Workshop – Systems Biology of Ageing
Jena, Germany   September 8-9, 2014

New FLI Lab Building
Leibniz Institute for Age Research - Fritz Lipmann Institute (FLI),
Beutenbergstr. 11, D-07745 Jena / Germany

Organised by the
Jena Centre for Systems Biology of Ageing – JenAge and
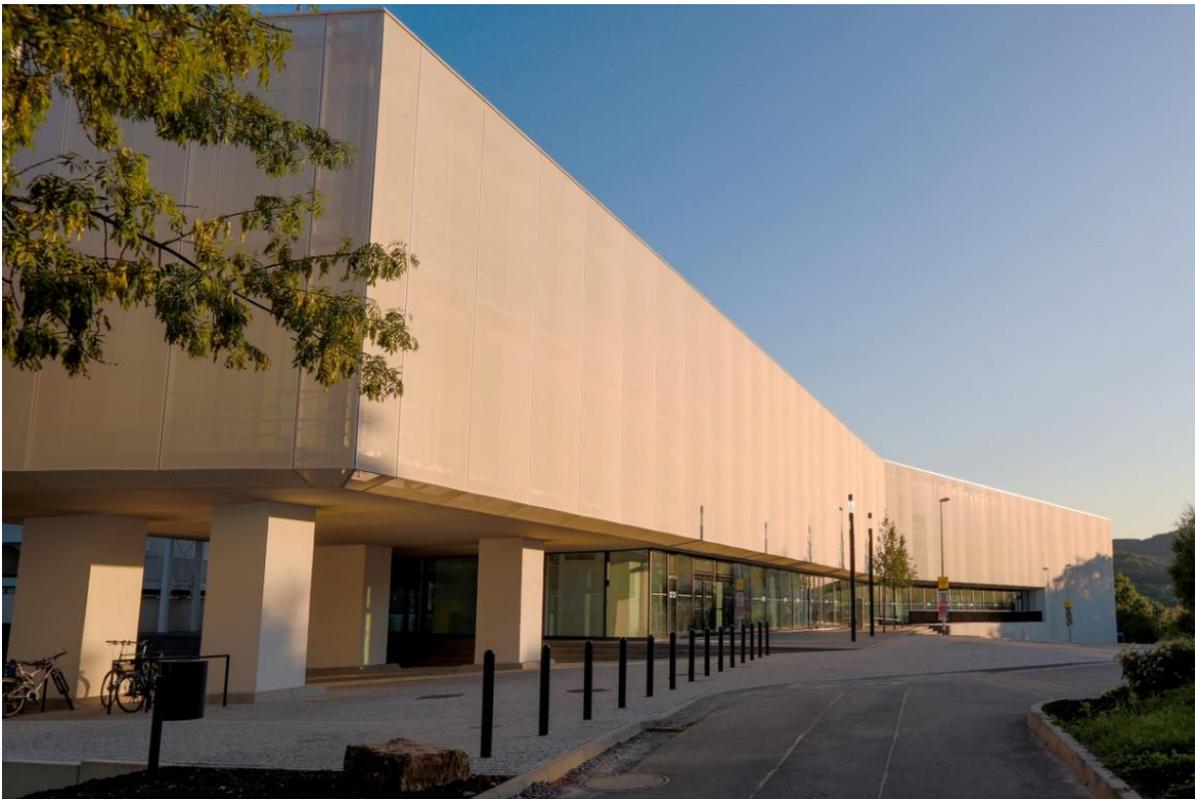**supported by the Leibniz Institute for Age Research, Fritz Lipmann Institute – FLI**

Photo: R. Hühne

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Programme

Monday, 08/09/2014

09:00   Registration

10:00   Introduction / Welcome

10:15-11:00 *A. Kriete (Philadelphia / PA, USA)*
Robustness and aging – The control gap. (Invited Talk)

11:00-11:30 *C. Dieterich and the SYBACOL consortium (Cologne, Germany)*
SYBACOL – Post-transcriptional gene regulation in worms and fly.

11:30-12:00 *M. Moeller, G. Fuellen and the ROSAge Consortium (Rostock, Germany)*
Longitudinal Data. Hard to obtain! Closest to truth?

12:00-12:30 *J. Mansfeld, K. Schmeisser, D. Kuhlow, S. Weimer, S. Priebe, I. Heiland, M. Birringer, M. Groth, A. Segref, Y. Kanfi, N. L. Price, S. Schmeisser, S. Schuster, A. Pfeiffer, R. Guthke, M. Platzer, T. Hoppe, H. Y. Cohen, K. Zarse, D. A. Sinclair, M. Ristow (Cologne, Fulda, Jena, Potsdam-Rehbrücke, Germany; Ramat-Gan, Israel; Zürich, Switzerland; Boston / MA, USA)*
Role of sirtuins in lifespan regulation is linked to methylation of nicotinamide.

12:30-13:30 Lunch

13:30-14:00 *M. A. Moni, P. Lio (Cambridge, UK)*
Multi omics methodologies to study the impact of infections on ageing associated comorbidities.

14:00-14:30 *T. Heinemann, P. Verbruggen, E. Manders, G. von Bornstaedt, R. van Driel, T. Höfer (Heidelberg, Germany; Amsterdam, The Netherlands)*
Robust DNA repair through collective rate control.

14:30-15:00 *M. Baumgart, M. Groth, S. Priebe, A. Savino, G. Testa, A. Dix, R. Ripa, F. Spallotta, C. Gaetano, M. Ori, E. Terzibasi Tozzini, R. Guthke, M. Platzer, A. Cellerino (Frankfurt/Main, Jena, Germany; Pisa, Italy)*
RNA-seq of the aging brain in the short-lived fish *N. furzeri* – Conserved pathways and novel genes associated with neurogenesis.

15:00-15:30 Coffee Break / Refreshments

15:30-16:00 *P. F. Thaben, K. Thurley, S. Lück, P. O. Westermark (Berlin, Germany)*
Loss of circadian rhythms in hundreds of genes in aged mouse skeletal muscle.

16:00-16:30 *J. Przybilla, T. Rohlf, J. Galle (Leipzig, Germany)*
Modeling DNA-methylation profiles in ageing and cancer.

16:30-18:00 Poster Session

18:00 Barbecue

Tuesday, 09/09/2014

09:00-09:45 *J. P. de Magalhães (Liverpool, UK)*
Bioinformatics, systems biology and ageing: Navigating the new oceans of data to discover the Fountain of Youth. (Invited Talk)

09:45-10:15 *G. Kollarovic, M. Studencka, L. Ivanova, C. Lauenstein, K. Heinze, A. Lapytsko, S. R. Talemi, A. S. Cabrita Figueiredo, J. Schaber (Magdeburg, Germany)*
To senesce or not to senesce: How primary human fibroblasts decide their cell fate after DNA damage.

10:15-10:45 *S. Marthandan, K. Klement, S. Ohndorf, P. Hemmerich, S. Diekmann (Jena, Germany)*
Quiescent fibroblast cells age.

10:45-11:15 Coffee Break / Refreshments

11:15-11:45 *C. Baldow, L. Thielecke, S. Gerdes, I. Glauche (Dresden, Germany)*
On the quantification of hematopoietic tissue remodeling in aging and disease: Simulations, measures and predictions.

11:45-12:15 *J. Galle, J. Przybilla (Leipzig, Germany)*
In vitro ageing of stem cells: A computational model approach.

12:15-12:45 *T. Rohlf (Leipzig, Germany)*
Connecting time scales – Modeling epigenetics and evolution of ageing.

12:45-13:45 Lunch

13:45-14:15 *S. Schuster, I. Heiland, T. Gossmann, J. Gebauer, L. de Figuereido, M. Ziegler, C. Kaleta (Jena, Germany; Tromsø, Bergen, Norway; Sheffield, Cambridge, UK; Odense, Denmark)*
Computer simulation of metabolism in the framework of age research.

14:15-14:45 *U. Hahn, E. Faessler (Jena, Germany)*
Natural language text analytics – Towards a novel type of content-driven information infrastructure for ageing research.

14:45-15:15 *R. Hühne, T. Thalheim, J. Sühnel (Jena, Germany)*
Towards data integration in ageing research: AgeFactDB - The JenAge Ageing Factor Database.

15:15-15:30 Closing Remarks

Poster Session
(08/09/2014, 16:30 – 18:00, Seminar room: Nucleus)

M. Baumgart, M. Groth, A. Savino, P. Sieber, S. Priebe, U. Menzel, R. Guthke, M. Platzer, A. Cellerino (Jena)
MicroRNA expression during ageing – A multi-species comparison.

M. Bens, K. Szafranski, M. Platzer (Jena)
High-quality assembly of the naked mole-rat transcriptome.

J. Gebauer, S. Schuster, L. F. de Figueiredo, C. Kaleta (Hinxton, Jena, Odense)
Detecting and investigating substrate cycles in a genome-scale human metabolic network.

A. Groß, B. Kracher, J. M. Kraus, K. Luckert, O. Pötz, T. Joos, L. de Raedt, M. Kühl, H. A. Kestler (Ulm, Reutlingen, Leuven)
Predicting the dynamic behavior of Wnt/β-catenin and Wnt/JNK signaling by a rule based probabilistic modeling approach.

N. Hartmann, C. Englert (Jena)
Alteration of mitochondrial activity with age in the short-lived fish *Nothobranchius furzeri*.

B. Hoppe, S. Pietsch, C. Englert (Jena)
Addressing kidney regeneration in *N. furzeri*, a novel model organism for ageing research.

A. Kadlecová, D. Schubert, J. Voller, M. Strnad (Olomouc)
***Caenorhabditis elegans* as a model organism for testing of anti-aging activity of plant hormone cytokinins.**

P. Koch, B. Downie, K. Reichwald, D. Chalopin, J.-N. Volff, M. Platzer (Jena, Lyon)
Using a novel NGS read-based method for the discovery and annotation of repetitive elements in the genome of the short-lived killifish *Nothobranchius furzeri*.

U. Menzel, S. Priebe, M. Baumgart, M. Groth, M. Baumgart, A. Cellerino, R. Guthke (Jena)
Identification of longevity biomarkers in *Nothobranchius furzeri* by transcriptome measurement and random forest analysis.

A. Petzold, P. Koch, K. Reichwald, M. Platzer (Jena)
Annotating the N. furzeri genome assembly.

S. Pietsch, B. Hoppe, L. Dong, C. Englert (Jena)
Exploring kidney homeostasis and regeneration using high-throughput analysis.

A. Sahm, K. Szafranski, M. Platzer (Jena)
Pipeline for identification of genes with an evolutionary history of positive selection.

P. Sieber, S. Schäuble, S. Schuster, S. Germerodt, C. Kaleta (Jena, Odense)
Caloric restriction and life-time expectancy – Insights from a generic agent-based model.

H. Stark, S. Schuster (Jena)
Blood is thicker than water – Calculating the optimal hematocrit.

J. Sühnel and the JenAge consortium (Jena, Pisa, Zürich)
Systems biology of mild stress in healthy ageing – A multi-species approach. Research profile of the Jena Centre for Systems Biology of Ageing – JenAge.

T. Thalheim, J. Schleicher, K. Wagner, R. Hühne, J. Sühnel (Jena)
The JenAge Information Centre – An information hub for ageing research and systems biology.

M. Thüne, J. Przybilla, T. Rohlf (Leipzig)
Analysis of time and space dependent high-dimensional epigenetic data.

Abstracts of Talks

Listed in order of programme

Invited Talk

Robustness and ageing – The control gap.

A. Kriete

School of Biomedical Engineering, Science and Health Systems, Drexel University, Philadelphia, USA

Robustness and aging have been discussed from two different perspectives. The first view addresses changes of robustness in aging organisms in response to perturbations. Hereby robustness is used as an aid to assess proximal mechanisms of aging. The second, more recently introduced aspect[1], considers the existence of robustness tradeoffs in biological designs and makes a contribution to the ultimate cause of aging.

Appealing to the latter view are limitations in control and regulation, which are fundamental to complex systems. It is in this area where both engineered technical systems, purposely designed, and evolved biological systems, are most alike. Control provides the ability of a system to maintain stability, perform tasks efficiently, reject perturbations, and restore structure and function if damaged. Such mechanisms are essential for open biological systems to provide survival and independence in fluctuating environments (nutrients, temperature, or perturbations by microorganisms). While optimized to perform such tasks robustly, both technical and biological systems can fail if exposed to unexpected perturbations. In this context aging appears as an inability of an organism to maintain function due to a lack of appropriate control, and this tradeoff is what is defined here as a control gap.

Limitations in molecular control mechanisms as it pertains to aging, supported by computer modeling, will be presented. Other performance and efficiency properties traded against longevity, along with a perspective on evolutionary theories, will be discussed.

[1] Kriete A. Robustness and aging – a system level perspective. *BioSystems.* **2013**;112:37-48.

Contributed Talk

SYBACOL – Post-transcriptional gene regulation in worms and fly.

C. Dieterich[1] and the SYBACOL consortium (www.sybacol.org)

[1] Computational RNA Biology Lab and Bioinformatics Core, Max Planck Institute for Biology of Ageing, Joseph-Stelzmann-Straße 9b, Cologne, Germany

Several signaling pathways have been discovered to prolong life span and health span in the roundworm *C. elegans* and other species, but the underlying mechanisms are poorly understood. Within SYBACOL, comprehensive OMICS data sets from wildtype and different mutant strains in longevity pathways have been generated. My presentation focuses on data processing, quality control and meta analysis of this comprehensive data resource. We identified common signatures of longevity in terms of differentially regulated gene sets and pathways across multiple mutants, which target different longevity pathways. We also report on the effect of aging on RNA metabolism, especially on processes such as RNA editing, splicing and post-transcriptional regulation by miRNAs.

Contributed Talk

Longitudinal Data. Hard to obtain! Closest to truth?

M. Moeller[1], G. Fuellen[1] and the ROSAge Consortium

[1] Institute for Biostatistics and Informatics in Medicine and Ageing Research, Rostock University Medical Center, Rostock, Germany

Ultimately and necessarily, ageing can only be understood as a longitudinal phenomenon, and longitudinal data, not cross-sectional data, should be at the heart of any analyses of molecular ageing processes. In reality, however, longitudinal data are almost impossible to obtain for human, and in the data from the few cohorts followed over decades, strong confounders are expected to be present. For example, neither measurement protocols nor environmental influences can be expected to be stable.

Model organisms allow some more longitudinal analyses, and we will first recapitulate work on data from the Nathan Shock Center at the Jackson Lab, describing mostly blood-based features of around 30 strains of mice (Moeller et al, Aging Cell 2014). These data were 'weakly longitudinal' in that blood-based features were obtained repeatedly from the same mouse, but its lifespan was not known and thus replaced by the average lifespan of the strain. In April 2014, the Jackson Lab published the data in a 'strongly longitudinal' fashion, assigning features and lifespan to individual mice. We found the overall trends in the data to be the same, no matter whether the data were paired or not. Surprisingly, the statistical confidence in the results from the paired data appears to be weaker, though. This may be an artifact: weakly longitudinal data necessarily feature more stability because lifespan is invariant for all mice of the same strain. But that stability would then just be based on missing true information.

Contributed Talk

Role of sirtuins in lifespan regulation is linked to methylation of nicotinamide.

J. Mansfeld[1,2,3,*], K. Schmeisser[1,*], D. Kuhlow[1,4], S. Weimer[2,4], S. Priebe[5], I. Heiland[6], M. Birringer[7], M. Groth[8], A. Segref[9], Y. Kanfi[10], N. L. Price[11], S. Schmeisser[1,12], S. Schuster[5], A. Pfeiffer[4], R. Guthke[5], M. Platzer[8], T. Hoppe[9], H. Y. Cohen[10], K. Zarse[1], D. A. Sinclair[11] and M. Ristow[1,2,4]

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[8] Genome Analysis, Leibniz Institute for Age Research, Fritz-Lipmann-Institute, Jena D-07745, Germany
[9] Department of C.elegans Genetics and Development, Institute for Genetics, University of Cologne, Cologne D-50674, Germany
[10] The Mina & Everard Goodman Faculty of Life Sciences, Bar-Ilan University, Ramat-Gan IL-52900, Israel
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* These authors contributed equally to the data presented.

Sirtuins, a family of histone deacetylases conserved in all higher organisms, have a fiercely debated role in regulating lifespan. Contrasting recent observations, we here find that overexpression of *sir-2.1*, the orthologue of mammalian SirT1, does extend *C.elegans* lifespan. Sirtuins mandatorily convert NAD+ into nicotinamide (NAM). We here find that NAM and its metabolite, 1-methylnicotinamide (MNA), extend *C.elegans* lifespan, also in the absence of *sir-2.1*. We identify *anmt-1* to encode a *C.elegans* orthologue of nicotinamide-N-methyltransferase (NNMT), the enzyme that methylates NAM to generate MNA. Disruption versus overexpression of *anmt-1* has opposing effects on lifespan independent of sirtuins. However and importantly, lack of *anmt-1* fully prevents *sir-2.1*-mediated lifespan extension. MNA serves as a substrate for a newly identified aldehyde oxidase, GAD-3, to generate hydrogen peroxide acting as a mitohormetic ROS signal to promote *C.elegans* longevity. The underlying mechanisms have been revealed by transcriptomics and subsequent promoter analysis. Taken together, sirtuin-mediated lifespan extension depends on methylation of NAM, providing an unexpected mechanistic role for sirtuins beyond histone deacetylation.

Schmeisser S *et al. Nat. Chem. Biol.* **2013**;9:693-700.

Contributed Talk

Multi omics methodologies to study the impact of infections on ageing associated comorbidities.

M. A. Moni and P. Lio

Computer Laboratory, University of Cambridge, Cambridge CB3 0FD, UK

Background: Comorbidity refers to the presence of multiple diseases or disorders in relation to a primary disease in an individual. Diseases are more likely to be comorbid if they share associated genes, proteins or molecular pathways. Infections and ageing are often associated to comorbidity that increases the risk of medical conditions which can lead to further morbidity and mortality.

Objective: This work presents a bioinformatics and statistical approach to estimate the prevalence, association and risk scores of the disease comorbidity. We have chosen Flu, SARS and HIV to quantify infection related comorbidity risk.

Methods: By using multi omics available data we have investigated pathways perturbation in HIV and SARS infections. First we have built a comorbidity profile map. Then based on the gene expression, protein-protein interactions (PPIs) and signaling pathways data, we investigate the comorbidity association of these 3 infective pathologies with other 7 diseases (heart failure, kidney disorder, breast cancer, neurodegenerative disorders, bone diseases, Type 1 and Type 2 diabetes). Phenotypic association is measured by calculating the Relative Risk and phi-correlation.

Results: The response to SARS seems to be mainly an innate inflammatory response; SARS dysregulates a large number of genes, pathways and PPIs sub-networks in different pathologies such as chronic heart failure (21 genes), breast cancer (16 genes) etc. HIV-1 induces comorbidities relationship with many other diseases, particularly strong correlation with the neurological, cancer, metabolic and immunological diseases. HIV significantly dysregulates a certain number of genes, pathways and PPIs sub-networks in pathologies such as kidney disorders (10 genes), bone diseases (7 genes) etc. It is notable that HIV and SARS similarly dysregulated 11 genes and 3 pathways. Only 4 significantly dysregulated genes are common between SARS-CoV and MERS-CoV, including *NFKBIA* that is a key regulator of immune responsiveness implicated in susceptibility to infectious and inflammatory diseases. Moreover, SARS and HIV infections dysregulates 4 genes (ANXA3, GNS, HIST1H1C, RASA3) and 3 genes (HBA1, TFRC, GHITM) respectively that affect the ageing process. These genes are involved in apoptosis, glycosaminoglycan degradation, lysosome, phagosome, endocytosis and metabolic pathways. We have discussed comorbidities of SARS and HIV in the context of Flu.

Conclusion: Bioinformatics and statistical approaches could provide important causality relationships and investigate hypotheses of ageing and senescence. Our integrated pipeline could elucidate aspects of disease progression mechanisms and disease comorbidity in a quantitative way.

Contributed Talk

Robust DNA repair through collective rate control.

T. Heinemann[1,2], P. Verbruggen[1,3], E. Manders[3], G. von Bornstaedt[1,2], R. van Driel[3] and T. Höfer[1,2]

[1] Division of Theoretical Systems Biology, German Cancer Research Center (DKFZ) Heidelberg, Germany
[2] BioQuant Center, Heidelberg, Germany
[3] Swammerdam Institute for Life Science, University of Amsterdam, Amsterdam, The Netherlands

DNA repair is indispensable for the intracellular protection against environmental and endogenous damaging agents. This is reflected in an increased susceptibility against cellular aging and cancer development as a consequence to impaired repair. Functional repair is carried out by enzymatic macromolecular complexes that assemble at specific sites on the chromatin fiber. How the rate of these molecular machineries is regulated by their constituent parts is poorly understood.

Here we quantify nucleotide-excision DNA repair (NER) in mammalian cells and find that, despite the pathways' molecular complexity, repair effectively obeys slow first-order kinetics. Theoretical analysis indicates that these kinetics are not due to a singular rate-limiting step. Rather, first-order kinetics emerge from the interplay of rapidly and reversibly assembling repair proteins, stochastically distributing DNA lesion repair over a broad time period. Based on this mechanism, the model predicts that the repair proteins collectively control the repair rate. Exploiting natural cell-to-cell variability, we corroborate this prediction for the lesion-recognition factors XPC and XPA.

Our findings provide a rationale for the emergence of slow time scales in chromatin-associated processes from fast molecular steps and suggest that collective rate control might be a widespread mode of robust regulation in DNA repair and transcription.

Contributed Talk

RNA-seq of the aging brain in the short-lived fish *N. furzeri* – Conserved pathways and novel genes associated with neurogenesis.

M. Baumgart[1,*], M. Groth[1,*], S. Priebe[2,*], A. Savino[3], G. Testa[3,4], A. Dix[2], R. Ripa[3], F. Spallotta[5], C. Gaetano[5], M. Ori[4], E. Terzibasi Tozzini[3], R. Guthke[2], M. Platzer[1], and A. Cellerino[1,3]

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*=equal contribution.

The brains of teleost fish show extensive adult neurogenesis and neuronal regeneration. The patterns of gene regulation during fish brain aging are unknown. The short-lived teleost fish *Nothobranchius furzeri* shows markers of brain aging including reduced learning performances, gliosis and reduced adult neurogenesis. We used RNA-seq to quantify genome-wide transcript regulation and sampled five different time points to characterize whole-genome transcript regulation during brain aging of *N. furzeri*.

Comparison with human datasets revealed conserved up-regulation of ribosome, lysosome and complement activation and conserved down-regulation of synapse, mitochondrion, proteasome and spliceosome. Down-regulated genes differ in their temporal profiles: neurogenesis- and extracellular matrix genes showed rapid decay, synaptic- and axonal-genes a progressive decay. A substantial proportion of differentially expressed genes (~40%) showed inversion of their temporal profiles in the last time point: spliceosome and proteasome showed initial down-regulation and stress response-genes initial up-regulation. Extensive regulation was detected for chromatin remodelers of the DNMT and CBX families as well as members of the polycomb complex and was mirrored by an up-regulation of the H3K27me3 epigenetic mark.

Network analysis showed extensive co-regulation of cell cycle/DNA synthesis genes with the uncharacterized zinc-finger protein ZNF367 as central hub. In situ hybridization showed that ZNF367 is expressed in neuronal stem cell niches of both embryonic zebrafish and adult *N. furzeri*. Other genes down-regulated with age, not previously associated to adult neurogenesis and with similar patterns of expression are AGR2, DNMT3A, KRCP, MEX3A, SCML4, and CBX1. CBX7, on the other hand, was up-regulated with age.

Contributed Talk

Loss of circadian rhythms in hundreds of genes in aged mouse skeletal muscle.

P. F. Thaben, K. Thurley, S. Lück and P. O. Westermark

Institute for Theoretical Biology, Charité – Universitätsmedizin, Berlin, Germany

The biological circadian clock generates rhythms with a period of ~24 hours that synchronize to our daily light-dark cycles. Genetic feedback loops in single cells are the primary generator of these rhythms, but they are also observable at the tissue and organ level, and all the way to the behavior of organisms. Thousands of genes in many cell types exhibit circadian rhythms in their expression. If and how these rhythms in gene expression are influenced by cellular and organismal aging is largely unexplored, as are the possible physiological consequences. We study circadian gene expression in different tissues in young and aged mice. Here, we report a study of circadian gene expression in hindlimb skeletal muscle of mice 8 and 80 weeks old, respectively. To assess changes in gene expression rhythms on a genome-wide scale, we developed a novel statistical method called differential rhythmicity. We found a widespread reduction of amplitudes in hundreds of genes. Many of these lost rhythms were in genes coding for components of signaling pathways involved in tissue homeostasis and muscle regeneration, whereas rhythms in genes coding for metabolic enzymes and their regulation were largely unaffected by age. The physiological significance of these findings are outlined in the context of circadian response analysis, a theoretical framework for understanding the circadian control of signaling and metabolic pathways that we have developed. Implications for muscle wasting and decreased muscle function in aged animals are discussed.

Contributed Talk

Modeling DNA-methylation profiles in ageing and cancer.

J. Przybilla[1] , T. Rohlf[1,2] and J. Galle[1]

[1] Interdisciplinary Center for Bioinformatics, Leipzig, Germany
[2] Max Planck Institute for Mathematics in the Sciences, Leipzig, Germany

The stem cell epigenome is characterized by a sensitive balance between chromatin modification- and demodification processes. During cancer development and ageing this balance becomes disturbed and results in characteristic changes of the epigenome that may also induce differences in gene expression patterns. In particular, characteristic drifts in DNA-methylation have been observed in both contexts.

Several experimental studies demonstrated that during both, cancer development and ageing, specific groups of CpGs become hyper-methylated while others become hypo-methylated compared to their normal methylation state. These changes are known to be functionally relevant as they can affect the transcription of associated genes: previously silenced genes can become active and active genes can become silenced. On the basis of differences in DNA-methylation patterns it is possible to classify cancer subtypes. In human blood, e.g., it was shown that the state of aging can be tracked by a couple of age-related CpGs (Weidner et al., 2014).

Here, we introduce a multi-scale model (Przybilla et al., 2013, Przybilla et al., 2014) that enables simulation of these epigenetic processes on the molecular, cellular and population level. We model cell populations where each cell contains an artificial genome encoding for round about 100 genes. Transcriptional regulation of these genes is controlled by cis-regulatory networks, trimethylation of lysine 4 (H3K4me3) and lysine 9 (H3K9me3) and DNA methylation.

We apply the model in order to provide a mechanistic explanation of parallel local hyper- and global hypo-methylation during cancer development and ageing. For this purpose, we analyze how persistent proliferation activity and mutations that change specific histone modifiers impact DNA methylation profiles.
We find that spontaneous loss of histone modifications during cell replication can induce hyper- and hypo-methlyation. These epigenetic drifts can be modified by mutations of chromatin modifiers. We present some typical simulation scenarios that are consistent with experimental findings.

Weidner CI *et al., Genome Biol.* **2014**;15(2):R24.
Przybilla J *et al., Front. Life Sci.* **2013**;7(1-2):31-43.
Przybilla J *et al., Aging Cell.* **2014**;13(2):320-8.

Invited Talk

Bioinformatics, systems biology and ageing: Navigating the new oceans of data to discover the Fountain of Youth.

J. P. de Magalhães

Department of Functional and Comparative Genomics, Institute of Integrative Biology, University of Liverpool, Liverpool, UK

Ageing is the major biomedical challenge of the 21st century, yet it remains largely mysterious, partly because the ageing process involves multiple genes and their interactions with each other and with the environment that remain poorly understood. In this talk, I will present genomic and computational approaches aimed at deciphering the genome and increasing our knowledge about how genes and pathways impact on ageing. We have also been employing whole transcriptome profiling (RNA-seq) to gather insights on ageing and its manipulation by diet. Moreover, I will present our work of integrating gene expression profiles with age-related changes at other biological levels and new online resources for integrative and systems biology of ageing. Lastly, I will discuss our recent work in sequencing and analyzing the genome of the longest-lived mammal, the bowhead whale, to identify longevity assurance mechanisms.

Contributed Talk

To senesce or not to senesce: How primary human fibroblasts decide their cell fate after DNA damage.

G. Kollarovic, M. Studencka, L. Ivanova, C. Lauenstein, K. Heinze, A. Lapytsko, S. Rastgou Talemi, A. S. Cabrita Figueiredo and J. Schaber

Institute for Experimental Internal Medicine, Medical Faculty, Otto von Guericke University, Magdeburg, Germany

Senescence is generally perceived as an important tumour-suppressor mechanism. It was suggested that the cell fate decision between permanent arrest, i.e. senescence, and transient arrest is mediated by a permanent DNA damage signal emanating from unrepairable telomeric DNA damage. Here, we address the question how cells discriminate between high initial and low background DNA damage including telomere associated DNA damage in order to make a cell fate decision. Specifically, we focus on the G1-S transition. To this end, we measured the dynamics of DNA double strand breaks after different doses of ionising radiation and corresponding dynamics of well-known players that mediate the DNA damage signalling and cell cycle arrest for MRC5 normal human diploid fibroblasts (HDFs). In order to come to a quantitative and mechanistic understanding of the underlying molecular network, we combined our experiments with mathematical models. We present the first parameterized mechanistic model for DNA-damage regulated G1-S transition in HDFs. The model well recapitulates DNA-damage dynamics for different ionizing radiation regimes. Moreover, the model is able to explain the dynamics of several key proteins involved in the G1-S checkpoint. The model predicts that G1-S arrest is regulated by a robust hysteresis-switch, whose bi-stable region, i.e. the region where the cell fate decision between proliferation and senescence is taken, is precisely between six and 12 double strand breaks. Thus, the model provides a quantitative and mechanistic explanation how cells count their double strand breaks and decide whether the amount of non-repairable DNA damage does not allow for proliferation. Accordingly, the model predicted that IR up to 10 Gy is not sufficient to permanently arrest the cells, which was experimentally corroborated. The model also suggests that, opposed to the commonly accepted opinion of a cyclin-regulated G1-S transition, it is the abundance of the cyclin-dependent kinases Cdk1 and Cdk2 that control DNA damage induced G1-S arrest. Cdk abundance in turn is mainly regulated by p21. Consequently, the model predicted that p21 silencing would not only rescue G1-S arrest, but up-regulate Cdk1,2, which was also experimentally corroborated.

Contributed Talk

Quiescent fibroblast cells age.

S. Marthandan, K. Klement, S. Ohndorf, P. Hemmerich and S. Diekmann

Leibniz Institute for Age Research - Fritz Lipmann Institute e.V. (FLI), Jena, Germany

Cellular senescence has been postulated to be a consequence of stepwise telomere shortening in each round of replication. If telomere attrition is the main mechanism triggering transition into senescence, quiescence should not contribute to this process and should extend lifespan to an amount the cells spent in the quiescent state. In contrast, we observed here that quiescent human fibroblast cells age and transit into senescence, clearly indicating that telomere attrition cannot be the main mechanism triggering senescence. Instead, cell maintenance due to oxidative stress might be the mechanism inducing senescence. Lowering oxygen levels from 20% to 3% indeed delays transition into senescence of MRC-5 however, not WI-38 fibroblasts, suggesting cell strain-specific stress responses. We also show that both, long-term quiescence and senescence are also protected against transition into apoptosis. Significant DNA damage accumulation over time occurs during senescence as well as quiescence and is also independent of oxygen levels suggesting primarily endogenous sources eliciting DNA damage accumulation. Since long-term quiescent fibroblasts do not transit into the irreversible states, terminal differentiation and apoptosis, quiescent cells are prone to tumor development. To counterbalance this risk, with time also quiescence cells transit into senescence, a state known to suppress tumor development.

Contributed Talk

On the quantification of hematopoietic tissue remodeling in aging and disease: Simulations, measures and predictions.

C. Baldow, L. Thielecke, S. Gerdes and I. Glauche

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Aging is most apparent on the tissue level as a progressive decline of functional ability. Although a wide range of molecular mechanisms has been identified that are closely connected to cellular aging, it is rarely understood how such intra-cellular aging processes translate on the phenotypic level of the overall tissue. The distinct age-related increase in the incidence of hematopoietic diseases, such as myeloproliferative neoplasms and leukemias, has long been associated with alterations of hematopoietic tissue structure with age. Novel methods for cell fate analysis, such as the use of genetic barcodes, allow assessing the clonal architecture of hematopoiesis even in *in-vivo* situations. However, it is still a challenging problem to quantify clonal contributions over time and, therefore, to estimate and predict future developments.

We developed a simple mathematical model of a self-stabilizing hematopoietic stem cell population to generate a wide range of possible clonal developments, reproducing typical, experimentally and clinically observed scenarios. We use the resulting model scenarios to suggest and test a set of statistical measures that should allow for an interpretation and classification of relevant clonal dynamics. In particular, we apply machine-learning approaches to identify measures for the reliable classification of clearly distinguishable scenarios, such as the early distinction between normal and potentially pathological developments. We report on our results to which extent these measures are suitable to prospectively predict atypical developments.

Additionally to the insights into structural principles of age-related tissue remodeling, our effort to establish a reliable classification of pathological and non-pathological clonal dynamics has a direct potential for clinical applications. Leukemogenesis is a well-known and severe problem in gene therapy patients. Based on the tight post-therapy monitoring of clonal developments in these patients our identified measures and the resulting categorization can aggregate time course data and provide estimates for the risk of atypical clonal developments and predict the manifestation of leukemia.

Contributed Talk

In vitro ageing of stem cells: A computational model approach.

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During ageing a decline in stem cell function is observed in many tissues. This decline is accompanied by complex changes of the chromatin structure among them are changes in histone modifications and DNA-methylation. Similar changes have been detected also during massive in vitro expansion, i.e. during in vitro ageing. In our opinion, understanding these age-associated processes is prerequisite to safe stem cell transplantation protocols. Using a computational tissue approach we study related questions on the molecular, cellular and population level.

For this purpose, we combine an individual cell-based model of stem cell populations with a model of epigenetic regulation of transcription. The combined model enables to simulate age-related changes of tri-methylation of lysine 4, 9 and 27 at histone H3 and of DNA methylation. These changes in chromatin structure entail expression changes of genes which induce age-related phenotypes of cells.

Applying the model in simulation studies, we demonstrate that massive in vitro expansion of stem cells can induce epigenetic reorganization which potentially affects the clonal competition in the culture and might facilitate expansion of transformed clones. Thereby, all the changes observed originate in the limited cellular capability to inherit epigenetic information, and thus are linked to cell proliferation. Whether significant drifts in regulatory states occur, depends on the ratio of time scales, e.g. of histone modification and cell replication. The time scale required for global chromatin re-organization and the degree to which the changes become stabilized depend on the properties of the histone and DNA modification machinery.

Contributed Talk

Connecting time scales – Modeling epigenetics and evolution of ageing.

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From the viewpoint of classical evolutionary biology, ageing represents a rather puzzling phenomenon, as it tends to limit individual reproductive success.
Furthermore, recent studies indicate an extreme diversity of ageing (and resulting mortality distributions) across biological clades [1], which clearly shows that the typical mammalian ageing pattern (e.g. approximately characterized by the Gompertz law) is far from being universal. Strong epigenetic drifts observed in mammalian (in particular, human) ageing suggest that chromatin-based phenomena, with probably high impact on self-renewal and differentiation of functional adult stem cells, play an important role for the emergence of aged phenotypes. We recently developed a mathematical model based on dynamic interactions between histone modifications and DNA methylation, showing that the observed epigenetic changes may originate in the limited cellular capability to inherit epigenetic information [2,3]. Spontaneous loss of histone modifications due to fluctuations on short time scales, e.g. caused by incorporation of de novo synthesized histones, can give rise to stochastic, and thus dysfunctional gene silencing by DNA methylation. In principle, these mechanisms are at work both in somatic and in germ line cells during the life time of an individual. In the latter, however, they can have evolutionary consequences e.g. through increased mutation (deamination) rates at methylated CpGs.

Here, we investigate effects on the evolution of CpG distributions resulting from epigenetic interactions. We simulate how different protective mechanisms (e.g. negative feedback between H3K4me3 and DNMT3a recruitment), and selective pressure on transcriptional states may stabilize regions of low methylation and resulting low, local CpG loss, and thereby explain empirical CpG distributions. A surprising prediction from this model is that CpG density and longevity of (mammalian) species should be correlated, which is indeed supported by publicly available data.

[1] Jones OR *et al.*, *Nature.* **2014**;505(7482):169-73.
[2] Przybilla J, Galle J, Rohlf T. *Bioessays.* 2012:34:841-848.
[3] Przybilla J *et al.*, *Aging Cell.* **2014;**13(2):320-28.

Contributed Talk

Computer simulation of metabolism in the framework of age research.

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Numerous metabolic processes are linked with ageing. Due to their complexity, computer simulation can help us enormously to understand the effects of changes in metabolism and even to predict hitherto unknown phenomena.

In this talk, the use of computer simulation is demonstrated by two examples. The first concerns nicotinamide adenine dinucleotide (NAD+), which is well known as a crucial cofactor in the redox balance of metabolism. Moreover, NAD+ is degraded in ADP-ribosyl transfer reactions which are important components of multitudinous signalling reactions. These include reactions linked to DNA repair and ageing. Proteins are modified by mono-ADP-ribosylation and poly-ADP-ribosylation. Histones are subject to NAD+-dependent deacetylation catalyzed by SIRTUINs, which have been shown to increase lifespan in a number of species. Several hypotheses on the relation of calorie restriction to NAD+ metabolism have been put forward.

Using the concept of elementary flux modes (EFMs), we determined all potential routes in a network describing NAD+ biosynthesis and degradation. All known biosynthetic pathways, which include *de novo* synthesis starting from tryptophan as well as the classical Preiss-Handler pathway and NAD+ synthesis from other vitamin precursors, are detected as EFMs. Moreover, several elementary modes are found that degrade NAD+, represent futile cycles or have other functionalities. A phylogenetic analysis of NAD metabolism in 45 species was performed, which documents significant differences between species. We critically examine the hypothesis that calorie restriction increases NAD+ turnover without altering steady-state NAD+ levels.

The second example concerns the detection of futile cycles in a genome-scale human metabolic network. While it is, so far, impossible to detect all of them due to combinatorial explosion, a representative sampling can be performed. We compared the extent of futile cycling in brain and liver cells with the expected extent in a generic human model. Moreover, we compared young and aged human brain cells.

Contributed Talk

Natural language text analytics – Towards a novel type of content-driven information infrastructure for ageing research.

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The JenAge project was probably the first German systems biology project that incorporated biomedical natural language processing (NLP) as crucial part of its information infrastructure. Text analytics were used to automatically unlock aging-relevant information from unstructured biomedical documents and thus complemented information available in manually created structured biological databases.

The range of functionalities which were available due to NLP techniques includes a wide variety of content-driven services. Among them are the support for semantic search of Medline documents, the extraction of ageing-relevant information to feed the central databases of JenAge (such as AgeFactDB: http://agefactdb.jenage.de/) as well as relational information incorporating RDF-style facts, e.g. relating to ageing-relevant protein-protein or mRNA interactions, the search for information at varying degrees of certainty of information and, finally, generating and modifying ageing-specific scientific hypotheses on the fly. All these different functionalities have been bundled into a common framework, the Semedico system, which provides an interactive graphic interface as well as visualization aids for document or fact retrieval.

These different functionalities are grounded in flexibly configurable NLP pipelines mainly based on automatically trained statistical models for named entity recognition (e.g. for recognizing instances of genes, mRNA, diseases) and relation extraction (e.g. linking specific proteins, mRNA, species or diseases). Complementing these machine learning-based approaches, the development of Semedico was also based on a large number of terminological resources publicly available in the biomedical community (e.g. the UMLS, BioPortal and OBO). The quality of these components has been demonstrated by outstanding results achieved on various benchmark data sets and the participation in international challenge competitions (such as BioCreative, BioNLP, CLEF-ER). Its usefulness for JenAge will be demonstrated by collaborative work with the Cellarino Lab on mRNA relation extraction.

Wermter J, Tomanek K, Hahn U. High-performance gene name normalization with GeNo. *Bioinformatics.* **2009**;25(6):815-821.
Buyko E, Faessler E, Wermter J, Hahn U. Syntactic simplification and semantic enrichment: trimming dependency graphs for event extraction. *Computational Intelligence.* **2011**:27(4):610-644.

Contributed Talk

Towards data integration in ageing research: AgeFactDB — the JenAge Ageing Factor Database.

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Research into ageing and age-associated diseases can greatly benefit greatly from a systems biology or integrative biology approach. One important aspect of this development would be integration of data taken either from existing databases or directly from the scientific literature. We have therefore developed AgefactDB (http://agefactdb.jenage.de) a database aimed at the collection and integration of ageing phenotype data including lifespan information. Ageing factors are considered to be genes, chemical compounds or other factors such as dietary restriction, whose action results in a changed lifespan or another ageing phenotype. Any information related to the effects of ageing factors is called an observation and is presented on observation pages. To provide concise access to the complete information for a particular ageing factor, corresponding observations are also summarized on ageing factor pages. In a first step, ageing-related data were primarily taken from existing databases such as the Ageing Gene Database—GenAge, the Lifespan Observations Database and the Dietary Restriction Gene Database—GenDR. In addition, we have started to include new ageing-related information. Based on homology data taken from the HomoloGene Database, AgeFactDB also provides observation and ageing factor pages of genes that are homologous to known ageing-related genes. These homologues are considered as candidate or putative ageing-related genes. AgeFactDB offers a variety of search and browse options, and also allows the download of ageing factor or observation lists in TSV, CSV and XML formats.

Poster Abstracts

Listed in alphabetical order by family name of first author

Poster

MicroRNA expression during ageing – A multi-species comparison.

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MicroRNAs (miRNAs) modulate mRNA translation and stability and therefore represent a novel layer in the regulation of gene expression, taking place after transcription. We used small RNA-seq (Illumina) to analyse the miRNA expression changes occurring during ageing in the model systems studied in our consortium (JenAge): *C. elegans*, *N. furzeri*, zebrafish, mouse, human fibroblast cell lines (MRC-5 and HFF), and human blood and skin biopsy samples donated by volunteers. We sequenced a total of 471 small-RNA libraries: between 2 and 3 different organs (brain, skin, liver, and blood) were analysed in up to 5 biological replicates at 4 to 5 different time points in each species. Bowtie and miRBase version 20 were used for annotation of miRNAs. Differentially-expressed miRNAs were identified by the use of DESeq2 (FDR < 0.05). Using Venn analysis, we could identify a small set of miRNAs consistently up- or down-regulated with age in all vertebrate species. We also analysed the effects of ageing on global miRNA editing in the brain of *N. furzeri*, zebrafish and mouse. In all three species, ageing is associated with increased editing of the 3' end of miRNAs and progressive enrichment of shorter isomiRs. Finally, we investigated the genome-wide effects of miRNAs by RNA-seq of zebrafish embryos injected with two different miRNAs up-regulated with age. The results were compared by generally applicable gene-set/pathway analysis (GAGE) with those obtained in the ageing brain, liver and skin of *N. furzeri* and all three comparisons showed that overexpression partially mimics the effects of ageing on gene regulation.

Poster

High-quality assembly of the naked mole-rat transcriptome.

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Knowledge of transcript sequences is a prerequisite to study qualitative and quantitative aspects of transcripts. Advances in next-generation sequencing technology, like growth in throughput, quality and read length, offer an affordable approach to near-complete snapshots of transcriptomes. Assembly programs were introduced to reconstruct mRNA molecules from next-generation sequencing data, but despite improvements in technology and algorithms, *de-novo* transcriptome assembly is still difficult. Difficulty arises from artifacts in RNA-seq data, like nonuniform transcript coverage, and from transcriptome complexity of mammalian cells with different alternative splice variants, highly similar paralogues and repeats. Therefore, reconstructed transcripts may be fragmented or misassembled (Martin and Wang, 2011; Grabherr et al., 2011).

Here we present a genome-independent (*de novo*) transcriptome assembly pipeline, that specifically addresses post assembly tasks, such as annotation of assembled transcript contigs, identification of misassembled contigs, scaffolding of fragmented contigs, coding sequence identification and 3' UTR clipping. We used our pipeline to *de novo* assemble and annotate the transcriptome of the naked mole-rat (NMR) (*Heterocephalus glaber*), a mouse-sized rodent with an exceptional long lifespan of >30 years in captivity and an important non-model organism in ageing research (Mele et al., 2010; Austad, 2009). Starting our pipeline with 352 million stranded reads obtained from 10 different tissues and using 19,178 human RefSeq mRNAs as a reference, we find NMR counterparts to 88% of human genes, covering 90% of genes annotated in GO Terms. Scaffolding was applied to 18% of all NMR counterparts, with a median increase in length of 20%. Comparison between human and identified NMR coding sequence length shows 71% of genes with ≥ 90% reconstructed CDS length.

Our data provides the basis for gene expression studies and qualitative analyses in NMRs investigating molecular causes leading to its long and healthy life.

Austad SN. Comparative biology of aging. *J Gerontol A Biol Sci Med Sci.* **2009**; 64:199–201.
Grabherr MG *et al.* Full-length transcriptome assembly from RNA-Seq data without a reference genome. *Nat Biotechnol.* **2011***;29:644–652.
Martin JA, Wang Z. Next-generation transcriptome assembly. *Nat Rev Genet.* **2011;**12:671–682.
Mele J *et al.* Mechanisms of aging in the naked mole-rat: The case for programmed aging. *Russ J Gen Chem.* **2010;**80:1455-1464.

Poster

Detecting and investigating substrate cycles in a genome-scale human metabolic network.

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Substrate cycles, also known as futile cycles, are cyclic metabolic routes which consume energy by hydrolysing cofactors such as ATP. In this work, we analyse a large number of futile cycles in human metabolism and discuss their statistics. For this purpose we use two recently published methods, the EFMEvolver and the K-shortest EFM method, to calculate samples of 100,000 and 15,000 substrate cycles, respectively. We find a surprisingly high number of futile cycles in human metabolism with up to one hundred reactions per cycle, flowing through up to six different compartments. An analysis of tissue specific models of liver and brain metabolism shows that there is a selective pressure acting against the uncontrolled dissipation of energy by avoiding the coexpression of enzymes belonging to the same futile cycle. This selective force is particular strong against futile cycles that have a high flux due to thermodynamical principles.

Poster

Predicting the dynamic behavior of Wnt/β-catenin and Wnt/JNK signaling by a rule based probabilistic modeling approach.

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Recent data indicates that a large number of proteins participate and interact in intracellular signal transduction forming large signaling networks. Due to the inherent complexity of such networks prediction of their behavior requires mathematical models and computational simulations. Here, we show that a static interaction network can be transformed into a semi-quantitative simulation model, which is able to reproduce the global behavior of the modeled signaling network. The model is based on the specification of probabilities for the actual occurence of so-called protein interactions including binding, enzymatic activation or phosphorylation. A set of local rules derived from experimental data and literature modifies these interaction probabilities according to the interdependencies between the different interactions. This enables the model to respond to external stimuli. Moreover, the model behavior can be observed under different conditions like knockout of network components or inhibition of specific interactions. The new rule-based probabilistic approach is able to represent dynamics of common network motifs found in signal transduction networks. We applied the presented computational method to Wnt/β-catenin and Wnt/JNK signaling. These signal transduction networks are involved in various biological processes ranging from development to aging. Our in-silico observations are in agreement with previously published findings as well as our own experimental data. These results suggest that protein-protein interaction maps augmented by local interaction rules can be a suitable means of predicting the global behavior of complex intracellular signaling networks under physiological and pathological conditions.

Poster

Alteration of mitochondrial activity with age in the short-lived fish *Nothobranchius furzeri.*

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Among vertebrates that can be kept in captivity the annual fish *Nothobranchius furzeri* has the shortest known lifespan. It also shows typical signs of ageing and is therefore an ideal model to assess the role of different physiological and environmental parameters on ageing and lifespan determination. Here we use *N. furzeri* to study whether ageing is associated with any mitochondrial alterations. We observed that mitochondrial DNA copy number, expression of mitochondria-related genes, and mitochondrial respiration significantly decreases with age in *N. furzeri*.

To manipulate the level of mitochondrial activity in *N. furzeri* and to study the effect on aging, we designed two approaches: i) Physical exercise and ii) over-expression of mitochondria-related genes. Physical exercise is achieved by forcing the fish to swim against a water current. Swimming performance and exercise conditions were tested in a swim tunnel. Long-term exercise experiments are performed in small tanks equipped with pumps that generate a defined water current. Preliminary experiments where fish were daily exercised (15 min per day) for six weeks showed a significant increase in the expression of numerous mitochondrial genes. We are currently analysing the effect of life-long exercise experiments on mitochondrial function and longevity.

To over-express mitochondrial genes, we have recently developed a protocol how to inject DNA transgenes into the 1-cell embryo of *N. furzeri*. At the moment we are generating several transgenic lines that over-express the *mitochondrial transcription factor A (Tfam)* and the *peroxisome proliferator-activated receptor γ coactivator-1α (Pgc-1α)* under different promoters. The amount of Tfam has been described to regulate mitochondrial DNA copy number and Pgc-1α has been shown to increase mitochondrial biogenesis in mammals. Overall, our findings suggest that despite the short lifespan, ageing of *N. furzeri* is associated with an impairment of mitochondrial function. Whether increased mitochondrial function is sufficient to delay ageing and extend lifespan is currently investigated.

Poster

Addressing kidney regeneration in *N. furzeri*, a novel model organism for ageing research.

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Loss of kidney function results in a number of diseases. Mammals can only partly repair the damaged functional units (nephrons) in the kidney. In contrast, fish and reptile nephrogenesis and renal regeneration persist throughout life. The mechanisms of kidney regeneration are not understood completely yet. Therefore it is important to clarify the underlying mechanisms of regeneration in this organ.

One focus of our research work is to study the kinetics of regeneration in the newly established ageing model *Nothobranchius furzeri*, to get an insight in the molecular mechanisms of this process. *N. furzeri*, which belongs to the group of killifish has an extremely short life span of less than 12 month and is found in ponds in the south of Africa, which desiccate during the dry season. To address a potential age-dependence, a renal injury was caused in young (16 week-old) and aged (42 week-old) *N. furzeri* by using nephrotoxic gentamicin. The regeneration process was followed over a period of eight days. To investigate kidney functionality FITC-conjugated dextran was injected, which is taken up only by the proximal part of intact nephrons and fails to be filtered after an injury occurred. While young fish have regenerated completely after 6 days, a delay of several days is visible in aged fish. Using proliferation assays we found that young fish showed an increased expression after 2 days while old fish show only a slight increase at day 6 after gentamicin injection. To study the functionality of the kidney an assay was developed to measure the glomerular filtration rate. With this assay we could show the regeneration on a functional level in young fish. Performing RNA-sequencing, we found a group of genes with a peak expression after two days in young fish and a delayed response in old fish. Among this group were genes, which are involved in mesenchymal-to-epithelial-transition (MET) and EMT, such as vimentin and occludin. These processes might play a role in the development of new nephrons. Furthermore we found miR-21 to be up-regulated in the regeneration process. To investigate the function of this microRNA in the regeneration process, we performed knock down experiments. In addition we want to gain an insight in the ability of renal cells to form new nephrons. Therefore a cell suspension of whole kidney marrow of transgenic fish of different ages was transplanted to middle aged fish and the number of newly formed nephrons/cells in this heterochronic background was quantified.

Poster

Caenorhabditis elegans as a model organism for testing of anti-aging activity of plant hormones cytokinins.

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Caenorhabditis elegans shares several conservative aging-related pathways with mammals including humans. It provides a more complex picture of biological processes than cell cultures and compared to mammal models, it is much easier and cheaper to maintain in the laboratory. This and also many other advantages make it an ideal organism to use in screening of compounds with anti-aging potential. Cytokinins, plant hormones derived from adenine, are one group of such compounds. Some of them previously delayed aging in cell cultures and also in flies. Moreover, they were able to suppress oxidative and glycation stress in a mouse model of brain aging. However cytokinins have never been tested in *C. elegans*. Our results suggest that some cytokinins including kinetin have anti-aging activity in this model. At present we are testing a library of synthetic cytokinin analogues that are being developed as drugs for skin diseases and as cosmeceuticals.

Poster

Using a novel NGS read-based method for the discovery and annotation of repetitive elements in the genome of the short-lived killifish *Nothobranchius furzeri.*

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Many *de novo* assemblies of complex eukaryotic genomes sequenced by next generation sequencing (NGS) technologies face the challenge of a high repeat content. These NGS reads are too short to span many types of repeats which may comprise several kilobases. In order to properly identify repeats (e.g. ~50% of the human, 55% of the zebrafish and 80% of the barley genome, respectively), species-specific repeat libraries are required. We have recently reported a k-mer based method ("RepARK") [1] for the discovery and annotation of the fraction of genomic NGS data sets representing repetitive elements.

We thoroughly analyzed the repeat content of the genome of the short-lived killifish *Nothobranchius furzeri*, which is a new model organism for aging research. This genome has an estimated size of 1.6-1.9 Gb [2], for which initial analyses showed a repeat content of 64% (21% of which are tandem repeats). Combining repeats identified by RepARK with those identified by reference based programmes produced a comprehensive library of repetitive elements (25,000 elements, 5.6 Mb). Using this library, we identified 62.4% of the NGS reads and 38% of the current 1.2 Gb genome assembly as repetitive. We also calculated the evolutionary history of individual repeat families which revealed a possible recent transposon activity in the *N. furzeri* genome. These potentially active DNA transposons (hAT & TcMar) and LINE retrotransposons (L2, REX-Babar & RTE) are currently further analyzed with regard to representation in transcriptome data and their potential role in development and aging.

[1] Koch P *et al.* RepARK - de novo creation of repeat libraries from whole-genome NGS reads. *Nucleic Acids Res.* **2014**;42:e80.
[2] Reichwald K *et al.* High tandem repeat content in the genome of the short-lived annual fish Nothobranchius furzeri: a new vertebrate model for aging research. *Genome Biol.* **2009**;10:R16.

Poster

Identification of longevity biomarkers in *Nothobranchius furzeri* by transcriptome measurement and random forest analysis.

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Fin biospies were obtained from 152 individuals of the shortlived teleost fish *Nothobranchius furzeri* (maximum lifespan ~ 60 weeks), at the age of 10 weeks and 20 weeks. Importantly, the biopsies were taken from the fish without sacrificing them, so that lifespan data were available for each individual.

Fishes could be selected from 3 lifespan groups: shortlived, middlelived and longlived, each including 10 individuals. Transcriptome data have been generated using RNASeq. In order to identify genes which are predictive for lifespan, a Random Forest analysis has been performed, considering the expression at both time points as well as the change of the expression between the two time points. Proximity values for the most predictive genes, derived from the Random Forest analysis, have been used to establish multidimensional scaling (MDS) plots. The MDSplots reveal that the individuals cluster very well according to the lifespan groups, therewith confirming the lifespanpredictive power of the identified biomarkers. Additionally, the MDSplots show that, for the majority of the samples, the individual sample is determining the transcriptome, rather than the age. A gene set enrichment analysis revealed Gene Ontology categories that are strongly affected by ageing, and KEGG pathways which are significantly different between the lifespan groups. Therefore, we conclude that individuals differing in longevity show differences in gene expression patterns already at young age.

Poster

Annotating the *N. furzeri* genome assembly.

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The African annual killifish *Nothobranchius furzeri* has been established as a new model organism for the studies of ageing and lifespan determination. To provide the genetic and genomic resources that are required for experiments and studies, its genome has been sequenced and assembled at the institute during the last few years. The resulting assembly is of a high quality, e.g. it consists of long sequences with sizes comparable to those of typical chromosomes. Subsequently, the next step in the *N. furzeri* genome project is to annotate protein-coding and non-protein-coding genes in the genome assembly.

Annotation of protein-coding genes was based on a number of different sources of evidence. Transcriptomes of five different organs (whole body, brain, embryo, kidney, liver and skin) were sequenced with Illumina and assembled de novo, i.e. genome-independent, as well as spliced-aligned on the genome. Additionally, known protein sequences from the UniProtKB database were aligned to the translated genome sequence. Finally, various gene predictors were trained based on available sequence data and applied to predict putative gene structures. Combining all generated evidence provided annotations for 26,142 protein-coding genes and 59,154 protein-coding transcripts.

Annotation of non-protein-coding genes relied on secondary structure prediction with miRDeep*, Infernal and other dedicated programs and also included additional MicroRNA-seq and RNA-seq data. In total, 1,411 non-protein-coding genes were predicted; the majority were miRNA (598) and tRNA (453) genes.

In summary, 27,553 *N. furzeri* genes were annotated, which is comparable to annotations of other already sequenced fish genomes. Future analyses will concentrate on the identification of gene symbols and products to complete the gene annotation of the *N. furzeri* genome.

Poster

Exploring kidney homeostasis and regeneration using high throughput analysis.

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To aim for new insights in regeneration and homeostasis of kidney we followed 2 approaches. We investigated regenerative processes after damage induction in the new model Organism *Nothobranchius furzeri* using RNA-Seq and to examine mechanisms of homeostasis we applied a Wt1 ChIP-Seq experiment in mouse.

The Wilms tumor suppressor gene 1 (Wt1) is known for its essential role in development of multiple organs, e.g. the kidney. In adult kidneys, Wt1 expression is restricted to podocytes, which are cells that keep the integrity of the glomerular filtration barrier and contribute to the maintenance of kidney homeostasis[1]. We applied the Wt1 ChIP-Seq approach from isolated adult glomeruli.

We could prove already known functions for Wt1 and show potential binding sites near known kidney disease genes using MACS[2] for the detection of enriched genomic regions. New targets for Wt1 could be found and confirmed in wet-lab experiments. In addition, we calculated a new binding motif for Wt1.

After renal injury with gentamycin injection, fish show the ability to form new nephrons, the functional units of the kidney. The regeneration process is mediated by a variety of yet unknown genes, involved in apoptosis and proliferation of nephrons. We used the newly established ageing model organism *Nothobranchius furzeri* to study renal regeneration and whether regeneration declines with age using different time points after damage induction for the RNA-Seq experiment in young and old fish.

We could identify a cluster of genes showing an expression pattern matching the observations from regenerating nephrons at 2, 4 and 8 days post injection of gentamicin. Some of these "regeneration pattern" genes show also a delayed expression in old fish. In a GO Term enrichment analysis[3] using human orthologs this cluster showed a significant relation to extracellular matrix disassembly.

1. Gebeshuber CA *et al.* Focal segmental glomerulosclerosis is induced by microRNA-193a and its downregulation of WT1. *Nat Med.***2013;**19:481-7.
2. Feng J, Liu T, Qin B, Zhang Y, Liu XS. Identifying ChIP-seq enrichment using MACS. *Nat Protoc.* **2012**;7:1728–40.
3. Bindea G *et al.* ClueGO: a Cytoscape plug-in to decipher functionally grouped gene ontology and pathway annotation networks. *Bioinformatics.* **2009**;25:1091-3.

Poster

Pipeline for identification of genes with an evolutionary history of positive selection.

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Positive selection describes the phenomena that beneficial mutations are distributed across a species. Genes under positive Darwinian pressure can be assumed to be gone through major structural and functional changes during these phases of their evolutionary history.
A pipeline for genome-wide detection of positively selected genes in specific branches of evolution will be presented. Our application case aims to identify genes which cause certain African mole rat species to outlive all other rodents.[1] Based on user-provided coding sequences of different species the pipeline constructs an orthologue-catalogue, calculates and processes alignments and performs genome-wide tests of positive selection.[2] Such a test compares a model H0, in which all sites are assumed to be under purifying or neutral selection, against a model HA, in which sites are allowed to be under positive selection in user-defined branch of evolution.

1. Lele J, Edrey YH, Lewis KN, Buffenstein R. Mechanisms of aging in the naked mole-rat: The case for programmed aging. *Russ J Gen Chem*. **2010**;80(7):1455-1464.
2. Yang Z. PAML 4: phylogenetic analysis by maximum likelihood. *Mol Biol Evol.* **2007**;24(8):1586-91.

Poster

Caloric restriction and life-time expectancy – Insights from a generic agent-based model.

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Aging is an ubiquitous phenomenon observed across most of the tree of life. However, still little is known about which of the physiological changes that occur during aging are due to the aging process itself (i.e. the continuous deterioration of an organisms' functions) and which are adaptations to the aging process. To investigate this important question from an evolutionary point of view, we use an individual-based resource allocation model in which organisms are constantly exposed to an external stressor. The accumulation of damage eventually leads to the death of the organism. Through an evolutionary algorithm we evolve organisms to determine how they should optimally allocate their resources between reducing stress levels through this stressor and reproduction. Analyzing the evolved organisms, we find a very particular regulatory program that sheds new light onto the relevance of previously reported physiological changes during aging for the survival and longevity of organisms. Moreover, this model allows us to explain previously observed links between obesity as well as dietary restriction with life-span and the onset of age-specific diseases.

Poster

Blood is thicker than water – Calculating the optimal hematocrit

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The oxygen flow in humans and other higher animals depends on the erythrocyte-to-blood volume ratio, the hematocrit. It is physiologically favourable when oxygen transport rate is maximum. If the hematocrit were too low, too few erythrocytes could transport oxygen. If it were too high, the blood would be very viscous, so that oxygen flow would again be reduced. We here calculate that hematocrit value that is optimal for transporting a maximum amount of oxygen per time. This can be done by using the Hagen-Poiseuille law and considering the dependence of blood viscosity on the hematocrit. Different empirical or theoretically derived formulas for the dependence of viscosity on the concentration in a suspension have been proposed in the literature. We check which formulas lead to the best agreement between the theoretical and observed values. We show that especially a formula proposed by Svante Arrhenius (1917) is very appropriate for this purpose, leading to an optimal value of 40 %. This conforms very well to the observed values in humans and many other species.

The results are valid and useful in spite of considerable simplifications such as considering blood as a Newtonian fluid and neglecting the deformation, orientation and aggregation of erythrocytes. Also the prediction that the ratio between the viscosities of the blood and blood plasma at high shear rates nearly equals Euler's constant (2.718) is in good agreement with observed values.

Finally, we discuss possible extensions of the theory. We give an explanation for the difference in hematocrit between genders. Moreover, we derive the theoretical optimal hematocrit for persevering divers among marine mammals such as seals to be around 65 %, in excellent agreement with the values observed in several species.

This theoretical analysis has important implications for understanding human and animal physiology since oxygen transport is a crucial factor for medicine and physical performance. In particular, our results imply that blood doping in sports is physiologically questionable.

Stark H, Schuster S. Comparison of various approaches to calculating the optimal hematocrit in vertebrates. *J Appl Physiol.* **2012;**113:355-367.
Schuster S, Stark H. What can we learn from Einstein and Arrhenius about the optimal flow of our blood? *Biochim Biophys Acta.* **2014;** 1840(1):271–276.

Poster

Systems biology of mild stress in healthy ageing – A multi-species approach. Research profile of the Jena Centre for Systems Biology of Ageing – JenAge.

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Research into ageing and age-associated diseases can greatly benefit from a systems biology approach. The aim of the Jena Centre for Systems Biology of Ageing – JenAge (www.jenage.de) is to join forces both in age research and systems biology. Previous work on model organisms demonstrated that mild stress can increase lifespan and delay ageing. The generally favorable biological response of an organism to low dose exposure of stressors, called hormesis, has been repeatedly suggested to be the biological mechanism underlying the effects of calorie restriction and other life-extending treatments. The JenAge Centre aims to identify conserved transcriptional and metabolic networks activated by mild stress and to investigate their role in preserving functional integrity in old age. JenAge adopts a multi-species approach to characterise network modulations by environmental, pharmacological and life-style perturbations. The JenAge groups are studying the effects of genetic, environmental and pharmacological perturbations on age-related networks in human tissue cultures, in various model organisms and in humans. The model organisms studied range from worm (*Caenorhabditis elegans*) over two fish models, the extremely short-lived turquoise killifish (*Nothobranchius furzeri*) and zebrafish (*Danio rerio*), to mice (*Mus musculus*). In an iterative process, experimental data are communicated to the analysis and modelling groups to generate testable hypotheses which in turn are validated by genetic and other manipulations in model organisms. Automatic text mining will be used to cope with the ever-increasing flood of age-related scientific documents in a systematic way and to generate plausible hypotheses on ageing and age-related diseases through text analytics. This information is used, together with data from other databases and from the JenAge Centre, to set up a new database on molecular, cellular and organismic aspects of ageing (agefactdb.jenage.de). The general JenAge objective is to gain new insights into the complex interplay of maintenance and repair networks that govern the accumulation of damage and finally lead to age-related diseases and death. Since the end of 2009 the JenAge Centre has obtained many interesting results described in more than 50 publications.

Poster

The JenAge Information Centre – An information hub for ageing research and systems biology.

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The JenAge Information Centre (info-centre.jenage.de) is an information hub that collects and provides information on ageing, age-related diseases and systems biology. Its aim is to assist researchers in these fields. The Information Centre is not community-driven but contributions and suggestions from the scientific community are welcome.

The examples shown on the poster have a focus on ageing research and not on systems biology. In this field the Information Centre offers information on ageing-related centres & institutes, interest groups, organisations, blogs, science news and a meetings calendar. It also includes a rather comprehensive collection of ageing-related databases, books, journals and papers. The paper subsection contains a hit list of most cited papers related to ageing research and also current and historical papers. Examples for historical papers are the Parkinson paper from 1817, the Alzheimer papers from the beginning of the 20th century, the classical ageing paper by Sir Peter Medawar entitled 'An unsolved problem of biology' from 1952 and the Hayflick papers from 1961 and 1965. The database collection currently includes 63 entries with information on biological, demographic and disease data, except cancer, as well as metadata. The JenAge Information Centre collection of biological databases has been the starting point for the development of the JenAge database AgeFactDB.

The website is regularly updated. According to a rather conservative statistics excluding robots and counting external visitors only once a day between 3400 and 4400 hosts per month have accessed the JenAge Information Centre in 2014.

Poster

Analysis of time and space dependent high-dimensional epigenetic data.

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We are interested in quantifying temporal changes of chromatin organization, as they are observed in cell differentiation as well as during ageing. Since epigenetic dynamics affect chromatin on many scales, the classical gene-centered view has to be broadened. Going beyond the restriction to preselected single loci of the genome, one is faced with genome-wide, parallel analysis of multiple factors, e.g. histone modifications, CpG distribution, chromatin insulators. Due to the amount and high dimensionality of these data, detection and intuitive visualization of patterns and correlations across different cell types, or different time points requires the development of specific tools.

Here, we study the global epigenetic (re-)organization of the human and murine genome by analysing publicly available ChIP-Seq experiments for several histone modifications (H3K4me1/2/3, H3K27ac, H3K27me3 and H3K36me3) and CTCF binding sites during adipogenic differentiation. The methods we present are very generic and are readily applicable to any (time dependent) data, e.g. for comparison between tissues from different age groups.

Specifically, we investigate the length distribution and the averaged positional distribution of CTCF binding sites in the vicinity of consecutive modified regions, confirming the important role of CTCF positions on a rather fine scale. We found a peak of CTCF enrichment on the boundaries of H3K4me1 and H3K27ac regions, homogeneous enrichment within regions of H3K4me2/3 and suppression within regions of H3K27me3 and H3K36me3. Considering the distribution of CpG, points were modified regions split up during differentiation are found to be clearly distinguished within a range of a few 100bp.
Further, we combine a straight forward genome-segmentation (based on the combinatorial pattern of histone modifications) with self-organizing maps and demonstrate the potential of this method for data reduction and intuitive visualization of the epigenetic landscape and its reorganization over time. Several key features, e.g. bivalent chromatin or mutual exclusion of H3K27ac and H3K27me3, can be seen at a glance.

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