirements and changes in working practices to be flexibly accommodated in house

EuroPhenome (Anne Marie Mallon) (www.europhenome.org)



MRC IMPC Will Harness This Capability

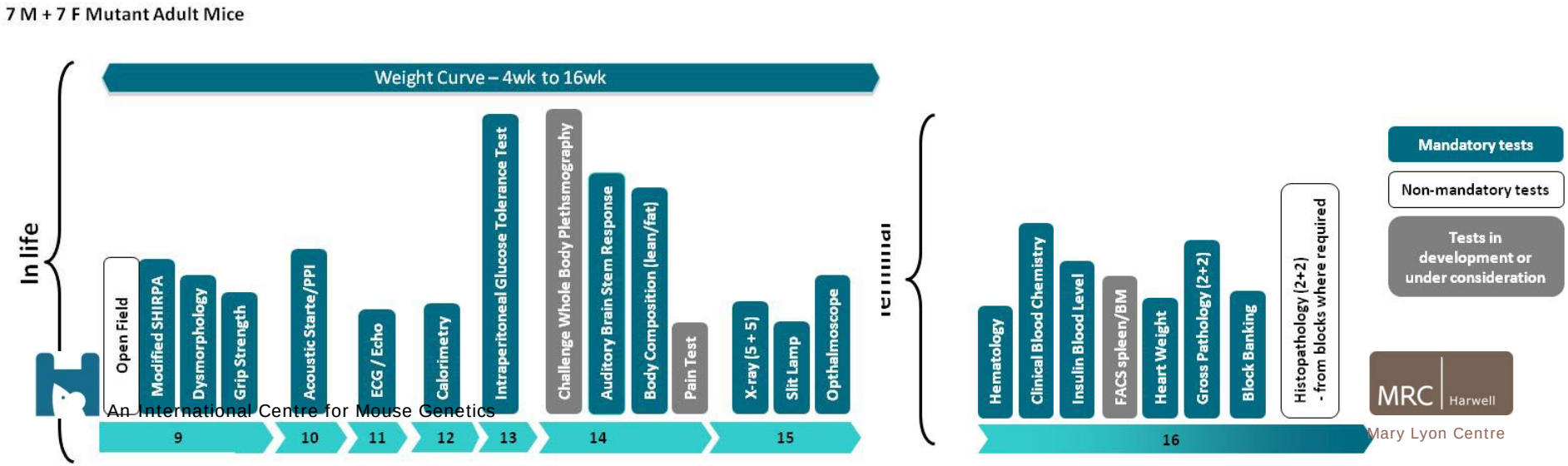
Develop and manage the DCC, building upon EUROPHENOME model

Produce and phenotype a min. of 530 lines, building upon EUMODIC

- MRC: 330 Lines
- BaSH: 200 Lines
- Reports will track progress for both programs

Production: GLT, Remove Neo, Archive, LacZ (E12.5, Adults)

Phenotyping: Lethality, Fecundity & IMPC Pheno Pipeline



Key Elements of MRC Strategy

1. Focus on core tests to bed down the IMPC pipeline. Manage expectations.

2. Evaluate, Improve, Enhance

3. Network with External Community – Life Science, Clinical and Industry

Our Success Will Be Measured By Active Scientific Engagement

(Not Terabytes of Data or Dewars of Embryos)

Funding Phased, Long Lead Time

Industry Liaison

Working Group

- Developing an Engagement Plan
- Collaborative/Partnering Opportunities

Workshop (London, March)

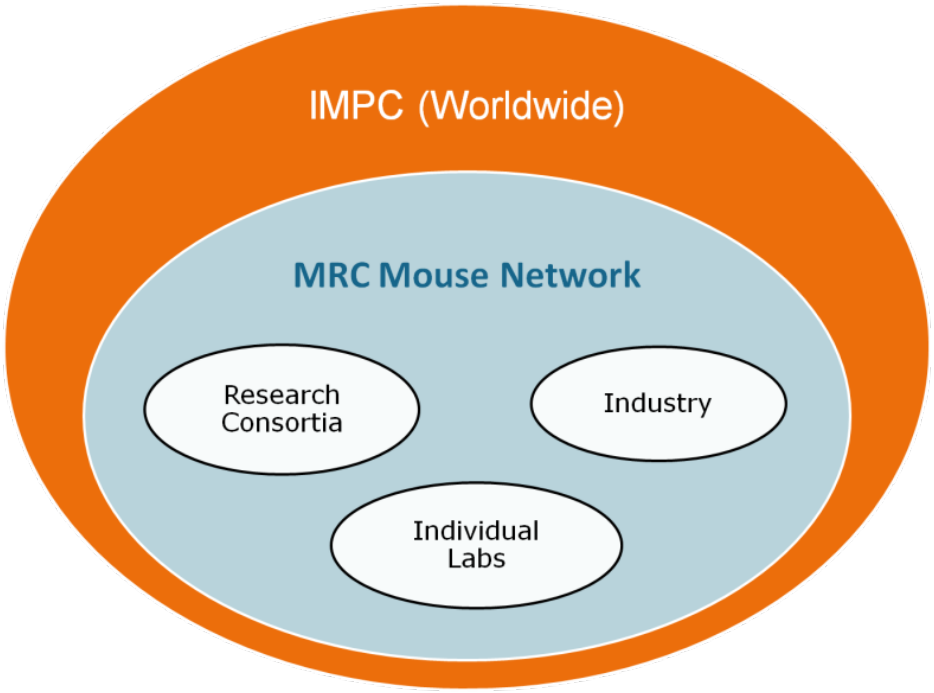
- 50 Delegates – Biotech, Pharma, Service Providers, IT, Consultants, Tech Transfer, Funders,…
- 10 Recommendations, including “Value Statement”
- Report available

Bio2011 (June)

- Largest WW Biopartnering event

Real Challenges:
MTA / Access; Speaking with one voice

Engagement Plan: MRC Mouse Network



MRC Mouse Network Will Be a Forum to:

Communicate about IMPC

Encourage Uptake of Resources

- Mice
- Phenotypic Data

Promote Use of Resource

- Training in Techniques
- Bioinformatics Tools

Facilitate Collaboration

- Research Consortia
- Expression of Interest Call

Get the Message Out

Road Shows (Jun-Aug)

- 14 Institutes, 7 Cities (red)
- Another 6 on phone (orange)

MRC Mouse Network Portal (in development)

- Hub of information concerning the MRC Mouse network.
- Communicate latest news, meetings, training opportunities, events, developments in new lines and interesting phenotypes.
- Encourage discussion and disseminate information between consortia via public and private threaded discussion forums and messaging.
- 220 Registrants, 8475 page views

Consortia Driving Our Science

14 Consortia Have Been Formed

- 400 PIs; 233 Profs; 76 Consultants; 8 FRS, Nobel
- Most Major UK Research Centres; 17 International Inst.

7 of the top 10 Therapeutic Areas

1200 Candidate Genes

Rank	Condition	MRC Network Consortium?	UK Male Morbidity (2009)	% of Total Deaths
1	Heart Disease	Yes	41,455	17.4
2	Lung Cancer	No	17,053	7.2
3	Stroke	Yes	16,888	7.1
4	Respiratory Disease	Yes	13,165	5.5
5	Flu and Pneumonia	Yes	11,108	4.7
6	Prostate Cancer	No	9,402	3.9
7	Colon Cancer	No	7,559	3.2
8	Neurodegenerative Disease	Yes	6,709	2.8
9	Haematopoietic Cancer	Yes	5,922	2.5
10	Liver Disease	Yes	4,604	1.9
	Total		133,865	56

Example: Mouse Models For Accelerated Drug Discovery

To accelerate drug discovery through the use of mouse knockout models,

beginning with the ubiquitylation system.

This class of genes is considered an untapped and rich source of novel drug targets against a broad range of therapeutic indications

The network will bring together academic, governmental, and industrial researchers.

- Dundee, Phosphorylation and Ubiquitylation (Alessi, Cohen)
- Technion University, Discoverer (Ciechanover)
- Nanjing, Production and Phenotyping (Gao)
- Toronto, Structural Genomics Consortium (Edwards)
- London, MRC HTP Screening & Bus Dev (Dalrymple)

MMADD Work Plan

- **Prioritization of ALL genes** of the deubiquitylases, E2 conjugating enzymes, F Box substrate targeting subunits of cullin containing E3 ligases and HECT E3-ligases
- **100 GLT knockout Mice** with phenotypic information – China and UK will contribute equally.
- **Discover novel 3D structural information** of candidate targets – with Structural Genomics Consortium
- **Advise and interpret** data to max value & improve pipeline.
- Use resources in **translational studies to validate target function in vivo, in vitro, and high throughput drug screening**.
- **Supplemental funding** will be secured, possibly through industry partnering.

Neuromouse

Six Themes (78 PIs): Director Kay Davies, 6 Theme Leaders

1. Ion channels and epilepsy
2. Mitochondrial Disease
3. Neurodegeneration
4. Neuromuscular Diseases
5. Neuropsychiatric disorders
6. Sensory systems

Work Plan

- **Gene Selection** (118 targets, expect approx 60 GLTs)
- **Assess outputs of the primary phenotyping** pipeline and to provide input into the pipeline's evolution.
- Each Neuromouse lab will **take responsibility for incorporating IMPC data and knockout lines into their labs**;
- Coordinate with **GWAS** studies;
- Theme leaders will meet every six-months to monitor progress of their groups;

Key Members of the Project Team


Martin F.


Lydia T.


Deen Q.


Sara W.


Tertius H.


Michael C.


Mark G.

Production

LacZ Profiling

Cohort Breeding

Necropsy

Quality Assurance

Archiving

Allele QC

Phenotyping

Pathology

Distribution

Genotyping

Histology

Training

LIMS, Data Portal & Bioinformatics


Alison W.


Andy B.


Annie M.

Finance, HR, Engagement


Liz M.


Hilary G.


Nanda R.

An International Centre for Mouse Genetics

MRC Harwell

Mary Lyon Centre

Project Management

MRC Working Groups

1. Production
2. Allele QC & Genotyping
3. LacZ
4. Phenotyping
5. IT
6. Engagement

Work Plans & Reporting

- ❑ Targets
- ❑ KPIs/Metrics
- ❑ Organization
- ❑ Resources
- ❑ Critical Path Activity
- ❑ Issue List

Oversight

- ❑ MRC IMPC Sr. Management Group
- ❑ BaSH PI Group & Contribution to BaSH Working Groups

Production Targets & Current Status

Oct 1st Kick-off

- Recruitment drive in place
- Cross skilling of existing staff

Production (GLT)	Yr 1	Yr 2	Yr 3	Yr 4	Yr 5	Total
MRC	53	79	79	79	40	330
NIH	40	50	50	50	0	190
Total	**93**	**129**	**129**	**129**	**40**	**520**

29 lines in production pipeline

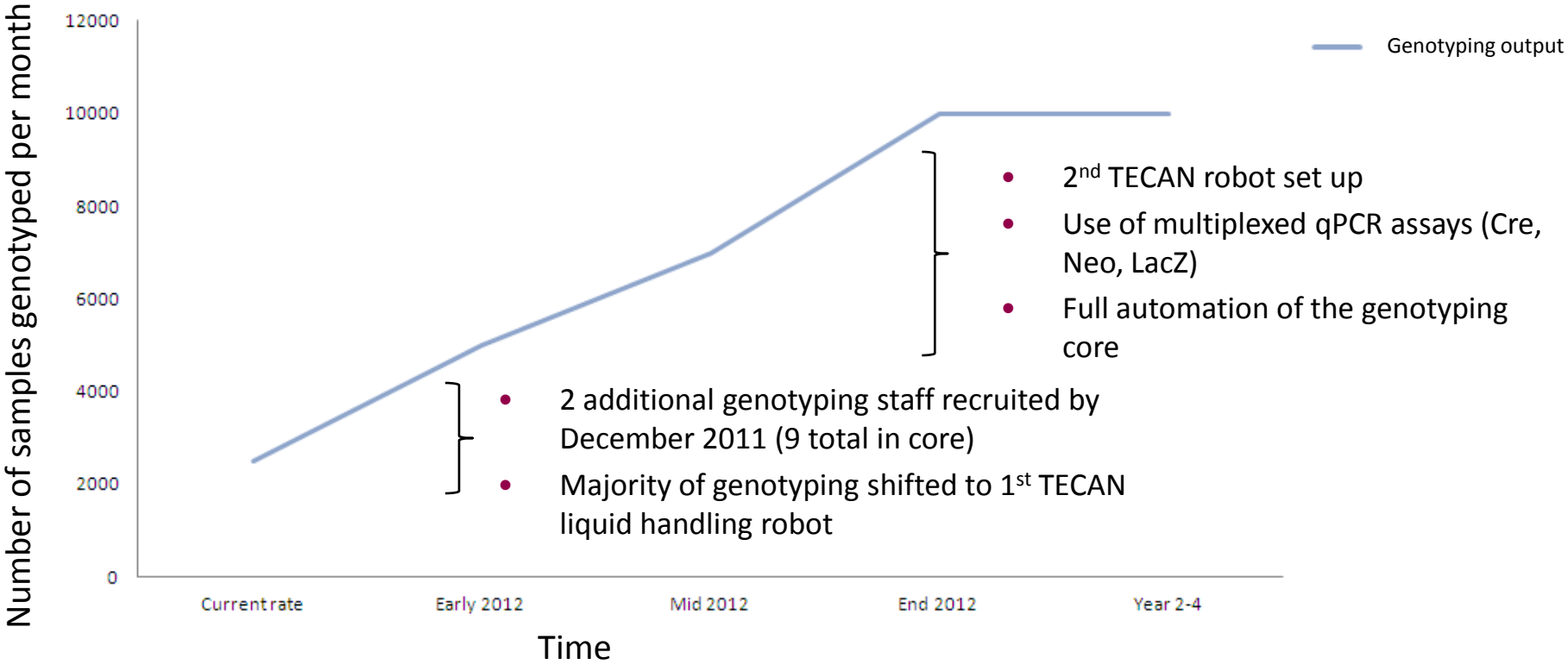
- ES cell distribution under way with WTSI
- Gene list complete (1200 from consortia)

Tech dev

- Boost GLT - 2i media, morula injection
- Laser assisted IVF, artificial insemination, improved sperm freezing conditions (Takeo & Nakagata)
- LacZ expression pilots

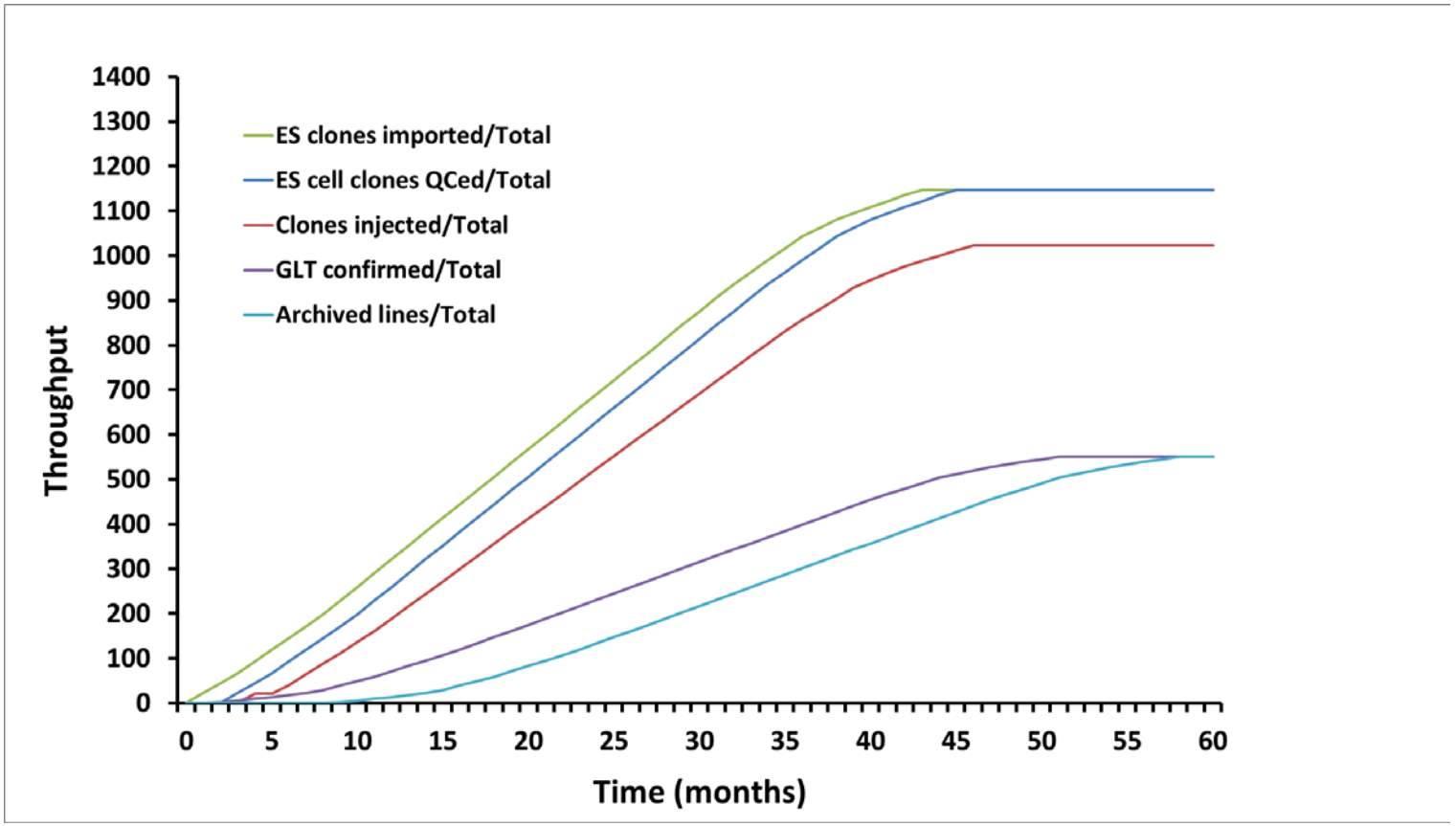
IMPC Genotyping Metrics (Deen Quwailid, Lydia Teboul)

Monthly Genotyping rate required for production of 550 IMPC mouse lines



Current rate	Early 2012	Mid 2012	End 2012	Year 2-4
2500	5000	7000	10,000	10,000

Mouse Conversion (Martin Fray)

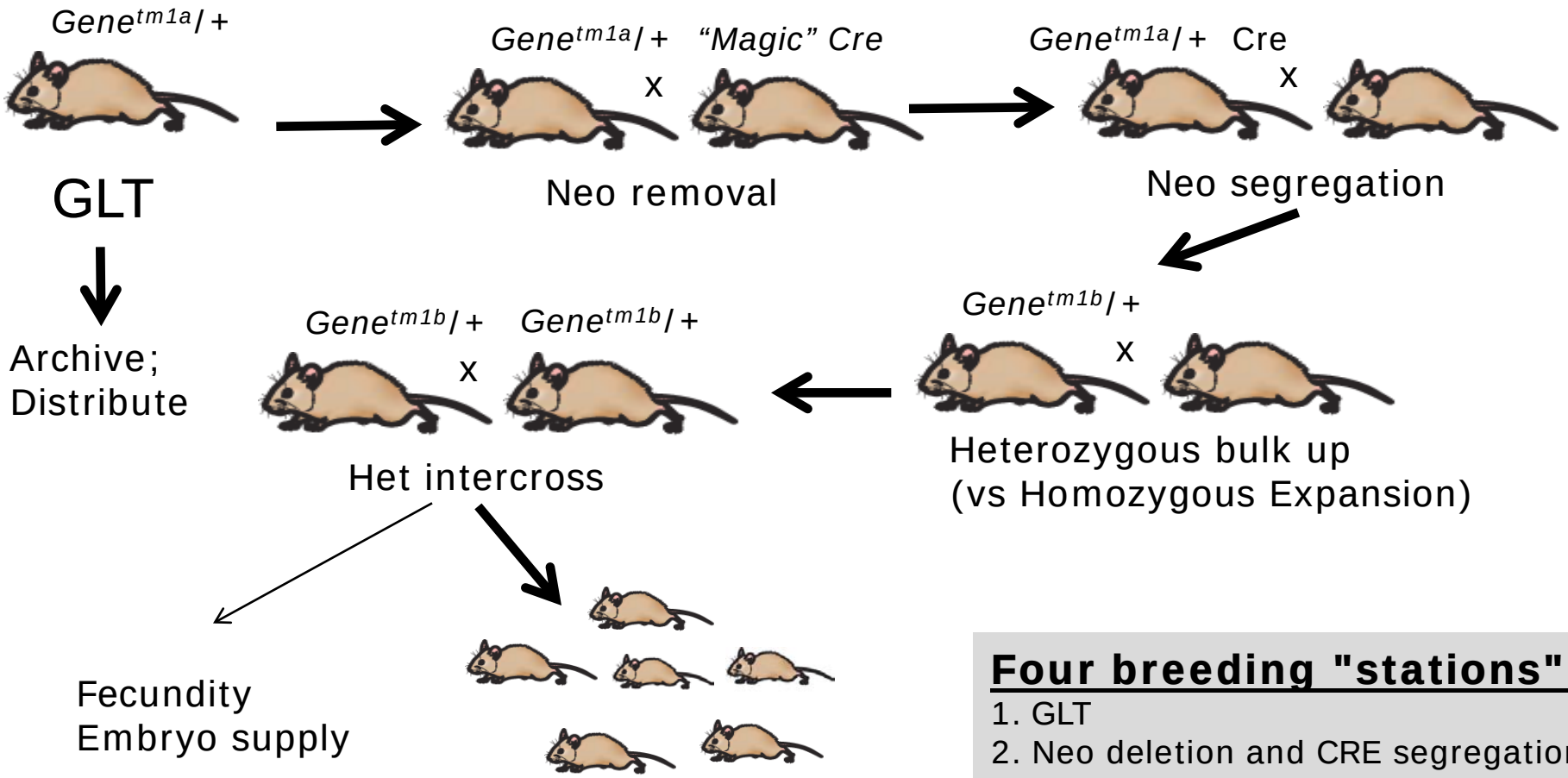


Total number of ES clones required: 550 x 2 x 1.1 = 1210

Require a sustained peak delivery of 30 clones/month

Maintain GLT at 50% Plus

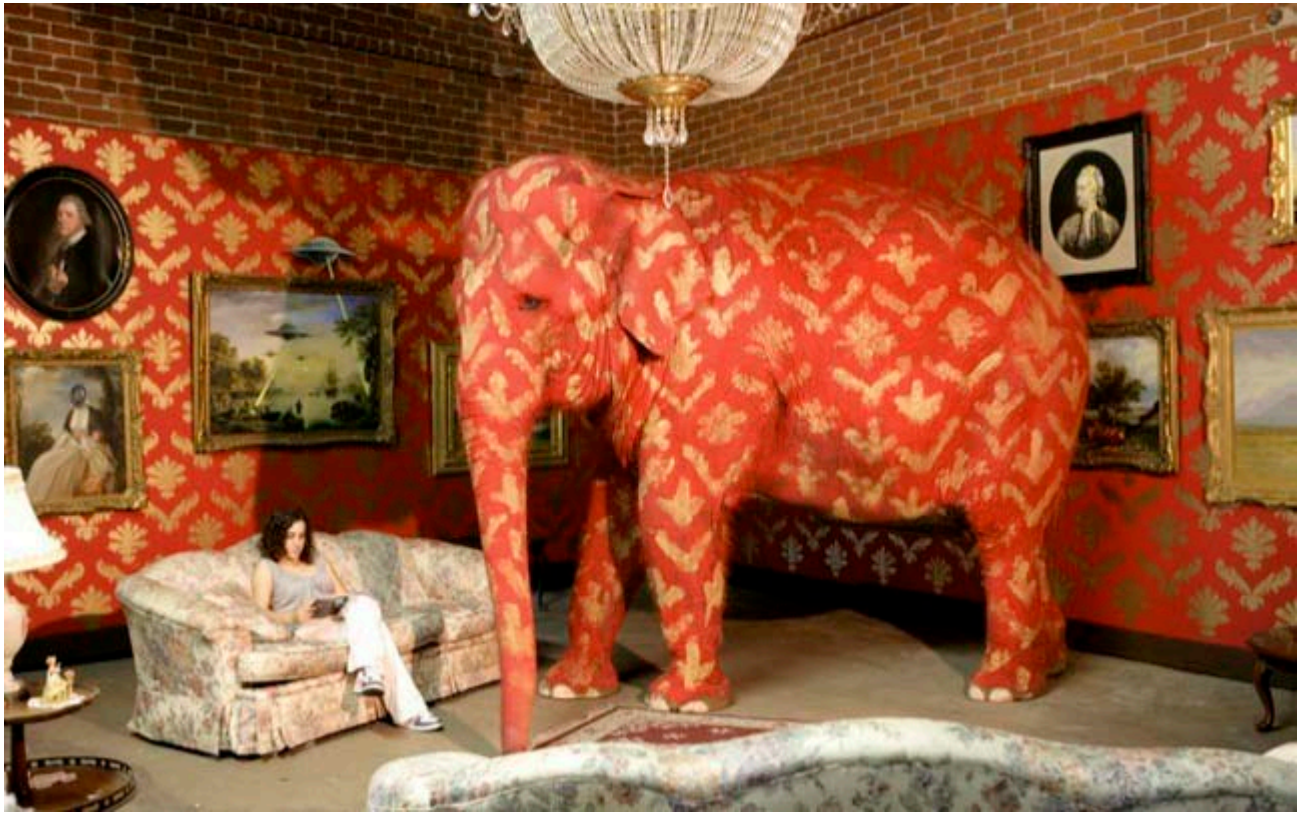
Breeding Scheme (Sara Wells)



Four breeding "stations":

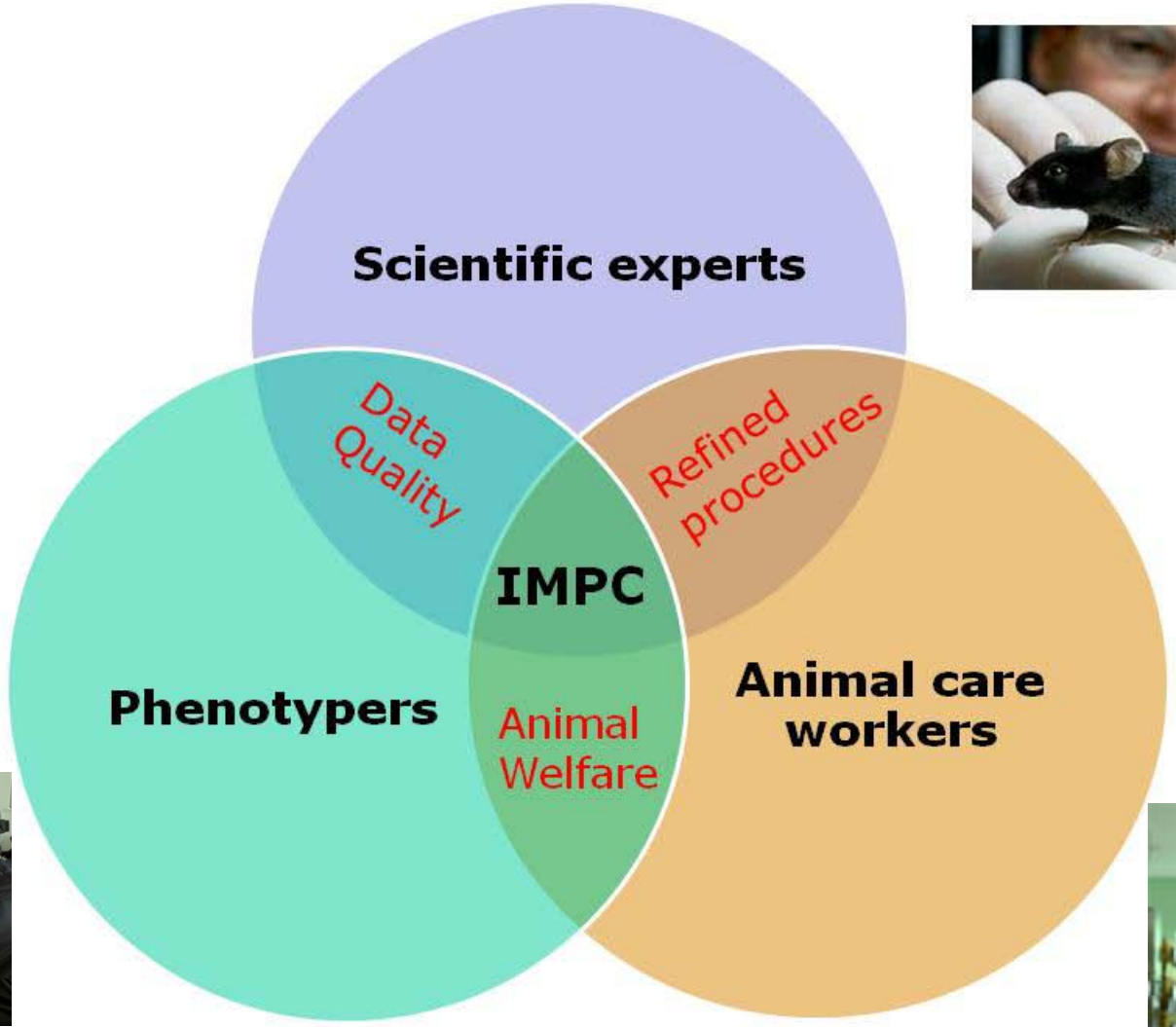
1. GLT
2. Neo deletion and CRE segregation
3. Heterozygous bulk up of tm1b
4. Heterozygous intercross (+fecundity).

Effective Management, Tracking & Reporting of Progress



Plans for Creating Conditional Alleles

Phenotyping



Phenotyping Plan
(Sara Wells, Tertius Hough)

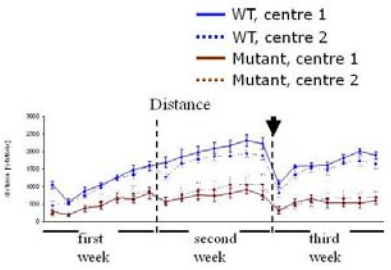
Focus on Core Tests in Yr 1

- 50 Lines Completed in 2010_11
- Review SOPs
- Parameter Sets

Existing Tests	Yr 1	Yr 2	Yr 3	Yr 4	Yr 5	Total
MRC	0	78	83	83	86	330
NIH	30	40	50	50	20	190
Total	**30**	**118**	**133**	**133**	**106**	**520**

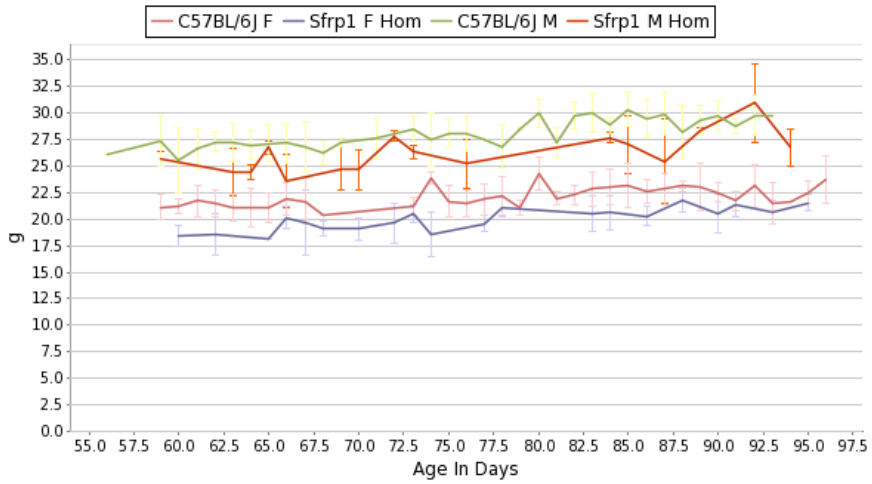
Improve With Time

- Refining the Basics
- Transition Tests to High Throughput
- Pilot New Tests



REFINING THE BASICS

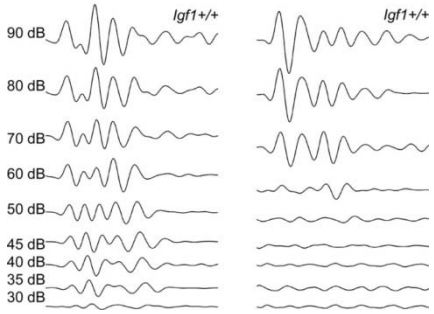
Weight Curve – 4wk to 16wk



EUMODIC	IMPC
At time of test	Regular time
Different time of day	Same time of day
Static balances	Dynamic balances
Running baseline	Colony mate controls

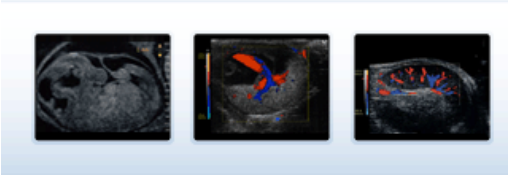
Transition Tests to High Throughput

Auditory Brainstem Response



- Refine test to reduce numbers
- Make HTP

Cardiovascular Phenotyping



- ECG / Echo
- 2D Short Axis Measurements
- Heart Wt, Hist.

Challenge Plethysmography

- Methacholine
- Hypoxia
- Hypercapnia

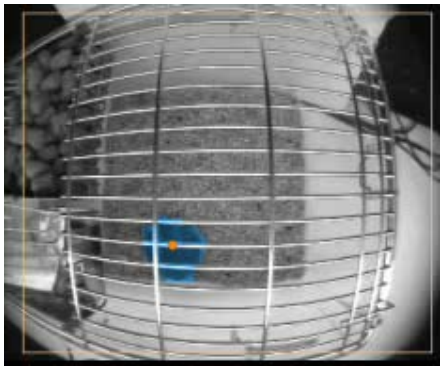
FOR THE FUTURE

Locomotor Tests

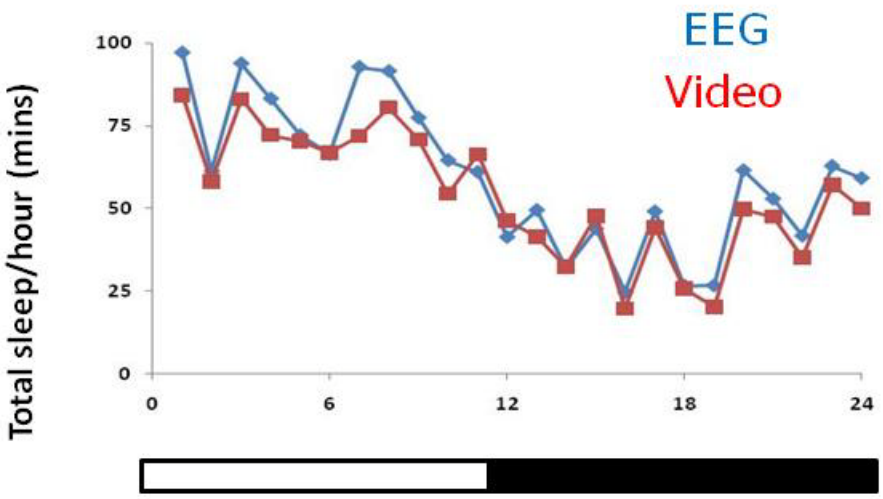
Pain and Immunological tests

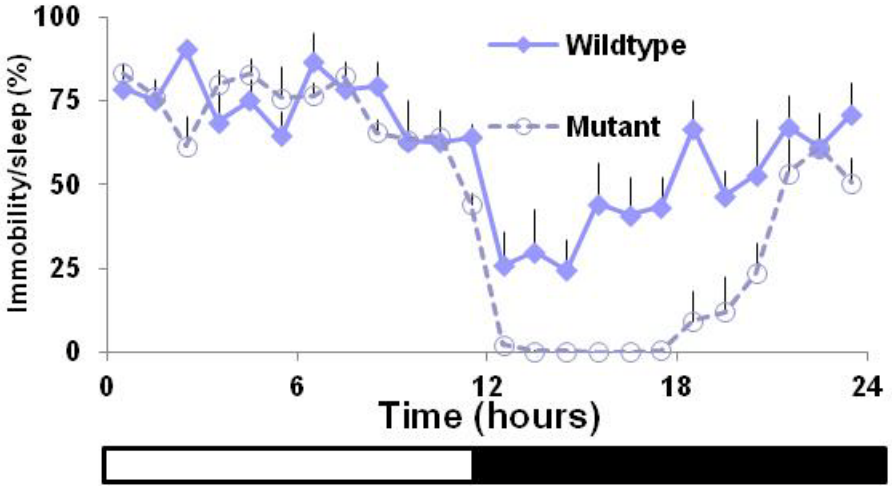
- **Improve reliability**
- **Possible replacement tests**

Sleep Assessment Using Home-Cage Video Tracking (Pat Nolan)



- Animals 'tracked' in home cage using infra-red cameras
- Tracking software used to analyse numerous parameters
- In this case, sleep, defined by periods of immobility with tracking software is highly correlated with EEG-defined sleep
- ADDITIONAL COGNATIVE & MOTOR FUNCTION TESTS ALSO BEING PILOTED





Micro-CT (Jurgen Schneider)

MRC Harwell is currently committed to purchase a micro-CT system

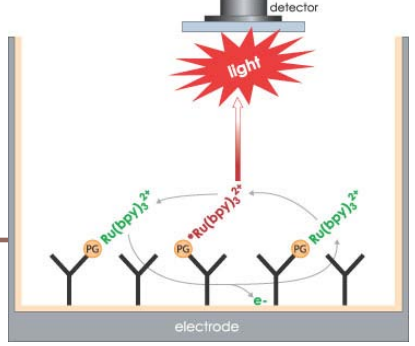
Potential to incorporate micro-CT as part of high-throughput screening programme

- 3D analysis of embryos, adult heart, brain

Method Development

- Contrast Agents, Atlases/Annotation

Multiplex Elisa Technology (Tertius Hough)



Electrochemiluminescence - MSD Platform

- Combination of electrochemiluminescence detection and patterned arrays
- Speed and high density of information through miniaturization
- Collect and quantitatively measure light emitted from microplate wells

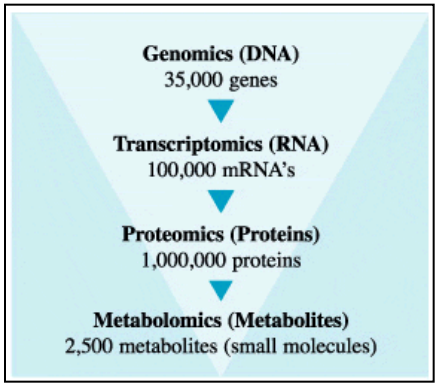
Advantages

- Parallel processing of biological assays – conserves sample volume
- Unique combination of sensitivity & large dynamic range, High precision
- No fluidics in measurement step

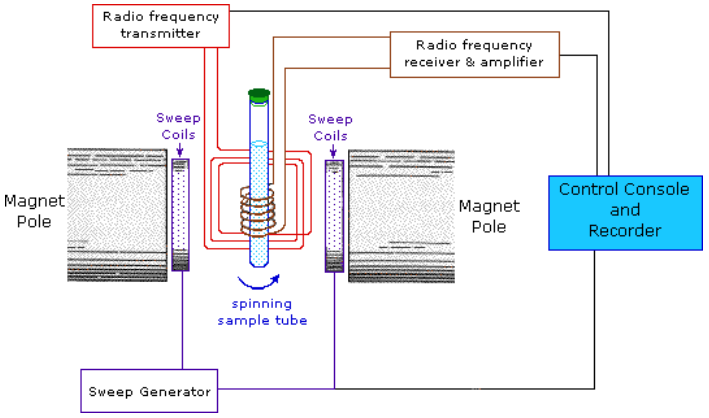
Broad range of applications for IMPC

- Cytokines; Gut Hormones; Kidney markers; Bone; Oncology/Phoshoprotein analysis; Neuroscience/ Alzheimer's markers; Fertility markers; IgGs

Biobanking & Metabolomics



- Opportunity for biobanking (urine, serum, tissues) to find early biomarkers of disease
- Profiles as surrogate markers for onset of disease (renal, liver, heart, ….)
- Metabolomics aim at quantifying & identifying all metabolites at the organ, tissue, cellular, or even subcellular level
- H-NMR spectroscopy produces a "metabolic fingerprint"
- We have refined protocol allows sample volumes as low as 10µl (urine)



Summary

MRC is Committed to IMPC:

Deliver => **500 Lines, High Quality Product**

MRC Mouse Network => **14 Consortia, 100's PIs**
Exploitation of Resource & Collaboration

Develop New Methods => **Richer, Higher Value Data**

Make a Real Difference => **Translational Research, New Discoveries Drives Funders Strategic Investment**

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An International Centre for Mouse Genetics

MEDICAL
RESEARCH
COUNCIL
TECHNOLOGY

MRC Harwell
Mary Lyon Centre

Thank You

Lets Rock and Roll

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QC standard for EUCOMM lines: Our 4 Criteria

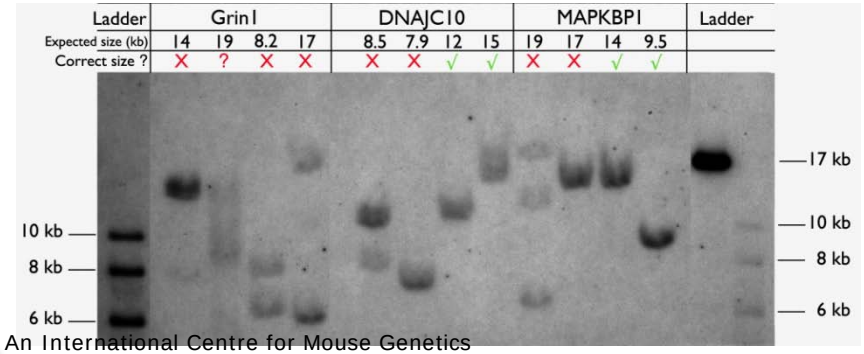
1. Confirm desired locus has been targeted

2. Confirm structure of the targeted allele

3. Confirm absence of additional vector insertions

4. Confirm presence/absence of downstream LoxP site

High throughput Southern blot with DIG-labelled universal probes



Gene specific or universal PCR & sequencing to detect the 3' LoxP

Controlling allele quality: capacity matching Harwell production targets

ES cells

✓ Amplification + sequencing target region (universal loxP PCR or locus specific for 15% of clones)

✓ Southern blotting, Karyotyping

✓ Current capacity (Harwell):
30 clones/month, WTSI from a date TBA

✓ **75% of received clones have the correct allele**

Mice

✓ Amplification + sequencing target region (universal loxP PCR or locus specific)

✓ Allele counting by qPCR:
Neo, lacZ and locus specific

✓ Covered by current capacity

✓ **All F1 carriers will be QC checked**

KPIs: Alleles Passed; Samples Processed