

Genomika a LZ

- ✓ Komparativní genomika
- ✓ Definice a standardizace kmenů na úrovni celých genomů

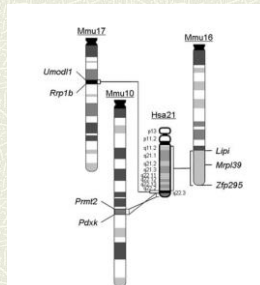
1

Komparativní genomika a LZ

- ✓ *Genomy LZ*
- ✓ *Genové manipulace a editace genomu LZ*

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Připomenutí: *Myš jako model Downova syndromu*



> <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC2893810/>

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Genomika a LZ

- ✓ Komparativní genomika
- ✓ Definice a standardizace kmenů na úrovni celých genomů

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Zvířecí (animální) laboratorní modely

- Nematoda
- Dáno
- Drosophila
- Ptáci, hlodavci, domácí zvířata, lidopii

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Netradiční laboratorní modely

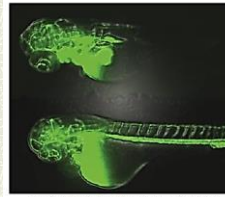
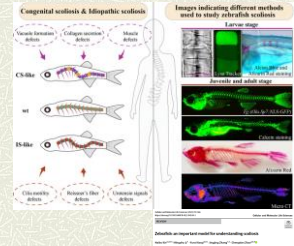


Image courtesy of Randall Peterson, Harvard Medical School and Massachusetts General Hospital, Boston, MA.

Fluorescent microangiograms of zebrafish homozygous for the gridlock mutation, which models human coarctation of the aorta. The vascular defect apparent in the upper embryo has been corrected in the lower embryo by treatment with the small molecule GS4012.



Single Cell/Nucleus Transcriptomics Comparison in Zebrafish and Human Nucleus Coaxons and Distal Molecular Responses to Alzheimer's Disease

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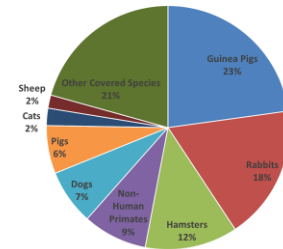
Netradiční laboratorní modely

| Model | Characteristics | Advantages | Disadvantages | Remarks |
|-------------------|---|---|---|---|
| Zebrafish | Classical invertebrate model | Fast to culture | 1. No nucleus, but an AI (AI) is present | Nonsequence microarray model |
| Gene editing | Method of genetic alteration | 1. The gene expression levels are stable and specific (CRISPR-Cas9) | 2. The research cycle is fast (2-3 weeks) | 1. CRISPR model 2. CRISPR-Cas9 model |
| Flu | Classical invertebrate model | 1. The gene expression levels are stable and specific (CRISPR-Cas9) | 2. The research cycle is fast (2-3 weeks) | 1. CRISPR model 2. CRISPR-Cas9 model |
| Mouse | Physical and chemical induction of disease | 1. The gene expression levels are stable and specific (CRISPR-Cas9) | 2. The research cycle is fast (2-3 weeks) | 1. CRISPR model 2. CRISPR-Cas9 model |
| Gene editing | Method of genetic alteration | 1. The gene expression levels are stable and specific (CRISPR-Cas9) | 2. The research cycle is fast (2-3 weeks) | 1. CRISPR model 2. CRISPR-Cas9 model |
| Gene modification | Method of genetic alteration | 1. The gene expression levels are stable and specific (CRISPR-Cas9) | 2. The research cycle is fast (2-3 weeks) | 1. CRISPR model 2. CRISPR-Cas9 model |
| Neurodegeneration | 1. The gene expression levels are stable and specific (CRISPR-Cas9) | 2. The research cycle is fast (2-3 weeks) | 3. The research cycle is fast (2-3 weeks) | 1. CRISPR model 2. CRISPR-Cas9 model |

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Využití LZ (USA)

Number of Animals used in research in the US in 2019 by Species



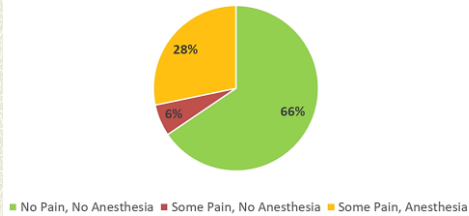
| Species | Number of Animals | % of Total | % Change from 2018 |
|-----------------------|-------------------|------------|--------------------|
| Guinea Pigs | 141,472 | 23.0% | 7% |
| Rabbits | 141,472 | 18.0% | 1% |
| Other Covered Species | 141,472 | 21.0% | 2% |
| Hamsters | 141,472 | 12.0% | 1% |
| Non-Human Primates | 141,472 | 9.0% | 1% |
| Dogs | 141,472 | 7.0% | 1% |
| Cats | 141,472 | 6.0% | 1% |
| Sheep | 141,472 | 2.0% | 1% |
| Other Covered Species | 141,472 | 20.7% | 1% |
| Total | 617,546 | 100% | 2.2% |

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Využití LZ (USA procedures)

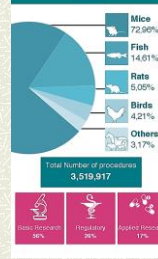
Proportion of Animals used in Research in the US in 2019 by Pain/Anesthesia category



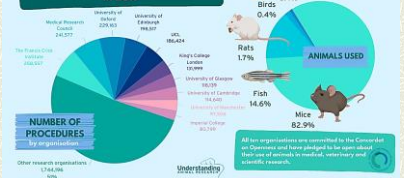
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Využití LZ (UK)

Statistics of Scientific Procedures on living animals 2019



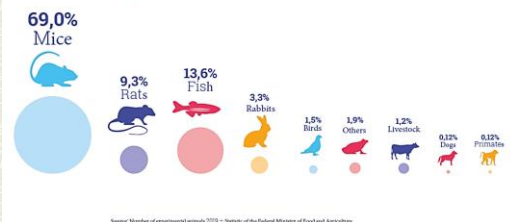
The 10 organisations that carry out the most animal research in Great Britain in 2019



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Využití LZ (DE)

What proportion do the different animal groups have in the experimental animals?



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Zvířecí (animální) laboratorní modely

- Nematoda
- Dánio
- Drosophila
- Ptáci, hlodavci, domácí zvířata, lidoopi

12

Laboratory Mouse

Education

Caltech, Oxford, Stanford, Harvard, MIT, Princeton, Cambridge, Imperial, Berkeley, Chicago, Yale, ETH Zurich, Columbia, UPenn, John Hopkins, UCL, Cornell, Northwestern, IMichigan, Toronto, Carving Melton, Duke, Washington, UTexas at Austin, CA Tech, Tokyo, Melbourne, Singapore, UBC, Wacomen-Madison, Edinburgh, McGill, Hong Kong, Santa Barbara, Karolinska Institute, Wisconsin, Manchester... and just about every other major university, medical school & research institution in the world.

Nobel Prizes

- 1905 - Transmission and treatment of TB
- 1906 - Structure of Nervous System
- 1907 - Role of proteins in disease
- 1908 - Immunity to infectious diseases
- 1928 - Investigations on typhus
- 1929 - Importance of dietary vitamins
- 1939 - Discovery of antibacterial agent, Penicillin
- 1945 - Discovery of penicillin
- 1951 - Yellow fever vaccine
- 1952 - Discovery of streptomycin
- 1954 - Culture of the polio virus
- 1960 - Understanding of immunity
- 1975 - Understanding of neurotransmitters
- 1974 - Structural & functional organization of cells
- 1975 - Tumour-viruses and genetics of cells
- 1977 - Hypothalamic hormones
- 1984 - Techniques of monoclonal antibody formation
- 1986 - Nervous growth factor and epidermal growth factor
- 1990 - Organ transplantation techniques
- 1992 - Regulatory mechanisms in cells
- 1996 - Immune-system detection of virus-infected cells
- 1997 - Discovery and characterization of prions
- 1999 - Discovery of signal peptides
- 2000 - Signal transduction in the nervous system
- 2004 - Odour receptors and organization of olfactory systems
- 2008 - Role of HIV and HIV in causing disease
- 2010 - Development of in vitro fertilisation
- 2011 - Discoveries around insulin and adaptive immunity



CV of a Lifesaver

Overview

- Involved in around 75% of research
- Short life-span and fast reproductive rate means mice are suitable for studying disease across whole life cycle
- 90% of genes have comparable genes in humans
- Similar reproductive and nervous systems and suffer many of the same diseases as humans including cancer, diabetes and anxiety
- Can be genetically modified to include human genes to enhance biological relevance
- Can act as an avatar for a human cancer to allow drug therapies to be trialled safely

Research Areas

- Alzheimer's disease, anaesthetics, AIDS & HIV, antiangioplasts, antidepressants, arthritis, blindness, bone and joint disease, brain injury, breast cancer, cardiac arrest, cystic fibrosis, deafness/hearing impairment, Down's syndrome, drugs for high blood pressure, transplant rejection, Hepatitis B, C & E, Huntington's disease, influenza, leukaemia, malaria, motor neurone disease, multiple sclerosis, muscular dystrophy, Parkinson's disease, prostate cancer, schizophrenia, spinal cord injury, stroke, testicular cancer, tuberculosis,

Contact

www.informaticsjax.org
www.jax.org
www.annualresearch.info
www.angegroup.org
www.gpnl.org/research.com

Myši model

- ✓ Druhý sekvenovaný savčí genom (2002)
- ✓ Pravidla a nomenklatura
<http://www.informaticsjax.org/>
- ✓ Databáze

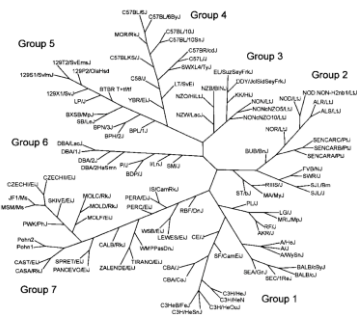
Významné stránky

TABLE 1 Selected Genetic and Strain Databases Available on the World Wide Web (WWW)

| Site | Contents | Web address (URL) |
|--|--|--|
| Mouse Genome Database | Mapping data (all techniques) genetic, cytogenetic, physical, and comparative mapping data | http://www.informaticsjax.org |
| MGC ^a Mammalian Genetics Unit | Comparative maps, strain list | http://www.mmu.har.ac.uk/ |
| The Whole Mouse Catalog (formerly Mice and Rats Home Page) | Links to web sites for mouse and rat research | http://www.zoedenta.com/wmc/ |
| Animal Genome Database in Japan | Mouse genetic mapping data, cytogenetic maps | http://www.ninia.affrc.go.jp/ |
| Human Genome Database ^b | Human gene symbols | http://bioinfo.sickkids.on.ca/http://afdbwww.afdb.org/ |
| Human Gene Nomenclature Database | Human gene symbols | http://www.genom.ucsf.ac.uk/cp2/ http://www.hugobone.com/nomenclature.html |
| National Center for Biotechnology Information (NCBI) | Mouse/human comparative maps, links to other databases | http://www.ncbi.nlm.nih.gov/Homology/ |
| Rat Genome Database | Rat genetics | http://ratmap.gsu.edu/ |
| Roslin Institute Bioinformatics | Pig, sheep, cattle, chicken | http://www.rz.lboro.ac.uk/bioinformatics/ |
| FlyBase | <i>Drosophila</i> genomics | http://flybase.bio.indiana.edu/ http://silico.jhu.edu/~ce/ce1701/ |
| Zebrafish Informatics | Zebrafish genomics | http://zfdb.bio.scripps.edu/ZFDB/ |

^a MGC: Medical Research Council
^b Note: At the time of this writing, the Human Genome Database is in transition between the two sites listed <https://www.informaticsjax.org/>
From: Genetic and Phenotypic Definition of Laboratory Mice and Rats / What Constitutes an Acceptable Genetic/Phenotypic Definition

Genetika LZ: myši kmeny



Petkov et al. 2021

Kmeny/linie LZ (myši/LZ)

International Mouse Strain Resource Center
(IMSR)

<http://www.findmice.org/>

- **Existující: >24.000**
- **Potenciál: 200.000**

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Genetika LZ

Základní genetické rozdělení

- *Outbrední LZ*
- *Inbrední LZ*

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Genetika LZ

Základní genetické rozdělení

- *Outbrední LZ*
- *Inbrední LZ*

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Neinbrední kmeny myši

- ✓ Outbred Stock (OUT)
- ✓ Noninbred Stock (NON)
- ✓ Closed Colony (CCO)

#

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Genetika LZ

Základní genetické rozdělení

- *Outbrední LZ*
- *Inbrední LZ*

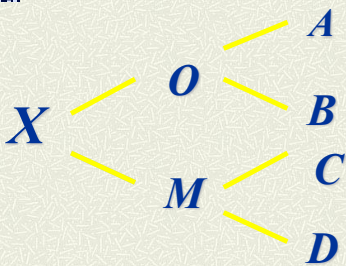
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Inbríding

Jev, který nastává po páření příbuzných jedinců

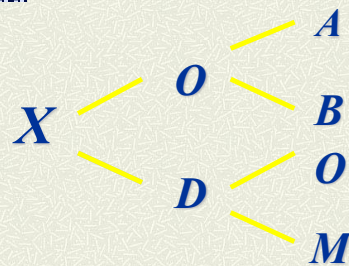
22

Inbríding



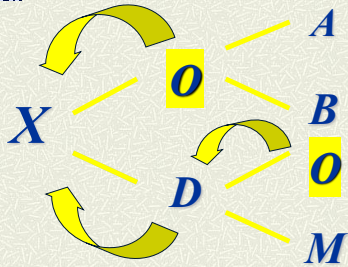
23

Inbríding



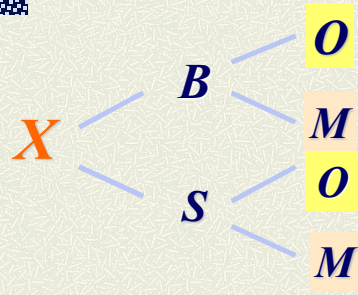
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Inbríding



25

Inbríding



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Inbríding

- Společný předek do 5. generace
- Homozygotnost

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Inbríding

- Účinky: *genotyp/genofond*
- Účinky: *fenotyp*

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Inbríding: účinky na fenotyp

kmen Vandoma, Zimbabwe (tzv. „Pštrosí lidé“): ektrodaktylie
 mormoni v Hilldale (Utah) a Colorado City (Arizona)
 amazonští Indiáni
 šlechtické rody



Karel II.: nepřirozeně velká hlava, deformovaná čelist, slabé tělo, potíže s chůzí a další defekty, mentální a psychické poruchy, impotence, neplodnost.

<https://slideplayer.cz/slide/564547/6/images/28/inbredn%C3%AD+depresive+u%C4%BDo%C4%B8ka%3A.jpg>

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Inbríding: účinky na fenotyp

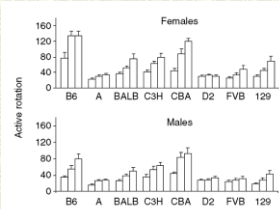


Figure 1: Rotorod performance of female and male inbred strains of mice, across three days, on a modified accelerating rotorod paradigm. Each bar represents active rotation behavior averaged over three trials for each of three consecutive test days. Error bars indicate standard error.

McFadyen et al. 2003

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Inbrední kmeny myší základní rozdělení

- ✓ Syngenní (isogenní)
- ✓ Rekombinantní
- ✓ Koisogenní
- ✓ Kongenní
- ✓ Konsomické, konplastické
- ✓ Mutantní
- ✓ Knock-out
- ✓ Transgenní/s editovaným genomem

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Syngenní (isogenní) kmeny (IS)

AaBb

AABB AAbb aaBB aabb

32

Syngenní (isogenní) kmeny (IS)

- Vzniklé opakovaným pářením bratr x sestra (>20 x)
- Geneticky identické - isogenní
- 100% (?) homozygotní

33

Rekombinantní kmeny (RI)

AA bb x aa BB

AaBb

AABB AAbb aaBB aabb

Segregating Inbred Strains (SEG)

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„Ko-“ kmeny

- Ko-izogenní: Coisogenic Strain (COI)
- Kongenní: Congenic Strain (CON)
- Konsomický: Consomic Strain (CSS)
- Konplastický: Conplastic Strain (CSS)

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Kongenní kmeny

- Vzniklé zpětnými kříženími
- Odlišnost v jediné oblasti (více lokusů)

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Kongenní kmeny

A x B

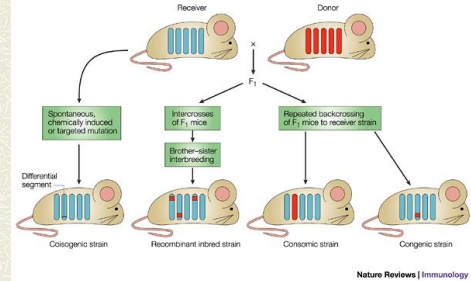
AB x B

ABB x B

ABBBBBBBBBBBBBBBBBBBBBBB

37

Typy kmenů



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Mutantní kmeny

- Mutant Strain (MSR)
- Mutant Stock (MSK)

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Jiné kmeny

- ✓ Wild-Derived Inbred Strains (WDS)
- ✓ Knock-out
- ✓ Transgenní/s editovaným genomem

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Specifické myší modely

Cellular and Molecular Biology | 10.1016/j.cel.2019.06.046

REVIEW

New developments in prion disease research using genetically modified mouse models

Johanna L. Suss¹, Glenn C. Telling²

Cells, 2019 Jun, 8(6): 546.
Published online 2019 Jun 6 doi: 10.1016/j.cel.2019.06.046

Mouse Models for Food Allergies: Where Do We Stand?

Stefan Schüler¹ and Melanie Albrecht

Figure 1 A transgenic mouse model of prion disease. The mouse genome contains a prion protein gene (Prn^C) that is flanked by loxP sites. Cre recombinase is used to excise the prion protein gene, which is then integrated into a new genomic location. The resulting mouse expresses the prion protein gene from a new promoter, leading to the production of PrP^C protein.

Figure 2 Mouse food allergy models. The mouse models are divided into two categories: (1) models for food allergy and (2) models for food allergy and food allergy. The models are based on the genetic background of the mouse, the type of allergen used, and the method of allergen administration.

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Specifické myší modely

The Journal of Neuroscience | 10.1523/JNEUROSCI.1111-19.2019

Review

Environmental Enrichment in Mouse Models and Its Translation to Human Factors Improving Conditions in Alzheimer Disease

M.E. Calabrese^{1,2}, L. Green^{1,2}, T.J. Rosene^{1,2}

Figure 1 In a mouse model of AD, mice exposed to standard housing show lower levels of dendritic arborization, paucity of dendritic spines and less robust synaptic transmission.

Table 1. Mouse wild type and transgenic models of AD used to test mouse and their human-toxicopathology.

| Strain | Pathology | Behavior | Interventions | Outcomes | Relevance |
|-----------|--|--|--|--|-----------|
| APP23 | Development of amyloid plaques and neurofibrillary tangles of A β and tau. Onset of disease and progression is dependent on age. | Deficits in learning and memory, reduced social interaction. | Enrichment of the environment, cognitive stimulation, physical exercise. | Enrichment of the environment, cognitive stimulation, physical exercise, and social interaction improve learning and memory, and reduce amyloid plaques and neurofibrillary tangles. | AD (95) |
| APP25 | APP of 25 kDa, onset of disease and progression is dependent on age. | Deficits in learning and memory, reduced social interaction. | Enrichment of the environment, cognitive stimulation, physical exercise. | Enrichment of the environment, cognitive stimulation, physical exercise, and social interaction improve learning and memory, and reduce amyloid plaques and neurofibrillary tangles. | AD (95) |
| APP25/PS1 | APP of 25 kDa and PS1, onset of disease and progression is dependent on age. | Deficits in learning and memory, reduced social interaction. | Enrichment of the environment, cognitive stimulation, physical exercise. | Enrichment of the environment, cognitive stimulation, physical exercise, and social interaction improve learning and memory, and reduce amyloid plaques and neurofibrillary tangles. | AD (95) |
| APP25/PS1 | APP of 25 kDa and PS1, onset of disease and progression is dependent on age. | Deficits in learning and memory, reduced social interaction. | Enrichment of the environment, cognitive stimulation, physical exercise. | Enrichment of the environment, cognitive stimulation, physical exercise, and social interaction improve learning and memory, and reduce amyloid plaques and neurofibrillary tangles. | AD (95) |
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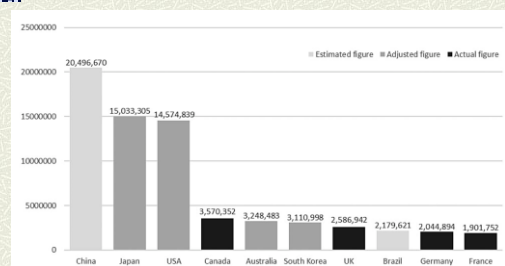
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Využití kmenů LZ

- ✓ Standardizace
- ✓ Biomodely
- ✓ Etika pokusů na zvířatech

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Využití LZ



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Etika pokusů na zvířatech



<https://www.ervivweb.cz/amp/thumbnails/712/f2/722419cca6ccefce6c47498ed8c1b298.jpg>