

THE 15th TRANSGENIC TECHNOLOGY MEETING (TT2019)



April 7-10, 2019

Kobe International Conference Center

(神戸国際会議場)

<http://www.tt2019.org/>

PROGRAM & ABSTRACTS

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The International Society for Transgenic Technologies (ISTT) seeks to foster communication and technology sharing, to enhance scientific research, to advance the field of animal transgenesis, particularly as it applies to useful experimental models in biology, medicine and biotechnology, and to represent the interests of an international body of professionals (scientists, technicians, and graduate students) working in the field of transgenic technologies.

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The benefits of membership include priority access/reduced registration fees to Transgenic Technology (TT) meetings, and free on-line access to *Transgenic Research*, as well as unique online content (pictures, videos, methods, talks, posters) through the members-only area of the ISTT website. Further information on membership can be found at:



www.transtechsociety.org

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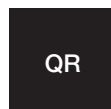
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Welcome Message

Welcome to Kobe, Japan!

It is a great honor and pleasure of the TT2019 Conference Committee and the ISTT to welcome you all to the 15th Transgenic Technology Meeting (TT2019). At the milestone of our 15th event to celebrate, the local staff of RIKEN Center for Biosystems Dynamics Research (BDR), the host institution of this Conference, also would like to offer the delegates from all over the world a warm welcome to Kobe, Japan.

The past four decades of animal transgenesis research have seen a number of technological breakthroughs, having paved the way to linking genes and their functions. We are now witnessing yet another wave of revolution in our field driven by genome editing technologies. This trend not only has tasked us to further enrich our technical expertise, but also has challenged us with many ethical questions. TT2019 starts off with the inaugural discussion session on ethics in the CRISPR era, followed by a number of innovative approaches using genome editing technologies in an expanding plethora of experimental model animal species, as well as those for translational and industrial applications. Yet it does, by no means, detract fundamentals from traditional approaches in animal genetic manipulations, underscored by many other excellent talks, including the ISTT Prize Lecture by Lluís Montoliu on transgenesis with large constructs, the Keynote Lecture by Hiroshi Hamada on elucidation of fundamental mechanisms underlying animal body plan by transgenic approaches and the Orbis Pictus Lecture by Gail Martin on historical perspectives of the embryonic stem cell technology. We will also hear from the recipients of Young Investigator Award and 3Rs Prize about innovative research accomplishments for which they were awarded.

Delegates will also feel the heat of new developments in our community in poster sessions where numerous emerging techniques and tools in transgenic technologies, including zygote electroporation, diverse animal models and bioresource development. The Post-TT2019 Hands-on Workshop will provide some of the delegates with a unique opportunity to exploit cutting-edge techniques in animal transgenesis.

We thank all the speakers and poster presenters in providing the seeds of exciting discussions. We are also deeply indebted to our sponsors and exhibitors from corporate and academic sectors for their generous support. Additionally, but not the least, the exhaustive efforts and support of the ISTT leadership, TT2019 Scientific Advisory and Organizing Committees and the local meeting staff warrant an enormous amount of gratitude for the realization of this Conference.

It is our great expectation that TT2019 will be a venue for the transgenic technology community to enjoy fruitful scientific exchange, to develop future collaborations and to explore possible future directions of our field!

Sincerely,



Yas Furuta
Chair of TT2019 Organizers



Wojtek Auerbach
ISTT President



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Yasuhide Furuta RIKEN Center for Biosystems Dynamics Research, Japan

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Scientific Program

I SUNDAY, APRIL 7, 2019

- 14:00 – 19:30 **REGISTRATION** *Main Hall Foyer, 1F ;
Poster Set-up International Conference Room, 3F*
- 17:00 – 17:10 **INTRODUCTION – TT2019 Committee** *Main Hall*
WELCOME ADDRESS
Eisuke Nishida Director, RIKEN Center for Biosystems Dynamics Research, Japan
- 17:10 – 19:30 **PRE-MEETING PANEL DISCUSSION:**
Ethical Aspects of Genome Editing *Main Hall*
Moderators: Ernst-Martin Füchtbauer & Jan Parker Thornburg
Mickey Gjerris University of Copenhagen, Denmark
“With great power comes great responsibility - Ethical aspects of gene-editing”
Ewa Maria Bartnik University of Warsaw, Poland
“The ethics of human genome editing”
Marion Abecassis INSERM Ethics Committee, France
“Genome Editing: A Global Legal Perspective”
- 19:30 – 21:00 **WELCOME RECEPTION; Poster Pre-View, Vendor Exhibits**
International Conference Room / Reception Hall / Lounge, 3F

I MONDAY, APRIL 8, 2019

8:30 – 8:35 **OPENING ADDRESS – Wojtek Auerbach (ISTT President) Main Hall**

8:35 – 9:25 **OPENING KEYNOTE LECTURE Main Hall**

Introduction: Hiroshi Kiyonari

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“How Left-Right Asymmetry is Established in the Mouse Embryo: Lessons we learned from Transgenic Mice”

Hiroshi Hamada RIKEN Center for Biosystems Dynamics Research, Japan

SESSION 1: Genome Editing – from Mechanisms to Applications Main Hall

Chair: Masato Ohtsuka

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9:25 – 9:50 Jonathan Wilde Massachusetts Institute of Technology, USA

“Efficient embryonic genome editing via RAD51-enhanced interhomolog repair”

9:50 – 10:05 Seiya Mizuno University of Tsukuba, Japan

“Modified-Cas9 improves genome editing efficiency in mouse zygote”

10:05 – 10:20 Gus McFarlane University of Edinburgh, Scotland, UK

“A CRISPR-Cas9 gene drive targeting female reproduction in mice”

10:20 – 10:40 **Coffee Break**

Main Hall Foyer, 1F; International Conference Room / Reception Hall / Lounge, 3F

SESSION 2: Transgenesis in Non-Mammalian Models Main Hall

Chair: Richard Behringer

10:40 – 11:05 Erin Alberstat Marine Biological Lab, Woods Hole, USA

“Unraveling crustacean body plan evolution through CRISPR/Cas9 genome editing”

11:05 – 11:30 Kiyoshi Naruse National Institute for Basic Biology, Japan

“Transgenic and genome editing technologies and medaka bioresource project”

11:30 – 11:55 Rachel Miller University of Texas, USA

“Tissue-Specific CRISPR knockouts in *Xenopus laevis*”

11:55 – 12:10 Anne Straume Institute of Marine Research, Norway

“Highly efficient CRISPR/Cas9 knock-in in Atlantic Salmon”

Scientific Program

12:10 – 12:35 **POSTER FLASH TALKS I : odd number posters** Main Hall

12:40- **Lunch Break**

12:40 – 13:25 **Luncheon Seminar** Main Hall

Presentation 1

Takayuki KOYANO Division of Molecular Genetics, Shigei Medical Research Institute, Japan
“GONAD -A simple method for genome-editing in Mice and Rats-”

Presentation 2

Masahito IKAWA Research Institute for Microbial Diseases, Osaka University, Japan
“CRISPR/Cas9 Mediated Genome Editing in Mice”

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13:25 – 14:40 **Poster Session I**
odd number posters; International Conference Room, 3F

12:40 – 14:40 **Vendor Exhibits** Reception Hall / Lounge, 3F

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SESSION 3: Panel Discussion – Running a Transgenic Core Main Hall

14:40 - 16:15 Moderator: Elizabeth Williams
Panelist: Martina Crispo Institut Pasteur de Montevideo, Uruguay
Judy Hallett Purdue University, USA
Yas Furuta RIKEN Center for Biosystems Dynamics Research, Japan
Geert Michel Charité Universitätsmedizin Berlin, Germany

16:15 – 16:40 **Coffee Break**
Main Hall Foyer, 1F; International Conference Room / Reception Hall / Lounge, 3F

SESSION 4: Development of Sex and Germ Line Main Hall

Chair: Aimee Stablewski & Toru Takeo

16:40 – 17:05 Tomoya Kitajima RIKEN Center for Biosystems Dynamics Research, Japan
“Causes of aneuploidy in eggs”

17:05 – 17:30 Jon Oatley Washington State University, USA
“Advancing reproductive capacity in livestock with gene editing”

17:30 – 17:55 Yumiko Saga National Institute of Genetics, Japan
“Germ cell-specific conditional knockout without making mouse lines”

17:55 – 18:10 Ella Thomson University of Queensland, Australia
"Understanding the structure/function relationship of the human sex determining gene, Sry, using a transgenic mouse model"

18:10 – 19:10 **ISTT PRIZE LECTURE (Main Hall)**

Introduction and Award Presentation: Wojtek Auerbach ISTT President

"Eventually, Size Did not Matter"

Lluís Montoliu National Center for Biotechnologies, Spain

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TUESDAY, APRIL 9, 2019

SESSION 5: Transgenic Technologies in the Rat Main Hall

Chair: Tomoji Mashimo & Lynn Doglio

8:30 – 8:55 Akiko Takizawa Medical College of Wisconsin, USA
"Establishment of a cryopreservation system for gene modified rat strains"

8:55 – 9:20 Ignacio Anegón Université de Nantes, France
"Generation of genome edited rats for the analysis of immune responses"

9:20 – 9:45 Hiromitsu Nakauchi Stanford University, USA
"Exploiting the organ niche for interspecies organogenesis"

9:45 – 10:00 Jeffrey Lee Regeneron Pharmaceuticals, Inc., USA
"Embryonic Stem Cell-Based Production of Large Targeted Genome Modifications in the Rat for the Purposes of Human Disease Modeling and Drug Discovery"

10:00 – 10:30 **Coffee Break**

Main Hall Foyer, 1F; International Conference Room / Reception Hall / Lounge, 3F

SESSION 6: Ethics and 3Rs

– Ethical Challenges in Genetically Engineered Animals Main Hall

Chair: Boris Jerchow & Branko Zevnik

10:30 – 10:50 Adrian Smith Norwegian Veterinary Institute, Norway
"PREPARE before you ARRIVE: how to plan animal experiments"

10:50 – 11:10 Anne Zintzsch Justus-Liebig-Universität Gießen, Germany
"Coping with the phenotype: Challenges of assessing severity"

11:10 – 11:30 Takashi Agui Hokkaido University, Japan
"Humane endpoints in generating and breeding genetically modified animals"

Scientific Program

11:30 – 11:40 Benjamin Davies Welcome Centre for Human Genetics, UK
"Replacement of surgical vasectomy by the use of wild-type sterile hybrids"

11:40 – 12:00 **3Rs Prize Presentation and Talk**

Sponsored by  JANVIER LABS
The 3Rs Research Institute
& Associated Services

Pawel Pelczar University of Basel, Switzerland
"Permanent companion females improve the welfare of vasectomized males without adversely affecting their plugging performance."

12:00 – 12:25 **POSTER FLASH TALKS II: even number posters**

12:30- **Lunch Break**

12:30 – 13:15 **Luncheon Seminar Main Hall**

Satoru Takahashi, Seiya Mizuno, Fumihiro Sugiyama
Department of Anatomy and Embryology, Faculty of Medicine, Laboratory Animal Resource Center (LARC),
Transborder Medical Research Center (TMRC), University of Tsukuba, Japan

Christopher Dowdy Charles River

"Generation services of genetically modified mice by using new modified Cas9 in
Laboratory Animal Resource Center, University of Tsukuba"

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of the way.™

13:15 – 14:30 **Poster Session II even number posters; International Conference Room, 3F**

12:30 – 14:30 **Vendor Exhibits Reception Hall / Lounge, 3F**

SESSION 7: New Developments in Transgenic Technologies Main Hall

Chair: Peter Hohenstein & Karen Brennan

14:30 – 14:45 Takaya Abe RIKEN Center for Biosystems Dynamics Research, Japan
"Fluorescent reporter mouse lines and image analyses reveal distinct region-specific
cell behaviors in the visceral endoderm"

14:45 – 15:00 Leslie Goodwin The Jackson Laboratory, USA
"Large-scale discovery of transgene insertion sites in mice"

15:00 – 15:15 Soren Warming Genentech, Inc., USA
"Targeted Locus Amplification (TLA) as a powerful tool for allele verification and
transgene integration site analysis"

- 15:15 – 15:30 Marie-Christine Birling CELPHEDIA, PHENOMIN, Institut Clinique de la Souris, France
“Generation of genomic structural variants and chromosomal manipulation by CRISPR/Cas9 genome editing in rodents”
- 15:30 – 15:45 Xiaofeng Liu Sun Yat-sen University, China
“Targeted editing of the ZBED6 binding site in intron 3 of IGF2 by base editor leads to enhanced muscle development in Liang Guang Small Spotted pigs”
- 15:45 – 16:00 Hiroshi Kiyonari RIKEN Center for Biosystems Dynamics Research, Japan
“Development of reproductive technology in Wistar rat strain”
- 16:00 – 16:30 **Coffee Break**
Main Hall Foyer, 1F; International Conference Room / Reception Hall / Lounge, 3F

SESSION 8: Transgenic Technologies in Large Animals/Livestock *Main Hall*

- Chair: Simon Lillico & Martina Crispo
- 16:30 – 16:55 Charlotte Brandt Sorensen Aarhus University, Denmark
“Genetically Modified Yucatan minipigs as models for hypercholesterolemia and atherosclerosis”
- 16:55 – 17:20 Adrian Mutto National University of General San Martin, Argentina
“Somatic Cell Nuclear Transfer and Its impact on equine industry”
- 17:20 – 17:45 Hideyuki Okano Keio University School of Medicine, Japan
“Disease modeling and brain mapping using genetically modified marmosets”
- 17:45 – 18:00 Hendrik Sake Friedrich-Loeffler-Institut, Germany
“Generation and characterization of SLA class I knockout pigs for xenotransplantation”
- 18:00 – 19:00 **ISTT GENERAL ASSEMBLY** *Main Hall*
- 19:30 – 21:30 **GALA DINNER** *Kobe Portopia Hotel*

Scientific Program

I WEDNESDAY, APRIL 10, 2019

SESSION 9: Genome Editing – Advanced Applications and Challenges (I) Main Hall

Chair: Benoît Kanzler & Cheryl Bock

- 8:40 – 8:55 Guochun Gong Regeneron Pharmaceuticals Inc., USA
“CRISPR expressing mice for tissue specific gene modulation”
- 8:55 – 9:10 Takuro Horii Gunma University, Japan
“Generation of epigenetic disease model mice by targeted demethylation of the epigenome”
- 9:10 – 9:25 Daniel Davis University of Missouri, USA
“Genome editing in an era of precision medicine: Multi-species approach to determine underlying mechanism of KCNT1-associated epilepsy”
- 9:25 – 9:40 Yang Zhou Chinese Academy of Sciences, China; Massachusetts Institute of Technology, USA
“Generation of a germline-transmissible macaque model of autism via CRISPR/Cas9”
- 9:40 – 10:05 Jaime Rivera University of Massachusetts Medical School, USA
“Dunkin' Transgenics: A Simple Method to Edit Mammalian Embryos”
- 10:05 – 10:35 **Coffee Break**
Main Hall Foyer, 1F; International Conference Room / Reception Hall / Lounge, 3F

SESSION 10: Genome Editing – Advanced Applications and Challenges (II)

Chair: Lluís Montoliu

- 10:35 – 10:50 Thomas Saunders University of Michigan, USA
“CRISPR/Cas9 Complement C3 Rat Knockout Model Implicates C3 in Neuropathy”
- 10:50 – 11:05 Lydia Teboul MRC Harwell Institute, UK
“Generation and validation of increasingly complex alleles introduced by genome editing”
- 11:05 – 11:30 Hui Yang Shanghai Institute for Biological Sciences, China
“Generation of genetically modified animals by CRISPR”
- 11:30 – 12:00 **YOUNG INVESTIGATOR AWARD**
Introduction and Award Presentation: Benoît Kanzler ISTT Vice President
“Light up the Embryos: Efficient Generation of Knock-in Reporter Mice by 2C-HR-CRISPR”
Bin Gu Hospital for Sick Children, Canada

ISTT Young Investigator Award is supported by an independent medical education grant from Regeneron Pharmaceuticals, Inc.

12:00- ***Lunch Break***

12:50 – 13:50 ***CLOSING KEYNOTE – Orbis Pictus Lecture***

Award Presentation: Czech Center for Phenogenomics

Introduction: Wojtek Auerbach (ISTT President)

Sponsored by  Czech Centre for Phenogenomics

“Twists and Turns on the Road to Gene Targeting”

Gail Martin University of California San Francisco, USA

13:50 – 14:15 ***CLOSING***

Poster Awards: ISTT Committee

Closing Remarks: Wojtek Auerbach (ISTT President) & TT2019 Committee

Presentation of TT2020: Rebecca Haffner-Krausz Weizmann Institute of Science, Israel

Abstracts

PRE-MEETING PANEL DISCUSSION

With great power comes great responsibility – Ethical aspects of gene-editing

Mickey Gjerris

University of Copenhagen, Denmark



The advent of novel technologies brings social challenges as they force us to revisit our values and examine if and how the new possibilities should be implemented in our lives. The emergence of gene-editing technologies such as CRISPR-Cas9 is the latest example of this. With the promise of revolutionizing plant and animal breeding and providing new tools to treat diseases and alleviate human suffering the benefits of gene-editing seems obvious, but they come with ethical concerns that needs to be addressed, if the implementation of the technology is to be socially robust.

With regard to plants and animals, discussions mainly revolve around risks to humans and the environment, but concepts such as naturalness, social justice and the integrity of plants and animals are just as important to discuss as the technology challenges the ethical values of many citizens. With regard to humans there has been widespread agreement in the international community that gene-technology should only be used on humans for therapeutic purposes to avoid that induced changes would be inheritable, but with the advent of gene-editing the discussion has resurfaced, not the least when a Chinese researcher claimed that he had already created "CRISPR-babies". The possibility of unintended side-effects cautions such a development, but there are other and broader ethical considerations to include in the discussion such as: How will it affect our understanding of human nature, if we begin editing our genome? The talk will give an overview of the ethical issues related to gene-editing and present ethical arguments for a cautionary approach to the new possibilities.

Invited and Short Talks

PRE-MEETING PANEL DISCUSSION

The ethics of human genome editing

Ewa Maria Bartnik

Institute of Genetics and Biotechnology
Faculty of Biology, University of Warsaw, Poland



Since the letter by Baltimore et al. “Biotechnology. A prudent path forward for genomic engineering and germline gene modification” appeared in Science almost exactly four years ago a lot of discussions have been taking place which mainly focus on the fundamental statement made in that publication, “Strongly discourage...any attempts at germline modification for clinical application in humans while societal, environmental, and ethical implications of such activity are discussed among scientific and governmental organizations....This will enable pathways to responsible uses of this technology, if any, to be identified”. The “if any” is fundamental, and especially so as genome editing had already been used on human embryos. The ethics issues as well as fundamental position statements made by leading institutions will be presented.

Abstracts

PRE-MEETING PANEL DISCUSSION

Genome editing: A Global Legal Perspective

Marion Abecassis

Attorney in Life Sciences –Member of the Board of the Association for Responsible Research and Innovation in Genome Editing (ARRIGE), France



The rapidity of scientific advances challenges experts' predictions... And the capacity of the law to adapt to the advent of new technology. "Be careful what you wish for" – What should we wish (and work) for with regards to genome-editing technology, from a global legal perspective?

The International Bioethics Committee of UNESCO raised, almost four years ago, that, "*it is important for States and governments to accept the principle of a shared global responsibility when the engineering of the human genome is involved.*" This takes on renewed importance now that gene-edited babies are born in China. Genome-editing technology can no longer be considered solely as a lab technology or a revolution of plants and animals transgenics, and its potential use on humans can no longer be thought as only occurring on the occasion of rare and highly controlled clinical trials.

This presentation will focus on existing international legal tools applicable to the use of genome-editing technology on living organisms and on-going initiatives advocating for a global framework for the use of genome editing.

Invited and Short Talks

OPENING KEYNOTE LECTURE

How left-right asymmetry is established in the mouse embryo: lessons we learned from transgenic mice

Hiroshi Hamada, Yayoi Ikawa, Eriko Kajikawa,
Katsura Minegishi, Katsutoshi Mizuno, Hiromi Nishimura
Riken Center for Biosystems Dynamics Research, Japan



In the past >20 years, my lab has studied how left-right (L-R) asymmetry is established in the mouse embryo. We now understand, in principle, how L-R symmetry is broken by rotating cilia and unidirectional fluid flow (how L-R asymmetry is originated), how this symmetry breaking leads to asymmetric gene expression in the lateral plate, and how asymmetric gene expression results in situs-specific organogenesis. However, there still remain several important questions unanswered. For example, how the fluid flow is sensed by embryo, and how L-R symmetry is broken in the absence of motile cilia in some vertebrates. In our past and ongoing work, we always addressed questions by taking advantage of transgenic mice. Thus, generating various transgenic and mutant mice has been the basic strategy of our work. Here we will introduce our recent work that would address some of the unanswered questions.

Supported by RIKEN Symposium Series



Abstracts

Plenary Session 3

Cutting-edge technologies in South America



Martina Crispo¹, G. Schlapp², M.N. Meikle², A.P. Mulet²

¹Institut Pasteur de Montevideo, Uruguay,

²Transgenic and Experimental Animal Unit, IPMon, Uruguay

Since 2006 the Transgenic and Experimental Animal Unit of Institut Pasteur de Montevideo has provided genetically modified (GM) tailor-made animal models for researchers of the region and the world, through standard techniques such as DNA microinjection, homologous recombination in ES cells, lentivirus and transposons. In 2014 the incorporation of CRISPR/Cas system into our portfolio allowed us to speed up the production of GM experimental animals and livestock, improving the efficiency of the techniques and reducing the costs. Since then, we have produced KO, KI and cKI animal models with applications in the fields of biomedicine and agriculture. In addition, our Unit provides mice and rats for different research groups working in cancer, neurodegenerative and metabolic diseases among others. Furthermore, we introduced IVF of ultrasuperovulated mouse oocytes using anti-inhibin serum and CARD method, which allowed us to obtain hundreds of zygotes that can be either vitrified or microinjected and transferred to foster females, reducing the number of animals used. We incorporated a molecular biology Lab that specializes in molecular design and genotyping analysis, offering a complete GM production service, from model design to detection of founders. All these cutting-edge technologies had positioned us as a reference core facility in South America, always keeping us at the forefront of GM animal generation techniques.

Abstracts

ISTT Prize Lecture

Eventually, size did not matter

Lluís Montoliu

CNB-CSIC and CIBERER-ISCIII, Madrid, Spain
National Center for Biotechnologies, Spain



During the first decade of animal transgenesis, researchers had to deal with the so-called chromosomal position effects, associated with altered expression of the injected transgenes upon integrating randomly in the host genome. This undesired variability was usually compensated by increasing the number of transgenic mouse lines generated. Therefore, only those phenotypes robustly found in several independent mouse lines were believed to be associated with the intended genetic modification.

In the 90's, an alternative approach was devised, to overcome those annoying position effects. The use of large genomic pieces of DNA, hundreds of kilobases in length, as transgenes was established. At first, yeast artificial chromosomes (YACs) were used for the production of transgenic mice. Subsequently, bacterial artificial chromosomes (BACs) and P1-derived artificial chromosomes (PACs) were tested similarly *in vivo*. These transgenes usually recapitulated the expression pattern of the endogenous genes they were carrying, thereby apparently eliminating the position effects. The size of these transgenes ensured the presence of relevant regulatory regions within the genomic region. Thus, the notion that “size matters” was adopted for artificial chromosome-type transgenic animals.

Twenty years later, CRISPR/Cas9-derived genome edited mice have been generated and used to functionally assess the role of relevant regulatory regions directly at the endogenous locus. These new experiments have revealed unexpected differences with analogous results generated through YAC transgenesis, suggesting that position effects had not been entirely eliminated. Therefore, 25 years later after establishing that sizes matter we probably have to conclude that, after all, size does not matter in animal transgenesis.

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Invited and Short Talks

Plenary Session 6

3Rs Prize Presentation and Talk

OP-7 Permanent companion females improve the welfare of vasectomized males without adversely affecting their plugging performance.

Pawel Pelczar, Heide Oller, Michael Bernstein

Center for Transgenic Models, University of Basel, Switzerland

Transgenic facilities require large numbers of vasectomized stud males to provide surrogate females necessary for their day to day operation. These males are mated as needed, but otherwise remain single-housed. The practice of single housing social animals such as laboratory mice is known to be stressful, however due to aggression it is frequently accepted when housing idle breeding or vasectomized males. Our lab has generated and distributed genetically sterile males that can serve as vasectomy replacements. We have now investigated whether providing a permanent companion female as a stress to sterile males will affect their ability to generate pseudopregnant females needed for embryo transfer procedures. Over the course of eight months, half of our colony of sterile males received a companion female, while the other half remained in single housing. We did not see a significant difference in plug rate between the two groups and therefore postulate that providing a sterile stud male with an additional permanent female companion improves animal welfare without reducing the plugging efficiency.

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Invited and Short Talks

Young Investigator Award

Light up the embryos: Efficient generation of knock-in reporter mice by 2C-HR-CRISPR

Bin Gu¹, Eszter Posfai², Janet Rossant²

¹Hospital for Sick Children, Canada,

²Program in Developmental and Stem Cell Biology, Hospital for Sick Children, Toronto, Canada



Large fragment knock-in, especially fluorescent reporter mouse models are important tools that could strongly promote the precise spatial-temporal analysis of developmental processes. Rapid and efficient generation of such models is still a major hurdle in mouse genetics. We developed 2C-HR-CRISPR, a highly efficient gene editing method based on introducing CRISPR reagents into mouse embryos at the 2-cell stage, taking advantage of the likely increase in HR efficiency during the long G2 phase and open chromatin structure of the 2-cell embryo. With 2C-HR-CRISPR and a modified biotin-streptavidin approach to localize repair templates to target sites, we rapidly targeted 20 endogenous genes that are expressed in mouse blastocysts with fluorescent reporters and generated reporter mouse lines with gene dependent efficiencies up to 95%. We showcase the first live triple-color blastocyst with all three lineages differentially reported. Additionally, we demonstrated efficient double targeting, enabling rapid assessment of the auxin-inducible degradation system for probing protein function in mouse embryos. These methods open up exciting avenues for exploring cell fate decisions in the blastocyst and later stages of development. We also suggest that 2C-HR-CRISPR can be a better alternative to random transgenesis by ensuring transgene insertions at defined 'safe harbor sites.

ISTT Young Investigator Award is supported by an independent medical education grant from Regeneron Pharmaceuticals, Inc.

CLOSING KEYNOTE: *Orbis Pictus* Lecture

Twists and turns on the road to gene targeting

Gail R. Martin

Department of Anatomy, University of California San Francisco, USA



The process of making specific changes in the genome of an embryo and creating animals that carry and can transmit those changes to their offspring has become commonplace. But how did we come to know that such a remarkable feat is possible? This talk will describe the milestones on the road that led to that knowledge, the first of which was the observation that certain male mice spontaneously developed a special type of testicular tumor. In addition, a major offshoot of the road to gene targeting – the development of in vitro model systems for studying various aspects of embryogenesis – will be discussed along with some of the lessons learned from experiments aimed at producing mice carrying specific mutations.

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Posters

Posters will be mounted in International Conference Room, Kobe International Conference Center 3F and will be available for viewing from 8:30 AM, Monday, April 8, 2019. The posters will remain on display for the duration of the meeting and can be viewed until the specified times:

Monday, April 8, 2019	5:00 PM
Tuesday, April 9, 2019	5:00 PM
Wednesday, April 10, 2019	1:00 PM

During the poster sessions, Tuesday, April 9, 13:15 – 14:30 (even numbers) & Monday, April 8, 13:25 – 14:40 (odd numbers), the authors of the posters will be present and available for more in-depth discussion.

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P-01 Detection of Mouse Pregnancy by 7.5 Days Post Coitum Using a Refined Weight Gain Method

Juan M Reyes, Xin Y Rairdan, Mariela D Pantle, Ben Grellman, Kristin D Evans
Genentech, Inc., USA

P-02 Re-Evaluating One-step Generation of Mice Carrying Conditional Alleles by CRISPR-Cas9-Mediated Genome Editing Technology

Channabasavaiah Gurumurthy¹ and 110 co-authors²

¹Mouse Genome Engineering Core Facility, Vice Chancellor for Research Office, University of Nebraska Medical Center, Omaha, NE, USA; ²42 other affiliations

P-03 Management of mouse sperm freezing and quality control

Nicole Kuepper, Julia Mock, Sonja Kropp, Klaus Pfeffer

Institute of Medical Microbiology and Hospital Hygiene, Heinrich-Heine-University Duesseldorf, Duesseldorf, Germany

P-04 Noninvasive multi-modality imaging in genetically engineered mouse (GEM) model of pancreatic cancer: 3R principle in cancer research

Ying Zhao^{1,2}, Rainer Heuchel³, Sandra Oerther⁴, Kristian Konigsson⁴, Moustapha Hassan^{1,2}

¹Preclinical Imaging Facility, Preclinical Laboratory; Clinical Research Center (KFC), Karolinska University Hospital; ²Experimental Cancer Medicine (ECM), Clinical Research Center (KFC), Department of Laboratory Medicine, Karolinska Institutet; ³Pancreatic cancer research laboratory, Department of Clinical Science, Intervention and Technology (CLINTEC), Karolinska Institutet; ⁴Preclinical Laboratory, Karolinska University Hospital, Sweden

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P-05 Development of the Cryosword for Vitrification of Mouse Embryos and Sperm

Barbara J Stone¹, K John McLaughlin²

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P-06 Zero Angle Microinjection (ZAM): A More Efficient Method of Mouse Zygote Pronuclear Injection

Xiaojun Xing, Timothy Nottoli, Suxia Bai

Yale Genome Editing Center, Department of Comparative Medicine, Yale School of Medicine, Yale University, USA

P-07 Motorization and automation of micromanipulation for genetic modification

Tomoo Eto¹, Hiroki Ueda², Ryoji Ito¹, Riichi Takahashi¹, Nobuaki Tanaka²

¹Central Institute for Experimental Animals; ²NSK Ltd., Japan

P-08 Modification strategies of recombinant BAC vectors for Sleeping Beauty mediated transgenesis

Mate Z.¹, Kovacs K.¹, Hevesi Z.², Harkany T.^{2,3}, Szabo G.¹, Erdelyi F.¹

¹Institute of Experimental Medicine, Hungarian Academy of Sciences, Hungary; ²Department of Molecular Neurosciences, Center for Brain Research, Medical University of Vienna, Austria; ³Department of Neuroscience, Karolinska Institutet, Sweden

P-09 qPCR as a versatile and sensitive tool for monitoring transgene stability and genetic quality control

DON P LIU

Charles River Laboratories, USA

P-10 Test for imprinted DNA methylation activity of the human H19-ICR in YAC transgenic mice by adopting a transgene co-placement strategy

Katsuhiko Hirakawa¹, Hitomi Matsuzaki², Keiji Tanimoto²

¹Graduate School of Life and Environmental Sciences, University of Tsukuba; ²Faculty of Life and Environmental Sciences, Life Science Center for Survival Dynamics (TARA), University of Tsukuba, Japan

P-11 Site-directed transgenic mice production using TARGATT™ knock-in technology in TMDU

Yoshikazu Hirate¹, Hitomi Takahashi¹, Yoshiaki Ito¹, Jinling Li², Chiho Ohtsuka¹, Hiroshi Asahara¹, Ruby Yanru Chen-Tsai², Masami Kanai-Azuma¹

¹Tokyo Medical and Dental University, Japan; ²Applied StemCell, Inc., USA

P-12 Manipulating the rodent genome: a comparison of traditional targeting vectors versus Cas9-mediated techniques

Susannah Diane Brydges, Jose Rojas, Jean Siao, Jeremy Rabinowitz, Yajun Tang, Qing Fang, Charleen Hunt, Guochun Gong, Sean Trzaska, Marine Prissette, Brian Zambrowicz
Regeneron Pharmaceuticals, Inc, USA

P-13 In vivo generation of the vascular-hematopoietic systems and cardiomyocytes from pluripotent stem cells

Giulia Coppiello¹, Marta Moya-Jodar¹, Gloria Abizanda¹, Javier Linares¹, Adrian Ruiz-Villalba¹, Ana Perez-Ruiz¹, Xabier Agirre¹, Juan Roberto Rodriguez-Madoz¹, Francisco Alberto Garcia-Vazquez², Xonia Carvajal-Vergara¹, Felipe Prosper^{1,3}, Xabier L. Aranguren¹
¹Laboratory of Regenerative Medicine, Foundation for Applied Medical Research, 31008 Pamplona, Spain; ²Group of Veterinary Physiology Teaching Innovation, Department of Physiology, Faculty of Veterinary Science, University of Murcia, Murcia, Spain; ³Department of Hematology, Clinica Universidad de Navarra, University of Navarra, Avenida Pio XII 36, E-31008 Pamplona, Spain

P-14 High-throughput Genome Editing in the Endogenous β -catenin Locus

Anagha Krishna¹, Martijn Kelder², Andrew Wood², Jeffrey Schoenebeck¹, Peter Hohenstein^{1,3}, Derya D Ozdemir¹
¹Roslin Institute, University of Edinburgh, UK, EH25 9RG; ²IGMM, University of Edinburgh, UK, EH4 2XU; ³Leiden University Medical Centre, Netherlands

P-15 Automation Applied for screening of Embryonic Stem Cells

Merone Roose Girma, Roger Caothien, Charles Yu, Lucinda Tam, Anna Pham, Soren Warming
Genentech, Inc., USA

P-16 Simple generation of transgenic mice by in vivo gene editing in preimplantation mouse embryos using rAAV vectors

Yeonsoo Yoon¹, Dan Wang², Phillip W.L. Tai², Alireza Edraki³, Raed Ibraheim³, Joy Riley¹, Judith Gallant¹, Erik J Sontheimer³, Guangping Gao², Jaime A Rivera-Perez¹
¹Department of Pediatrics, Division of Genes and Development, University of Massachusetts Medical School; ²Department of Microbiology and Physiological Systems and Horae Gene Therapy Center, University of Massachusetts Medical School; ³RNA Therapeutics Institute, University of Massachusetts Medical School, USA

P-17 Comparison of Efficiency of CRISPR targeting: PNI vs ZEN vs GONAD

Susan Jenifer Tamowski, Kyle O'Connor, He Lan, Wenhua Li, Nicholas Black
University of Utah, USA

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P-18 One step to achieve non-mosaic gene edited porcine embryo by CRISPR/Cas9

Peiqing Cong, Junjiu Huang, Xiaohu Su, Wei Chen
Sun Yat-sen University, P. R. China

P-19 Efficient 2C-HR-CRISPR Gene Editing Using Nuclear Injection of IVF-Generated 2-cell Embryos

Kevin A Kelley¹, Kasper Bonderup¹, Philippe Soriano²

¹Mouse Genetics and Gene Targeting CoRE, Icahn School of Medicine at Mount Sinai, New York, NY 10029; ²Department of Cell, Developmental and Regenerative Biology, Icahn School of Medicine at Mount Sinai, New York, NY, USA

P-20 Generation of Large Knock-In alleles with Double Stranded Plasmid templates using the 2C-HR CRISPR method

Mitra Cowan^{1,2,3}, Jade Desjardins^{1,2,3}, Janice Penney^{1,2,3}, Erzsebet Nagy-Kovacs^{1,2,3}, Nobuko Honma-Yamanaka^{1,2,3}, Yojiro Yamanaka^{1,2,3}

¹McGill Integrated Core for Animal Modeling, McGill University; ²Research Institute McGill University Health Centre; ³Department of Human Genetics, Goodman Cancer Research Centre, Canada

P-21 HDAC inhibitor increase gene targeting efficiency on the bovine β -casein locus in MAC-T cells using TALEN system

Man-Jong Kang¹, Da Som Park¹, Se Eun Kim¹, Deog-Bon Koo²

¹Department of Animal Science, Chonnam National University, Republic of Korea; ²Department of Biotechnology, Daegu University, Republic of Korea

P-22 Efficient floxing via embryo electroporation of two RNPs & two single stranded oligo donors

Xiaoxia Cui¹, Monica Sentamanat¹, Mike White², Colin Florian¹

¹Genome Engineering & iPSC Center, Department of Genetics, Washington University in St. Louis, St. Louis, MO, USA; ²Microinjection Core, Department of Immunology & Pathology, Washington University in St. Louis, St. Louis, MO, USA

P-23 Using embryo electroporation with long DNA templates and ribonucleoprotein complexes for advanced genome editing in mouse

Marie TEIXEIRA¹, Severine CHAUTARD², Emilie MICHEL², Benedicte F PY², Antoine MARCAIS², Farida HENRY¹, Denise AUBERT^{1,3}, Suzy MARKOSSIAN^{1,3}

¹SFR BioSciences, Plateau de Biologie Experimentale de la Souris (AniRA-PBES), Ecole Normale Supérieure de Lyon, Université Lyon1, CNRS UMS3444, INSERM US8, France; ²CIRI, INSERM U1111, Université Claude Bernard Lyon 1, CNRS UMR 5308, Ecole Normale Supérieure de Lyon, Université Lyon1, France; ³Institut de Genomique Fonctionnelle de Lyon, Ecole Normale Supérieure de Lyon, Université, Lyon, France

P-24 CRISPR/Cas9 murine zygote electroporation: combining efficiency improvement with animal reduction

Nicole Elenter, Ana Paula Mulet, Geraldine Schlapp, Maria Noel Meikle, Martina Crispo
Transgenic and Experimental Animal Unit. Institut Pasteur de Montevideo. Uruguay

P-25 Electroporation as an efficient mean of delivering genome editing reagent into mouse zygotes for CRISPR-mediated mouse genome editing

Pinghu Liu, Hideko Takahashi, Jingqi Lei, Carl Haugen, Yan Li, Lijin Dong
Genetic Engineering Core, National Eye Institute, NIH, USA

P-26 CRISPR/Cas9 electroporation can replace microinjection for in vivo genome editing in mice

Alberto Cebrian-Serrano¹, Florian Giesert², Wolfgang Wurst²
¹Institute of Diabetes and Obesity, Helmholtz Diabetes Center, Helmholtz Zentrum Munchen; ²Institute of Genetic and Development, Helmholtz Zentrum Munchen, Germany

P-27 Simple electroporation for efficient CRISPR/Cas9 genome editing in murine zygotes

Simon E. Troder¹, Lena K. Ebert², Linus Butt², Sonja Assenmacher¹, Bernhard Schermer², Branko Zevnik¹
¹Cologne Excellence Cluster for Cellular Stress Responses in Aging-Associated Diseases (CECAD), University of Cologne, Cologne, Germany; ²Department II of Internal Medicine and Center for Molecular Medicine Cologne (CMMC), University of Cologne, Cologne, Germany

P-28 Highly efficient gene targeting and homologous recombination using Cpf1 electroporation

Lisa Jane Garrett, Jun Cheng, Gene Elliott, Dawn Watkins-Chow
National Institutes of Health, USA

P-29 CRISPR genome editing based on electroporation of rat and mouse zygotes

Petr Kasparek, Irena Jenickova, Jana Kopkanova, Radislav Sedlacek
Institute of Molecular Genetics, ASCR, Prague, Czech Republic

P-30 Optimization of i-GONAD electroporation conditions for genome editing in the C57BL/6 strain

Masato Ohtsuka^{1,2}, Ayaka Nakamura³, Hiromi Miura^{1,2}, Channabasavaiah B Gurumurthy⁴, Masahiro Sato⁵
¹School of Medicine, Tokai University, Kanagawa, Japan; ²Center for Matrix Biology and Medicine, Graduate School of Medicine, Tokai University, Japan; ³Support Center for Medical Research and Education, Tokai University, Japan; ⁴Mouse Genome Engineering Core Facility, University of Nebraska Medical Center, USA; ⁵Frontier Science Research Center, Kagoshima University, Japan

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P-31 Creating knockdown mouse models using CRISPR-Cas system

Hiromi Miura¹, Masahiro Sato², Channabasavaiah B Gurumurthy³, Masato Ohtsuka¹

¹Tokai University School of Medicine; ²Kagoshima University, Japan; ³University of Nebraska Medical Center, USA

P-32 Intentional somatic mosaicism induced by 2-cell stage injection can circumvent embryonic/perinatal lethality for CRISPR mediated genome modification in mice

Mitra Cowan^{1,2,3}, Jade Desjardins^{1,2,3}, Janice Penney^{1,2,3}, Erzsebet Nagy-Kovacs^{1,2,3}, Nobuko Honma-Yamanaka^{1,2,3}, Yojiro Yamanaka^{1,2,3}

¹McGill Integrated Core for Animal Modeling, McGill University; ²Research Institute McGill University Hospital Centre; ³Department of Human Genetics, Goodman Cancer Research Centre, Canada

P-33 Analysis of individual indels in KO and KIN mice after CRISPR/Cas9 gene editing

Lev Fedorov, Yingming Wang

Oregon Health & Science University, USA

P-34 Genome-wide Profiling of Adenine Base Editor Specificity by EndoV-seq

Puping Liang, Xiaowei Xie, Shengyao Zhi, Hongwei Sun, Xiya Zhang, Junjiu Huang, Zhou Songyang

The First Affiliated Hospital, Sun Yat-sen University; MOE Key Laboratory of Gene Function and Regulation and Guangzhou Key Laboratory of Healthy Aging Research, SYSU-BCM Joint Research Center, School of Life Sciences, Sun Yat-sen University, Guangzhou, P. R. China

P-35 The application of CRISPR/Cas9 technology in generating complex mouse models

Andrew Kueh

Walter and Eliza Hall Institute, Australia

P-36 Triple-target CRISPR enabled almost perfect whole-body bi-allelic knockouts at first generation

Kenta Sumiyama¹, Genshiro Sunagawa¹, Maki Tadenuma Ukai¹, Perrin Dimitri^{1,2}, Hiroki Ueda¹

¹RIKEN Center for Biosystems Dynamics Research, Osaka, Japan; ²Queensland University of Technology, Brisbane, Australia

P-37 Application of CRISPR cas9 based genome editing system in the spontaneous mutant mouse STR/Ort

Tadashi Okubo¹, Kentaro Uchida², Masashi Takaso²

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P-38 Novel artificial intron technologies to generate conditional knock-out mice using CRISPR/Cas9

Jan Parker-Thornburg^{1,2}, Amber Thomas-Gordon^{1,2}

¹MD Anderson Cancer Center; ²UT Health Graduate School of Biomedical Sciences, USA

P-39 A novel strategy for efficient gene knock-in in rodents

Kazuto Yoshimi^{1,2}, Yoshiki Miyasaka², Yoshihiro Uno², Yuko Kotani², Kosuke Hattori², Yuko Yamauchi², Arisa Tanigawa², Tomoji Mashimo^{1,2}

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P-40 Splicing donor site sgRNAs enhance CRISPR/Cas9-mediated knock-out efficiency

Manuel Sanchez-Martin^{1,2,3}, Ignacio Garcia-Tunon^{1,4}, Elena Vuelta-Ramos², Sandra Perez-Ramos², Maria Herrero², Raquel Saldana⁵, Fermin Sanchez-Guijo^{1,6}, Lucia Mendez², Veronica Alonso-Perez^{1,4,6}, Jesus Maria Hernandez-Rivas^{1,4,6}

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P-41 SED [Single strand Ended Double strand] DNA donors as an inexpensive, easy to make, and efficiently inserted, long CRISPR donor

Elena McBeath¹, Yuka Fujii², Leonardo Cruz¹, Jun-ichi Abe¹, Marie-Claude Hofmann¹, Keigi Fujiwara¹

¹University of Texas, MD Anderson Cancer Center; ²Houston Methodist Research Institute, USA

P-42 Optimization of in vitro manipulation techniques to improve rat genome editing efficiency

Hongsheng Men¹, Daniel J Davis², Barbara J Stone³, Elizabeth C Bryda^{1,2}

¹Rat Resource and Research Center, University of Missouri, Columbia, MO 65201, USA; ²Animal Modeling Core, University of Missouri, Columbia, MO 65201, USA; ³ParaTechs Corporation, Lexington, KY 40505, USA

P-43 Comparison of CRISPR methods and reagents for the generation of a KI mice

Anna Pujol¹, Sandra Turon¹, Anna Arbos¹, Miquel Garcia², Fatima Bosch²

¹Transgenic Animal Unit (UAT), CBATEG, Universitat Autònoma de Barcelona, Bellaterra, Spain; ²CBATEG, Universitat Autònoma de Barcelona, Belaterra, Spain

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P-44 Harnessing the CRISPR/Cas9 System in Mouse Genome Engineering @ LARGE, RIKEN BDR

Takaya Abe¹, Kenichi Inoue¹, Hiroshi Kiyonari¹, Yas Furuta^{1,2}

¹LARGE, RIKEN Center for Biosystems Dynamics Research, Japan; ²MGCF, Sloan Kettering Institute, Memorial Sloan Kettering Cancer Center, USA

P-45 Improving a pipeline for the generation of genetically altered mice by genome editing

Adam Caulder, Gemma Codner, Alasdair Allan, Elker Malzer, Skevoulla Christou-Smith, Matthew Mackenzie, Jorik Loeffler, Francesca Pike, Dan Archer, Susan Varley, Marie Hutchison, Janet Kenyon, Michelle Stewart, Sara Wells, Lydia Teboul

The Mary Lyon Centre, MRC Harwell Institute, Harwell Campus, Didcot, Oxon, OX11 0RD, UK

P-46 Improving Efficiency of CRISPR/Cas9-mediated Genome Editing in Mice

Lin Wu, Ying Chen, Zhenjuan Wang, Laurie R. Chen, Sarah J. Johnson

Genome Modification Facility, Harvard University, USA

P-47 Genome editing activity at CNRS, Toulouse, France: Efficiency for generating KO, KI and large deletions, a mixed picture.

Pascalie Mercier¹, Christophe Audouard², Jacques Auriol², Severine Ethuin², Laurence Lepourry², Alice Davy²

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P-48 Optimizing efficiencies of large knockin integration by CRISPR/Cas in mouse zygotes

Sagrario Ortega¹, Jaime Munoz¹, Pierfrancesco Vargiu¹, Estefania Ayala¹, Martina Reiss², Harald Kranz²

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P-49 The efficiency of generating large DNA knock-in mice by CRISPR/Cas9 : a report from a single core facility

I-Shing Yu^{1,2}, Chia-Lu Hung¹, Hong-Yu Jhou¹, Shih-Fan Wu¹, Cheng-Ju Wang¹, Kuei-Liang Chen¹, Wei-Nien Chen¹, Yung-Shun Wang¹, Po-Yuen Wu¹, Feng-Lin Pu¹, Huang-Yi Ling¹, Chien-Hui Wu¹, Shu-Wha Lin^{1,2,3}

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P-50 Analyses of on-target mutations in 632 founder mice generated by using CRISPR/Cas9 to target 17 genomic loci

HY Shin^{1,2}, C Wang¹, HK Lee^{1,3}, KH Yoo^{1,4}, X Zeng¹, T Kuhns¹, CM Yang¹, T Mohr¹, Y Du⁵, C Liu⁵, L Hennighausen¹

¹Laboratory of Genetics and Physiology, National Institute of Diabetes and Digestive and Kidney Diseases, US National Institutes of Health, Bethesda, Maryland 20892, USA; ²Department of Biomedical Science and Engineering, Konkuk University, Seoul 05029, Republic of Korea; ³Department of Cell and Developmental Biology & Dental Research Institute, Seoul National University, Republic of Korea; ⁴Department of Life Systems, Sookmyung Women's University, Seoul 140-742, Republic of Korea; ⁵Transgenic Core, National Heart, Lung, and Blood Institute, US National Institutes of Health, Bethesda, Maryland 20892, USA

P-51 Optimizing Precise Genome Modification of the Rat Embryo

Mayumi Isaka, Daniel Kennedy, Michael Brown, Anthony Gagliardi, Katharina Lutter, Jingjing Wang, Roxanne Ally, Kevin Mitchell, Brian Morse, Jeffrey Lee, Joseph Hickey, Eric Chiao, Wojtek Auerbach, Brian Zambrowicz

Regeneron Pharmaceuticals, Inc., USA

P-52 CRISPR editing for mouse models: new developments and unexpected findings

Paul Quinton Thomas^{1,2,3}, Melissa Ashleigh White^{1,2,3}, Sandra Gaye Piltz^{1,2,3}

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P-53 Gene Editing Rat Resource Center (GERRC): Rat models for heart, lung, blood, and sleep disorder studies

Jason Klotz^{1,2}, Aron Geurts^{1,2}, Melinda Dwinell^{1,2}

¹Medical College of Wisconsin, Genomic Sciences and Precision Medicine Center; ²Medical College of Wisconsin, Physiology, USA

P-54 Transgenic rat mediated by sleeping beauty transposon and development of embryo cryopreservation system

Yun-kyong Jin¹, Hee-kyoung Kim¹, Kyeong-Min Kim¹, Muhammad Ailla Joan¹, Sung-hyu Koo^{1,2}, Goo Jang^{1,2}

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P-55 rGONAD: A new simple method for genome-editing in rats

Takayuki Koyano¹, Masumi Numba¹, Tomoe Kobayashi¹, Masato Ohtsuka², Makoto Matsuyama¹

¹Division of Molecular Genetics, Shigei Medical Research Institute, Okayama, Japan; ²Department of Molecular Life Science, Division of Basic Medical Science and Molecular Medicine, School of Medicine, Tokai University, Isehara, Kanagawa, Japan

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P-56 Sexing in pigs by using Gene Editing

Stefanie Kurtz¹, Antje Frenzel¹, Stoyan Petkov², Andrea Lucas-Hahn¹, Petra Hassel¹, Roswitha Becker¹, Heineke Eylers¹, Heiner Niemann³, Bjorn Petersen¹

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P-57 Expression of human IL2 recombinant protein in milk and germ-line transmission from transposon mediated transgenic dairy cattle

Goo Jang¹, Song-Jeon Lee², Soo-Young Yum¹, Kyeong-Min Kim¹, Shin-Ki Park³, Ji-Young Kim⁴, Seong-Hun Bae², Hyeong-Jong Kim², Hee-Kyeong Kim¹, Yun-Kyeong Jin¹, Seong-Jin Kim³, Kyung-Woon Kim⁴, Goo Jang^{1,5}

¹College of Veterinary Medicine, Seoul National University; ²Seoul Milk Coop; ³Theragen Etx; ⁴National Institute of Animal Science; ⁵LARTBIO Inc., Republic of Korea

P-58 Generation of a polled phenotype in Holstein-Friesians and Brown Swiss using DNA-nucleases

Felix Schuster¹, Bjorn Petersen¹, Jens Boch², Heiner Niemann³

¹Institute of Farm Animal Genetics, Friedrich-Loeffler-Institute; ²Institute of Plant Genetics, Leibniz University Hannover; ³Hannover Medical School, Germany

P-59 Assessment of PDX-1-deficient pigs generated using the CRISPR/Cas9 system introduced into porcine zygotes via electroporation

Fuminori Tanihara, Maki Hirata, Nhien Thi Nguyen, Quynh Anh Le, Takayuki Hirano, Takeshige Otoi
Faculty of Bioscience and Bioindustry, Tokushima University, Japan

P-60 Efficiency of gene editing by electroporation of Cas9 protein (GEEP) to generate GGTA1-modified pigs

Nhien Thi Nguyen, Fuminori Tanihara, Maki Hirata, Takayuki Hirano, Quynh Anh Le, Takeshige Otoi
Faculty of Bioscience and Bioindustry, Tokushima University, Japan

P-61 Effect of Cas9 protein levels on genomic mutations using the gene editing by electroporation of Cas9 protein (GEEP) system in putative zygotes

Quynh Anh Le, Maki Hirata, Fuminori Tanihara, Nhien Thi Nguyen, Takayuki Hirano, Takeshige Otoi
Faculty of Bioscience and Bioindustry, Tokushima University, Japan

P-62 Effects of CRISPR/Cas9-mediated gene targeting of porcine endogenous retrovirus on the developmental competence of porcine embryos

Maki Hirata, Fuminori Tanihara, Nhien Thi Nguyen, Quynh Anh Le, Takayuki Hirano, Takeshige Otoi
Faculty of Bioscience and Bioindustry, Tokushima University, Japan

P-63 Correction of the structural mutations of 450 kb KIT locus by CRISPR/CRISPR/Cas9 increases red blood cells in Yorkshire pigs

Zuyongh He, Ke Qin, Xinyu Liang, Kai Zhang, Xiaohong Liu, Yaosheng Chen
Sun Yat-sen University, P. R. China

P-64 Generation of a CLN7 KI sheep model of Batten disease

Fabien Delerue^{1,2}, Nicole Morey^{1,2}, Charley Lea Pollard³, Christopher G Grupen³, Imke Tammen³, Lars Matthias Ittner^{1,2}

¹Genome Editing at Macquarie University (GEM), Sydney - Australia; ²Dementia Research Centre, Macquarie University, Sydney - Australia; ³School of Veterinary Science, the University of Sydney, Sydney - Australia

P-65 CNS transduction and expression of AAV9 genes after systemic delivery

James Pickel¹, Nick Flytzanis², Nick Goeden², Viviana Gradinaru²

¹National Institutes of Mental Health / NIH; ²California Institute of Technology, USA

P-66 Development of a transposon transfection technology for making transgenic chicken embryo at day 0 of incubation

Shahin Eghbalsaied, Sabine Klein, Ute Beermann, Ronald Wittig, Meike Stunkel, Maren Ziegler, Wilfried Kues

Institute of Farm Animal Genetics, Friedrich-Loeffler-Institut (FLI), Mariensee, Neustadt am Rubenberge, Germany.

P-67 Transgenic resources for the study of human disease and development at the National Xenopus Resource

Marcin Wlizla, Sean McNamara, Nikko-Ideen Shaidani, Marko Horb

National Xenopus Resource, Marine Biological Laboratory, USA

P-68 Exploring beneficial genetic variants in Atlantic salmon using gene editing

Erik Kjarner-Semb, Rolf Edvardsen, Anna Wargelius

Institute of Marine Research, Norway

P-69 Generation of a Cancer Gene Allelic Series in Mice by CRISPR/Cas9 System

Hsiang-Hsian Lori Fan¹, I-Shing Yu², Yin-Hung Lin¹, Shin-Yu Wang¹, Ying-Hsuan Liaw¹, Pei-Lung Chen^{1,3}, Tsung-Lin Yang⁴, Shu-Wha Lin⁵, You-Tzung Chen¹

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P-70 Challenges in modelling human diseases ? mice are not men

Benedikt Wefers^{1,2}, Xianyuan Xiang³, Regina Kneutinger¹, Sarah Passon¹, Christian Haass^{1,3}, Wolfgang Wurst^{1,2}

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P-71 Altering organelle localization of mitochondrial protein show lethal impairment of sulfur respiration

Masanobu Morita¹, Tomoaki Ida¹, Tomohiro Tanaka², Tetsuro Matsunaga¹, Akira Nishimura¹, Motohiro Nishida², Hozumi Motohashi³, Takaaki Akaike¹

¹Tohoku University Graduate School of Medicine; ²National Institute for Physiological Sciences; ³IDAC, Tohoku University, Japan

P-72 PA28 $\alpha\beta$ OVEREXPRESSION ENHANCES LEARNING AND MEMORY OF FEMALE MICE WITHOUT INDUCING PROTEASOME ACTIVITY

Julia Adelfof¹, John Wiseman¹, My Andersson², Michelle Porritt¹, Anne Petersen³, Madeleine Zetterberg³, Malin Hernebring^{1,3}

¹AstraZeneca, UK; ²University of Lund, Sweden; ³University of Gothenburg, Sweden

P-73 Transgenic mice with genetically evoked progressive degeneration of noradrenergic neurons as a novel tool to study presymptomatic phase of Parkinson's Disease.

Justyna Barut¹, Agnieszka Jurga¹, Katarzyna Rafa-Zablocka¹, Monika Baginska¹, Rosanna Parlato^{2,3}, Andrii Domanskyi⁴, Irena Nalepa¹, Grzegorz Kreiner¹

¹Dept. of Brain Biochemistry, Institute of Pharmacology PAS, Krakow, Poland; ²Institute of Applied Physiology, University of Ulm, Ulm, Germany; ³Institute of Anatomy and Cell Biology, University of Heidelberg, Heidelberg, Germany; ⁴Institute of Biotechnology, University of Helsinki, Helsinki, Finland

P-74 Generation of Fam83h Knockout Mice by CRISPR/Cas9-mediated Gene Engineering

Fardin Fathi, Sherko Naseri, Sara Parsa, Farzad Soleimani, Bahram Nikkho, Karim Rahimi

Cellular and Molecular Research Center, Research Institute for Health Development, Kurdistan University of Medical Sciences, Sanandaj, Iran

P-75 Inactivation of glyoxalase 1 in cell lines and mice reveals compensatory mechanisms for methylglyoxal detoxification

Rebekka Medert¹, Dagmar Schumacher¹, Jakob Morgenstern², Yoko Oguchi¹, Nadine Volk², Stefan Kopf², Jan Benedikt Groner², Peter Paul Nawroth², Thomas Fleming², Marc Freichel¹

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P-76 Disease relevant mutations introduced by genome editing in a humanised Barth Syndrome mouse model

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P-77 Gut Microbiota may influence your Mouse Model Phenotype

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P-78 Altered spermatogenesis in rats lacking the specific thyroid hormone transporter, monocarboxylate transporter 8

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P-79 A coagulation factor IX-deficient rat as a translational pre-clinical model of severe hemophilia B

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P-80 CRISPR genome-edited animal models with patient-specific mutations

Almudena Fernandez, Santiago Josa, Diego Munoz, Andrea Montero, Ana Maria Guardia, Marta Sanchez Diez, Marcos Rubio, Gema Garrido, Marta Cantero, Julia Fernandez, Lluís Montoliu CNB-CSIC and CIBERER-ISCIII, Madrid, Spain

P-81 Next-Gen large collaborative Alzheimer's Disease model consortium: MODEL-AD

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P-82 Application of reporter mice for live imaging in embryology

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P-83 A single reporter mouse line for Vika, Flp, Dre, and Cre recombination

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P-84 A novel creation of reporter mouse model for assessing germ layer formation at gastrulation

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P-85 Cryopreservation of embryos using two protocols: A comparison study

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P-86 A Novel Undifferentiated Spermatogonia-specific Surface Protein 1 (USSP1) in Neonatal Mice

Junju Huang

Sun Yat-sen University, P. R. China

P-87 N-acetyl cysteine improves the fertilizability of vitrified-warmed mouse oocytes by recovering the thiol levels of zona pellucida post oxidation

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P-88 Possums and primordial germ cells- a novel method of pest control

Melanie Kate Laird, Tim Hore, Neil Gemmell

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P-89 Research & Development of Mouse Reproductive Technology in CARD

Satohiro Nakao, Ayumi Mukunoki, Hidetaka Yoshimoto, Kana Tamura, Chihiro Sugahara, Koharu Kirikihira, Kotono Ito, Mei Yamada, Serina Kuroshima, Ryusei Maeda, Jorge Sztejn, Toru Takeo, Naomi Nakagata

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P-90 Nuclear DNA fragmentation as a measure of sperm fertility in mutant mice

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P-91 Staging estrous by vaginal impedance in hormonally primed rats

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P-92 Building up Nation-wide Infrastructure of Mouse Biology in Korea

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P-93 Applying genome editing technology to mutant mouse model creation in RIKEN BRC

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P-94 Rat Resource and Research Center

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P-95 The NCI Mouse Repository: Cancer models and miRNA-ES cell resource

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P-96 Data management systems for CARD mouse bank in Kumamoto University

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P-97 Setting up rat embryo cryopreservation in a transgenic animal unit

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P-98 Sanitation and move of a large animal facility

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P-99 Progress update of the mouse resource program at RIKEN BioResource Research Center

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P-100 Extending the Benefits of goGermline™ Chimeras: Expediting time to First Heterozygotes and First Experimental Cohorts

Jonathan Gauntlett, Maree Hagan, Jacqui Watts, Frank Koentgen, Roger Askew

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