7th International *Macrostomum* Meeting

November 29 – December 1, 2013
European Research Institute for the Biology of Ageing
Groningen, The Netherlands

Program, participants, abstracts
and general information

Organizers:
Eugene Berezikov, Lukas Schärer, Peter Ladurner

Sponsored by:
Stichting Medicorum
ERIBA

Program

Friday, November 29, 2013

16:00-18:00 Registration

18:00-19:00 **Peter Lansdorp.** Guanine-rich DNA and stem cells

19:00- Welcome drinks and dinner at ERIBA

Saturday, November 30, 2013

8:30-9:00 Coffee

9:00-9.40 **Willi Salvenmoser.** Microscopic anatomy of *Macrostomum lignano*

9:40-10.00 **Roland Hoffman.** Use of nanotomy for *Macrostomum lignano*

10.00-10.30 **Kira Zadesenets.** The variation in chromosome number in karyotype of free-living flatworm, *Macrostomum lignano* (Platyhelminthes, Turbellaria)

10.30-11.00 Coffee break

11.00-11.30 **Kaja Wasik.** Building molecular genetic tools in a new model organism – *Macrostomum lignano*

11.30-12.00 **Eugene Berezikov.** *Macrostomum lignano* genomic resources

12.00-12.30 **Jochen Rink.** Heads or tails: Polarity in Planarians

12.30-13.30 Lunch

13.30-16.30 Free time (sightseeing opportunity, lab tour, collaborator meetings)

16.30-18.00 Poster session, combined with refreshments and small bites

18.00-18.30 Walk to a restaurant in a city center

18.30- Joint dinner in the restaurant NI HAO **(**Gedempte Kattendiep 122)

Program posters

Belinda Artes, Frederic Pacho, Sabrina Folie, Giada Carta, Constanze Ettl, Peter Ladurner. *M. lignano* projects in brief: TALENS, stem cells, gene complexity, antibodies.

Magda Grudniewska, Stijn Mouton and Eugene Berezikov. The flatworm *Macrostomum lignano* is a powerful model for stem cell and ageing research.

Georgina A. Rivera-Ingraham, U. Bickmeyer, D. Abele. Nitric oxide: a modulator of the hypoxic response in *Macrostomum lignano*?

Stella S. Schukies, Sigmund V. Sperstad and Matthias Leippe. A potential Toll-like receptor of *Macrostomum lignano.*

Sigmund V. Sperstad and Matthias Leippe. Evolutionary conserved immune effector proteins in *Macrostomum lignano.*

Valeria Vavilova, Irina Sormacheva, Evgeniy Brusentsev, Sergei Amstislavsky, Eugene Berezikov and Alexandr Blinov. Cryopreservation of free-living flatworm *Macrostomum lignano.*

Jakub J. Wudarski, Daniel Olivieri, Philipp M. Weissert, Eugene Berezikov. Towards transgenesis in *Macrostomum lignano*

Program

Sunday, December 1, 2013

9:00-9:30 Coffee

9:30-10:00 **Steven Ramm.** Functional and evolutionary genetics of seminal fluid in *Macrostomum*

10:00-10:30 **Birgit Lengerer.** Fundamentals of biological adhesion in *Macrostomum lignano*

10:30-11:00 **Robert Pjeta, Julia Wunderer.** Posterior end specific genes in *Macrostomum lignano*

11:00-11:30 Coffee break

11.30-12:00 **Stijn Mouton.** Studying ageing and rejuvenation in *Macrostomum lignano*

12:00-12:30 **Lukas Schärer.** General discussion and closing remarks

12.30- Lunch and departure

Abstracts

Talk: Guanine-rich DNA and stem cells

Peter Lansdorp

European Research Institute for the Biology of Ageing, University of Groningen, University Medical Center Groningen, Groningen, The Netherlands

Talk: Microscopic anatomy of *Macrostomum lignano*

Willi Salvenmoser[1], Bernhard Egger[1], Michael W. Hess[2] and Peter Ladurner[1]

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Here we describe the major tissues and cell types of the model organism *Macrostomum lignano* with light and electron microscopical methods. We will discuss in detail the epidermis with epitheliosomes, lysosomes, Golgi fields and the polymorph structured nucleus; the musculature and their interactions with the extra cellular matrix; a new cell type in the gut; two new cell types in the male gonad and also parenchymal cells types. In addition we will show the protonephridial system, the male copulatory organ and the stem cell system. Furthermore the nervous system and the different gland cell types will be discussed. Finally we will show a comparison between high pressure frozen and chemical fixed animals.

Abstracts

Talk: Use of nanotomy for *Macrostomum lignano*

Roland Hoffmann

European Research Institute for the Biology of Ageing, University of Groningen, University Medical Center Groningen, Groningen, The Netherlands

New developments in transmission electron microscopy (TEM) allow large area imaging of *Macrostomum lignano* at macromolecular resolution.

Talk: The variation in chromosome number in karyotype of free-living flatworm, *Macrostomum lignano* (Platyhelminthes, Turbellaria)

Kira S. Zadesenets[1], Irina D. Sormacheva[1], Alexander G. Blinov[1], Eugene Berezikov[2], Nikolay B. Rubtsov[1]

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2 - European Research Institute for the Biology of Ageing, Groningen, The Netherlands

In this study preliminary results of cytogenetic analysis of chromosomes of *M. lignano* are presented. Earlier, the karyotype of *M. lignano* was described as 2n=8 (one pair of large chromosomes and 3 pairs of small chromosomes) (Egger and Ishida, 2005). In this study the cytological analysis was performed for individual worms of DV1 line. The chromosome slide preparation technique was optimized for obtaining metaphase spreads from single worms. It was shown that karyotype of diploid cells in different individuals can contain 8, 9 or 10 chromosomes. In all cases karyotype contained 6 small metaphase chromosomes, but the number of large metaphase chromosomes could vary (1, 2 or 3). The chromosome rearrangement could be explained by chromosomal duplication event which led to tetrasomy 1. In this presentation the chromosome number variability in different individuals of *M. lignano* is discussed.

Abstracts

Talk: Building molecular genetic tools in a new model organism – *Macrostomum lignano*

Wasik K.A.[1], Battistoni G. [1], Gurtowski J. [1], Schatz M.C. [1], Ladurner P., Schärer L., Berezikov E. [3], Hannon G.J. [1,2]

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Macrostomum lignano has an impressive regenerative ability and presents an interesting opportunity to study stem cells (neoblasts) in detail. However, the molecular tools for manipulating *Macrostomum* remain limited. Therefore, we sought to develop a minimal set of tools to aid in the study of *Macrostomum*.

First, we have sequenced *Macrostomum's* genome and, in collaboration with the Berezikov (ERIBA) and Schatz (CSHL) labs, are currently preparing a draft genome assembly. Additionally, we have sequenced and assembled the transcriptome from whole worms and sorted neoblasts. As an additional layer to the transcriptome, we have sequenced the small RNA populations from whole worms and sorted neoblasts. Finally, we have generated a *Macrostomum* BAC library consisting of 60,000 20Kb BACs and 40,000 60Kb BACs. The BAC backbone used has been optimized for genomic recombineering and the library should prove useful in the development of transgenic worms. Taken together, these advances should prove an important resource for the *Macrostomum* community.

Macrostomum neoblasts express PIWI proteins, which seem to be crucial for the worms' regenerative properties. PIWI proteins together with their associated PIWI-interacting-RNAs (piRNAs) are responsible for preventing the mobilization of transposable elements in the germline in many organisms. Genetic ablation of PIWIs or piRNAs in these organisms leads to uncontrolled transposition and sterility. In *Macrostomum* PIWI proteins appear to play a role outside of their normal germline context. We are interested in studying PIWI proteins in *Macrostomum* to understand their role in this stem-cell specific context. Towards this goal, we have generated antibodies against 4 presumptive PIWI proteins in *Macrostomum* (MacPIWIs). These antibodies can be used for immunofluorescence, western blot and immunoprecipitaiton. Using these antibodies, we have observed that MacPIWI1/2 associate with a class of presumptive piRNAs (29-31nt long, 1U bias). We are currently characterizing this piRNA population as well as other small RNA populations.

Abstracts

Talk: Macrostomum lignano genomic resources

Eugene Berezikov

European Research Institute for the Biology of Ageing, Groningen, The Netherlands

The progress towards integrating transcriptome and genome information and development of genome editing tools for *M. lignano* will be presented.

Talk: Heads or tails: Polarity in Planarians

Jochen Rink

Max Plank Institute of Molecular Cell Biology and Genetics Dresden, Germany

The regenerative abilities of planarians present body plan morphogenesis from arbitrary starting points, thus providing a unique experimental system for studying the mechanisms that shape and proportion tissues. Our lab has so far concentrated on the mechanisms patterning the planarian A/P-axis. The striking RNAi phenotypes of beta-Catenin-1/Canonical Wnt signaling pathway components clearly implicate this pathway in A/P-axis patterning, but whether beta-Catenin acts directly as cell fate determinant or indirectly via the selection of other patterning cues has remained unclear. We have examined gene expression patterns under inhibition or overactivation of the pathway and we succeeded in directly measuring beta-Catenin activity along the planarian A/P-axis. Our findings suggest that Wnt/beta-Catenin signaling directly patterns the tail half of planarians, but that a second, currently unknown patterning system, specifies cell identities in the head region. A further conceptual problem is the coordination between beta-Catenin activity in preexisting tissues with the likewise beta-Catenin dependent choice of blastema identity during regeneration. The fact that tissue fragments always regenerate along their original A/P axis further demonstrates that this process involves polarity cues from the preexisting tissues. Our preliminary analysis of cilia polarity in the epithelium further reveals that global body plan polarity feeds back on local tissue polarity. We are currently trying to elucidate the molecular mechanisms that are at the core of the self-organizing properties of the planarian coordinate system.

Abstracts

Poster: M. lignano projects in brief: TALENS, stem cells, gene complexity, antibodies

Belinda Artes[1], Frederic Pacho[2], Sabrina Folie[1], Giada Carta[1], Constanze Ettl[1], Peter Ladurner[1]

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We (BA, FP, PL) are attempting to generate a non-adhesion phenotype by knock-out of the *Macrostomum lignano intermediate filament gene 1 (macif1)*, a gene known to be expressed in the anchor cells. Previous studies have shown that this gene is essential for *M. lignano* adhesion. TALENS were generated containing a restriction enzyme cutting site at the proposed TALENS cutting region. *Macif1* TALENs mRNA was produced and injected into 150 one-cell stage eggs. After hatching, the posterior end was amputated and the tails of 10 animals were pooled. PCR was performed using the lysate as PCR template. So far no TALEN-induced modifications were found in 80 animals.

In the next project we (SF, PL) are aiming to isolate stem cells of *M. lignano* by disintegration into single cells. Various protocols to produce live single cell suspensions are currently tested and culture media are being evaluated for short term maintenance. Different combinations of nuclear and cytoplasmic labeling are tested to adapt for the available lasers at the in-house FACS machine. It is the goal to isolate stem cell pools and characterize them using immunohistochemical, ultrastructural, and transcriptomic approaches.

In order to obtain full length adhesion candidate genes we (GC, PL) try to isolate the full ORF of the genes RNA815_13121.1, RNA815_13121.2, RNA815_13121.3. Three primer pairs were used to amplify fragments form cDNA. 24 clones were sequenced and revealed additional variations that were not present in the transcriptomes. However, apparently the hypothetical genes RNA815_13121.2, RNA815_13121.3 collapse to one gene, a situation also suggested by the newly available transcriptome. RACE experiments were performed but did so far not yield to elongation of the genes.

In the next project we plan to generate polyclonal antibodies against adhesion candidates. We have produced antibodies against six proteins using 15AA peptides for immunization. The antisera were tested by immunohistochemistry. Initially, by conventional PFA fixation no staining was observed. Antigen retrieval and various fixatives were applied. Finally, one the antibodies generated against the intermediate filament gene 1worked well using antigen retrieval.

Poster: The flatworm *Macrostomum lignano* is a powerful model for stem cell and ageing research

Magda. Grudniewska, Stijn Mouton and Eugene Berezikov

European Research Institute for the Biology of Ageing, University Medical Center Groningen, Groningen, The Netherlands

Determining the causes of ageing still remains one of the central questions in biology. To answer this question, different model systems are being used. One of the emerging models is the free-living flatworm *Macrostomum lignano*. This species is convenient for *in vivo* research of how stem cells are regulated to replace missing, damaged, and aged tissues, as it has a pluripotent population of stem cells, called neoblasts. Despite the high regenerative capacity and high cellular turnover during homeostasis, *M. lignano* ages gradually and has a 90^{th} percentile lifespan of about one year. Interestingly, repeatedly regenerated animals were observed to have a longer lifespan than non-regenerated individuals. This led to the hypothesis that regeneration in *M. lignano* induces rejuvenation.
To screen which pathways play important roles in stem cell ageing and rejuvenation we will compare the libraries of aged non-regenerated and aged regenerated (and thus potentially rejuvenated) worms.
This poster will introduce *Macrostomum lignano* as a new model organism and illustrate how it can be used by presenting ongoing projects.

Poster: Nitric oxide: a modulator of the hypoxic response in *Macrostomum lignano?*

Georgina A. Rivera-Ingraham*, U. Bickmeyer, D. Abele

Alfred Wegener Institute Helmholtz Centre for Polar and Marine Research. Dept. Functional Ecology. Am Handelshafen 12, 27570 Bremerhaven, Germany.

Nitric oxide (NO) is an important messenger molecule and physiological modulator not only in mammals but also in invertebrates. Presence of nitric oxide synthase (NOS) has been reported for parasitic and free—living flatworms, evidencing that NO pertains to an evolutionary ancient signaling mechanism. All the available information on NO formation in flatworms comes from neuronal studies. However, NO has a wide variety of roles in cellular physiology. When studying the physiological adaptations of organisms to environmental hypoxia, the interaction of NO with cytochrome C oxidase (CytOX, the final oxygen acceptor of the mitochondrial electron transport chain) is of special interest, since NO is able to compete with O_2 for the CytOX binding site, altering respiration rates. We hypothesized that intertidal flatworms such as *Macrostomum lignano*, which must cope with variable environmental oxygenation in sedimentary habitats, may be using NO as a respiratory modulator to adjust to variable hypoxic conditions. To test this hypothesis, we carried out respiration measurements in the presence of L-NAME (a NOS inhibitor) and spermineNONOate (an NO donor) and using batches of 20 individuals per well of a glass microtiter plate, equipped with optical oxygen sensor spots (Presens, Regensburg, Germany). Under normoxic conditions, between 21 kPa and 18 kPa pO_2, *M. lignano* respires oxyconformingly, i.e. in a pO_2-dependent manner. Below 18 kPa, the flatworms switch to oxyregulation and keep respiration rates constant at 0.027 ± 0.002 nmol $O_2 \cdot h^{-1} \cdot ind^{-1}$ down to 3 kPa pO_2 (lower critical pO_2) (p_c), suggestive of a capacity for metabolic regulation in hypoxia. Application of SpNONOate abolished the oxyregulatory capacity in *M. lignano*, and respiration rates decreased linearly with decreasing pO_2. Contrary, L-NAME, changed the respiratory behaviour of *M. lignano* by shifting its lower p_c from 3 kPa to ≈1 kPa. Animals stained with the NO-specific dye DAF-2AM to investigate NO production with confocal microscopy showed a significant increase of NO formation in near anoxic conditions. Our results suggest that NO formation is mediating the respiratory response of *M. lignano* under conditions of severe hypoxia, and that NOS may be active below 3 kPa. NO production by NOS would decrease the worms respiration rates in an oxyconforming manner below 3 kPa, inducing a state of metabolic slow down. Without the mediation of NO, *M. lignano* respiration continues undiminished down to as low as 1 kPa pO_2 which would cause more rapid depletion of the hypoxic oxygen reserve. Below 1 kPa respiration rates decline also in the absence of NO, indicating that the worms have reached their anaerobic limit of respiration. The role of NOS in these worms is thus to reduce respiratory oxygen uptake and prolong survival under sever hypoxia.

Poster: A potential Toll-like receptor of *Macrostomum lignano*

Stella S. Schukies, S. V. Sperstad & M. Leippe

Zoological Institute, Zoophysiology, University of Kiel, Germany

Toll-like receptors (TLRs) are known to be involved in central biological processes throughout the animal kingdom. Nevertheless, their functional origin remains uncertain and ancestry in development, innate immunity, and cell adhesion are currently under consideration. To reconstruct their path of evolution and to allow inference on their original functionality, additional data are needed especially within the lophotrochozoan lineage. Previously, TLRs of annelids and molluscs were reported to be involved in pathogen recognition; however, until now, a TLR has not been identified among the Platyhelminthes.
Here, we present the first potential TLR within the group of flatworms: *M. lignano* possesses a single TLR of which the presence of a transcript has been verified and localised to the gut. A transcriptional monitoring throughout different developmental stages and various immune challenges, complemented by an RNA interference-based knockdown, are undertaken to elucidate its role in the development and/or the immune system of the worm.

S. Schukies is a recipient of a stipend of the International Max-Planck Research School for Evolutionary Biology.

Poster: Evolutionary conserved immune effector proteins in *Macrostomum lignano*

Sigmund V. Sperstad and Matthias Leippe

Zoological Institute, Zoophysiology, University of Kiel, Germany

Multicellular organisms live in close proximity to a vast diversity of microorganisms. Invertebrates rely on their innate immune responses for protection against pathogens and have developed mechanisms both for utilization of and protection against microbes. They depend on microorganisms as a food source, while they also have to avoid pathogens. Among the evolutionary conserved immune defense molecules are the humoral effector families/superfamilies that are widely distributed in the animal kingdom. Three such families are the lysozymes, the membrane-attack-complex-perforins (MACPFs), and the saposin-like-proteins (SAPLIPs). Lysozymes have been shown to have a role in both immunity and digestion, while the functions of MACPF proteins include immune defense and development. The SAPLIPs are lipid-binding and membrane-interacting proteins, and some members have been shown to participate in antimicrobial defense. We have surveyed the EST database and the genome of *M. lignano*, and here, we give an overview on the members of these three protein families. Moreover, transcriptional profiling of the aforementioned genes upon immune challenge will be presented.

Poster: Cryopreservation of free-living flatworm *Macrostomum lignano*

Valeria Vavilova[1], Irina Sormacheva[1], Evgeniy Brusentsev[1], Sergei Amstislavsky[1], Eugene Berezikov[2] and Alexandr Blinov[1].

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Cryopreservation is the perspective method for the maintaining of mutant, transgenic, and wild-type lines of different model organisms. Development of a reliable, high-throughput and standardized cryopreservation methods for storage of the new model organism *Macrostomum lignano* is an actual problem. We investigated the feasibility of cryopreservation of *M. lignano* embryos and whole worms at the different developmental stages by vitrification and slow freezing. Initially we examined embryo's membrane permeability for various substances. The penetrative cryoprotectants (DMSO, ethylene glicol (EG), MeOH, glycerol (Gly)) in concentrations from 5% to 25% and its combinations with non-penetrative cryoprotectants (sucrose, trehalose) were used for cryopreservation trials. Embryos and whole worms toxicity tests were performed for all combinations of cryoprotectants and the lethal concentrations were determined. The survival of the embryos was assessed by their ability to hatch, while the survival of worms was determined by the ability to move, feed, and produce viable offspring. Different cryopreservation methods were used with *M. lignano* embryos and whole worms on different developmental stages using cryovials and straws. The ongoing research will hopefully allow us to develop the safe and reliable method for maintaining different *M. lignano* lines for future biomedical studies.

Poster: Towards transgenesis in *Macrostomum lignano*

Jakub J. Wudarski, Daniel Olivieri, Philipp M. Weissert, Eugene Berezikov

European Research Institute for the Biology of Ageing, University Medical Center Groningen, Groningen, The Netherlands

Generating transgenic animals is a very important step towards establishing a species as a good model organism. We have undertaken the challenge of developing *M. lignano* transgenesis techniques. One prerequisite for many transgenesis techniques in *M. lignano* is the ability to successfully inject one-cell stage eggs with high success rate. Here, we show advances in optimizing one-cell stage microinjection techniques that allow us to successfully inject *M. lignano* eggs. We also present an outlook on the transgenesis techniques we are currently adapting to use in *M. lignano*.

Talk: Functional and evolutionary genetics of seminal fluid in *Macrostomum*

Steven Ramm

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In addition to sperm, the male ejaculate typically contains a complex suite of other components that are transferred to the female reproductive tract at the time of mating. These ejaculate components are known collectively as seminal fluid, and are produced in the prostate and other accessory reproductive glands. Because seminal fluid-mediated effects on female reproductive physiology and behaviour could have profound consequences for determining male reproductive success, the study of seminal fluid has become a central concern of sexual selection and sexual conflict research. Nevertheless, despite the presumed importance of seminal fluid in the fields of reproductive and evolutionary biology, there are currently very few experimental systems where it has been feasible to systematically study seminal fluid function and evolution. In this presentation I will introduce a new multi-disciplinary research project that aims to investigate the functional and evolutionary genetics of seminal fluid proteins in the free-living, simultaneously hermaphroditic flatworm genus *Macrostomum*. The programme aims to test the working hypothesis that post-mating sexual selection is a major force shaping seminal fluid function and evolution, and to test unique predictions about seminal fluid that apply to simultaneous hermaphrodites. I will describe preliminary results from ongoing collaborations with the Schärer, Ladurner and Berezikov labs that help to define the scope of the project, and set out our planned goals and methodology.

Abstracts

Talk: Fundamentals of biological adhesion in *Macrostomum lignano*

Birgit Lengerer[1], Robert Pjeta[1], Julia Wunderer[1], Marcelo Rodrigues[1], Lukas Schärer[2], Eugene Berezikov[3], Michael Hess[4], Kristian Pfaller[4], Bernhard Egger[5], Willi Salvenmoser[1], Peter Ladurner[1,*]

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Free-living flatworms, in both marine and freshwater environments, are able to adhere to and release from a substrate several times within a second. However, nothing is currently known about the molecules that are involved in this adhesion process. We are investigating *Macrostomum lignano* in order to identify its adhesive- and releasing- related factors. We analysed in detail the morphology of its adhesive organs using light-, confocal- and electron microscopy. Each organ consists of strictly three different cell types, two gland cells and one modified epidermal cell. One gland cell is supposed to produce and secrete glue and the second gland cell presumably expels a substance to release from the attachment. The gland necks of both secretory cells run parallel toward one modified epidermal cell, through which they expel their vesicles. In a whole mount in situ hybridization screen we identified a gene encoding for an intermediate filament protein which was found to be essential for adhesion. We refer to this gene as macif1 (Macrostomum intermediate filament). RNA interference mediated knock-down resulted in the first experimentally induced non-adhesion phenotype of a marine animal. Specifically, the absence of intermediate filaments in the anchor cells led to papillae with open tips, a reduction of the cytoskeleton network, a decline in hemidesmosomal connections, and to shortened microvilli containing less actin. For further developmental analysis a polyclonal antibody against *macif1* was produced. Due to their simplicity and fast

regeneration time, the adhesive organs of *M. lignano* provide an ideal system to analyse de novo organ regeneration. We now try to unravel the question if the three interacting cells emerge together and migrate to their final position, or if they develop separately and connect in later stages of differentiation. Therefore, we analyse their morphology during regeneration and are currently producing comparative transcriptoms at different stages of regeneration.

Supported by FWF grant P25404-B25 and COST action TD0906

Talk: Posterior end specific genes in *Macrostomum lignano*

Robert Pjeta[1], Julia Wunderer[1], Birgit Lengerer[1], Marcelo Rodrigues[1], Carta Giada[1], Willi Salvenmoser[1], Roberto Arbore[2], Eugene Berezikov[3], Lukas Schärer[2], Peter Ladurner[1]

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In *Macrostomum lignano* many genes are exclusively expressed in the tail, as organs of the copulatory and adhesive system are located only in this region. The main focus of our research is the identification and characterization of adhesion related genes. The adhesive system of *Macrostomum lignano* allows the animals to repeatedly adhere and release from the substrate and it is comprised of about 130 adhesive organs. Each of them is composed of two gland cells which are secreting the adhesive- and the releasing factor respectively, and one modified epidermal cell, the anchor cell.

In order to identify genes related to the adhesive system as well as other posterior end specific genes, we performed a whole-mount *in situ* hybridization screen. Using data from a region specific transcriptome, about 400 genes were identified to be tail specific. We selected 240 genes for our screen, based on several parameters like sequence length and fold change. So far, we were able to identify distinct expression patterns for 163 genes. The majority was related to the reproductive system and comprised prostate (39 %) - and antrum specific glands (15 %). We were also able to detect 24 genes which had expression in the area of the adhesive organs. We are currently analyzing their function by RNA interference.

A set of 3 adhesion candidate genes was found, that showed a highly comparable expression pattern. All these genes showed a hydrophilic amino acid repeat region. Interestingly, at least two genes are located in close proximity within a scaffold of the genome assembly.

It is our goal to analyze the full complement of posterior end specific genes and isolate the full open reading frame of all adhesion candidate genes for antibody production and further analysis.

Talk: Studying ageing and rejuvenation in *Macrostomum lignano*

Stijn Mouton, Magda Grudniewska, Eugene Berezikov

European Research Institute for the Biology of Ageing, Groningen, The Netherlands

Determining the causes of ageing remains one of the central questions in biology. To answer this question, different model systems have been used. All have their advantages and disadvantages and allow focusing on specific aspects of the ageing process. One of the emerging models is the free-living flatworm *Macrostomum lignano*. This species is convenient for *in vivo* research of how stem cells are regulated to replace missing, damaged, and aged tissues as it has a pluripotent population of mesodermal stem cells. Despite the high regenerative capacity and high cellular turnover during homeostasis, *M. lignano* ages gradually and has a 90^{th} percentile lifespan of about one year. Interestingly, repeatedly regenerated animals were observed to have a longer lifespan than non-regenerated individuals, leading to the hypothesis that regeneration in *M. lignano* induces rejuvenation.

During the last months, we set up an ageing project with the aim to determine a gene-expression ageing profile and the effect of regeneration on it. To do so, we are performing RNA-Seq of non-regenerated, single regenerated, and multiple regenerated worms of different ages. In this talk, we will present the project, show the first results, and discuss the potential of *M. lignano* as a new ageing model.

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Meeting information

Venue

The meeting will be held at the conference room of the **European Research Institute for the Biology of Ageing (ERIBA)**. The welcome reception, coffee breaks and poster sessions will be organized in the ERIBA atrium. A joint dinner in the city center will be held on Saturday night (Restaurant Ni Hao, Gedempte Kattendiep 122).

Address

ERIBA – UMCG
Antonius Deusinglaan 1
Building 3226
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Transport

Train

Groningen is best accessible by train. Please use the Journey planner at http://www.ns.nl/en to plan your trip.

Air

Amsterdam Schiphol airport is the closest to Groningen large airport. There are direct trains from Schiphol to Groningen, the journey time is two hours (see Journey planner at http://www.ns.nl/en).

Car

As soon as you enter Groningen, please follow the road signs to the UMCG. Once you have left the ring road and crossed the bridge over the Eemskanaal, choose at the traffic lights direction UMCG Noord. (You will pass the UMCG main entrance on your left hand side). From here, continue to follow UMCG Noord. When driving along the Petrus Campersingel, turn left at the traffic lights to the Vrydemalaan. After approx. 150 meters turn left to enter the UMCG premises. Follow the signs to car park Noord.

Local transport

ERIBA is located close to the Groningen city center. From Groningen Central Station you can either take a taxi to the UMCG or travel by bus 3 (to Lewenborg) or bus 6 (to Beijum). The bus and taxi will stop at Bloemsingel (UMCG Noord).