

**13. Jahrestagung der DG-GT
&
4th Annual Workshop
"Viral Vectors and Gene Therapy" of the GfV
Düsseldorf, 12.-14.07.2006**



Mittwoch, 12.07.2006

15.00-18.00 Uhr PhD student course
Moderation: Hildegard Büning, Jens Bohne

• **Vektoren: Ad**

Florian Kreppel



Adenovirus vectors belong to the most frequently used viral gene transfer vectors. They offer several advantages like a well characterized virus biology, high titer vector production and high genomic stability. The educational session will give an overview over the different types of adenovirus vectors and their potential applications in gene therapy. Finally, an overview on chemical capsid modification techniques will be provided.

AAV

Hildegard Büning



Among the variety of viruses currently under development as vectors for gene therapy adeno-associated viruses (AAV) are some of the most promising candidates. AAV vectors have demonstrated long-term expression in numerous animal models and an excellent safety profile in human clinical studies. An overview of the current knowledge of different serotypes, infectious biology, vector production, integration and capsid engineering will be provided.

Retroviren, Lentiviren

Jens Bohne



Für die Therapie einiger monogenetischer Erkrankungen hat sich der Gentransfer mit retroviralen Vektoren als erfolgreich erwiesen. Eine sichere und effiziente Handhabung der Vektoren baut auf einem grundlegenden Verständnis des retroviralen Lebenszyklus auf. Der modulare Aufbau der verschiedenen Vektortypen soll aufgezeigt werden, besonders im Hinblick auf ein zukünftiges „Plentivirus“, welches die positiven Eigenschaften der Retrovirus-Familie zusammen führt.

Produktion, Sicherheit, GMP, Standards

Klaus Kühlcke



Die Herstellung von retroviralen Vektoren für klinische Studien in der Gentherapie erfordert das upscaling von Methoden aus dem Labormaßstab in einen pharmazeutischen Maßstab. Außerdem müssen aufgrund der regulatorischen Vorgaben eine Vielzahl von Qualitätskriterien beachtet werden. Die Entwicklung von aus diesen Notwendigkeiten resultierenden Methoden wird vorgestellt und diskutiert werden. Insbesondere wird auf eine Methode zur Erzeugung von retroviralen Vektoren in einem Maßstab von bis zu 200 Litern und eine anschließende Konzentrierung der Vektoren eingegangen werden.

Rainer Löw



Vorstellung der verschiedenen Techniken zur Herstellung retroviraler Vektoren im Labormaßstab. Die verschiedenen Techniken werden bezüglich der notwendigen Etablierungsarbeit aber auch Sicherheit der produzierten Vektoren verglichen. Ein besonderer Schwerpunkt wird die Herstellung von

selbst-inaktivierenden retroviralen (SIN) Vektoren sein. Insbesondere deren stabile Produktion als Grundlage für die Herstellung klinisch relevanter Vektoren wird diskutiert.

16.30 Uhr Kaffee, Kuchen und Getränke

• Neue Methoden:

Viral libraries

Luca Perabo

Combinatorial strategies for the optimization of protein function have been employed and refined for more than a decade. Recently these technologies have been applied to engineer viral particles for human gene transfer. This session will provide an overview on the state of the art techniques in this field and consider promises and challenges of the use of viral libraries for directed evolution of gene therapy vectors.

Microarrays

Sandeep Nambiar



High-density DNA microarray measures the activities of several thousand genes simultaneously and the gene expression profiles have recently been used to distinguish biological or patho-physiological states from one another. This new approach promises to give better therapeutic measurements to patients by diagnosing disease stages with improved accuracy. One of the challenges in applying data analysis techniques for classifying gene expression data is to extract accurate, readily interpretable rules providing biological insight as to how classification is performed. Current methods generate classifiers that are accurate but difficult to interpret. The lecture will give a comprehensive overview of microarray platforms and various supervised and unsupervised analytical approaches taking molecular classification of cutaneous melanoma as a sample data set.

DNA methylation

Alireza Mirmohammadsadegh



DNA methylation of cytosine at position C5 in CpG dinucleotides is a crucial epigenetic modification of the genome that is involved in the regulation of transcription, genomic imprinting and inactivation of tumor suppressor genes. Aberrant DNA methylation of CpG dinucleotides in promoter regions has been found to be associated with a growing number of cancer diseases. The majority of studies of DNA methylation is semiquantitative and positional CpG methylation, which is crucial for gene inactivation, has only recently been described. This so called pyrosequencing technology is a method for sequencing-by-synthesis in real time. This convenient method can use a simple blood sample and will most likely contribute to the molecular profiling of tumors and the early detection of aberrant DNA methylation.

LAM-PCR

Manfred Schmidt



Retro- and lentiviral based gene transfer vectors integrate stably and semi-randomly into the cellular genome. The generated proviral-genomic fusion sequence can be used as a clonal marker that is characteristic for each transduced cell and its clonal progeny. The fusion sequences can be identified down to the single cell level using linear amplification mediated (LAM-) PCR. Thus, monitoring of transduced cells in pre-/clinical gene therapy is feasible and helps to uncover insertional side effects.

18.00-19.00 Uhr Meet the expert

19.00 Uhr Abendveranstaltung

Donnerstag, 13.07.2006

09.00 Uhr **Opening remarks by Ulrich Hengge**
Welcome address by Christopher Baum (DG-GT),
Toni Cathomen (GfV)
Introduction by Ulrich Hengge

Keynote lecture Prof. Kathy High:
“What we have learned from gene therapy of hemophilia
in mice, dogs and man”

10.00-11.20 Uhr **Viral vector meets cell surface (I)**
Moderation: Toni Cathomen, Christopher Baum

Hildegard Büning: Retargeting of AAV2

Freie Vorträge

- 1) Philip Raake et al.: Coapplication of vascular permeability factor significantly enhances long-term transduction of cardiomyocytes by retroinfusion of adeno-associated virus serotype 2 into the coronary vein
- 2) Miriam Böckmann et al.: Selective targeting of orthotopically growing medullary thyroid carcinoma in RET transgenic mice by a newly identified peptide: new implications for systemic cancer gene therapy
- 3) Lydia J. Duerner, Klaus Cichutek, and Christian J. Buchholz: Selection for tumour homing of retroviruses displaying a human antibody library

11.20-11.40 Uhr **Kaffeepause und Posterwalk**

11.40-13.00 Uhr **Viral vector meets cell surface (II)**
Moderation: Michael Hallek, Axel Rethwilm

Stephan Urban: Anti-viral peptides against HBV infection

Freie Vorträge

- 1) Marlon R. Veldwijk et al.: Generation and application of an efficient Chronic Myelogenous Leukemia (CML)-targeted recombinant adeno-associated virus (rAAV) vector: New therapeutic options?
- 2) Jörg Schuettrumpf et al.: Coagulation and viral transduction: Protease-activated receptors modulate AAV and adenoviral gene transfer efficacy
- 3) Christian Jogler et al.: Replication properties of human adenovirus in vivo and in primary cell cultures from different animal species
- 4) Klaus Mantwill et al.: Potential of the oncolytic adenovirus Delo3 RGD in the treatment of gliomas

13.00-14.00 Uhr **Lunch Break und Posterwalk**

14.00-15.20 Uhr In the cell

Moderation: Bertram Opalka, Helmut Hanenberg

Michael Kann: Capsid-Import bei HBV

Freie Vorträge

- 1) Axel Schambach, Melanie Galla, and Christopher Baum: Upstream polyadenylation enhancers in the U3 region of self-inactivating retroviral vectors improve titer and gene expression, and prevent transcriptional read-through
- 2) A. Wolfstein et al.: The inner tegument promotes herpes simplex virus capsid motility along microtubules in vitro

Urs F. Greber: Virus trafficking in cells

15.20-16.20 Uhr Genotoxicity (I)

Moderation: Regine Heilbronn, Manuel Grez

V. Piguet: Analysis of HIV-1 replication and integration in dendritic cells identifies restrictions early in the viral life cycle

Freie Vorträge

- 1) Anja Ehrhardt et al.: Transposition from a gene-deleted adenoviral vector results in phenotypic correction in a canine model for hemophilia B
- 2) Daniel Wicke et al.: Gene therapy of CAMT: A difficult balance to cure MPL deficiency

16.20-16.40 Uhr Kaffeepause

16.40-17.30 Uhr Genotoxicity (II)

Moderation: Stefan Frühauf, Bernd Gänsbacher

W. Hammerschmidt: EBV-Replicons

Freie Vorträge

- 1) Cynthia C. Bartholomae et al.: Lentiviral SIN-vectors affect target site selection and clone survival
- 2) Peter A. Horn et al.: Establishment of multilineage hematopoietic readout systems in a nonhuman primate model (*Callithrix jacchus*) for comparative in vivo/in vitro analysis

17.30-18.30 Uhr Mitgliederversammlung DG-GT und Posterwalk

19.00 Uhr Social event

Freitag, 14.07.2006

09.00-10.40 Uhr News from the clinics (I)

Moderation: Rainer Haas, Ulrich Hengge

Clinical section (I)

Christoph Klein: Genterapie bei Wiskott-Aldrich-Syndrom

Christof von Kalle: Update zu X-SCID

Manuel Grez: Update zu Chronischer Granulomatose

Freie Vorträge

- 1) Sylvia Borchers et al.: Graft versus leukemia with little graft versus host disease? A clinical phase I/II study using transfusion of transduced donor T-cells for donor leukocyte transfusion
- 2) Isabel Vogler et al.: Improvement of DLI safety in pediatric leukaemia patients by combining two independent suicide strategies

10.40-11.00 Uhr Kaffeepause und Posterwalk

11.00-12.30 Uhr News from the clinics (II)

Moderation: Christof von Kalle, Wolfgang Uckert

Clinical (II) and Preclinical section

Tomáš Hanke: Clinical experience with DNA-MVA-vectored vaccines against HIV-1

Freie Vorträge

- 1) Mohamed Hassan et al.: Induction of apoptosis by ectopic expression of BH3-only member Noxa in melanoma cell lines
- 2) V. Kleff et al.: Correction of β c-deficient Pulmonary Alveolar Proteinosis (β c-PAP) by hematopoietic stem cell gene transfer: Studies in a murine knockout model
- 3) Oliver Ebert, Katsunori Shinozaki, and Savio LC Woo: Eradication of advanced hepatocellular carcinoma by transarterial infusions of oncolytic vesicular stomatitis virus in rats
- 4) Tsanan Giroglou et al.: Gene therapy of glioblastoma with adult bone marrow derived tumor infiltrating cells (BM-TIC)
- 5) Melanie Wurm et al.: Foamyviral vectors efficiently transduce nonhuman primate hematopoietic and embryonic stem cells

12.30-13.30 Uhr News from the clinics (III)

Moderation: Dorothee van Laer, Wolfgang Hammerschmidt

Immunotherapy section

Karin Mölling: DNA Vakzinierung gegen Tumoren

Freie Vorträge

- 1) Josef Köstler et al.: Designed new vaccines like the equine herpesvirus (EHV) type 1 based viral vectors effectively transduce and activate monocyte derived dendritic cells (MDDC)
- 2) Elisa Kieback et al.: Depletion of gene-modified T cells as a safety mechanism in adoptive T cell therapy
- 3) Felix Bohne et al.: Redirection of T cells using chimeric T cell receptors against HBV surface proteins

13.30-14.30 Uhr Lunch Break und Posterwalk

14.30-14.40 Uhr Verleihung der Posterpreise

**14.30 Uhr DFG-Schwerpunktprogramm 1230
Christopher Baum et al.**

Christian Buchholz, Langen:	Dissecting and targeting retroviral vector cell into human hematopoietic stem cell
Hildegard Büning, Köln:	Entry and genotoxicity of AAV vectors in the lympho-hematopoietic system
Jürgen Kleinschmidt, Heidelberg:	Structure-function relationships of AAV capsids in post entry trafficking and gene transfer
Florian Kreppel, Ulm:	Analysis of post-receptor restriction in adenovirus vector entry and targeting
Manfred Ogris, München:	Receptor-targeting and intracellular disassembly of gene vectors
Hans-Joachim Lipps, Witten:	Establishment and mitotic stability of a non-viral episome for mammalian cells
Wolfgang Hammerschmidt, München:	Extrachromosomal plasmids and conditionally regulated gene vectors
Jochen Bodem, Würzburg:	Influence of nuclear architecture on gene vector persistence and expression
Manuel Grez, Frankfurt:	Vector-Chromatin Interactions: Molecular basis of retroviral integration clusters
Zoltan Ivics, Berlin:	Mechanism of target site selection in Sleeping Beauty transposition and its experimental manipulation for targeted transgene insertion in human cells
Anja Ehrhardt, München:	Evaluation and delivery of various improved recombinases into mammalian cells with viral vectors for long-term and stable transgene expression levels
Toni Cathomen, Berlin:	Gene repair in hematopoietic stem cells of a SCID mouse model
Siegfried Weiß, Braunschweig:	Fate of plasmid DNA in host cells after bacteria mediated gene transfer
Armin Baiker, München:	Development of a non-viral episomal vector system enabling tissue-specific replication
Klaus Cichutek, Langen:	A lentiviral vector transducing G0-arrested cells: Analysis of genetic determinants
Christopher Baum, Hannover:	Origin and evaluation of insertional self-renewal mutants in the hematopoietic system
Stefan Frühauf, Heidelberg:	Retroviral vector insertions in hematopoietic cells
Manfred Schmidt, Heidelberg:	Stability, integration properties and vertical transmission of integrase deficient lentiviral vectors and their innovative uses in hematopoietic cells
Helmut Hanenberg, Düsseldorf:	Avoiding MDS/AML in genetic therapy for FA with complex retroviral vectors

Boris Fehse, Hamburg:

Do retroviral vector insertions influence the fate of terminally differentiated cells such as T lymphocytes by insertional mutagenesis?

Wolfgang Uckert, Berlin:

Risk analysis of T cell receptor gene-modified T lymphocytes

Ingo Röder, Leipzig:

Mathematical modeling of individual clone dynamics for genetically modified stem cells within the hematopoietic system

17.00 Uhr Verabschiedung und Ende der Tagung

Wir danken allen Sponsoren:



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