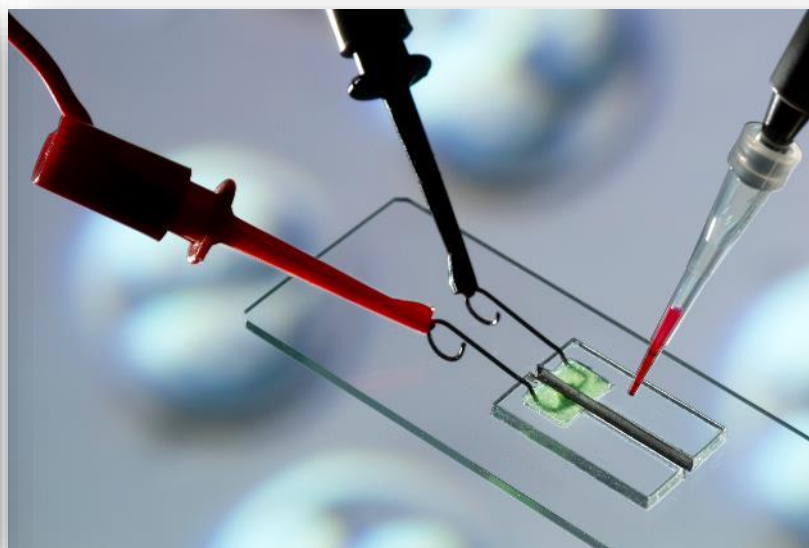
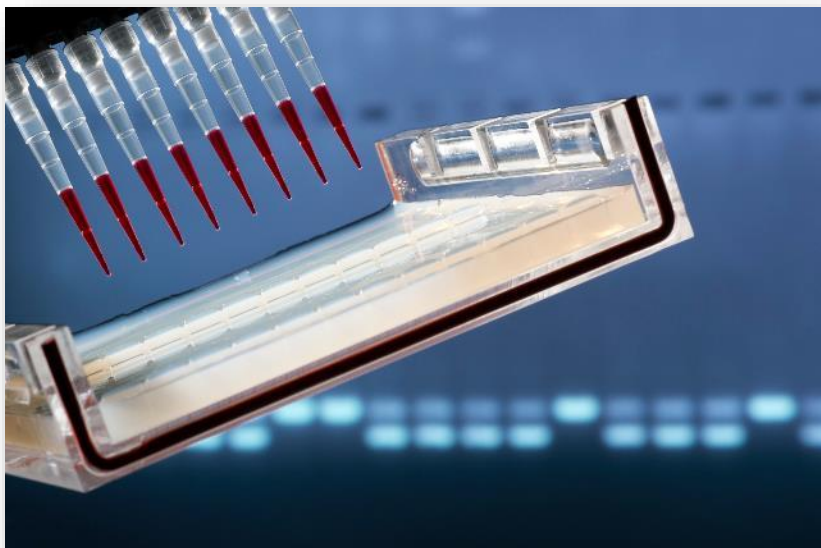




The Infrafrontier RI

Julia Fernández & Lluís Montoliu (CNB-CSIC & CIBERER-ISCI)



30 May 2022

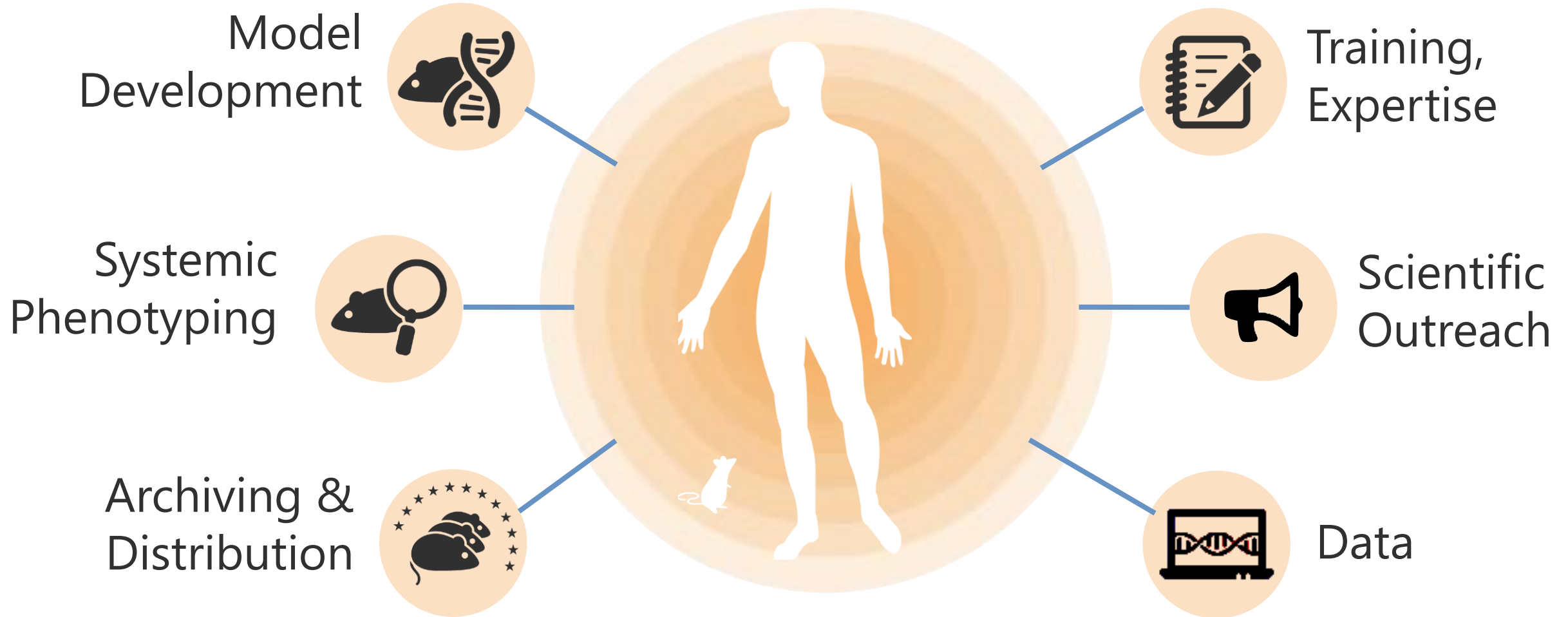
The INFRAFRONTIER Research Infrastructure

2007



Providing
unique scientific resources, services, and expertise
to advance the understanding and treatment of **human diseases using rodent models**

Access to physical resources, analytical platforms, knowledge and data



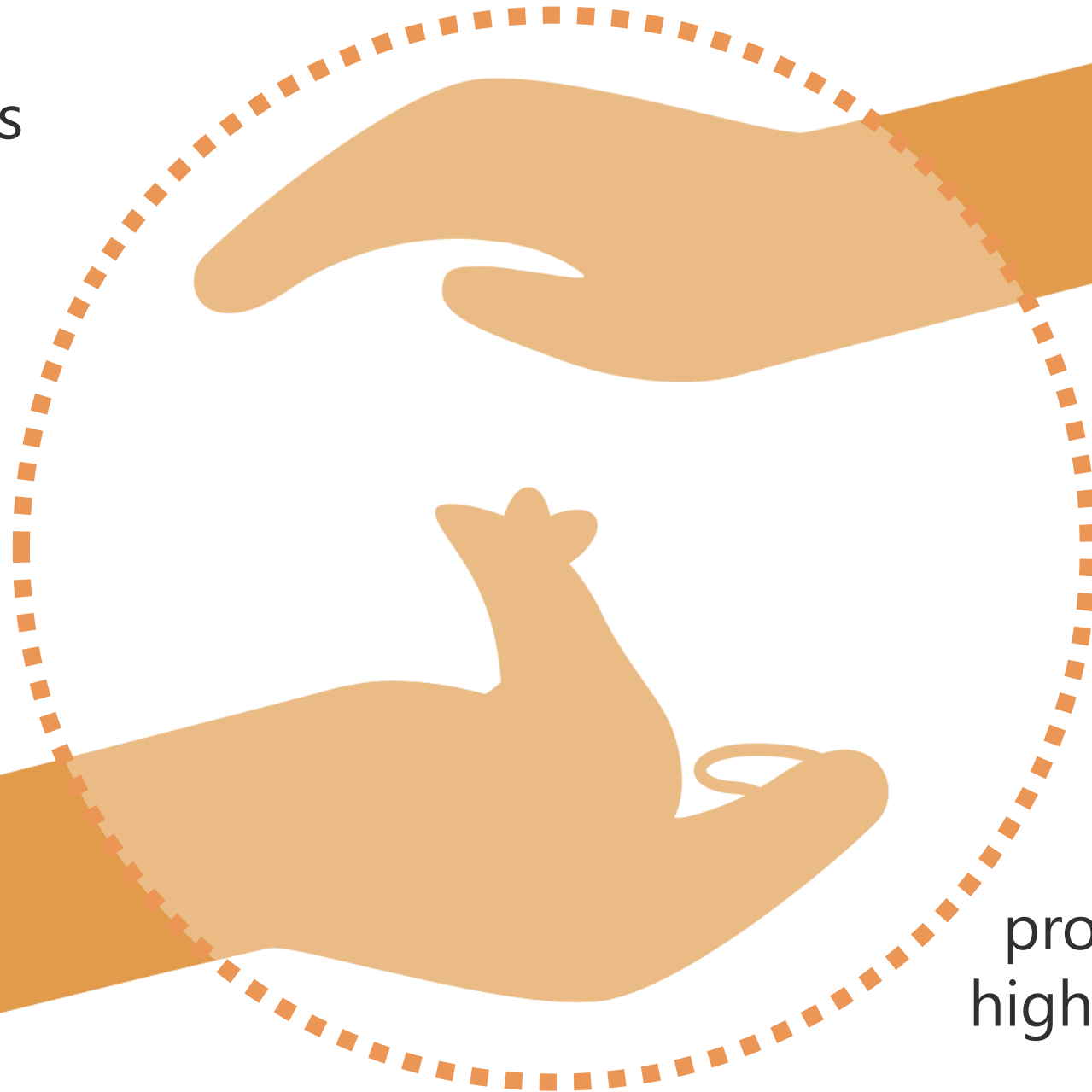
Promoting responsible and reproducible research

Promoting the 3Rs

Replacement

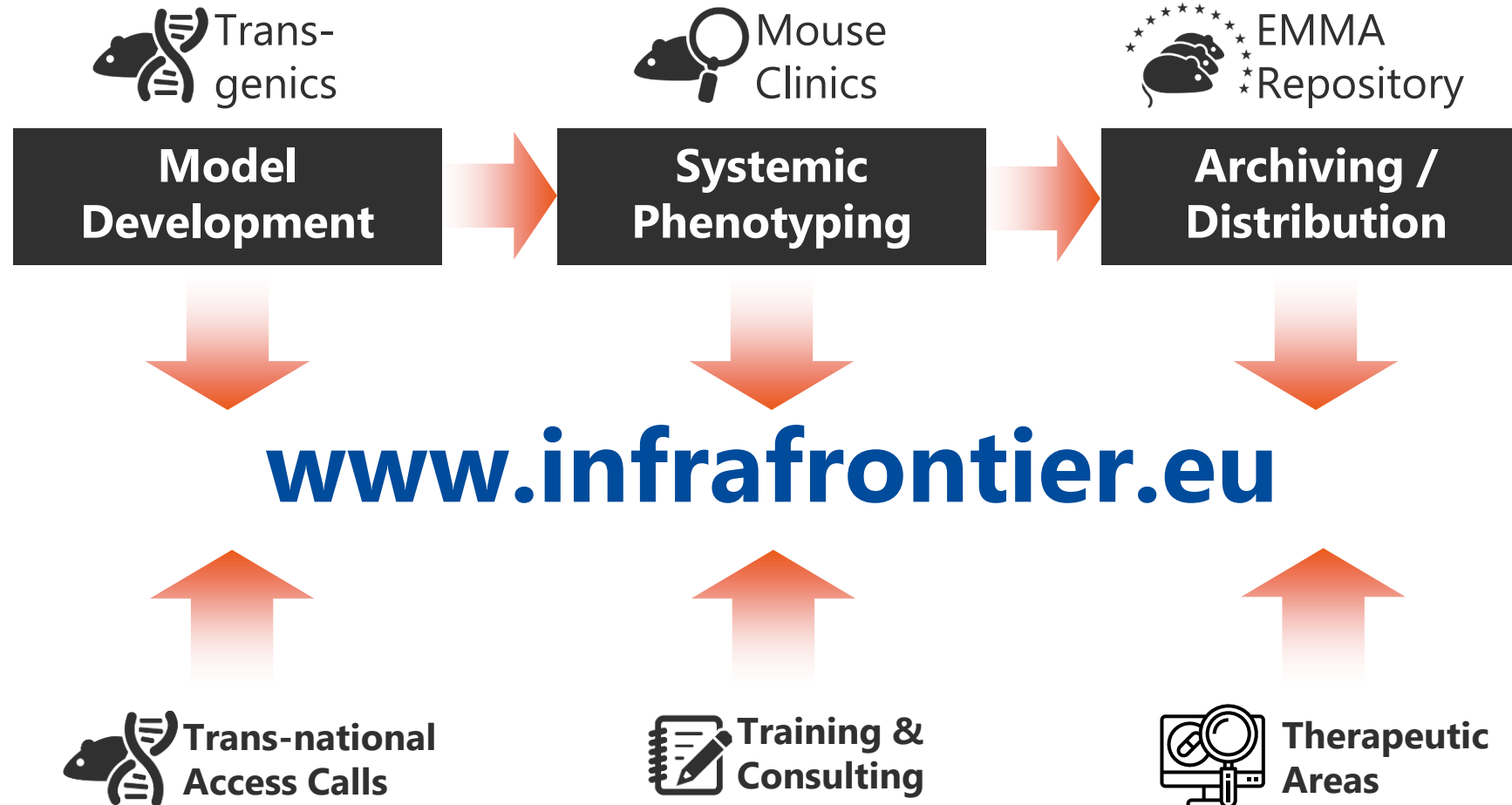
Reduction

Refinement



Minimising
duplication by
providing centralised
high-quality resources

INFRAFRONTIER Resources and Services



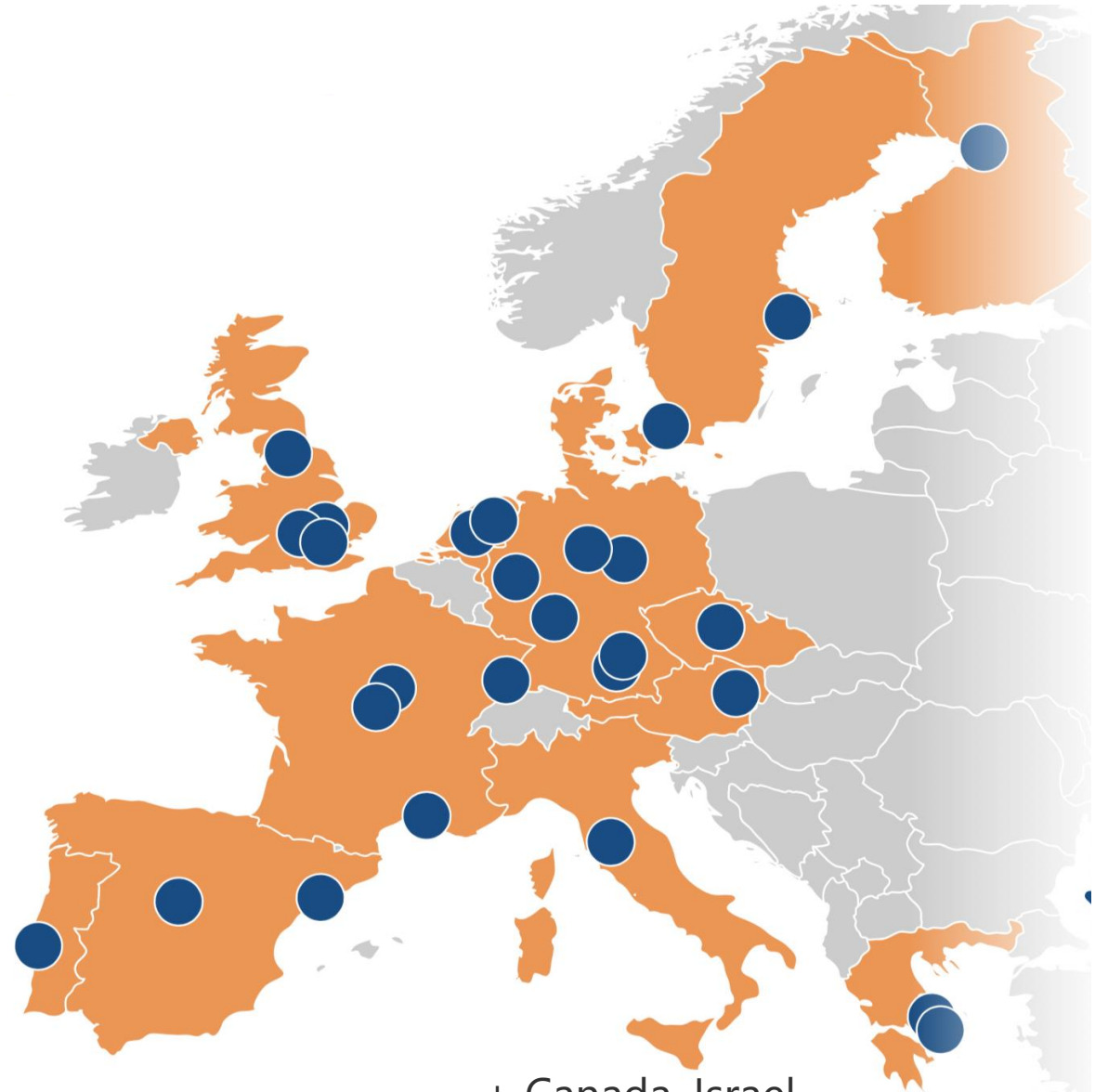
Distributed research infrastructure – one face to the customer



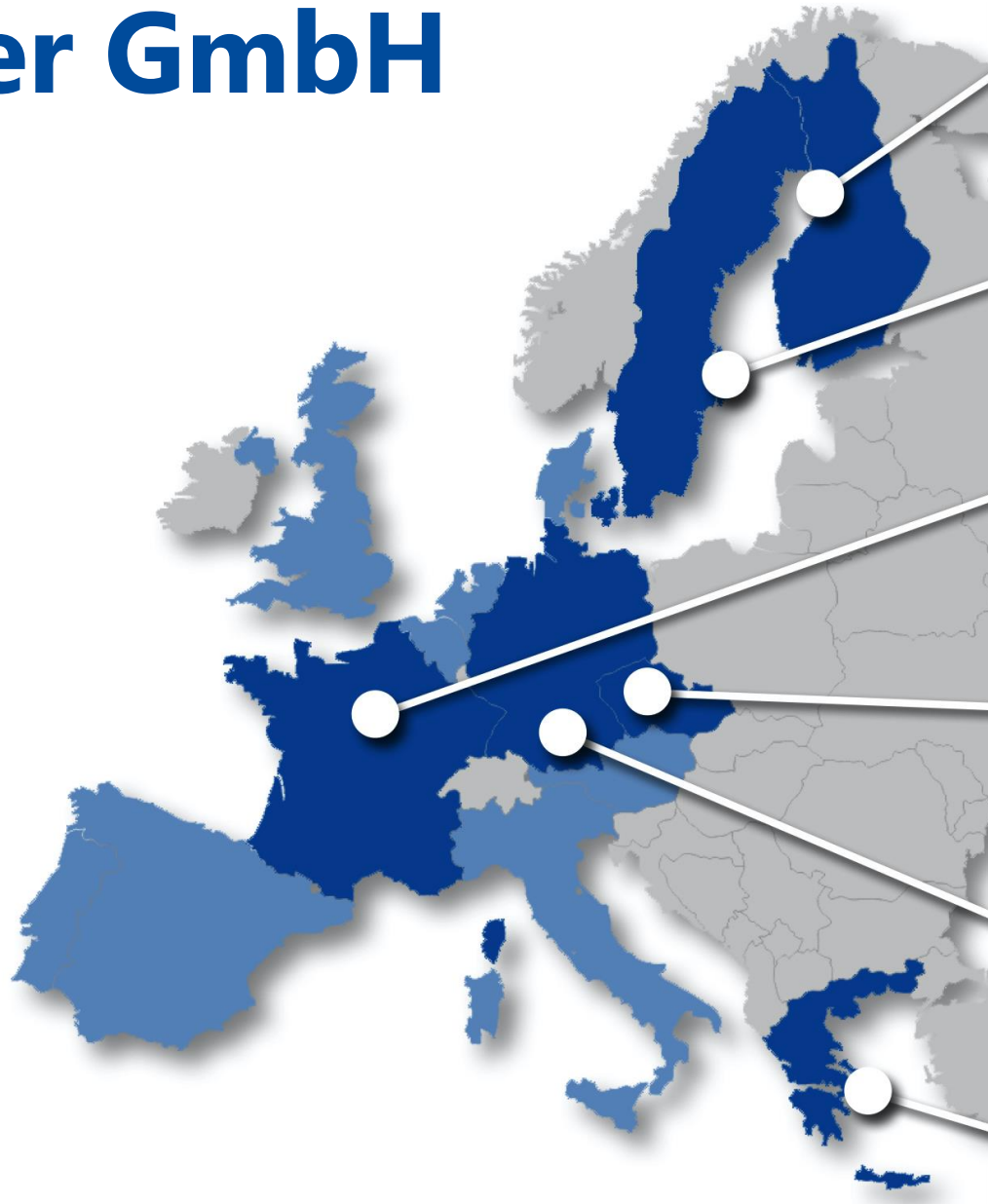
INFRAFRONTIER
mouse disease models

A European Research Infrastructure
for the **generation, phenotyping,**
archiving and **distribution** of
mammalian models

A consortium of leading European
research institutions - **29 partners in**
15 countries



Infrafrontier GmbH



UNIVERSITY of OULU
OULUN YLIOPISTO



**Karolinska
Institutet**



Le Centre nationale de la
recherche scientifique



Institute of Molecular Genetics of the ASCR, v.v.i.

HelmholtzZentrum münchen
German Research Center for Environmental Health



"ALEXANDER FLEMING"
Biomedical Sciences Research Center

2013

Resources and Services

Deposit mice



EMMA archives your mouse mutant free of charge >

Order mice



Access to 7600 mouse mutants and research tools >

Axenic service



INFRAFONTIER produces germ-free mice free of charge >

NKI GEMM-ESC archive



Rapid target gene validation in complex cancer mouse models >

Mouse production and phenotyping



INFRAFONTIER open calls >

Training and consulting



Hands-on courses in cryopreservation and phenotyping >

EMMA strains

Search EMMA strains

Search by: Gene/Allele symbol, strain name, EMMA ID, DO name/ID, phenotype, ...

Browse EMMA strains

Collections

Human Diseases

? Help




Useful information for EMMA users

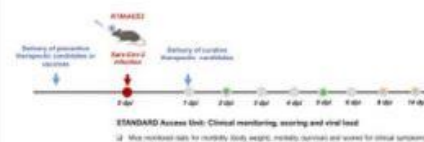
Learn more >

- EMMA repository conditions
- SOPs and FAQs
- Major collections archived at EMMA
- EUCOMM/KOMP-CSD allele conversion service

Tweets by @InfrafrontierEU

 **INFRAFONTIER**
@InfrafrontierEU

Happy to announce an overwhelming response to our latest #opencall on #COVID19 Therapeutics Pipeline. More than 20 fascinating proposals from 8 countries for 5 slots. Looking forward to the evaluations and outcomes! bit.ly/3be0Q80



EMMA: current members

16 nodes / 13 countries



CNR/IBC

Istituto di Biologia Cellulare, Monterotondo, Italy

CNRS/CDTA

Centre de Distribution, de Typage et d'Archivage animal, Orléans, France

MRC/MGU

Mammalian Genetics Unit, Harwell, UK

Karolinska Institutet

Karolinska Institutet, Stockholm, Sweden

FCG/IGC

Instituto Gulbenkian de Ciência, Oeiras, Portugal

HMGU/IEG

Institute of Experimental Genetics, Munich, Germany

EMBL/EBI

European Bioinformatics Institute, Hinxton, UK

GIE-CERBM/ICS

Institut Clinique de la Souris, Illkirch/Strasbourg, France

Sanger Institute

Wellcome Trust Sanger Institute, Hinxton, UK

CNB/CSIC

Centro Nacional de Biotecnología, Madrid, Spain

Fleming

Biomedical Sciences Research Center Al. Fleming, Athens, Greece

OULU

University of Oulu, Oulu, Finland

BIAT

Vetmeduni Vienna, Biomodels Austria, Vienna, Austria

IMG

Institute of Molecular Genetics, Prague, Czech Republic

TAU

Tel Aviv University, Tel Aviv, Israel

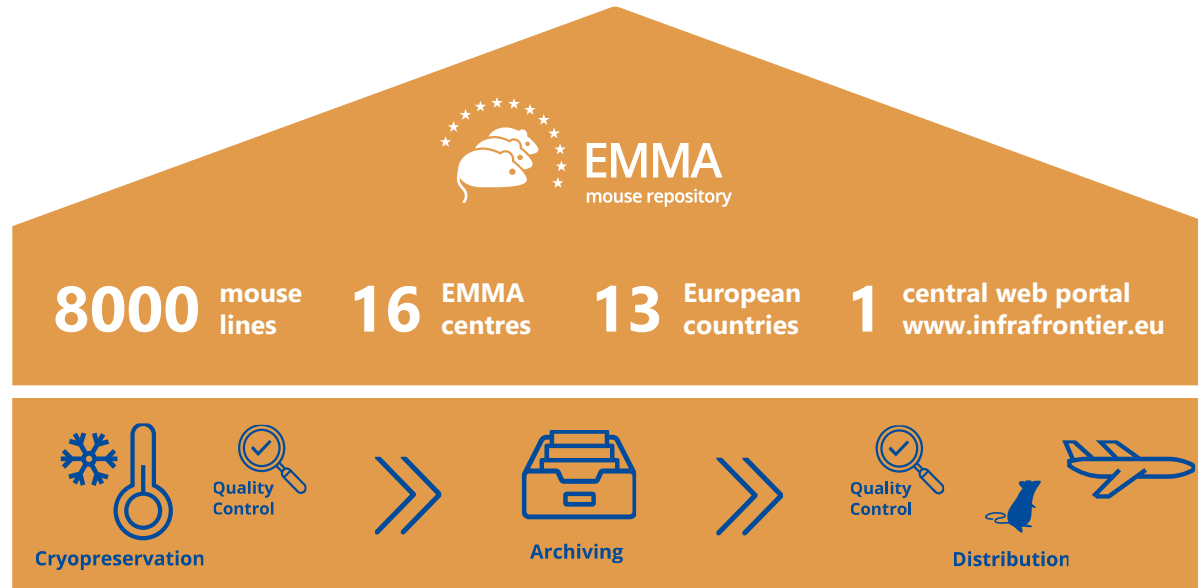
NKI

Netherlands Cancer Institute, Netherlands, Amsterdam

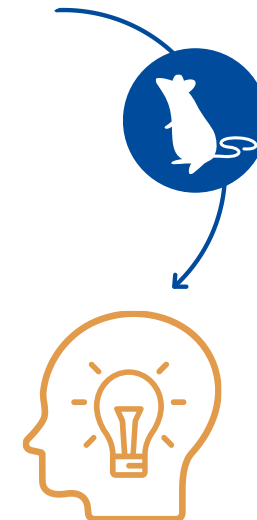


www.infrafrontier.eu

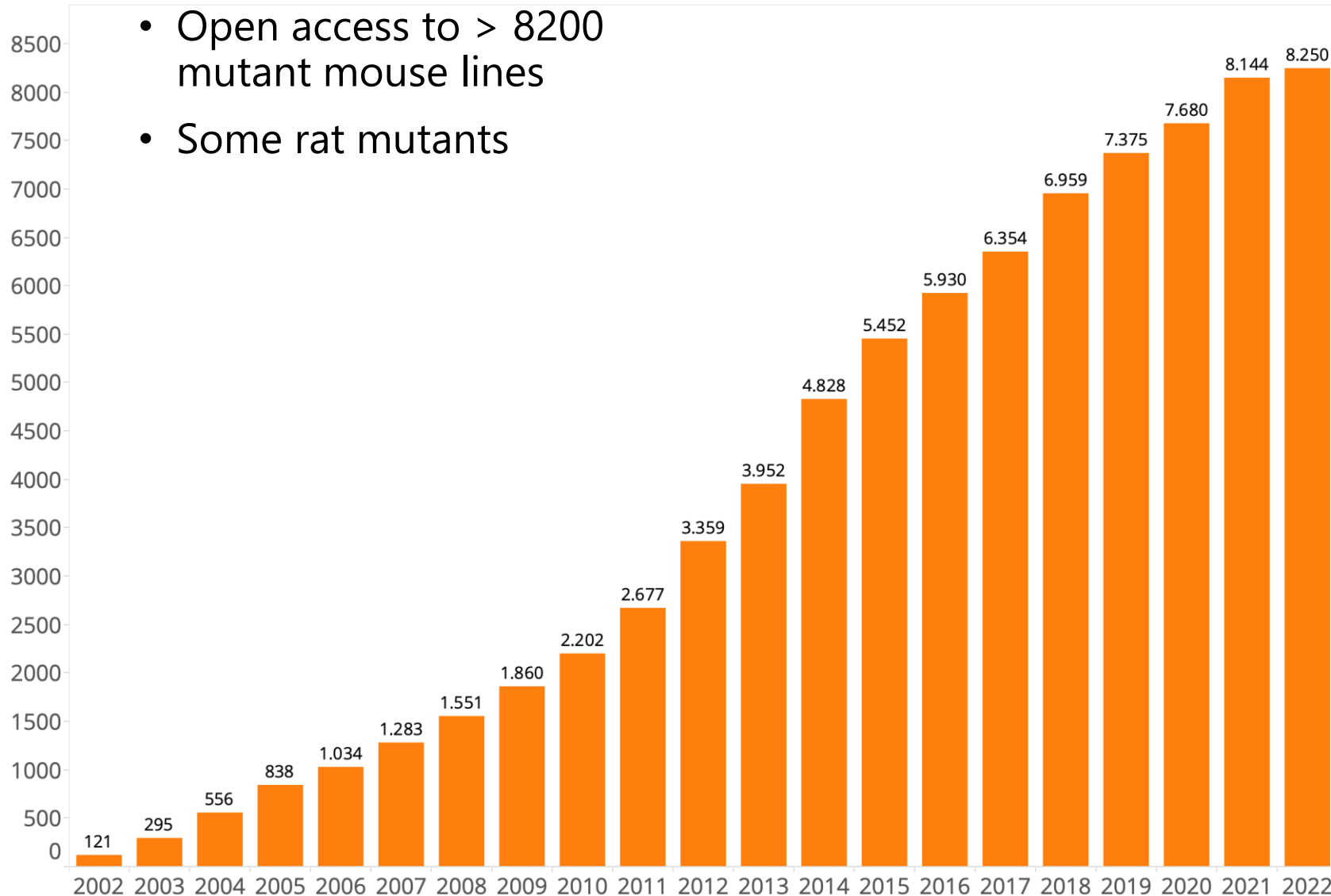
The European Mouse Mutant Archive (EMMA)



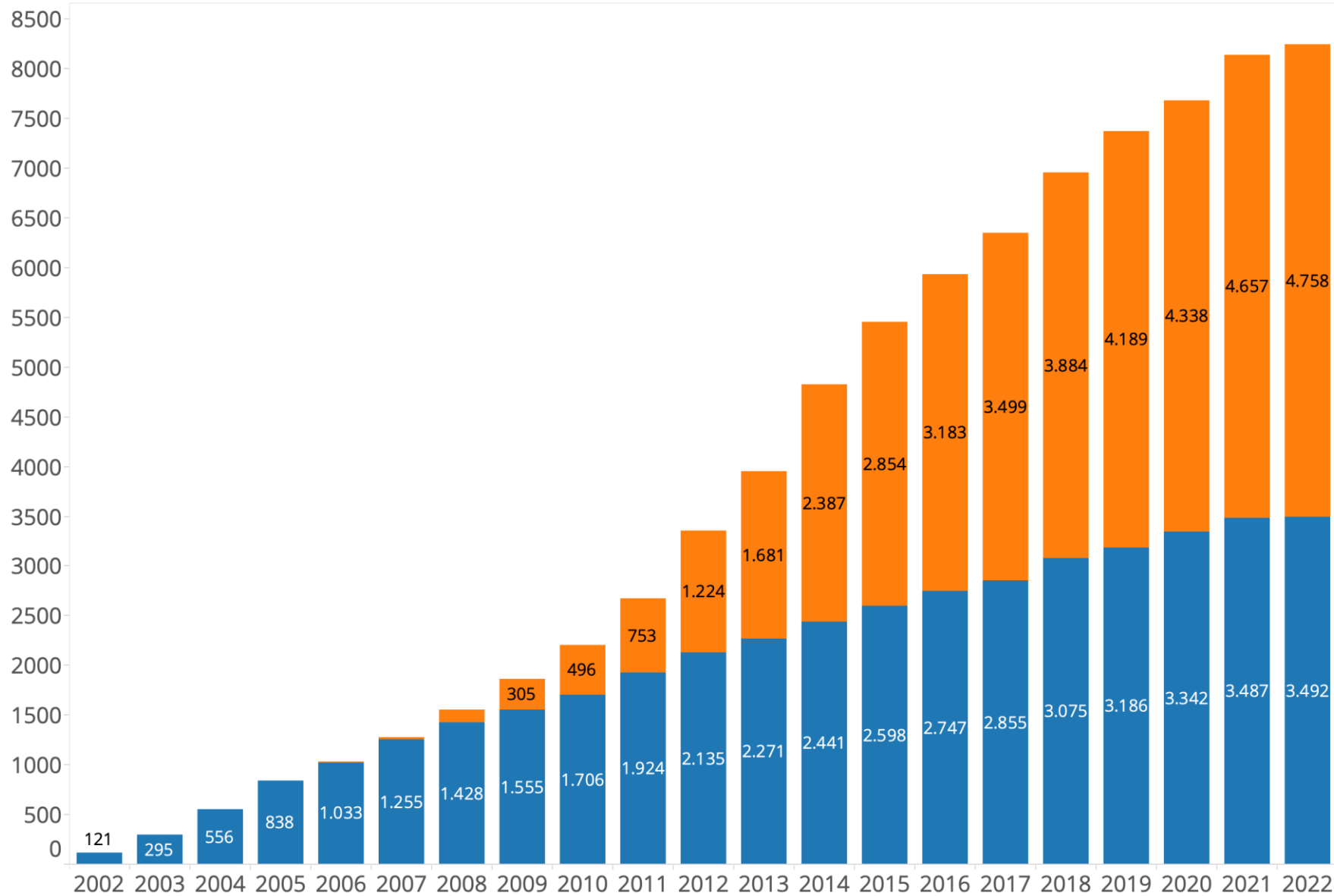
- **Open access to more than 8000 mouse lines**
- High-quality health controlled mice with defined and verified genotype
- EMMA ships globally



3rd largest non-profit mouse repository worldwide



Community vs IMPC strains

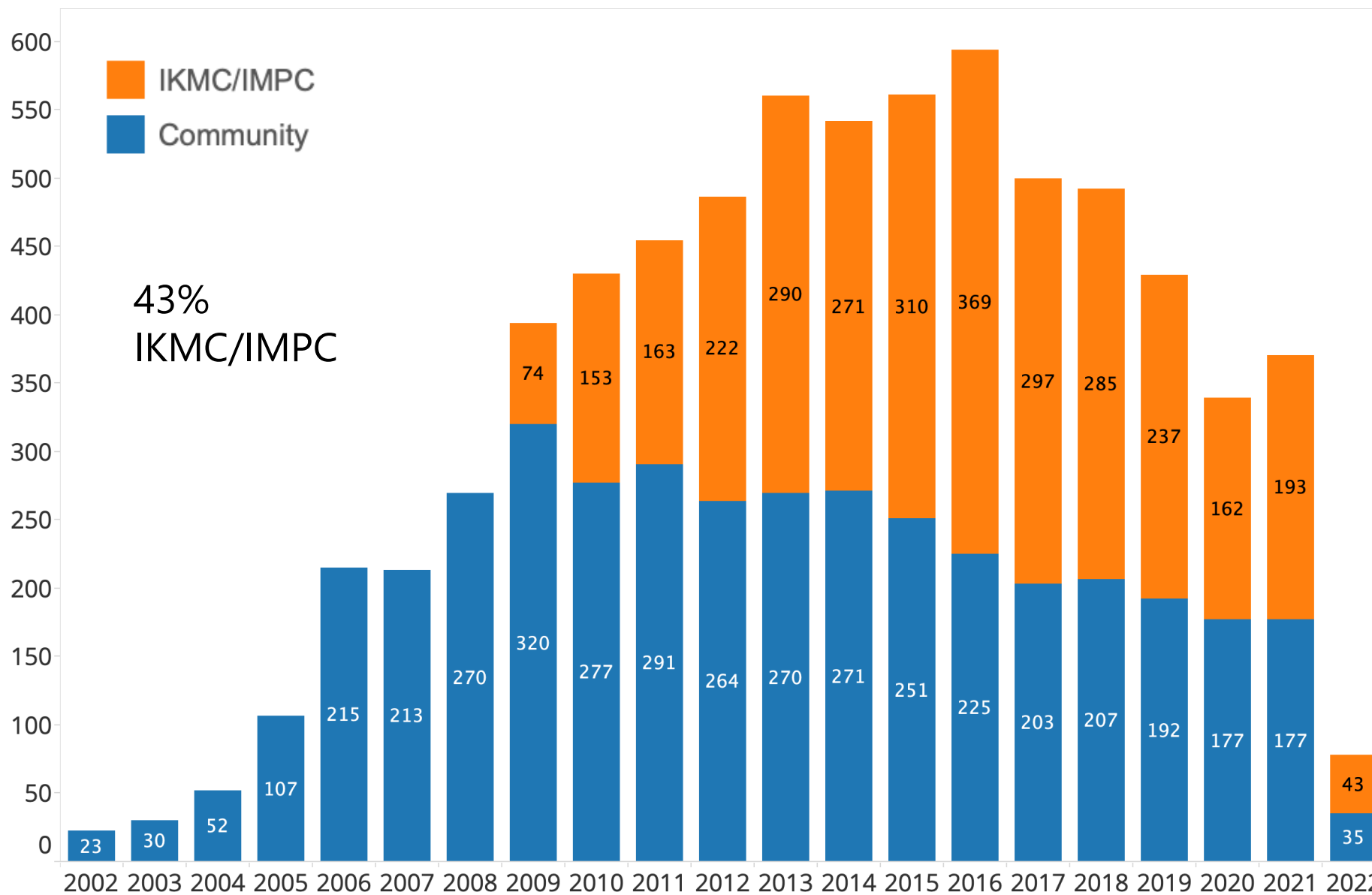


58%

42%

as of 2022-03-22

Requests from EMMA



**Total
numbers**

3069

4076

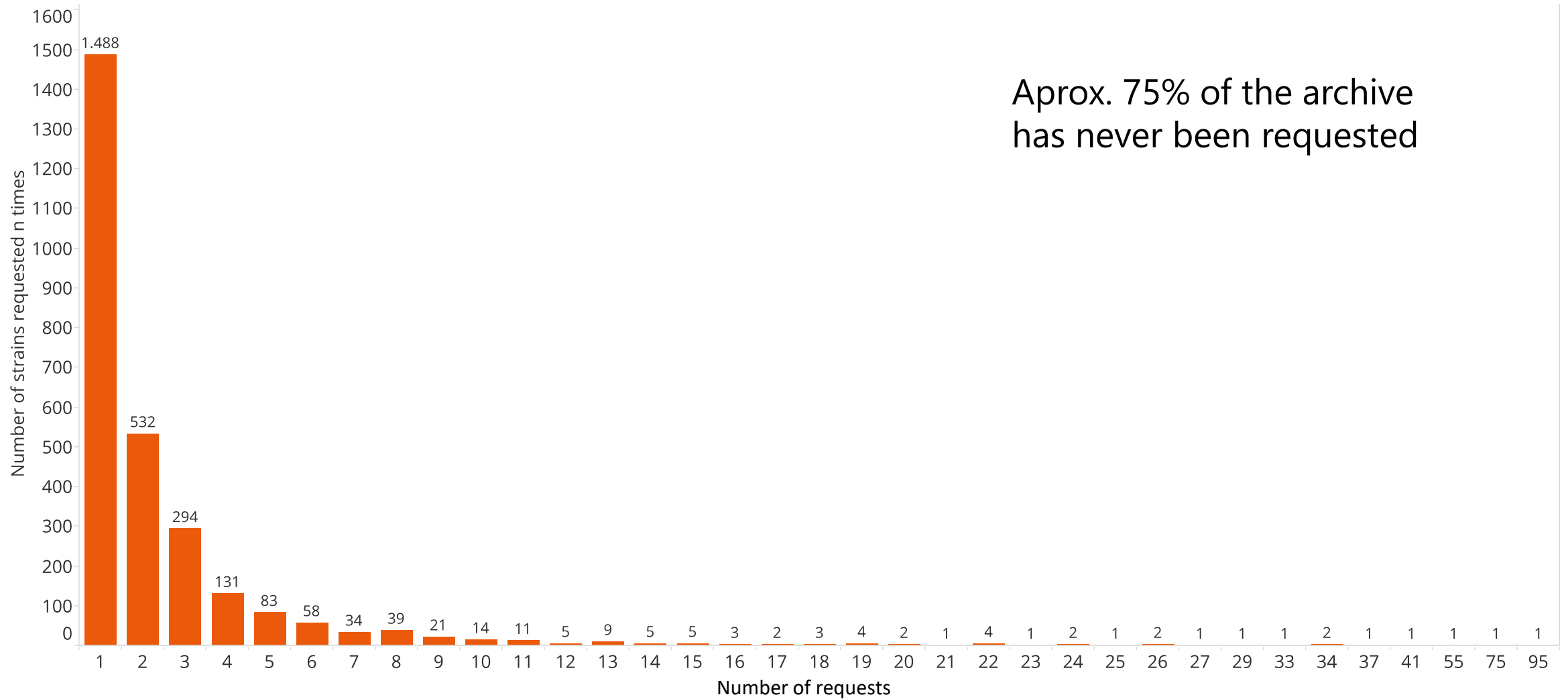
as of 2022-03-22

EMMA Shipping Destinations



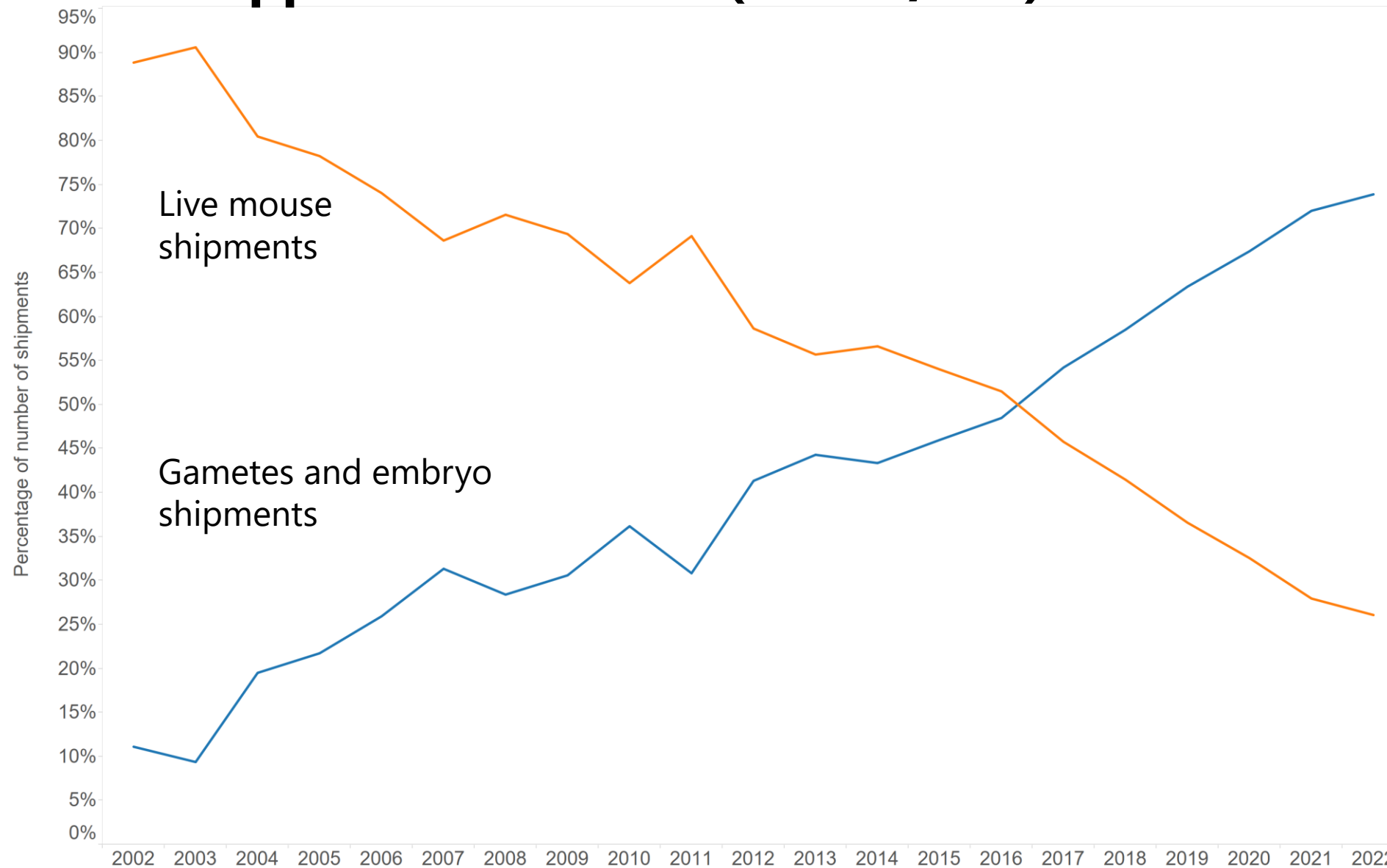
700 unique
destinations
(cities)
worldwide

Number of strains requested N times



Aprox. 75% of the archive
has never been requested

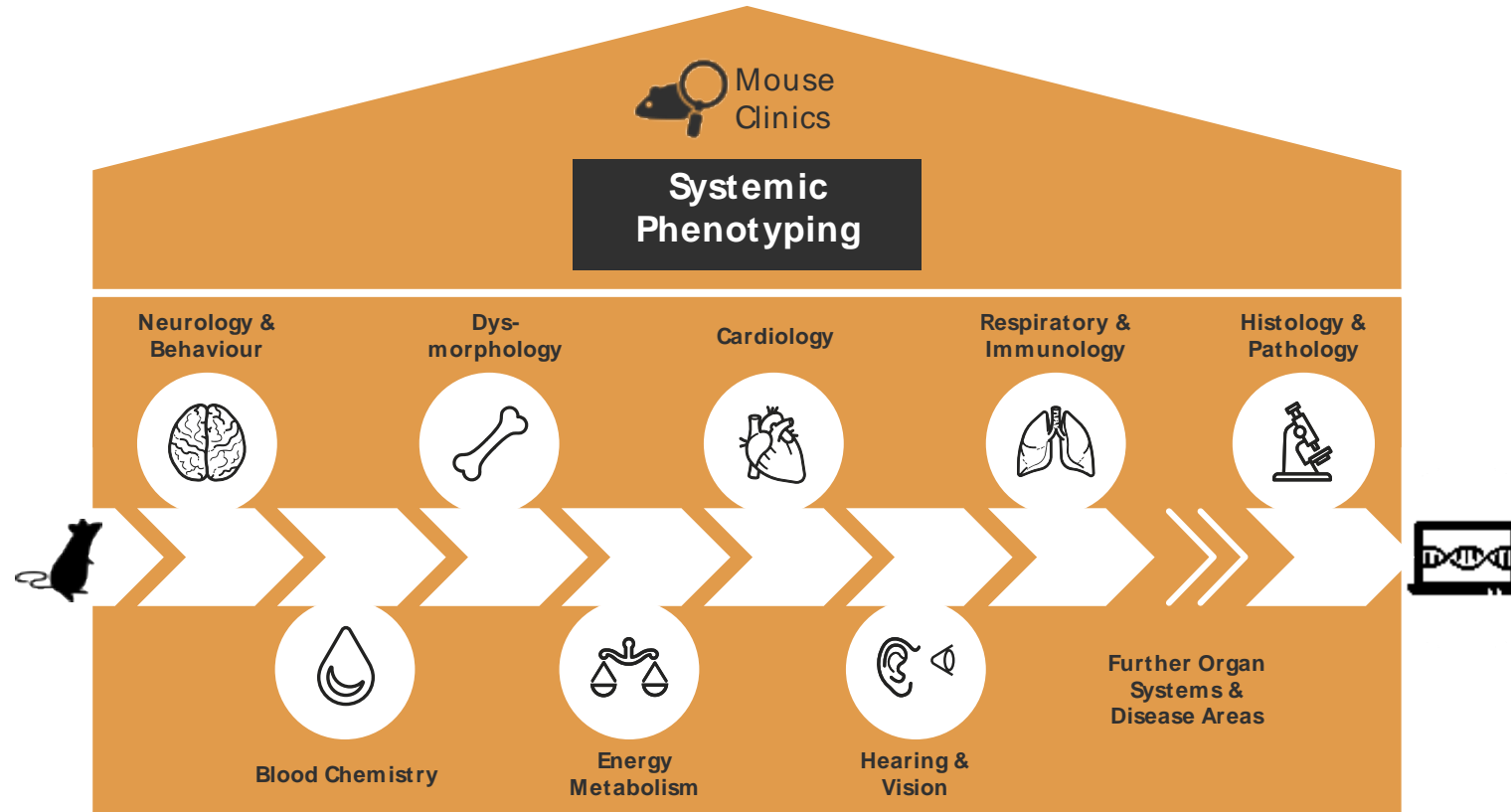
Material shipped from EMMA (frozen/live)



Impact of INFRAFRONTIER on ANIMAL WELFARE

- Number of animals “saved” (not bred, not born, not used) because they are cryopreserved
- ~50 animals/year needed to maintain a colony
- ~ 8.000 mouse lines archived in EMMA-INFRAFRONTIER
- **Up to 400.000 mice are saved (= not born) per year**

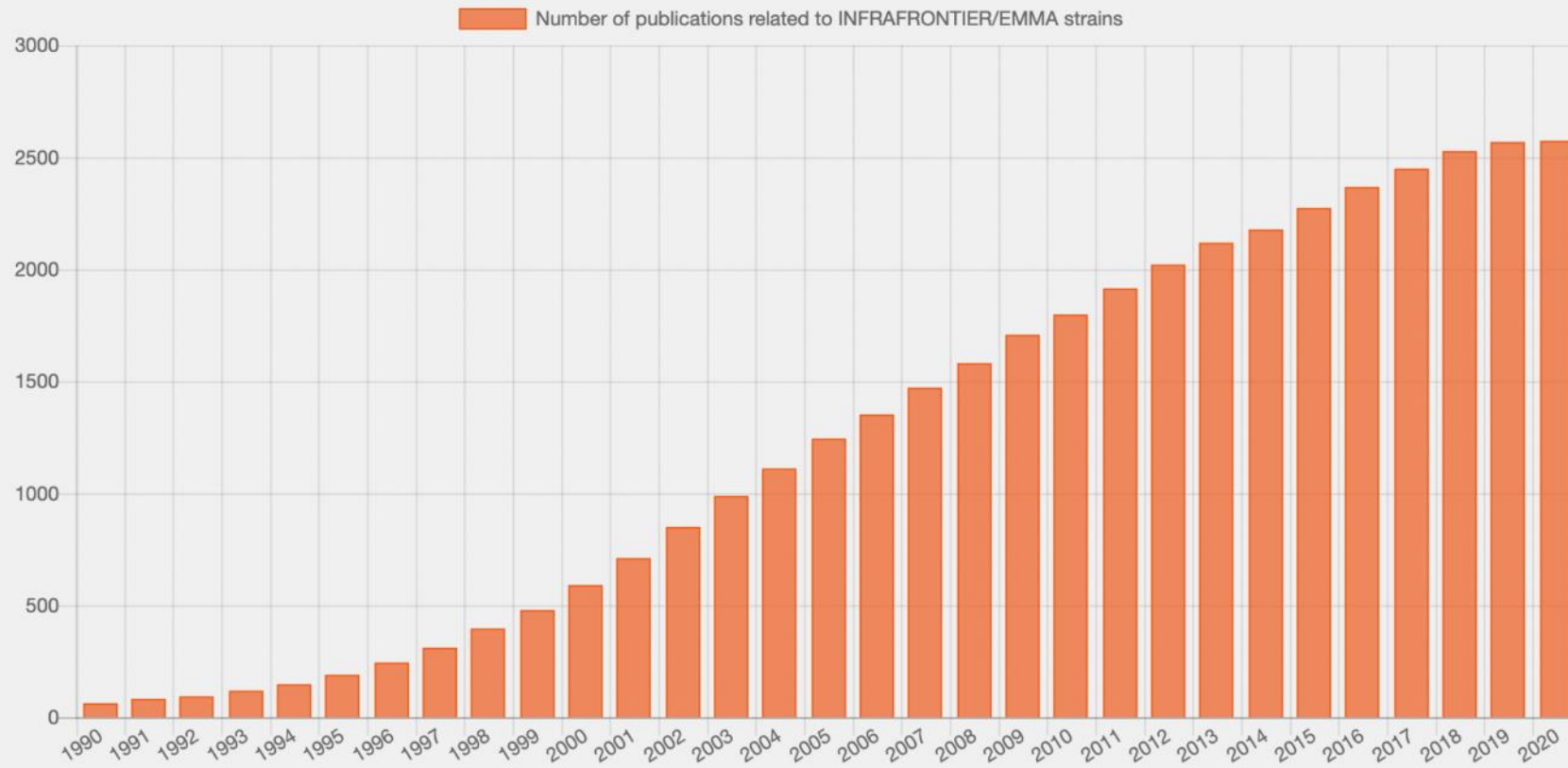
Systemic phenotyping and mouse clinics



- Mouse clinics offer a **whole–system screen** covering more than 500 parameters in different disease areas and organ systems
- Added benefit of obtaining **maximum information** using a **minimum amount of animals**

Publications related to INFRAFRONTIER / EMMA

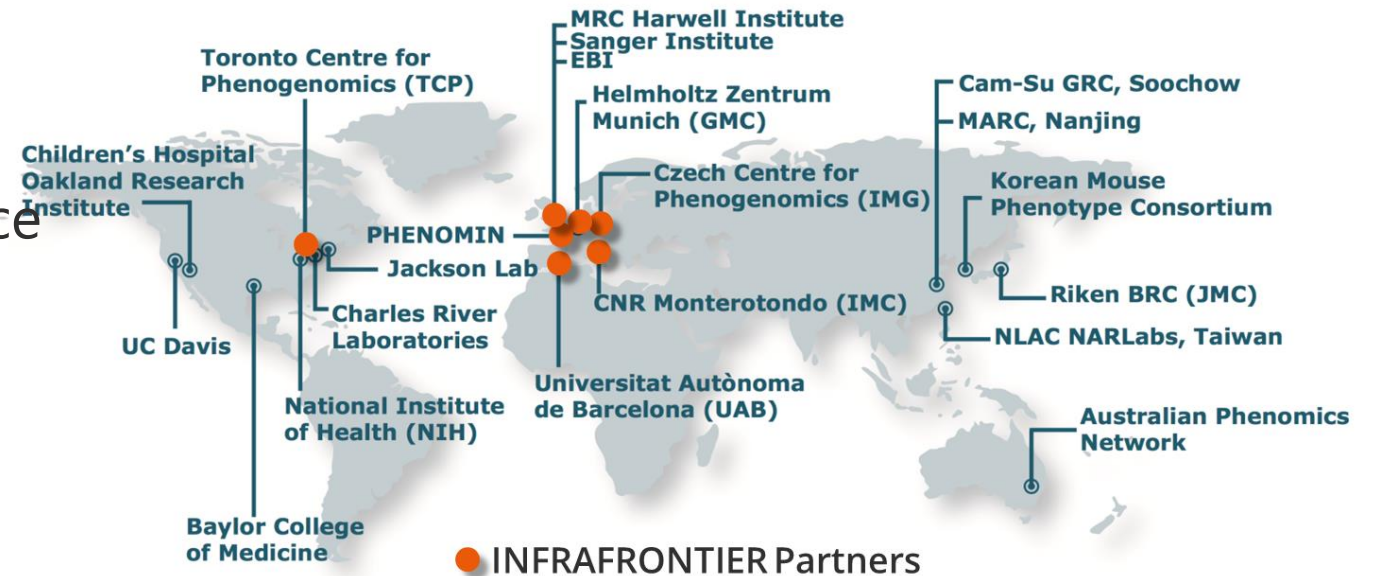
INFRAFRONTIER bibliography



- **2574** publications; both from strain submissions and strain requests
- **49** Cell publications
- **43** Nature Genetics publications
- **82** Nature publications

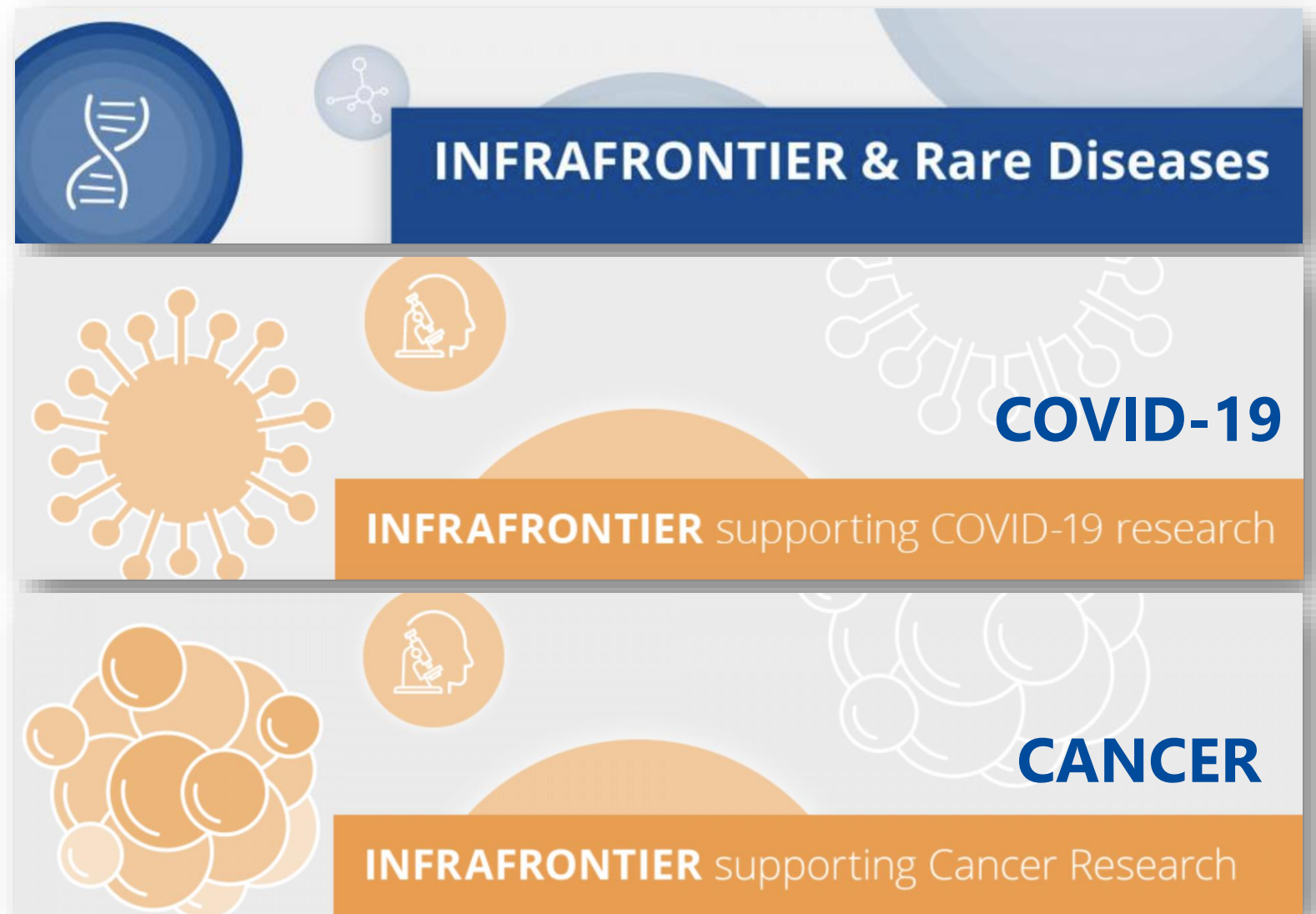
INFRAFRONTIER and IMPC

- International **M**ouse **P**henotyping Consortium
- Systemic phenotyping of knock-out mice for all ~20.000 protein coding genes → **comprehensive catalogue of gene function**
- Phenotyping data and mice are made **openly available** to the biomedical community
- **INFRAFRONTIER is the largest distributor of IMPC lines worldwide - 350 shipments per year**



INFRAFRONTIER Therapeutic Areas

Collection of
INFRAFRONTIER resources
and services related to a
therapeutic area in one
page

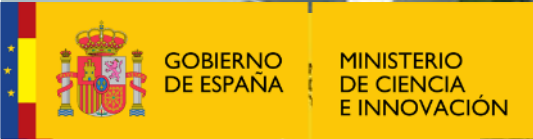


The image displays three horizontal banners, each representing a therapeutic area. The top banner is blue and features a DNA double helix icon on the left and a molecular structure icon in the center. The text 'INFRAFRONTIER & Rare Diseases' is written in white on a dark blue background. The middle banner is orange and features a virus particle icon on the left and a microscope icon in the center. The text 'COVID-19' is written in blue on the right, and 'INFRAFRONTIER supporting COVID-19 research' is written in white on an orange background. The bottom banner is light blue and features a cluster of cells icon on the left and a microscope icon in the center. The text 'CANCER' is written in blue on the right, and 'INFRAFRONTIER supporting Cancer Research' is written in white on an orange background.

INFRAFRONTIER & Rare Diseases

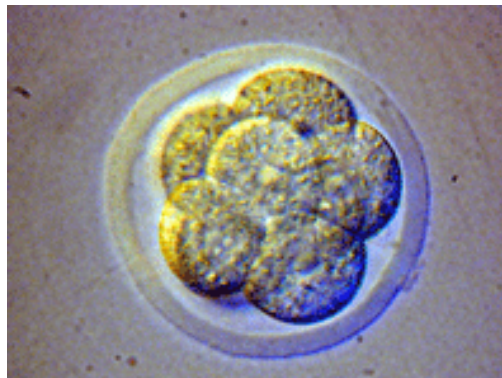
COVID-19
INFRAFRONTIER supporting COVID-19 research

CANCER
INFRAFRONTIER supporting Cancer Research



The EMMA-Infrafrontier Spanish node at CNB

Julia Fernández & Lluís Montoliu



EMMA-Infrafrontier Services provided from CNB

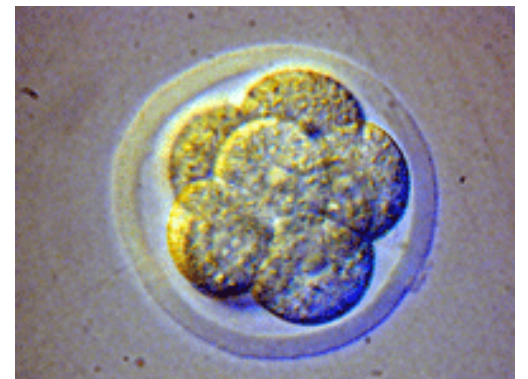


- **Depositing mouse lines into EMMA**
 - Depositing requests evaluated by external committee
 - Approved requests are cryopreserved for **free**
 - Depositor covers shipping costs
 - Cryopreserved mouse lines freely available to everyone
 - Grace period possible (up to 2 years), MTAs accepted
- **Requesting mouse lines from EMMA**
 - Access to EMMA archive requires payment of **repository fee**
 - Frozen sperm/embryos: 1,600 Euros + shipping costs
 - Live mice: 3,200 Euros + shipping costs



EMMA procedures at CNB - I

- **Cryopreservation of mouse strains**
 - Cryopreservation of mouse embryos, sperm, oocytes
 - Genotyping (single embryos) and quality control
- **Rescue of mouse strains from frozen material**
 - Thawing of mouse embryos, sperm, oocytes
 - In vitro fertilization and embryo transfer
 - Genotyping and quality control



EMMA procedures at CNB - II

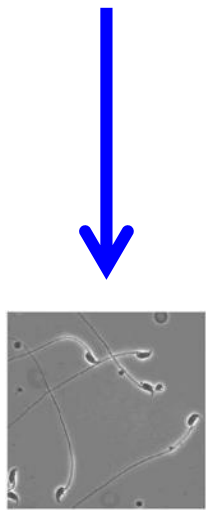
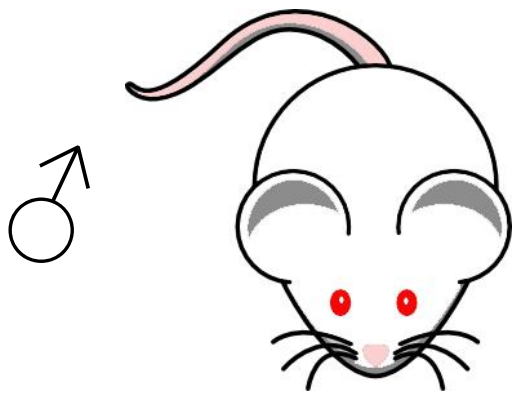
- **Importing mouse lines**

- Receiving frozen mouse embryos (LN₂)
- Receiving frozen mouse sperm (LN₂ and dry ice)
- Receiving refrigerated mouse embryos (+4/+8°C)
- Receiving refrigerated mouse epididymes (+4/+8°C)
- Receiving live mice

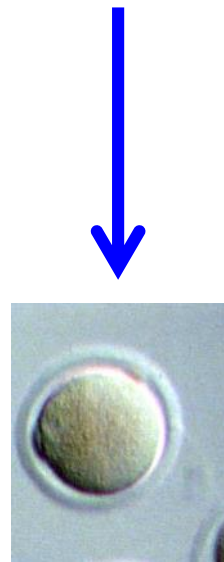
- **Exporting mouse lines**

- Shipping frozen mouse embryos (LN₂)
- Shipping frozen mouse sperm (LN₂ and dry ice)
- Shipping refrigerated mouse embryos (+4/+8°C)
- Shipping refrigerated mouse epididymes (+4/+8°C)
- Shipping live mice

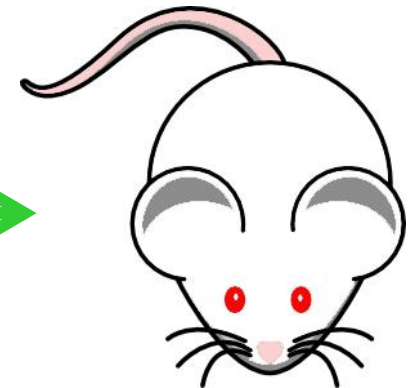




IVF



QC OK



FREEZE ↓ ↑ **THAW**

SPERM CRYOPRESERVED

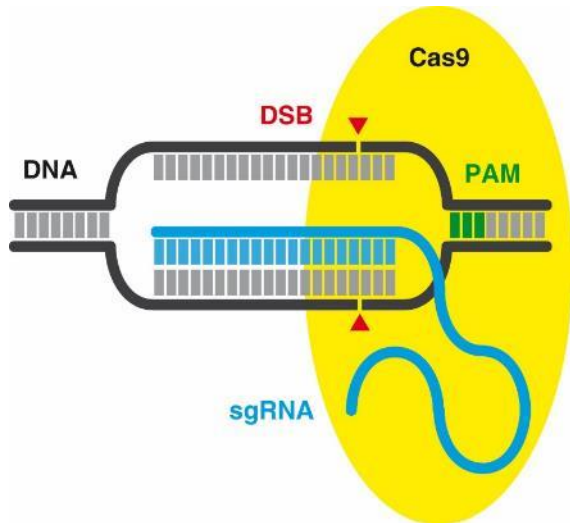
FREEZE ↓ ↑ **THAW**

EMBRYOS CRYOPRESERVED

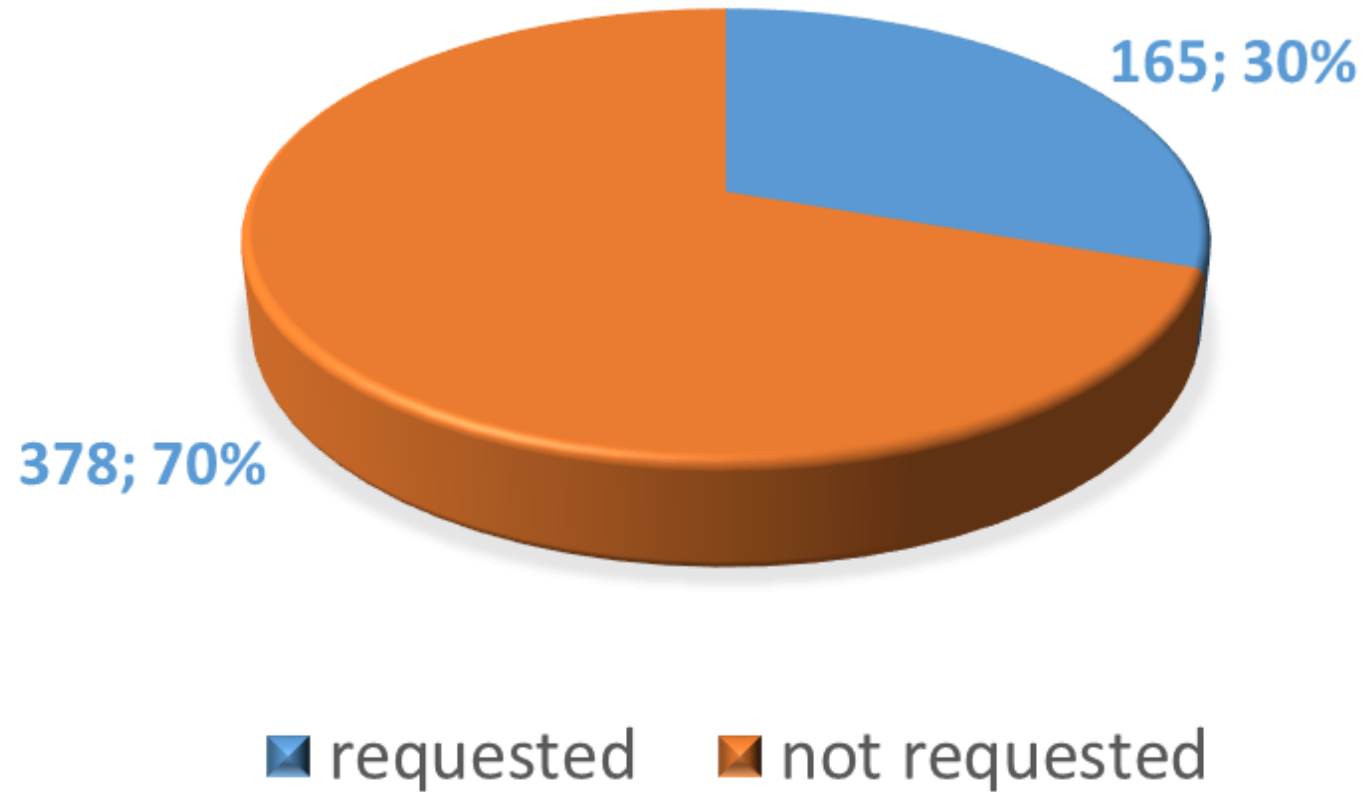
RESCUE

EMMA procedures at CNB - III

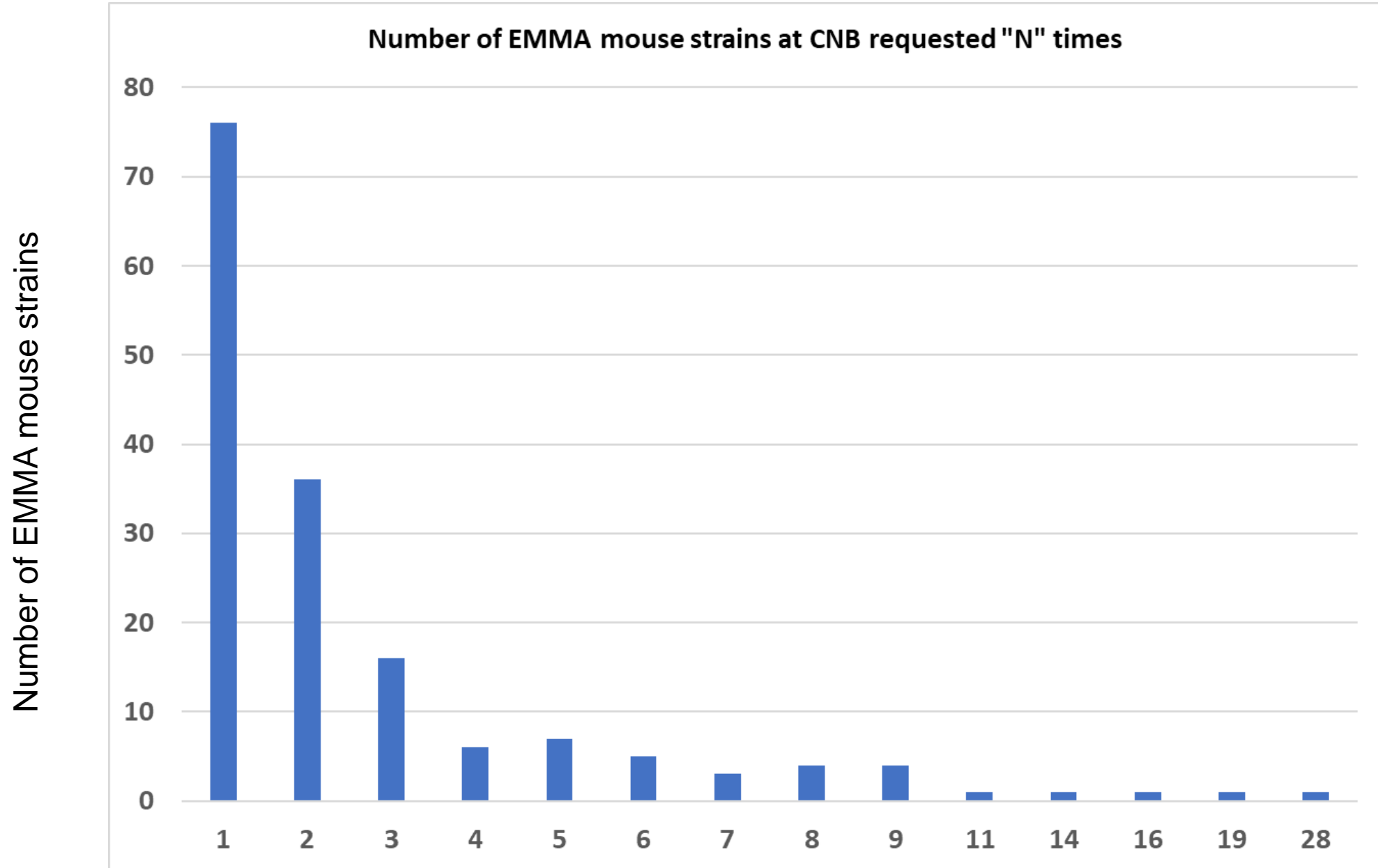
- Rederivation of mouse strains (into SPF zone)
- Generation of knockout mice from ES cell clones (TNAs)
- Generation of genome edited (CRISPR) mice (TNAs)
 - Advice / experimental design / guide RNA selection
 - Gene disruptions
 - Gene deletions/inversions
 - Gene substitutions/mutations (ssDNA oligos)
 - Electroporation of mouse embryos
 - Embryo transfer into pseudopregnant females



EMMA MOUSE LINES AT CNB-CSIC



EMMA lines in total: 8000
EMMA lines at CNB: 543



165 mouse lines requested

Number of times requested

INFRAFRONTIER & CRISPR

- Helmholtz Munich licensed the legal distribution of CRISPR mouse lines from **Caribou** and **BROAD**
- CRISPR licenses are paid annually and are distributed among INFRAFRONTIER participating partners
- 10% of INFRAFRONTIER repository fee is added for CRISPR mouse lines to compensate the royalties paid
- CNB-CSIC is offering the generation of CRISPR mouse lines for third parties as long as they are deposited in and obtained from INFRAFRONTIER

Services provided	CNB	CSIC Centres	Public Centres	Private Centres
Cryopreservation and validation of a mouse line as embryos (3xE + 3xA + 1xC) (1 unit = 1 mouse line cryopreserved as embryos)	596,24€	699,38€	1170,96€	1282,48€
Cryopreservation and validation of a mouse line as sperm (B + D) (1 unit = 1 mouse line cryopreserved as sperm)	485,95€	570,01€	746,46€	781,56€
Revitalizing a mouse line from frozen embryos (C + 3xF) (1 unit = 1 mouse line revitalized from embryos)	242,96€	284,99€	428,97€	469,84€
Revitalizing a mouse line from frozen sperm (D + 3xF) (1 unit = 1 mouse line revitalized from sperm)	561,88€	659,08€	899,05€	984,69€
Generation of a mouse model genetically edited with CRISPR-Cas9 technology through EMMA-INFRAFRONTIER as frozen embryos or sperm (L + <u>1600€ service fee</u> + 160€ CRISPR license) (1 unit = 1 mouse line edited with CRISPR-Cas9 and obtained from EMMA-INFRAFRONTIER as frozen embryos or sperm)	3353,61€	3629,30€	3929,31€	4135,92€
Generation of a mouse model genetically edited with CRISPR-Cas9 technology through EMMA-INFRAFRONTIER as live animals (L + <u>3200€ service fee</u> + 320€ CRISPR license) (1 unit = 1 mouse line edited with CRISPR-Cas9 and obtained from EMMA-INFRAFRONTIER as live animals)	5113,61€	5389,30€	5689,31€	5895,92€



New!



New!

A- Cryopreservation of mouse embryos (1 unit = 1 session)	52,55€	61,64€	133,05€	145,72€
B - Cryopreservation of mouse sperm (1 unit = 1 session)	140,16€	164,40€	233,15€	255,36€
C - Thawing mouse embryos (1 unit = 1 session)	26,87€	31,52€	43,23€	47,35€
D - In Vitro Fertilization from frozen mouse sperm (1 unit = 1 session)	345,79€	405,61€	513,31€	562,20€
E - In Vitro Fertilization from fresh mouse sperm (1 unit = 1 session)	137,24€	160,98€	242,86€	265,99€
F - Embryo Transfer to pseudopregnant females (1 unit = 1 female)	72,03€	84,49€	128,58€	140,83€
G - Importing refrigerated sperm/epididymis (1 unit = 1 service)	255,21€	299,37€	376,90€	412,79€
H - Maintaining cryopreserved vials in liquid nitrogen (1 unit = 1 vial/year)	3,69€	4,33€	9,93€	10,87€
I - Mouse genotyping (1 unit = 1 mouse)	3,20€	3,75€	6,09€	6,67€
J - Lending vapour phase nitrogen containers (1 unit = 1 service)	158,19€	185,56€	230,83€	252,82€
K - Mouse embryos for validating equipment and procedures (1 unit = 1 straw)	147,64€	173,19€	256,36€	280,77€
L - Generation of a mouse model with CRISPR* (will be deposited in EMMA-INFRAFRONTIER, mandatory) (1 unit = 1 mouse line edited with CRISPR deposited in EMMA-INFRAFRONTIER)	1593,61€	1869,30€	2169,31€	2375,92€

EMMA – INFRAFRONTIER fees at CNB-CSIC – May 2022



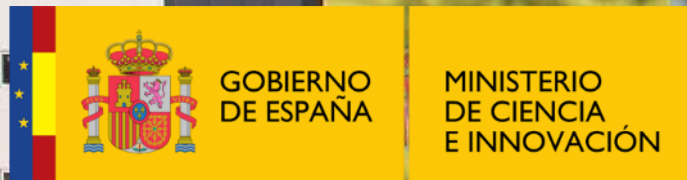
**CNB Mouse Embryo
Cryopreservation
Facility
Spanish EMMA node**

Julia Fernández
María Jesús del Hierro
Marta Castrillo
Cristina Bernal
Inés Arroba
Lluís Montoliu



www.user.cnb.csic.es/criocnb/

CNB Mouse Cryopreservation Facility



**Spanish EMMA-Infrafrontier
node at CNB-CSIC in Madrid**

criocnb@cnb.csic.es

www.user.cnb.csic.es/criocnb/