Deutsche Gesellschaft für Parasitologie

Fraunhofer IME

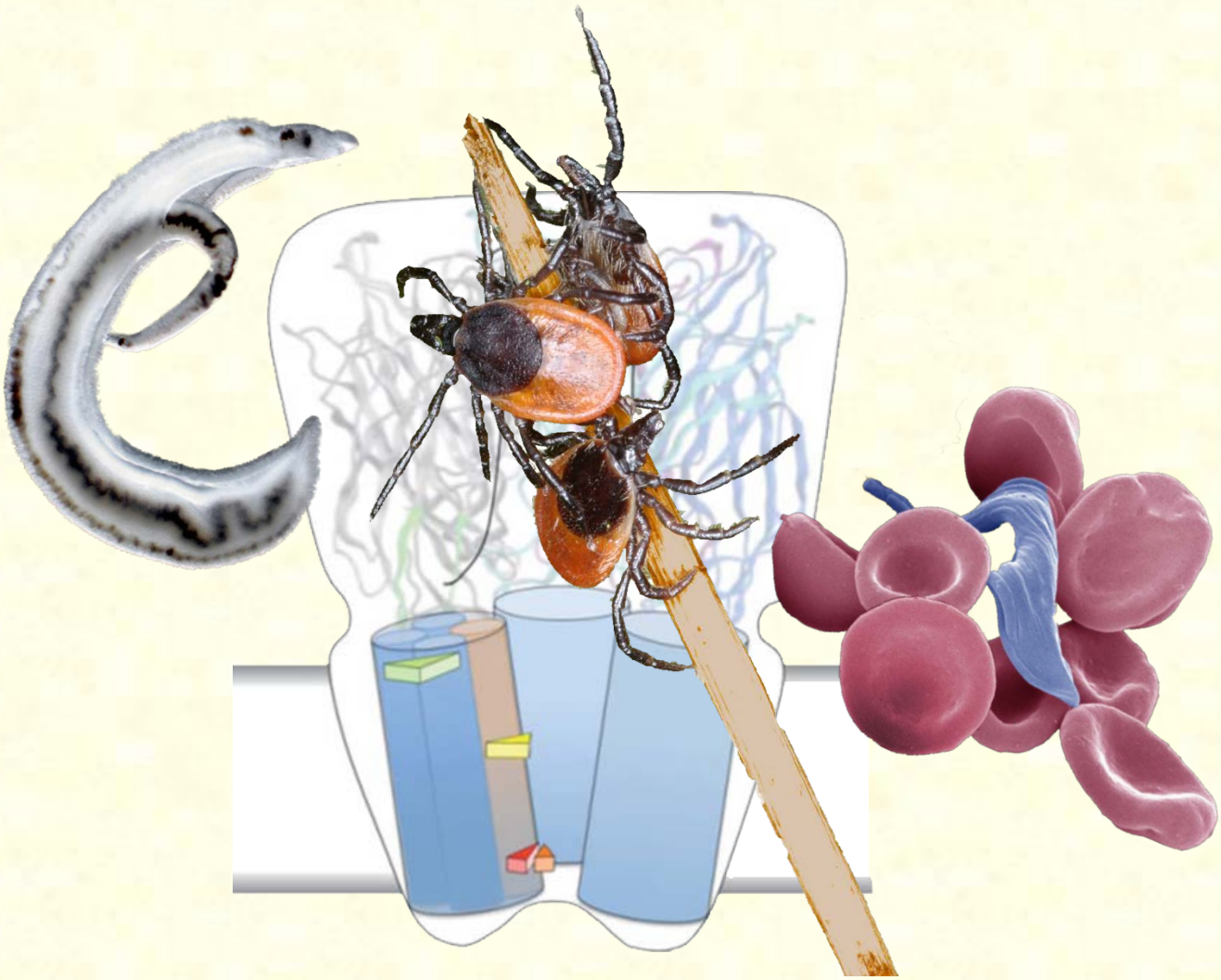
18th Drug Design & Development Seminar (DDDS)

of the German Society for Parasitology (DGP)

March 30th – 31st, 2017

Forschungszentrum Borstel
Leibniz-Zentrum für Medizin und Biowissenschaften

Parkallee 1-40
23845 Borstel, Germany



Sponsors:

Boehringer Ingelheim

JUNG-Stiftung für Wissenschaft und Forschung

MERCK

Cover legend

From left to right:

Schistosoma mansoni – courtesy of Conor Caffrey, Skaggs School of Pharmacy and Pharmaceutical Sciences, UCSD, San Diego, USA

Ixodes ricinus - courtesy of Jan Erhart and Petr Kopáček, Institute of Parasitology, BC CAS, Prague, Czech Republic

Trypanosoma brucei among red blood cells – courtesy of Michael Duszenko, Interfaculty Institute of Biochemistry, University of Tuebingen, Germany

In the back: schematic representation of the CysLGCC sectional view - courtesy of Tina Weber, Merck Darmstadt & Paul M. Selzer, Boehringer Ingelheim Animal Health GmbH, Ingelheim, Germany

Hosted by: Fraunhofer IME-SP, Hamburg

Scientific Committee:

Dr. Sheraz Gul
Fraunhofer IME-SP
Hamburg

Dr. Helmut Haas
*helmin*Guard
Sülfeld/Borstel

Prof. Dr. Paul M. Selzer
Boehringer Ingelheim Animal Health GmbH
Ingelheim

Dr. Sandra Noack
Boehringer Ingelheim Animal Health GmbH
Ingelheim

About the Drug Design & Development Seminar (DDDS)

The Drug Design & Development Seminar (DDDS) was founded in 1999 as an active working group of the German Society for Parasitology, by Prof. Dr. Peter Köhler (University of Zürich, CH), Prof. Dr. Rolf Walter (BNI, Hamburg, DE), and Prof. Dr. Heiner Schirmer (Uniersity of Heidelberg, DE). Since 2004, Prof. Dr. Paul M. Selzer (Boehringer Ingelheim Animal Health GmbH, Ingelheim, DE) is the sole coordinator of the DDDS, transferring the meeting into an international well recognized scientific forum. Exchange of scientific information about anti-parasitic chemotherapy between universities, industry, and other research organizations continues to be important to accelerate anti-parasitic drug development. The DDDS is open to all scientists and professionals interested in the field of anti-parasitic research. The DDDS aims at connecting human and veterinary health by complementary approaches in medical and veterinary parasitology and medicinal chemistry to stimulate One-Health approaches to combat parasitic diseases. The main topics include but are not limited to:

- Target identification and validation
- Identification of modulators
- Synthesis and optimization of lead compounds towards marketable drugs
- Delivery of active compounds to infected hosts

Venue

Forschungszentrum Borstel
Leibniz-Zentrum für Medizin und Biowissenschaften

Parkallee 1-40
23845 Borstel (close to Hamburg)
http://www.fz-borstel.de

How to get there

By car via Hamburg:

Head for Norderstedt and take the main road B432 towards Bad Segeberg until the turn-off Borstel (approximately 28 km). Then turn to the right. The Center lies on your right hand side.

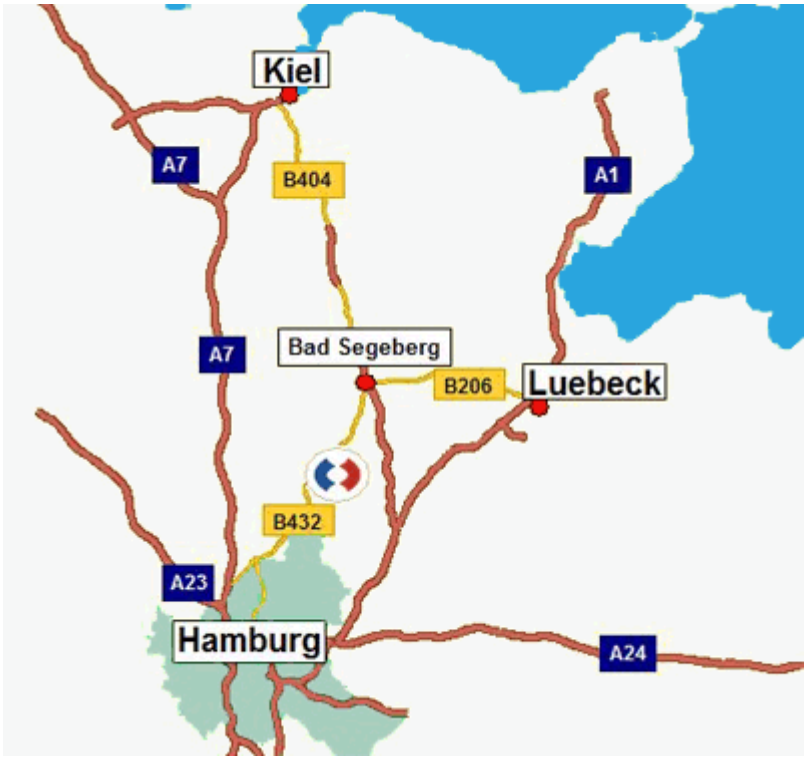
For navigation systems: please enter 'Sülfeld (Borstel)'

By train:

Destination Bad Oldesloe. It is advisable to take a taxi from Bad Oldesloe to the Research Center Borstel (approximately 15 Euro).

By plane:

Destination Hamburg. Take the bus to the underground stop U1, then the underground U1 until the stop Ochsenzoll. From Ochsenzoll by bus/line 7550 towards Bad Segeberg until the bus stop "Abzweig Borstel". The Center lies on your right hand side.



Attention:

There will be a complimentary bus shuttle from hotels “Schloß Tremsbüttel” and TRYP Bad Oldesloe to the Borstel venue. For shuttle schedule, see agenda. A bus shuttle will also be provided after the seminar to the Hamburg airport, from where the main railway station is easily accessible via public transportation (S1).

Registration desk opening hours

Wednesday, 29.03.2017:	17:00 – 20:00	Hotel TRYP Bad Oldesloe
Thursday, 30.03.2017:	8:00 – 18:00	Forschungszentrum Borstel
Friday, 31.03.2017:	8:00 – 9:00	Forschungszentrum Borstel

Wednesday, 29.03.2017		
17:00	20:00	Registration
18:00	20:00	Welcome Reception at the hotel TRYP by Wyndham Bad Oldesloe

Thursday, 30.03.2017		**Name**	**Title**	**Institution**
08:00		Bus shuttle from hotels Schloß Tremsbüttel & TRYP Bad Oldesloe		
08:00		Registration & Poster set-up		
09:00	09:20	Welcome and Introduction		
09:20	10:00	Keynote	Loïc Le Hir de Fallois	Antiparasitic drug discovery and research in Animal Health: Overview & development of Afoxolaner isoxazoline	Boehringer Ingelheim Animal Health Duluth, US
10:00	12:50	Short Talks (incl. break)		
12:50	14:15	Lunch break & Poster session		
14:15	14:55	Keynote	Collette Britton	Parasitic helminth small RNAs: regulators of development, host-parasite interactions and as potential therapeutic targets	University of Glasgow, UK
14:55	16:15	Short Talks		
16:15	16:45	Coffee break & Poster session		
16:45	17:25	Keynote	Georg von Samson-Himmelstjerna	More complicated than imagined - what mechanisms lead to benzimidazole resistance in helminthes?	Freie Universität Berlin, DE
17:25	18:45	Short Talks		
18:45	18:55	Wrap-up		
19:00	21:30	Dinner & Poster session		
21:30		Bus shuttle to hotels TRYP Bad Oldesloe & Schloß Tremsbüttel		

Friday, 31.03.2017		**Name**	**Title**	**Institution**
08:00		Bus shuttle from hotels Schloß Tremsbüttel & TRYP Bad Oldesloe		
09:00	09:40	Keynote	Jeremy C. Mottram	Targeting cell signalling and proteolysis for trypanosomatid drug discovery	University of York, UK
09:40	11:20	Short Talks		
11:20	11:50	Coffee break & Poster session		
11:50	12:30	Keynote	Fiona Tomley	Coccidiosis in poultry: challenges for disease control in today's global industry	University of London, UK
12:30	13:50	Short Talks		
13:50	14:00	Wrap-up & Closing		
14:00	14:30	Boxed Lunch		
14:30		Bus shuttle to Hamburg Airport – connection to S1 to main railway station		

Wednesday, 29.03.2017			
17:00	20:00	Registration	at TRYP by Wyndham Bad Oldesloe
18:00	20:00	Welcome Reception	at TRYP by Wyndham Bad Oldesloe

Thursday, 30.03.2017		**Name**	**Title**	**Institution**
08:00	09:00	Registration & Poster set-up		
09:00	09:20	Welcome and Introduction	Stefan Ehlers, CEO, Research Center Borstel Carsten Claussen, Department Head, Fraunhofer IME-ScreeningPort Paul Selzer, Head of Antiparasitics R&D, Boehringer Ingelheim AH	
09:20	10:00	Keynote	Loïc Le Hir de Fallois	Antiparasitic drug discovery and research in Animal Health: Overview & development of Afoxolaner isoxazoline	Boehringer Ingelheim Animal Health, Duluth, US
10:00	10:20	Session Chair: Heinz Sager, Elanco Animal Health	Jan Perner	Molecular targets to impair blood meal processing in ticks	Czech Academy of Sciences, České Budějovice, CZ
10:20	10:40		Christoph Krull	Artificial tick feeding and its potential use in acaricide efficacy screening	Freie Universität Berlin, DE
10:40	11:00		Sandra Schorderet-Weber	Blocking Transmission of Vector-Borne Diseases	Neuchâtel, CH
11:00	11:30	Coffee break		
11:30	11:50	Session Chair: Helmut Haas, *helmin*Guard	Koen Dechering	Barcoded live mosquito screens for high throughput discovery of compounds that interrupt transmission of malaria	TropIQ Health Sciences, Nijmegen, NL
11:50	12:10		Andrew Hemphill	*In vitro* screening of the open source MMV pathogen boxes reveals novel compounds with profound activities against *Neospora caninum* infection	University of Bern, CH
12:10	12:30		Manu De Rycker	Discovery and lead optimisation of a promising new antileishmanial compound series within an academic-industry partnership	Drug Discovery Unit Dundee, UK
12:30	12:50		Theodora Calogeropoulou	Heteroaryl- and aryl-substituted miltefosine derivatives	National Hellenic Research Foundation Athens, GR
12:50	14:15	Lunch break & Poster session		

Thursday, 30.03.2017			Name	Title	Institution
14:15	14:55	Keynote	Collette Britton	Parasitic helminth small RNAs: regulators of development, host-parasite interactions and as potential therapeutic targets	University of Glasgow, UK
14:55	15:15	Session Chair: Manu De Rycker, Drug Discovery Unit Dundee	Helmut Haas	Engaging worms for health - drug discovery on live schistosomes	*helmin*Guard, Sülfeld/ Borstel, DE
15:15	15:35		Christoph G. Grevelding	BACADs and AASs: different concepts, novel compounds, common anti-schistosomal activities	Justus Liebig University Gießen, DE
15:35	15:55		Jacob Golenser	Inhibition of *Schistosoma mansoni* development in mice by slow release of artemisone	Hebrew University of Jerusalem, IL
15:55	16:15		Gehan Labib Elenain	Etanercept: Initiative Study on the drug potentiality in murine *Schistosomiasis mansoni*	Abu Dhabi University, AE
16:15	16:45	Coffee break			
16:45	17:25	Keynote	Georg von Samson-Himmelstjerna	More complicated than imagined - what mechanisms lead to benzimidazole resistance in helminthes?	Freie Universität Berlin, DE
17:25	17:45	Session Chair: Ronald Kaminsky, para.C Consulting	Heinz Sager	Drug Resistance in the Cattle Tick *Rhipicephalus (Boophilus) microplus* – Monitoring Tools and search for new Compounds	Elanco Animal Health, Basel, CH
17:45	18:05		Pasquale Linciano	Fragment-based approach in the discovery of highly active antiparasitic PTR1 Inhibitors	University of Modena and Reggio Emilia, IT
18:05	18:25		Oliver Koch	*Mycobacterium tuberculosis* thioredoxin reductase inhibitors with activity on mycobacterial growth in infected macrophages based on *in silico* molecular design	Dortmund University, DE
18:25	18:45		Martin Folger	Development of an *in vitro* drug release method for a veterinary modified-release dosage form as a tool for quality control	Boehringer Ingelheim Animal Health, Ingelheim, DE
18:45	18:55	Wrap-up			
19:00	21:30	Dinner & Poster session			

Scientific Program

Friday, 31.03.2017			Name	Title	Institution
09:00	09:40	Keynote	Jeremy C. Mottram	Targeting cell signalling and proteolysis for trypanosomatid drug discovery	University of York, UK
09:40	10:00	Session Chair: Michael Boshart, LMU München	Jurgen R. Haanstra	Targeting pathogen metabolism without collateral damage to the host	University of Groningen & University Amsterdam, NL
10:00	10:20		Sabine Bachmaier	A cyclic AMP-independent PKA from *Trypanosoma brucei* as promising drug target	LMU München, DE
10:20	10:40		Alec O'Keeffe	The effect of media perfusion on the infection and drug activity against *Leishmania major*	London School of Hygiene & Tropical Medicine, UK
10:40	11:00		Daria Monaldi	Antiparasitic drug discovery in Epigenetics	University of Freiburg, DE
11:00	11:20		Asaad Khalid	Discovery of natural bioactive compounds: The Road to El Dorado	Jazan University, SA
11:20	11:50	Coffee break			
11:50	12:30	Keynote	Fiona Tomley	Coccidiosis in poultry: challenges for disease control in today's global industry	University of London, UK
12:30	12:50	Session Chair: Sheraz Gul, Fraunhofer IME-SP	Julien Furrer	Ruthenium complexes for the treatment of protozoan diseases	University of Bern, CH
12:50	13:10		Kristina Haeussler	The pentose phosphate pathway of *Plasmodium* parasites as a drug target	Justus Liebig University Gießen, DE
13:10	13:30		Anita Cohen	Design, synthesis and antimalarial activity of novel bis{N-[(pyrrolo[1,2-a]quinoxalin-4-yl)benzyl]-3-aminopropyl}amine derivatives	Aix-Marseille Université, FR
13:30	13:50		Eva-Maria Schäfer	Ferredoxin-Ferredoxin-NADP+-Reductase – a gem amongst drug-targets	Philipps-University Marburg, DE
13:50	14:00	Wrap-up & Closing	Sheraz Gul, Fraunhofer IME-ScreeningPort Paul Selzer, Boehringer Ingelheim Animal Health Helmut Haas, *helming*uard Sandra Noack, Boehringer Ingelheim Animal Health		
14:00	14:30	Boxed Lunch			

Posters	Name	Title	Institution
P1	Amir Reza Bagheri	Unlocking nanocarriers for the programmed release of the antimalarial artemison	University of Bayreuth, DE
P2	Jacob Golenser	Imaging of experimental cerebral malaria using the liposomal, FDA approved indocyanine green	Hebrew University of Jerusalem, IL
P3	Sheraz Gul	Successful transnational EU funded neglected disease drug discovery projects	Fraunhofer IME-SP Hamburg, DE
P4	Simone Häberlein	Aldehyde dehydrogenase, a potential drug target in *Schistosoma*	Justus Liebig University Gießen, DE
P5	Annette Kaiser	EIF-5A controls apoptotic damage of myocytes in experimental cerebral malaria	University Duisburg-Essen, DE
P6	Eric R. Kalkman	Developing a high content screening assay to identify invasion and proliferation inhibitors of *Toxoplasma gondii*	Wellcome Centre for Molecular Parasitology, University of Glasgow, UK
P7	Tanja Corinna Knaab	3-Hydroxy-N'-arylidenepropanehydrazonamides: Novel analogues of arylamino alcohols cure *Plasmodium berghei*-infected mice after peroral administration	University of Düsseldorf, DE
P8	Maxime Madder	Clinvet South Africa and Morocco: two contract research organisations offering unique opportunities for drug development and research	Clinvet International, ZA

Antiparasitic drug discovery and research in Animal Health: Overview & development of Afoxolaner isoxazoline

Loïc Le Hir de Fallois, Diane Larsen

Merial Inc. now part of Boehringer Ingelheim, Duluth, GA-USA

E-mail: loic.lehir@merial.com

After a general overview of Antiparasitic Drug Discovery in Animal Health and its associated Research and Development process, we will review the work that led to the discovery and development of Afoxolaner isoxazoline used in NexGard®.

Afoxolaner is a new antiparasitic isoxazoline compound characterized by a good safety profile and extended effectiveness against fleas and ticks on dogs following a single oral administration [1]. The isoxazoline starting point seemed to have originated from the insecticidal phthalic diamides from Nissan and Nihon Nohyaku and the anthranilic diamides from DuPont [2]. The excellent ectoparasitic activity of the novel isoxazoline chemotype was demonstrated against flea and tick species both in laboratory on whole organism and in dose confirmation in dogs followed by comprehensive field studies [3a, b].

The flea and tick efficacies are directly related to the rapid absorption of afoxolaner and a plasma peak reaching the lethal concentration (LC90) in the blood, around 23 ng/mL for fleas and 100 ng/mL for ticks [4]. Extensive formulation optimization work led to the design of a highly palatable soft chew used in (NEXGARD®).

The speed of kill of afoxolaner against experimental infestations by *Ctenocephalides felis* was evaluated after oral administration of afoxolaner at a dose to achieve 2.5 mg/kg bodyweight. The efficacy of afoxolaner compared to control dogs increased to 99.5% at 8 h post-treatment and 100% at 12 and 24 h post-treatment [5].

[1] Wesley L. Shoop et al., Discovery and mode of action of afoxolaner, a new isoxazoline parasiticide for dogs, Vet. Parasitol. 2014, 201, 179-189

[2] George P. Lahm et al., 4-Azolyphenyl isoxazoline insecticides acting at the GABA gated chloride channel, Bioorg. Med. Chem. Lett., 2013, 23, 3001-3006

[3a] James S. Hunter III et al., Evaluation of the curative and preventive efficacy of a single oral administration of afoxolaner against cat flea *Ctenocephalides felis* infestations on dogs, Vet. Parasitol., 2014, 201, 207–211

[3b] Pascal Dumont, Curative and preventative efficacy of orally administered afoxolaner against dog flea *Ctenocephalides canis* (Curtis, 1826) infestation in dogs. Vet. Parasitol. 2014, 201, 212–215

[4] Laura Letendre et al., The intravenous and oral pharmacokinetics of afoxolaner used as a monthly chewable antiparasitic for dogs. Vet. Parasitlol. 2014, 201, 190–197

[5] B.N. Kunkle et al., Assessment of the onset of action of afoxolaner against existing adult flea (*Ctenocephalides felis*) infestations on dogs, Vet. Parasitol. 2014, 201, 204–206

Parasitic helminth small RNAs: regulators of development, host-parasite interactions and as potential therapeutic targets

Collette Britton, Alan Winter, Neil Marks, Henry Gu, Kirsty Maitland, Victoria Gillan, Eileen Devaney

Institute of Biodiversity, Animal Health and Comparative Medicine, College of Medical, Veterinary and Life Sciences, University of Glasgow, Glasgow, UK

E-mail: Collette.Britton@glasgow.ac.uk

The mechanisms regulating development and survival of parasitic helminths within their hosts are not well understood. We are examining microRNAs in parasitic nematodes to investigate their regulatory roles in development and immune modulation. miRNAs are small (22 nucleotide), non-coding RNAs that regulate gene expression post-transcriptionally and are important in controlling cell proliferation and differentiation in diverse organisms. We previously sequenced the miRNAs expressed by the highly pathogenic veterinary nematode *Haemonchus contortus* and the filarial nematode *Brugia pahangi*, which is closely related to filarial nematodes of humans and companion animals (1). Using microarray analysis we have profiled the expression pattern of miRNAs in the parasitic stages of both species. This identified miRNAs significantly enriched in the L3 stages pre- and post-infection, suggesting roles in regulating larval arrest and activation in response to host signals (2). Mimics and inhibitors of differentially expressed miRNAs are being tested for their effects on interfering with nematode development. miRNAs are also released in the excretory-secretory (ES) products of *H. contortus* and have been sequenced from both the ES supernatant and ES microvesicles. Secreted parasite miRNAs can be detected in gut tissue from *H. contortus* infected sheep and we speculate that these may modulate immune outcome. Our results are important in understanding parasite development and host-parasite interactions and highlight the potential of miRNAs and the pathways they regulate as novel drug targets for parasite control.

[1] Winter, A.D. et al., 2012. BMC Genomics 13: 4
[2] Winter, A.D. et al., 2015. BMC Genomics 16: 331

More complicated than imagined - what mechanisms lead to benzimidazole resistance in helminths?

Georg von Samson-Himmelstjerna and Jürgen Krücken

Institute for Parasitology and Tropical Veterinary Medicine, Freie Universität Berlin, Germany

E-mail: gvsamson@fu-berlin.de

Only since the start of the area of broad spectrum anthelmintics in the 1960s safe, affordable and effective treatment and control of helminth infections has become available. As the first real broad spectrum antiparasiticides, the benzimidazoles (BZ) are in use against gastro-intestinal nematodes (GIN) as well as some cestode and even protozoan parasite species for now more than five decades. The frequent and intensive use of anthelmintics can lead to drug resistance as also known for other antiinfectives. For the BZs this has already been reported during the early 1970s in small ruminant GIN and since then became a widespread problem in this important group of parasites as well as in equine cyathostomins. More recently, BZ resistance has been seen also in cattle GIN [1] and our own recent investigations indicate also a sub optimal response of *Ascaris* sp. in African school children. BZs act by binding with high affinity to the β-tubulins of pathogens which leads to a blocking of microtubule polymerization and due to the ongoing depolymerization thus subsequently to a complete loss of these essential cell structure. By comparison of the β-tubulin isotype 1 coding sequences from BZ-susceptible and –resistant populations of the sheep GIN *Haemonchus contortus* it was found that a single nucleotide polymorphism at the codon 200 leading to the expression of tyrosine instead of phenylalanine correlates with BZ resistance. Subsequently the similar P167Y and an E198A polymorphism were also described to be associated with BZ-resistance in this and other GIN species. In addition to these drug target related BZ resistance mechanisms, subsequently also unspecific resistance mechanisms involving an increased drug efflux mediated by P-glycoprotein transmembrane pumps were encountered. However, it is currently unclear which of the many P-glycoprotein genes of parasitic nematodes are involved in this process. Furthermore, also the modification of cytochrome P450 based drug metabolism was recently suggested to potentially be associated with BZ resistance in nematodes [2]. Noteworthy, parasite species and even population specific differences concerning the relevance of the various potential BZ resistance mechanisms have already been demonstrated. Future research should address the question if certain selectional factors contribute to the evolution of specific resistance mechanism, which fitness costs (if any) are associated with the different resistance mechanisms and if for example combinations of resistance mechanisms also occur.

[1] Bullen et al. Aust Vet J. 2016; 94:35-41

[2] AlGusbi et al. Int. J. Parasitol. 2014; 44:647-58

Targeting cell signalling and proteolysis for trypanosomatid drug discovery

Jeremy C Mottram

Department of Biology, University of York, UK

E-mail: Jeremy.mottram@york.ac.uk

Enzymes that play roles in regulating post-translational modifications of proteins, such as protein kinases (PKs) and peptidases are promising drug targets in kinetoplastid parasitic protozoa. For African trypanosomes, a combination of RNAi and inducible over-expression have proved to be useful tools for investigating essential genes and we have used these to characterise PKs and peptidases from *T. brucei*; however the RNAi pathway has been lost in many *Leishmania* species. We have addressed these limitations by developing an inducible gene knock-out system based on dimerised Cre recombinase (diCre) in *L. mexicana*. The application of this technology to characterise *Leishmania* genes involved in the ubiquitination pathway will be discussed in comparison with in vitro and in vivo kinome-wide RNAi screens in *T. brucei*. Further, I will describe how functional genetics can be applied to mechanism-based phenotypic screening in parasitic protozoa.

Coccidiosis in poultry: Challenges for disease control in the modern poultry industry

Fiona Tomley

Department of Pathobiology and Population Sciences, The Royal Veterinary College, University of London, UK

E-mail: ftomley@rvc.ac.uk

Sustainability and security of food sources is a major priority with the global human population set to exceed nine billion by 2050. Poultry production has increased ~6 fold since the 1950's and will double again by 2050 to keep pace with predicted demand. Rapid and continued expansion of the poultry industry requires the careful balancing of many factors including (1) selective breeding of broiler and layer lines of chickens that massively out-perform earlier counterparts in terms of meat and egg production; (2) the use of high-density, often fully integrated, production systems to maintain efficiency and minimise costs and losses; (3) the effective control of production diseases through biosecurity, husbandry, vaccination and chemoprophylaxis.

Coccidiosis, caused by *Eimeria* parasites, is a significant threat especially to chickens, causing enteritis that results in high morbidity and occasionally high mortality. Control relies mainly on prophylactic in-feed ionophores and chemicals, but drug-resistant parasites are globally ubiquitous and these anticoccidials are no longer totally effective. Thus coccidiosis costs the poultry industry an estimated $3 billion per year, compromising efficient production and bird welfare. Live and live-attenuated vaccines are used successfully in some sectors however there are significant issues with the relative cost and logistics of vaccine production, particularly for the safest live-attenuated vaccines.

Understanding prevalence, diversity, gene flow and population structure in *Eimeria* parasites is invaluable to understand disease epidemiology (including drug resistance and vaccine efficacy) and to predict how future subunit vaccines, based on a small number of immunogenic antigens, will perform in the field. Seven *Eimeria* species are recognised to infect chickens. These all have a global distribution and recent high throughput sequencing and genetic fingerprinting has shown much higher levels of genetic (and to a lesser extent antigenic) polymorphism than previously recognised. In addition, three cryptic Operational Taxonomic units (OTUs x, y and z), originally described in Australia, are now known to be circulating in chicken houses from several major poultry-producing countries of the southern hemisphere. The precise relationship of these OTUs to recognised species, as well as their prevalence, pathology and associated risks remain largely unknown.

Thorough understanding of *Eimeria* pathogenesis and the hosts' response to infection and vaccination chicken also provides knowledge that can be exploited for development of effective, sustainable control across all sectors of meat and egg production. Relevant examples from our ongoing studies include dissection of the genetic basis of natural resistance to *Eimeria* in chickens (with partners at the Roslin Institute), development of new vaccine 'platforms' for the delivery of coccidial antigens to the gut of the chicken, and the study of early events in parasite invasion of host cells leading to further identification of likely therapeutic targets (with partners at the University of Liverpool).

In this keynote talk, I will present data from recent and ongoing studies where we are using genetics, multi-omics, reverse-genetics and bioimaging approaches to address these important challenges.

Molecular targets to impair blood meal processing in ticks

Jan Perner, Daniel Sojka, Radek Šíma, Ondřej Hajdušek, Petr Kopáček

Institute of Parasitology, Biology Centre, Czech Academy of Sciences, České Budějovice, Czech Republic

E-mails: perner@paru.cas.cz, kopajz@paru.cas.cz

Feeding and digestion of host blood are key physiological processes providing essential nutrients for the development and fecundity of ticks. Components of ingested host blood, which exceeds the weight of unfed females by more than one hundred times, are gradually taken up by tick midgut digest cells and intracellularly digested by a multi-enzyme network of acidic aspartic and cysteine endo- and exo-peptidases [1]. Enzymatic hydrolysis of haemoglobin, the major protein component of blood, results in the intracellular release of a vast excess of haem. In most eukaryotic cells, haem and iron homeostasis is based on a balanced flux between haem biosynthesis and degradation. In contrast, ticks are not capable of synthesising or degrading haem [2]. Therefore, ticks have evolved unique molecular mechanisms for haem acquisition and also molecular measures that prevent the cellular toxicity of obtained haem. As haem is not further catabolised in ticks, iron ions are obtained from a non-haem source, likely host serum transferrin. RNA interference and pilot vaccinations were exploited to validate proteinaceous targets in ticks, which participate in host protein hydrolysis and haem or iron metabolism [3]. Utilising a membrane feeding system for ticks, medium-throughput drug screen experiments were initiated.

[1] Sojka, D. et al.: New insights into the machinery of blood digestion by ticks. Trends in Parasitology 29, (2013)

[2] Perner, J. et al.: Acquisition of exogenous haem is essential for tick reproduction. eLife 5, (2016)

[3] Hajdušek, O. et al.: Knockdown of proteins involved in iron metabolism limits tick reproduction and development. Proceedings of the National Academy of Sciences 106, (2009)

Artificial tick feeding and its potential use in acaricide efficacy screening

Christoph Krull, Bettina Böhme, Ard Nijhof

Institute for Parasitology and Tropical Veterinary Medicine, Freie Universität Berlin, Berlin, Germany

E-mail: christoph.krull@fu-berlin.de

The artificial feeding of ticks could reduce the use of experimental animals required for tick feeding and also offer a technique of interest for various applications in research on ticks and tick-borne diseases. However, the complexity of tick feeding behavior and the long duration of their blood meal has impeded the development of a feeding technique which is comparable to natural tick feeding regarding tick attachment, engorgement- and rearing success.

In the 'Optimization and Automation of Artificial Tick Feeding' (OAKS) project financed by the German Federal Ministry for Education and Research (BMBF), efforts are undertaken to optimize different parameters in the artificial feeding of ticks such as environmental conditions and attachment stimuli. It was demonstrated that the *in vitro* feeding and fecundity of adult *Dermacentor reticulatus* ticks could be improved by feeding ticks at 5% CO_2 concentrations and through supplementation of the blood meal with extra glucose (4 g glucose / litre heparinized blood). All life stages of *Ixodes ricinus* could also be fed *in vitro* under these conditions providing successful post-feeding development.

Two pilot experiments performed in triplicate in which blood containing Fipronil (10 ppm) was fed to *Ixodes ricinus* ticks *in vitro* showed that Fipronil killed all ticks within 4 days (1st experiment) or 9 days (2nd experiment). The observed variation in efficacy was associated with differences in the tick attachment rates between the two experiments, with lower attachment and thus decreased exposure to Fipronil in the second experiment. The results demonstrate that artificial tick feeding assays may find use in the efficacy screening of systemic acaricides, but further optimization to obtain consistently high tick attachment rates will be required.

Blocking Transmission of Vector-Borne Diseases

S. Schorderet-Weber[1], S. Noack[2], P. M. Selzer[2], R. Kaminsky[3]

[1]Sablons 30, 2000 Neuchâtel, Switzerland; [2]Boehringer Ingelheim Animal Health GmbH, Ingelheim, Germany; [3]paraC Consulting for Parasitology and Drug Discovery, Altenstein 13,79685 Haeg-Ehrsberg, Germany

E-mail: sandra.schorderet@hotmail.com

Vector-borne diseases are responsible for significant health problems in humans, as well as in companion and farm animals. Killing the vectors with ectoparasitic drugs before they have the opportunity to pass on their pathogens could be the ideal way to prevent vector borne diseases. Blocking of transmission might work when transmission is delayed during blood meal, as often happens in ticks. The recently described systemic isoxazolines [1] have been shown to successfully prevent disease transmission under conditions of delayed pathogen transfer [2, 3]. However, if the pathogen is transmitted immediately at bite as it is the case with most insects, blocking of transmission becomes only possible if ectoparasiticides prevent the vector from landing on or, at least, from biting the host. Chemical entities exhibiting repellent activity in addition to fast killing, like pyrethroids, could prevent pathogen transmission even in cases of immediate transfer [4]. Successful blocking depends on effective action in the context of the extremely diverse life-cycles of vectors and vector-borne pathogens of medical and veterinary importance [5]. With representative examples, we shall highlight important parameters to consider for ectoparasiticide research, when considering the ideal drug profile for preventing disease transmission.

[1] Weber & Selzer, 2016. ChemMedChem 11, 270-276
[2] Beugnet, F. et al., 2014. Parasite Vector 7, 283-289
[3] Taenzler, J. et al., 2016. Parasite Vector 9, 234-236
[4] Brianti, E. et al., 2014. Parasite Vector 7, 327-336
[5] Schorderet-Weber S. et al., 2017, Int J Parasitol Drugs Drug Resist., in press

Barcoded live mosquito screens for high throughput discovery of compounds that interrupt transmission of malaria

Angelika Sturm[1], Martijn Vos[1], Marie Miglianico[1], Maarten Eldering[1], Rob Henderson[1], Sijia Li[2], Geert-Jan van Gemert[3], Robert Sauerwein[1,3], Matthew Tremblay[2], Koen Dechering[1]

[1]TropIQ Health Sciences, The Netherlands; [2]California Institute for Biomedical Research, United States of America; [3]Radboud University Medical Center, The Netherlands

E-mail: k.dechering@tropiq.nl

Elimination of malaria requires a new type of drug that targets the gametocytes, the parasite stages that underlie transmission of the disease through the mosquito vector. To accelerate the search for such transmission-blocking agents we have developed a high throughput method for phenotypic screens on live mosquitoes. This new method relies on DNA-barcoding to trace individual insects during experiments. The DNA barcodes are mixed with compounds and luciferase expressing *Plasmodium falciparum* gametocytes prior to membrane feeding on multiwell plates. Using a luciferase assays on individual mosquitoes eight days after infection, infected and uninfected mosquitoes are identified. The barcodes from the uninfected mosquitoes reveal compounds with transmission blocking activity. Screening of various focused chemical libraries identified a number of compounds with potent activity against the malaria transmission stages. Interestingly, some of these compounds cross-react against other parasites, including kinetoplastids and schistosomes. On top of the anti-parasitic compounds, we identified compounds that kill the mosquito vector. Two of these, afoxolaner and fluralaner, belong to the isoxazoline class of endectocides that are marketed as veterinary drugs. Modeling of their potential impact on malaria transmission showed that, thanks to the potent mosquitocidal activity and exceptional long in vivo half-life of these molecules, a single, seasonal, mass drug administration of either drug is predicted to dramatically reduce the number of clinical cases and may lead to local malaria elimination in low transmission settings. Isoxazolines are, therefore, promising oral malaria transmission-blocking agents that are suitable for single dose treatment.

In vitro screening of the open source MMV (Medicines for Malaria Venture) pathogen boxes reveals novel compounds with profound activities against *Neospora caninum* infection

Joachim Müller, Adriana Aguado-Martinez, Andrew Hemphill

Institute of Parasitology, Vetsuisse Faculty, University of Bern, Switzerland

E-mail: andrew.hemphill@vetsuisse.unibe.ch

*Neospora cani*num is a major cause of fetal infection and abortion in cattle, and constitutes an important veterinary health problem of high economic significance. To date, there are no chemotherapeutic treatments against neosporosis. It is therefore tempting to repurpose compounds effective against other intracellular parasites, most notably other apicomplexans. Here, we present the results of a screen of 400 compounds included in the Pathogen box made accessible by the Medicines for Malaria Venture (MMV) on Human foreskin fibroblasts infected with a *N. caninum* beta-galactosidase reporter strain. A first screening has yielded 44 compounds that were effective against *N. caninum* at 1 µM. 34 of these compounds affected, however, the vitality of the host cells. The 10 remaining compounds were then further characterized, and their IC_{50} values were determined. These included buparvaquone, a naphtoquinone, and the calcium-dependent kinase inhibitor BKI-1294. Four compounds, namely MMV011765, MMV671636, MMV676602, and MMV688762, had IC_{50}s below 10 nM. MMV676602, and MMV688762 inhibited proliferation of *N. caninum* tachyzoites only when added already during infection, but not when added after infection, while MMV671636 inhibited proliferation to background level even when added 24 h post infection. MMV676602 and MMV671636, were characterized in more detail regarding their effects on the ultrastructure of tachyzoites. Furthermore, they were investigated in a non-pregnant neosporosis mouse model, comprised of male BALB/c mice infected with $5x10^5$ *N. caninum* Nc-Spain7 tachyzoites. Two days post infection, treatments with the compounds (10 mg/g bw in corn oil), or with corn oil alone (placebo control) were initiated and continued over 5 days. All mice were euthanized at day 21 pi. None of the mice showed clinical signs at this stage. 9 of 10 mice having received the placebo control and 10 of 10 mice having received MMV676602 were PCR positive for *N. caninum* in their brains, and 4 of 10 mice were lung positive for *N. caninum*. In the group treated with MMV671636, however, only 5 of 10 mice were brain positive and 0 lung positive. All tested mice were seropositive for *N. caninum*. Interestingly, the serum titers were significantly lower in mice treated with MMV671636 than in mice from the other groups. MMV676602 appears to be an interesting candidate to be further investigated in the pregnant neosporosis mouse model.

Discovery and lead optimisation of a promising new antileishmanial compound series within an academic-industry partnership

Manu De Rycker[1], Michael Thomas[1], Tim Miles[2], Paul G. Wyatt[1], Jose Maria Fiandor-Roman[2], David G. Gray[1], Kevin D. Read[1] and Ian H. Gilbert[1]

[1]Drug Discovery Unit, Dundee University, Dundee, UK; [2]Kinetoplastid Discovery Performance Unit, GlaxoSmithkline, Tres Cantos, Spain

E-mail: m.derycker@dundee.ac.uk

The Drug Discovery Unit at the University of Dundee and the GlaxoSmithKline Kinetoplastid Discovery Performance Unit, with support from the Wellcome Trust, have formed a five year partnership to conduct drug discovery within kinetoplastid diseases. This collaboration has made significant progress, highlighted by the identification of a lead optimisation series for visceral leishmaniasis (VL). Here we describe the initial identification of the series through transitioning a *Trypanosoma brucei* active series into a VL active series. The series was profiled extensively in a panel of in vitro *Leishmania* assays including promastigote, axenic amastigote and intracellular assays as well as rate-of-kill and clinical isolate assays, an effort which helped us develop our *Leishmania* screening cascade. An overview of the lead-optimisation campaign will demonstrate how a focus on balancing potency and solubility eventually delivered a compound with candidate-level properties. Current standard of care for VL suffers from multiple issues (lack of efficacy, safety, drug resistance, stability, cost, parenteral administration only) and as a community there is a limited pipeline for VL. The new chemical series presented here is a significant step forward towards the development of a new oral drug for visceral leishmaniasis. This work illustrates the substantial benefits that working in an academic-industry partnership brings for the development of new drugs for neglected diseases.

Heteroaryl- and aryl-substituted miltefosine derivatives

M. Roussaki[1]; K. C. Prousis[1]; T. Fotopoulou[1]; G. Magoulas[1], P. Afroudakis[1],C. Borsari[2], N. Santarem[3], A. Cordeiro-da Silva[3]; Sheraz Gul[4]; E. Bifeld[5]; J. Eick[5]; P. Tejera Nevado[5]; J. Clos[5]; M. J. Corral[6], J. J. Torrado[6], J.M. Alunda[6], M. P. Costi[2], Theodora Calogeropoulou[1]

[1]National Hellenic Research Foundation, Institute of Biology, Medicinal Chemistry and Biotechnology, Athens, Greece; [2]Dipartimento di Scienze della Vita, Università degli Studi di Modena e Reggio Emilia, Modena, Italy; [3]Instituto de Biologia Molecular e Celular (IBMC) da Universidade do Porto, Portugal; [4]Fraunhofer Institute for Molecular Biology and Applied Ecology IME, Hamburg, Germany; [5]Bernhard Nocht Institute for Tropical Medicine, Hamburg, Germany; [6]Universidad Complutense de Madrid, Madrid, Spain

E-mail: tcalog@eie.gr

Ether phospholipid derivatives possess a broad pharmacological spectrum including anticancer, antifungal and antiprotozoal activity. Miltefosine (hexadecylphosphocholine) is an alkylphosphocholine with demonstrated activity against various parasite species and is currently the only oral drug available for the treatment of visceral (VL) and cutaneous leishmaniasis (CL), a neglected tropical infection caused by unicellular parasites. Miltefosine is administered as first-line treatment for VL in India (28 day regimen, 2.5 mg/kg/day) and has been adopted in several national VL elimination programmes (e.g. in India, Bangladesh and Nepal). However, at the therapeutically effective doses, severe gastrointestinal side effects and serious weight loss were observed while teratogenicity remains a problem.

As a continuation of our studies on ring-substituted ether phospholipid derivatives [1-3] we investigated the presence of various heteroaromatic rings or a dinitroaniline moiety in the lipid portion of alkylphosphocholines. The effect of the new derivatives against *T. brucei* (blood stream form) as well as against *L. infantum*, *L. donovani and T. cruzi* amastigotes, was investigated. Furthermore, a wide range of *in vitro* ADME-Tox studies revealed that the resulting derivatives were less toxic than Miltefosine. Selected derivatives exhibiting potent activity against *L. Infantum* amastigotes were studied *in vivo* in BALB/c mice.

Acknowledgement

This project has received funding from the European Union's Seventh Framework Programme for research, technological development and demonstration under grant agreement n° 603240 (NMTrypI - New Medicine for Trypanosomatidic Infections). http://www.nmtrypi.eu/ and by COST CM1307 (STSM for Chiara Borsari).

[1] Calogeropoulou et al (2012) US 8,097,752
[2] Calogeropoulou. et al. 2010 Bioorg. Med. Chem. Lett. 20: 5484-5487
[3] Calogeropoulou et al. 2008 J. Med. Chem.51:897-908

Engaging worms for health - drug discovery on live schistosomes

H. Haas[1], T. Spangenberg[2], N. Reimers[1], C. G. Grevelding[3], M. Sombetzki[4], C. H. Hokke[5], M. J. Doenhoff[6], G. Schramm[1,7]

[1]*helmin*Guard, Sülfeld/Borstel, Germany; [2]Merck Global Health Institute, Switzerland; [3]Institute for Parasitology, University of Giessen, Germany; [4]Department for Tropical Medicine and Infectious Diseases, University of Rostock, Germany; [5]Leiden University Medical Center, The Netherlands, [6]Faculty of Medicine & Health Sciences, University of Nottingham, UK; [7]Experimental Pneumology, Research Center Borstel, Germany

E-mail: hhaas@helminguard.de

Schistosomes are parasitic worms endemic in sub/tropical countries. Worldwide at least 218 million people required preventive treatment against schistosomiasis in 2015 and more than 200,000 deaths occur each year in Sub-Saharan Africa alone [1]. The drug of choice for treating schistosomiasis is praziquantel. However, praziquantel has only limited efficacy in immunocompromised hosts and resistance is emerging. Thus, new drugs are required.

We have established an *in vitro* culture protocol that mimics the *in vivo* environment of the worms and allows growing large numbers of *Schistosoma mansoni* larvae into adults. Using this approach, schistosome life cycle stages (schistosomula, juveniles and adults) were exposed to compounds from various drug libraries for anti-schistosomal agents. Drug effects on the parasites were microscopically assessed and recorded at several time points. Thus, changes in morphology and motility of the worms as well as the time course of drug action could be precisely characterised and saved by photo/video documentation.

This approach revealed drug/group-specific patterns of action and morphological/functional changes such as early vs. late onset of effects, hyper-activity vs. paralysis, shrinkage vs. extension, empty vs. filled guts, circular contractions vs. ballooning of the worms. As a secondary finding, compounds with anti-neoplastic/cytostatic activity became apparent due to adverse effects on the host cells present in the culture. Notably, the analysis of approved drug libraries revealed several drugs with so-far unknown activity against schistosomes with at least one of them being potentially equivalent to praziquantel.

[1] WHO Schistosomiasis Fact sheets www.who.int/mediacentre/factsheets/fs115/en/

BACADs and AASs: different concepts, novel compounds, common anti-schistosomal activities

C.G. Grevelding[1], A. Blohm[1], P. Mäder[2], T. Quack[1], S. Beckmann[1], K. Lange-Grünweller[2], A. Grünweller[2], R.K. Hartmann[2], M. Schlitzer[2], R. Krieg[3], B. Schönecker[4], S. Rahlfs[5], K. Becker[5]

[1]Institute of Parasitology, BFS, Justus Liebig University Giessen; [2] Institute of Pharmaceutical Chemistry, Philipps University Marburg; [3]Institute of Anatomy II, University Hospital Jena; [4]Institute of Organic and Macromolecular Chemistry, Friedrich Schiller University Jena; [5]Biochemistry and Molecular Biology, IFZ, Justus Liebig University Giessen, Germany

E-mail: christoph.grevelding@vetmed.uni-giessen.de

As neglected tropical disease schistosomiasis has significant medical and socio-economic impact in tropical and subtropical countries. A vaccine is not available, and praziquantel is widely used as drug of choice against all *Schistosoma* species causing the disease. This justifies the fear of emerging resistance and motivates the search for alternative treatments.

We followed different approaches to find novel schistosomicides. Based on an *in vitro* culture system for schistosomes and on genome-data indicating the presence of drug targets for conserved genes/proteins, we previously found that certain inhibitors used to treat cancer in human affected schistosome reproduction and vitality [1]. In an analogous approach we used biarylalkyl carboxylic acids (BACADs), which were developed as aldose-reductase inhibitors in the context of diabetes long-term effects. Of 32 BACADs tested on adult worms, 18 caused abnormal egg production in vitro; 12 of these additionally affected vitality and/or tegument and gut integrity [2, 3]. The variety of phenotypes indicated various targets. In a different and novel approach our cooperation partners identified arylmethylamino steroids (AASs) as potent antiparasitics against chloroquine-sensitive and -resistant *Plasmodium falciparum* (IC50 1-5 nM *in vitro*). The lipophilic character of AASs may facilitate membrane permeation and bioavailability. Due to the dual-lipid bilayer nature of the schistosome surface, a physiologically active tegument, we assumed that AASs penetrate also these parasites. Indeed, already in low concentrations (1-5 µM) we observed lethal effects on adult worms *in vitro* with specific AAS derivatives [4].

[1] Dissous, C. & Grevelding C.G., 2011. Trends Parasitol. 27, 59-66
[2] Blohm, A.S. et al., 2016. Parasitol. Res. 115(10), 3831-42
[3] Mäder, P. et al., 2016. ChemMedChem. 11, 1459-68
[4] Krieg, R. et al., 2017. Nat. Commun. 8, 14478

Inhibition of *Schistosoma mansoni* development in mice by slow release of artemisone

D Gold[1,3], M Alian[2], A Domb[2], Y Karawani[3], M Jbarien[3], N Salaymeh[3], J Chollet[4], R K Haynes[5], V Buchholz[6], A Greiner[6], J Golenser[3]

[1]Department of Clinical Microbiology and Immunology, Sackler Faculty of Medicine, Tel Aviv University; [2]School of Pharmacy, Faculty of Medicine, Hebrew University of Jerusalem; [3]The Kuvin Center for the Study of Infectious and Tropical Diseases, The Department of Microbiology and Molecular Genetics, Hebrew University of Jerusalem-Hadassah Medical School; Israel; [4]Swiss Tropical Institute, P.O. Box, CH-4002 Basel, Switzerland; [5]Centre of Excellence for Pharmaceutical Sciences, School of Health Sciences, North-West University, Potchefstroom 2520, South Africa; [6]Macromolecular Chemistry II, University of Bayreuth, Germany

E-mail: Golenser@md.huji.ac.il

Schistosomes are parasitic helminths, most important in terms of socio-economic and public health in tropical and subtropical areas. Schistosomiasis causes skin allergies, intestinal, liver and urinary pathologies. Chronic disease may also lead to cancer. In addition, there are often systemic symptoms, such as retarded growth, slowing of cognitive development and continuing low-level blood loss. The current treatment is based on praziquantel (PZQ) that affects only the adult stages of schistosomes. Also, following its widespread use, resistance is emerging.

It is our purpose to test a drug, which could serve as a potential alternative or complement to PZQ, and also as a means of treating infections at an earlier, pre-granuloma stage. Derivatives of the peroxidic antimalarial drug artemisinin have been indicated as potential alternatives, because both plasmodia and schistosomes are blood-dwelling and blood-feeding parasites. The mechanism of action of artemisinins is ascribed inter alia to oxidative effects of the peroxide on intracellular reductants leading to formation of cytotoxic reactive oxygen species. We used the newer artemisinin derivative artemisone, which has improved pharmacokinetics and anti-plasmodial activity, and reduced toxicity compared to other artemisinins that are in clinical use against malaria. We infected adult mice by subcutaneous injection of *S. mansoni* cercariae and treated them at various times post infection by the following methods: i. artemisone suspension administered by gavage; ii. subcutaneous injection of a gel containing a known concentration of artemisone or praziquantel; iii. subcutaneous insertion of the drug incorporated in a solid polymer; iv. intraperitoneal injection of the drug solubilized in DMSO. Drug administration in polymers was performed to enable slow release of the artemisone that was verified in vivo and in vitro bioassays using drug-sensitive malaria parasites. We found superior strong anti-schistosome effects, mainly following repetitive treatments with the drug absorbed in the polymers. The results indicate that artemisone has a potent anti-schistosome activity even at an early stage of the infection. Its main importance in this context is its effectiveness in treating hosts harboring juvenile schistosomes, before egg-deposition and induction of deleterious immune responses.

Etanercept: Initiative Study on the drug potentiality in murine *Schistosomiasis mansoni*

G. Labib Elenain[1,2]

[1]Natural Sciences Department, Abu Dhabi University, UAE; [2]Immunology and Drug Evaluation, Theodor Bilharz Research Institute, Egypt

E-mail: gehan.elenain@adu.ac.ae

Schistosomiasis is a worldwide health problem. There is an alarming of Praziquantel drug resistance [1] and a demand for new treatment. This initiative aimed at studying the efficacy of soluble Tumor Necrosis Factor Receptor II:Fc protein as an alternative drug in murine schistosomiasis *mansoni*. Of eleven therapeutic Fc-fusion proteins, Etanercept is the most successful one [2]. Female BALB/C mice were infected subcutaneously with 40 *S. mansoni* cercariae. Five and half week later, those mice were divided into three groups, each contained 9 mice. Group I served as a control. The others were treated intraperitoneally (i.p.) with Etanercept. Group II received an initial dose of 100µg Etanercept, whereas group III was administered by a 300µg. One week later, each experimental group was injected by a 100µg-dose of Etanercept for 5 days. All mice were sacrificed at week 7.5 post-infection. Compared to the infected controls, the liver weight, hepatic egg-load and granuloma size were sharply less in the experimental groups. Yet, the number of worms was comparable. Unlike IL-4, the mRNA of IL-2 was not detected in the experimental mice.

[1] Campelo YD, et al., 2017. Biomed Pharmacother 23; 88:488-499
[2] Jafari R et al., 2017. Curr Med Chem

Drug Resistance in the Cattle Tick *Rhipicephalus (Boophilus) microplus* – Monitoring Tools and search for new Compounds

Heinz Sager, Léonore Lovis

Elanco Animal Health, Basel, Switzerland

E-mail: heinz.sager@elanco.com

The cattle tick *Rhipicephalus* (*Boophilus*) *microplus* is widespread in tropical and subtropical countries. Because of its economic impact on the cattle industry, different measures – of which acaricide-treatments are the most relevant – are taken to reduce the tick burden on the animals as well as to control the spread of the ticks. The use of chemical compounds started at the end of the nineteenth century with the introduction of arsenicals. In the meantime at least eight new acaricide classes have been introduced to the market, most of them followed by reports of treatment failure and finally the identification of resistance.

The cattle tick *R. (B.) microplus* uses predominantly modifications at the target site and increased metabolic detoxification to obtain resistance against acaricides, at least to what is known so far. Probably best described are the mechanisms of resistance for synthetic pyrethroids where four single nucleotide substitutions in the gene coding for the voltage-gated sodium channel were identified that could be assigned to different geographic areas.

The detection of resistance against drugs (or acaricide classes) is a prerequisite for successful control strategies, and a variety of assays and methods exist to test the sensitivity of ticks to drugs. However, all of them are either slow, labor intensive, or can assess only a limited number of acaricides. Still, it is worth to evaluate their suitability for use in screening systems to identify new acaricide-compounds. This is discussed for phenotypic as well as for target-based approaches.

Fragment-based approach in the discovery of highly active antiparasitic PTR1 Inhibitors

Pasquale Linciano[1], Luca Costantino[1], Cecilia Pozzi[4], Rebecca Wade[2], Stefano Mangani[4], Anabela Cordeiro da Silva[5], Sheraz Gul[6], Maria Paola Costi[1]

[1]University of Modena and Reggio Emilia, Modena, Italy; [2]HITS gGmbH, Heidelberg Germany; [4]University of Siena, Siena , Italy; [5]IBMC - Instituto de Biologia Molecular e Celular IBMC - Instituto de Biologia Molecular e Celular and Universidade do Porto, Porto, Portugual; [6]Fraunhofer- IME SP, Hamburg, Germany

E-mail: p.linciano@unimore.it

Parasitic neglected tropical diseases caused by trypanosomatids such us *T. brucei* (Human Sleeping Sickness), *T. cruzi* (Chagas Disease) and several species of *Leishmania* represent an actual severe burden. Trypanosomatids lack the ability to synthesize pterins (i.e. folic acid and biopterin) de novo. However, antifolates cannot be used in therapy of trypanosomatidic infections because dihydrofolate reductase (DHFR) inhibition is compensated by another enzyme, the pteridine reductase-1 (PTR1). PTR1 is mainly involved in the reduction of biopterine but can also reduce folates, thereby safeguarding the cell survival. Therefore, PTR1 could be a promising target for the design and the development of new non-folate drugs to be used in combination with classic antifolates. To address these issues, we started a fragment-based drug design (FBDD) approach to improve the quality and the efficiency of the drug discovery process. Starting from a library of fifteen pteridine-like substrate analogues, the X-ray crystallography screening provided eight complexes that were used for further FBDD elaboration. The affinity (K_i) of the starting fragments was in the range of 10^{-3}-10^{-4} M. Two of them were suitable for synthetic modification and generated 4 chemical sub-classes with high affinity and selectivity against the target enzymes (K_i: 10^{-7}-10^{-8} M) and good ligand efficiency (LE: 0.5-0.8). 15 final compounds gave x-ray crystal structures supporting the FBDD rationale. All the fragments and compounds were evaluated for their capability to inhibit parasitic PTR1 and *Ki* values were obtained to comply with the FBDD principle [1]. The final compounds were screened against three different trypanosomatids (*T. brucei*, *T. cruzi* and *L. infantum*) resulting very potent toward *T. brucei* (EC_{50}: 90-300 nM) and selective with the respect of human macrophages (SI: 80-140 times). Early tox studies support the absence of early toxicity. The progress of the compounds in the pipeline followed the Target Profile guideline (TPP) that reduces the liabilities of the future drug candidates [2]. Six compounds were selected for snap-PK on BALB/c mice studies and one of them showed a suitable profile (oral $t_{½}$ of 3h and C_{max} of 2.5 uM, 30-times the antiparasitic EC_{50}) [2] for *in vivo* studies on *T. brucei* infection model. The results obtained are in line with the FBDD approach achieving, with swift and focused scaffold improvement, a sustainable delivery of high-quality lead candidates.

This work was supported by NMTrypI (New medicine for Trypanosomatidic Infections) and received funding from the European Union's Seventh Framework Programme under grant agreement no.603240. http://cdm.unimo.it/home/dipfarm/costi.mariapaola/NMTrypI_Home.html

[1] Top Curr Chem. 2012;317:1-32. doi: 10.1007/128_2011_180

[2] Nature Reviews Drug Discovery 14, 751–758 (2015) doi:10.1038/nrd4683

Mycobacterium tuberculosis thioredoxin reductase inhibitors with activity on mycobacterial growth in infected macrophages based on *in-silico* molecular design.

L. Bering[1], J. Pretzel[1], M. Schneefeld[2], N. Reiling[3], F.-C. Bange[2], O. Koch[1]

[1]TU Dortmund University, Dortmund, Germany; [2]Medical School Hannover, Hannover, Germany; [3]Research Center Borstel, Borstel, Germany

E-mail: oliver.koch@tu-dortmund.de

The resurgence of tuberculosis, caused primarily by *Mycobacterium tuberculosis* (*Mtb*), and the appearance of multi-drug and extensively drug resistant strains strengthen the need for new drugs with alternative modes of action [1]. The interaction between the mycobacterial thioredoxin reductase (TrxR) and its substrate thioredoxin (Trx) is a promising new drug target for the treatment of tuberculosis, since *Mtb* lacks the common glutathione system and its TrxR shows a substantial difference in sequence, mechanism and structure to human TrxRs. The mycobacterial TrxR is essential for thiol redox homeostasis and genetic inactivation killed and lysed *Mtb* [2]. Although the target mechanism is a protein-protein interaction, the first known inhibitors with different scaffolds could be identified using an exhaustive high-throughput docking based on the available TrxR X-ray structures [3]. By means of structure-based design, the activity of the most promising candidate could be increased to an IC_{50} up to the low nanomolar range that also showed an influence on the growth of *M. tuberculosis*.

In order to further improve the bioactivity of the promising compounds, we focused on optimizing the physicochemical properties that are important for permeability, since *M. tuberculosis* shows an unusual thick and impermeable cell wall. An analysis of calculated polar surface area and logP indicated an influence of these properties on the minimum inhibitory concentration. Based on computational molecular design, compounds with improved properties were synthesized. In fact, these compounds showed an improved antimycobacterial activity, which underlines the assumption regarding optimized properties.

The most promising compound was also tested on infected human macrophages and showed a clear dose-dependent activity on mycobacterial growth, without affecting macrophage viability. I will present and discuss the design and the results of our improved compounds.

[1] World Health Organization Tuberculosis Programme; http://who.int/tb/
[2] Lin, K. et al. PLoS Pathog. 2016, 12(6): e1005675
[3] Koch, O. et al. J. Med. Chem. 2013, 56: 4849-4859

Development of an *in vitro* drug release method for a veterinary modified-release dosage form as a tool for quality control

Timothy S. Priddy[1], Ronald R. Roush[1], Lawrence Bryson[2], Martin Folger[3]

[1] Department of Vaccine Assay Development, Boehringer Ingelheim Vetmedica, Inc., Saint Joseph MO, USA;[2] Department of Biometrics and Statistics, Boehringer Ingelheim Vetmedica, Inc., Saint Joseph MO, USA;[3] Global CMC Development Pharmaceuticals, Boehringer Ingelheim Vetmedica GmbH, Ingelheim am Rhein, Germany

E-Mail: martin.folger@boehringer-ingelheim.com

In the field of antiparasitics for veterinary application, long-acting active pharmaceutical ingredients (APIs), as well as modified-release dosage forms ensuring a prolonged pharmacological activity of the API, are frequently applied. One of the most important characteristics of an API is the release or exchange profile of the API from the drug product matrix, especially for that of a long-acting or modified-release formulation. Besides the requirement to develop a robust and discriminating in vitro dissolution / release method for quality control purposes, in vitro release methods may also help to understand *in vivo* behaviour of different formulations depending on their composition and release behaviour. In the former case, the impact of the quality of raw materials, including the API and the excipients chosen, as well as the variable effects of excipient quantities and manufacturing processes are assessed.

The development and validation of an *in vitro* drug exchange (IVDE) method is presented that is capable of reproducibly detecting changes in qualitative and quantitative composition of a long-acting veterinary formulation. The major three development steps a) identification of a suitable medium to extract the API over an acceptable amount of time without chemical degradation, b) choice of a suitable dissolution apparatus and design of the analytical method to be used in a quality control (QC) laboratory, as well as c) determination of the discriminatory power of the IVDE method with respect to various other formulations containing the same API, are outlined.

Furthermore, the retrospective characterization of different formulations that were also tested in vivo during early clinical development revealed that the IVDE method could presumably be suitable for prospective evaluations of long-acting dosage forms that contain the same API with, perhaps, a different targeted release profile. The feasibility of such an approach can be realized by attributing differential in vivo pharmacokinetics for drug products that demonstrate differential IVDE profiles, and vice versa, if necessary.

Targeting pathogen metabolism without collateral damage to the host

Jurgen R. Haanstra[1,2], Albert Gerding[1], Amalia M. Dolga[3], Freek J. H. Sorgdrager[4], Manon Buist-Homan[5], François du Toit[6], Klaas Nico Faber[5], Hermann-Georg Holzhütter[7], Balázs Szöör[8], Keith Matthews[8], Jacky L. Snoep[2,6,9], Hans V. Westerhoff[2,9,10] and Barbara M. Bakker[1,2]

[1]University of Groningen, University Medical Center Groningen, Department of Pediatrics and Systems Biology, Centre for Energy Metabolism and Ageing, Center for Liver, Digestive and Metabolic Diseases, Groningen, The Netherlands; [2]Department of Molecular Cell Physiology, Faculty of Earth and Life Sciences, VU University, Amsterdam, The Netherlands; [3]University of Groningen, Department of Molecular Pharmacology, Groningen, The Netherlands; [4]University of Groningen, University Medical Center Groningen, European Research Institute for the Biology of Ageing, Groningen, The Netherlands; [5]University of Groningen, University Medical Center Groningen, Department of Hepatology and Gastroenterology, Centre for Liver, Digestive and Metabolic Diseases, Department of Laboratory Medicine, Groningen, The Netherlands; [6]Department of Biochemistry, Stellenbosch University, South Africa; [7]Charité - Universitätsmedizin Berlin, Institut für Biochemie, Berlin, Germany; [8]Centre for Immunity, Infection and Evolution, Institute for Immunology and Infection Research, School of Biological Sciences, University of Edinburgh, UK; [9]Manchester Centre for Integrative Systems Biology, School of Chemical Engineering and Analytical Science, University of Manchester, Manchester, UK; [10]Swammerdam Institute for Life Sciences, Faculty of Science, University of Amsterdam, Amsterdam, The Netherlands

Email: j.r.haanstra@vu.nl

The development of drugs that can inactivate disease-causing cells (e.g. cancer cells or parasites) without causing damage to healthy or to host cells is complicated by the fact that many proteins are very similar between organisms. Nevertheless, due to subtle, quantitative differences between the biochemical reaction networks of target cell and host, a drug can limit the flux of the same essential process in one organism more than in another. We identified precise criteria for this 'network-based' drug selectivity, which can serve as an alternative or additive to structural differences. We combined computational and experimental approaches to compare energy metabolism in the causative agent of sleeping sickness, *Trypanosoma brucei*, with that of human erythrocytes, and identified glucose transport and glyceraldehyde-3-phosphate dehydrogenase as the most selective antiparasitic targets. Computational predictions were validated experimentally in a novel parasite-erythrocytes co-culture system. Glucose-transport inhibitors killed trypanosomes without killing erythrocytes, neurons or liver cells. This exemplifies that the arsenal of potential selective drug targets can be broadened beyond the pathogen-specific proteome through a differential network-based approach integrating *in silico* modelling and *in vitro/in vivo* experimentation.

This work is published as: Haanstra, J. R. et al. Targeting pathogen metabolism without collateral damage to the host. Sci. Rep. 7, 40406; doi: 10.1038/srep40406 (2017)

cyclic AMP-independent PKA from *Trypanosoma brucei* as promising drug target

Sabine Bachmaier[1], Susanne Kramer[1], George Githure[1], Yuri Volpato[1], Julia Pepperl[1], Jean-William Dupuy[2], Marc Bonneu[2], Hans-Gottfried Genieser[3], Frank Schwede[3] and Michael Boshart[1]

[1]Biocenter, Section Genetics, Ludwig-Maximilians-Universität München, Martinsried, Germany;
[2]Centre de Génomique Fonctionnelle Bordeaux, Université Bordeaux, F-33076 Bordeaux, France;
[3]BIOLOG Life Science Institute, Bremen, Germany

E-mail: boshart@lmu.de

The protein kinase A (PKA) signaling pathway is highly conserved throughout the eukaryotic kingdom. Activation by cyclic AMP and regulation of a plethora of cellular processes such as metabolism, differentiation or cell cycle progression, are hallmarks of PKA in unicellular as well as multicellular organisms. Here we show that the PKA signaling pathway has been retooled in the evolutionary early branching eukaryotic parasite *Trypanosoma brucei*. The parasite PKA is not activated by the second messenger cAMP, while otherwise exhibiting characteristic PKA features. Instead of cAMP, we identified several 7-deazapurine derivatives as potent activators of *T. brucei* PKA by a small-scale chemical screen. Compound optimization led to improvement in specificity and potency. The most potent compounds were subsequently used as tools to identify downstream components of PKA signaling in *T. brucei.* Sequence comparison revealed the presence of highly similar PKA orthologues in the related kinetoplastids *Trypanosoma cruzi* and *Leishmania donovani* indicating conservation of this unconventional pathway within kinetoplastids. The presence of a conserved PKA with unconventional properties in kinetoplastids combined with the essentiality of the kinase in *T. brucei* suggests it as an optimal drug target for treatment of kinetoplastid-caused diseases.

The effect of media perfusion on the infection and drug activity against *Leishmania major*

A.O'Keeffe[1], S.Murdan[2], S.Croft[1]

[1]London School of Hygiene & Tropical Medicine, London, [2]UCl School of Pharmacy, London, UK

E-mail: alec.o'keeffe@lshtm.ac.uk

Leishmaniasis is a neglected tropical disease caused by protozoan parasites of the genus *Leishmania*. These parasites infect, survive and multiply in macrophages in the skin causing cutaneous leishmaniasis [1. All current treatments and drugs have significant limitations and the search for novel drugs is a major focus of research in industry and academia.

As part of a program to improve the predictiveness of *in vitro* and *in vivo* (rodent) models to identify novel drugs, reduce attrition rates and accelerate progress, we are developing novel *in vitro* assays that more closely simulate the *in vivo* situation.

We have concentrated on cutaneous leishmaniasis and firstly simulating interstitial fluid flow found in the skin, using the Kirkstall QV900 system. It is estimated that up to 20% of the body's mass is interstitial fluid, and much of this fluid is in constant motion, albeit slowly [2]. Our investigation into the effect of interstitial fluid flow on the infection rate has shown the application of flow reduces the infection caused by *Leishmania* in comparison to a static control. In further experiments we observed a change in the systems response to drugs under flow conditions. The EC_{50} of miltefosine shifted from 9.6µM to 19.3µM and again to 36.2µM, when the cells were raised on an 3D printed insert, and the EC_{90} shifted from 36.6µM to 120µM and 285µM under the same three conditions. This pattern was seen repeated across all 4 drugs tested. To further investigate the effects seen, drug accumulation studies and cell phenotyping was carried out.

[1] Croft, S. L. et al (2011) Clinical Microbiology and Infection 17(10), 1478-1483
[2] Swartz, M. A et al (2007) Annual Review of Biomedical Engineering 9, 229-256

Antiparasitic drug discovery in epigenetics

D. Monaldi[1], M. Marek[2], J. Lancelot[3], W. Sippl[4], R. Pierce[3], C. Romier[2], M. Jung[1] and the A-ParaDDisE Consortium[5]

[1]Institute of Pharmaceutical Sciences, Albert-Ludwigs-Universität Freiburg, Germany; [2]Département de Biologie Structurale Intégrative, Institut de Génétique et Biologie Moléculaire et Cellulaire (IGBMC), Université de Strasbourg (UDS), CNRS, INSERM, Illkirch, France; [3]Center for Infection and Immunity of Lille (CIIL), INSERM U1019—CNRS UMR 8204, Université Lille Nord de France, Institut Pasteur deLille, Lille, France; [4]Institute of Pharmacy, Martin-Luther-Universität Halle-Wittenberg,Halle, Germany; [5]a.paraddise.cebio.org

E-mail: Daria.monaldi@pharmazie.uni-freiburg.de

The development of novel antiparasitic agents is the purpose of the A-ParaDDisE project, funded by EC(FP7) and involving 17 research teams in 5 european countries, Brazil and Australia, that work on four parasitic diseases: Schistosomiasis, Leishmaniasis, Chagas´ disease and Malaria. Targeting histone modification enzymes, able to introduce or remove epigenetic marks on chromatin, these research groups aim to develop parasite-selective and species-specific drugs, starting from the identification of histone modification enzymes in the parasitic genome until preclinical *in vivo* testing.

As part of this project, our interest is focused on Schistosomiasis and, more exactly, on one of its major causative agents: *Schistosoma mansoni*. Although one drug, praziquantel, is already in the market, several reasons, including resistence development, made urgent the necessity of novel therapeutic agents [1]. Within the A-ParaDDisE project, it was possible to identify five *S.mansoni* Sirtuins (NAD+-dependent lysine deacetylases) and, due to strong effects of human Sirtuin 2 inhibitors (hSirt2i) on parasitic life span and reproduction, to define the parasitic isoform 2 (smSirt2) as potential therapeutic target [2].

As consequence of that, we aim to find novel smSirt2 inhibitors as lead candidates by the use of a focused library screening approach [3]. Furthermore since *in vitro* and *in vivo* studies have reported the ability of hSirt2 to catalyze long chain deacylation [4], results of the investigation on the ability of smSirt2 in removing long chain acyl groups are reported.

[1] Wilson, M.S. et al.; 2007. Immunol. Cell. Biol. 85, 148-154
[2] Lancelot, J. et al.; 2013. PLoS Negl. Trop. Dis. 9, e2428
[3] Schiedel, M. et al.; 2015. J. Biomol. Screening. 20, 112-121
[4] Feldman, J.L. et al.; 2015. Biochemistry. 54, 3037-3050

Discovery of natural bioactive compounds: The Road to El Dorado

Asaad Khalid

Substance Abuse Research Center, Jazan University, Jazan 45142, Saudi Arabia

E-mail: drasaad@gmail.com

Just like *Eldorado* (legendary lost city of gold), the road to the discovery of bioactive compounds and drug hits from plants could be full of challenges and adventures. Natural products are characterized by its unorthodox and often unanticipated chemical structures that offer novel leads of clinically useful drugs. The myriad of structurally diverse compounds found in nature makes them play an important role as a unique source for drug discovery, but they often play hard-to-get. Even though, most of the FDA approved drugs are either natural products or natural product-derived compounds. Studies have demonstrated that the hit rate of natural products is on average 3-10%, compared with ~ 0.03% of that of compounds from synthetic origin [2, 3].

On the other hand drug discovery could follow any of the two approaches i.e. cell-based and/or target-based where enzymes represent the major class of drug targets. Recent reports show that about 50% of small molecule drugs are enzyme inhibitors [1].

This lecture will give an overview of our research on the discovery of bioactive hits and from interesting bioactive plants. Our cell-based and target based research will also be highlighted. This research has led to the identification of very interesting bioactive compounds from many plants.

[1] Copeland, R. A. 2005. Evaluation of enzyme inhibitors in drug discovery: a guide for medicinal chemists and pharmacologists. John Wiley & Sons, Inc., Hoboken, N.J.

[2] Kinghorn AD. In: Gullo V, editor. The Discovery of Natural Products with Therapeutic Potential. Stoneham: Butterworth-Heinemann; 1994. p. 81-108

[3] Koch MA, Wittenberg LO, Basu S, Jeyaraj DA, Gourzoulidou E, Reinecke K, et al. Compound library development guided by protein structure similarity clustering and natural product structure. Proc Natl Acad Sci U S A 2004;101(48):16721-6

Ruthenium Complexes for the Treatment of Protozoan Diseases

Afonso Basto[1], Joachim Müller[1], Riccardo Rubbiani[2], Andrew Hemphill[1], Gilles Gasser[2], Julien Furrer[3]

[1]Institute of Parasitology, Vetsuisse Faculty, University of Bern, Switzerland; [2]Department of Chemistry, University of Zurich, Switzerland; [3]Departement of Chemistry and Biochemistry, University of Bern, Switzerland

E-mail: julien.furrer@dcb.unibe.ch

Recently, some of us have shown that dinuclear thiolato-bridged arene ruthenium complexes of the type $[(\eta^6\text{-}p\text{-cymene})_2Ru_2(\mu_2\text{-SR})_3]^+$ were among the most cytotoxic ruthenium complexes reported so far, with nanomolar IC_{50} values against both A2780 human ovarian cancer cells and their cisplatin-resistant mutant variant A2780cisR [1]. Interestingly, *in vivo* studies of *diruthenium-1*, $[(\eta^6\text{-}p\text{-cymene})_2Ru_2(\mu_2\text{-}SC_6H_4\text{-}p\text{-}Bu^t)_3]^+$, demonstrated a significant increase in survival of the treated mouse group [2-3].

Motivated by the impressive results obtained in cancer therapy, we decided to test some of those compounds against *Neospora caninum* and *Toxoplasma gondii*. Two complexes, namely $[(\eta^6\text{-}p\text{-cymene})_2Ru_2(\mu_2\text{-}SC_6H_4\text{-}p\text{-}CH_3)_3]^+$ and *diruthenium-1*, displayed IC_{50} values on *T. gondii* of 34 and 62 nM, respectively, with a high selectivity on *T. gondii* over human foreskin fibroblasts (HFF) as expressed by a selectivity index of around 20'000. Transmission electron microscopy (TEM) showed that treatment with these compounds has a dramatic impact on the parasite mitochondrion. Moreover, upon treatment of human ovarian carcinoma A2780 cells with these drugs, between 71 and 97% of the ruthenium was found to accumulate in the mitochondria. This strongly suggests that these compounds are targeting the mitochondria in both parasites and cancer cells. *Giardia lamblia* that is devoid of functional mitochondria is not affected by treatments with these complexes up to a concentration of 5 µM, thus further supporting this statement.

[1] J. Furrer, G. Süss-Fink, *Coord. Chem. Rev.* **2016**, *309*, 36-50.
[2] P. Tomšik, D. Muthná, M. Řezáčová, S. Mičuda, J. Ćmielová, M. Hroch, R. Endlicher, Z. Ćervinková, E. Rudolf, S. Hann, D. Stibal, B. Therrien, G. Süss-Fink, *J. Organomet. Chem.* **2015**, *782*, 42-51.
[3] A. Koceva-Chyła, K. Matczak, P. Hikisz, K. Durka, K. Kochel, G. Süss-Fink, J. Furrer, K. Kowalski, *ChemMedChem*, **2016**, *11*, 2171-2187.

The Pentose Phosphate Pathway of *Plasmodium* parasites as a drug target

K. Haeussler[1], J. Preuss[1,2,3], E. Jortzik[1], S. Rahlfs[1], A. Pinkerton[2], L. Bode[3], K. Becker[1]

[1]Biochemistry and Molecular Biology, Interdisciplinary Research Center. Justus Liebig University Giessen. Germany; [2]Conrad Prebys Center for Chemical Genomics, Sanford Burnham Prebys Medical Discovery Institute, USA; [3]Department of Pediatrics, University of California, San Diego, USA

E-mail: Kristina.Haeussler@ernaehrung.uni-giessen.de

Malaria is still one of the most important diseases, caused by the parasite *Plasmodium*. WHO currently recommends an artemisinin-based combination therapy. However, due to the increasing occurrence of resistant *Plasmodium* strains, the development of new drugs is of essential importance.

Inside the erythrocytes, *Plasmodium* is massively exposed to reactive oxygen species. To deal with this oxidative stress, the parasites possess a complex antioxidative system that depends on the supply of NADPH [1]. The pentose phosphate pathway represents the major source of NADPH for the parasites and contributes substantially to the maintenance of the cellular redox balance. In *Plasmodium*, the first two steps are catalyzed by a unique bifunctional enzyme, the glucose 6-phosphate dehydrogenase 6-phosphogluconolactonase (GluPho). This NADPH-producing enzyme differs structurally and functionally from the human homologs and – as we showed recently in gene knock out studies – is essential for blood stages of *P. falciparum*. For these reasons, we consider PfGluPho to be an excellent target for the development of new antimalarial drugs [2].

High-throughput screenings of different compound libraries revealed several hits. Improving these first hits resulted in lead compounds with IC_{50}s in the lower nanomolar range against recombinant PfGluPho and *P. falciparum* 3D7 in cell culture. Counterscreening the human homolog hG6PD, with IC_{50}s over 80 µM, underscores the enormous specificity of the compounds. Our most potent compound is currently tested in mouse models.

[1] Jortzik, E. and Becker, K., 2012. Int J Med Microbiol. 302, 187-194
[2] Allen, S. M. et al., 2015. FEBS Journal. 282, 3808–3823

Design, synthesis and antimalarial activity of novel bis{N-[(pyrrolo[1,2-a] quinoxalin-4-yl)benzyl]-3-aminopropyl}amine derivatives

Jean Guillon[1,2], Anita Cohen[3], Nassima Meriem Gueddouda[1,2], Rabindra Nath Das[1,2], Stéphane Moreau[1,2], Luisa Ronga[1,2], Solène Savrimoutou[1,2], Louise Basmacyian[3], Alix Monnier[1,2], Myriam Monget[1,2], Sandra Rubio[1,2], Timothée Garnerin[4], Nadine Azas[3], Jean-Louis Mergny[1,2], Catherine Mullié[4], and Pascal Sonnet[4]

[1]UFR des Sciences Pharmaceutiques, ARNA Laboratory, University of Bordeaux, France, [2]INSERM U1212, UMR CNRS 5320, ARNA Laboratory, Bordeaux, France, [3]Laboratory of Parasitology, Faculty of Pharmacy, Aix-Marseille UIniversity, France, [4]Laboratoire de Glycochimie, des Antimicrobiens et des Agroressouces, Université de Picardie Jules Verne, Amiens, France

E-mail: Anita.COHEN@univ-amu.fr

The pyrrolo[1,2-*a*]quinoxaline heterocyclic framework constitutes the basis of an important class of compounds possessing interesting biological activities, including antiparasitic ones [1, 2]. Novel series of bis- and tris-pyrrolo[1,2-*a*]quinoxaline derivatives, already defined as quinoline-like bio-isosteres [1], were synthesized and tested for *in vitro* activity upon the intra-erythrocytic stage of W2 and 3D7 *Plasmodium falciparum* strains. Biological results showed good antimalarial activity with IC50 in the µM range. In attempting to investigate the large broad-spectrum antiprotozoal activities of these new derivatives, their properties toward *Leishmania donovani* were also investigated and revealed their selective antiplasmodial profile. In parallel, the *in vitro* cytotoxicity of these molecules was assessed on the human HepG2 cell line. Structure-activity relationships of these new synthetic compounds are here discussed.

Two bis-pyrrolo[1,2-*a*]quinoxalines were identified as the most potent antimalarial candidates with selectivity index (SI) of 40.60 on W2 strain, and 39.25 on 3D7 strain, respectively. As the telomeres of the parasite could constitute an attractive target, we investigated the possibility of targeting *Plasmodium* telomeres by stabilizing the *Plasmodium* telomeric G-quadruplexes through a FRET melting assay by our new compounds [3]. Indeed, telomerase activity has been identified in gametocytes and during the transition to erythrocytic stage of *P. falciparum* [4]. The telomeric 3' G-overhang region of P. falciparum is comprised of repeated degenerate unit 5'GGGTTYA3' (where Y may be T or C) [5] which can fold into intramolecular G-quadruplex [6].

These results led us to conclude that two bis{*N*-[4-(methoxypyrrolo[1,2-*a*]quinoxalin-4-yl)benzyl]-3-aminopropyl}piperazines seemed to be able to discriminate between *Plasmodium* and human telomeric quadruplexes, which could be now considered as a promising starting point for the further development and optimization of new and potent antimalarial compounds.

[1] (a) Guillon J et al. J Med Chem 2004;47:1997-2009
(b) Guillon J et al. Bioorg Med Chem 2007;15:194-210
(c) Guillon J et al. J Enzyme Inhib Med Chem 2007;22:541-549
(d) Guillon J et al. Bioorg Med Chem 2008;16:9133-9144
(e) Guillon J et al. Eur J Med Chem 2011;46:2310-2326
[2] Ronga L et al. J. Eur J Med Chem 2014;81:378-393
[3] Guillon Jet al. J Enzyme Inhib Med Chem 2017;32:547-563
[4] Bottius E. Bakhsis N, Scherf A. Mol Cell Biol 1998;18:919-925
[5] Raj DK et al. Biochem Biophys ResCommun 2003;309:685-688
[6] De Cian A et al. ChemBioChem 2008;9:2730-2739

Ferredoxin-Ferredoxin-NADP+-Reductase – a gem amongst drug-targets

Schäfer, E.-M.[1]; Volkamer, A.[2]; Seeber, F.[3]; Baumeister, S.[4]; Lechner, M.[1]; Schlitzer, M.[1]

[1]Institut für pharmazeutische Chemie, Fachbereich Pharmazie, Philipps-Universität Marburg, Marburg, Germany; [2]BioMed X Innovation Center, Heidelberg, Germany; [3]Robert-Koch-Institut, FG 16: Erreger von Pilz- und Parasiteninfektionen und Mykobakteriosen, Berlin, Germany; [4]Institut für Entwicklungsbiologie und Parasitologie, Fachbereich Biologie, Philipps-Universität Marburg, Germany

E-mail: schaef62@staff.uni-marburg.de

Among infectious diseases Malaria still is one of the biggest problems for public health especially in developing countries [1]. Due to rising resistance the need for new high potent drugs is urgent. Therefore, it is not only necessary to enhance yet known drugs, but also to find new promising targets. A promising new target is the redox-system ferredoxin/ferredoxin-$NADP^+$-reductase. It is located in the apicoplast and is from plant origin. As mammalian cells lack of this plant-like system, less side effects of designed inhibitors are expected [2]. This system is essential for the synthesis of iron-sulphur clusters and its distribution to enzymes of various essential pathways like the lipoic-acid synthesis or the non-mevalonat isoprenoid synthesis. Therefore, an inhibition of this system would cause an inhibition of these essential pathways and therefore lead to the parasites' death. There are three approaches towards inhibition. As ferredoxin-ferredoxin-$NADP^+$-reductase is a complex of two proteins, each protein can be addressed separately. Thus we designed and synthesised inhibitors for each, the Fd and the FNR part of the active enzyme. In future compounds that lead to an interruption of protein-protein-interaction are planned [3].

[1] World Malaria Report 2013, World Health Organisation, Genf, Schweiz
[2] Komoto-Ariga, Y.; Saito, T.; Ikegami, T.; J. Biochem., 2007, 142, 715-720
[3] Seeber, F.; Aliverti, A.; Zanetti, G.: Curr. Pharm. Des. 2005, 11, 3159-3172

Unlocking nanocarriers for the programmed release of antimalarial artemisone

A. R. Bagheri[1], S. Agarwal[1], J. Golenser[2], A. Greiner[1]

[1]Macromolecular Chemistry II, University of Bayreuth, Germany; [2]Department of Microbiology and Molecular Genetics, The Kuvin Center for the Study of Infectious and Tropical Diseases, The Hebrew University of Jerusalem, Israel

E-mail: greiner@uni-bayreuth.de

Programmed delivery of hydrophobic drugs from nano-carriers and its clinical translation is still fraught with many limitations and therefore an unresolved challenge. We outperform the limitations by the use of polymer nanofiber nonwovens (NFN) prepared by electrospinning technique which has been utilized in the past for immobilization of several drugs in NFN [1,2].

We conducted a fundamental study on drug delivery of artemisone (ART) from NFN, following a new paradigm based on suitable ART-loaded NFN, demonstrating a reliable release from an infusion system for the therapy of malaria (and other pathogens that are sensitive to artemisinins). In our system, the adjustment of drug release is achieved outside physiological environment by preparation of ART-loaded NFN which can release ART within the infusion system. The release of ART could be programmed by choice of a hydrophobic polymer for the electrospinning NFN, the amount of drug loaded NFN, the rate of infusion, and composition of the infusion medium [3]. ART loaded NFN integrated into an infusion system provides a convenient tool, fulfilling the clinical demands towards a programmable and easy-to-use administration of ART with improved stability of the encapsulated ART in the formulations, without actual patient contact. This system could be transferred to numerous other hydrophobic drugs and also be used when drug combinations are needed e.g. artemisinin drug combinations.

[1] S. Agarwal, J. H. Wendorff, A. Greiner, 2008. Polymer 49, 5603-5621
[2] X. Hu, S. Liu, G. Zhou, Y. Huang, Z. Xie, X. Jing, 2014. J. Controlled Release 185, 12-21
[3] A. R. Bagheri, S. Agarwal, J. Golenser, A. Greiner, 2017, global challenges, accepted

Imaging of experimental cerebral malaria using the liposomal, FDA approved indocyanine green

J Golenser[1], E Portnoy[2], N Vakruk[1], S Magdassi[3], S Eyal[2]

[1]Department of Microbiology and Molecular Genetics, The Kuvin Center for the Study of Infectious and Tropical Diseases, Faculty of Medicine, The Hebrew University of Jerusalem, Israel; [2]School of Pharmacy, Israel; [3]Casali Institute, Institute of Chemistry and Center for Nanoscience and Nanotechnology, Israel

E-mail: Golenser@md.huji.ac.il

Cerebral malaria (CM) is a severe complication and a leading cause of death by *Plasmodium falciparum* infection. CM is the result of interrelated events: mechanical obstruction due to parasite sequestration in the microvasculature and up-regulation of Th1 immune responses. In parallel, blood-brain-barrier (BBB) breakdown, and damage or death of microglia, astrocytes and neurons occur. CM is diagnosed by presence of parasites and neurological dysfunction. Misdiagnosis often leads to treatment delay and mortality. Neuroimaging has been used only in elucidation of CM mechanism in experimental animal models.

We used the following methods: synthesis of liposomal indocyanine green (ICG); continuous imaging (Ivis Kinetic®); experimental *P. berghei* ANKA-mouse model for CM (ECM); brain histology. Using ECM, based on the BBB breakdown, we attempted at ICG imaging for developing diagnostic method for CM. ICG is the only FDA-approved near-infrared (NIR) fluorescent dye for intravenous injection. However, its poor aqueous stability and short half-life in plasma limit its utility in diagnosing disease. We found enhanced emission intensity of liposomal ICG in comparison with free ICG, probably, owing to increased stability and to the dye's embedding within the liposomal bilayer in its monomeric form. The Liposomal ICG's emission was greater in the brains of sick mice compared to naïve mice and drug treated mice (where CM was prevented). Histological analyses suggest that the increased accumulation of ICG-liposomes in brain blood vessels is due to extensive cerebral uptake by activated phagocytes.

In conclusion, ICG characteristics, together with its known biocompatibility, make liposomal ICG a valuable delivery form for in vivo NIR imaging of CM. ICG-liposomes may be further developed as a valuable diagnostic tool and a biomarker for effectiveness of CM treatment, as well as other diseases that involve inflammation and blood vessel occlusion.

Successful Transnational EU funded Neglected Disease Drug Discovery projects

Sheraz Gul, Pasquale Linciano, Anabela Cordeiro-da-Silva, Rosaria Luciani, Gesa Witt, Bernhard Ellinger, Maria Kuzikov, Philip Gribbon, Jeanette Reinshagen, Markus Wolf, Nuno Sanarem, Stefania Ferrari, Ina Pöhner, Rebecca Wade, Lucio Freitas, Carolina Borsoi Moraes, Bruno S. Pascoalino, Vanessa Fontana, Ulrike Wittig, Wolfgang Müller, Stefano Mangani, M. Paola Costi, Rob Leurs, Iwan de Esch, Marco Siderius, Antoni Blaazer, Erik de Heuvel, Maarten Sijm, Geert Jan Sterk, Harry de Koning, David Bailey, Paul England, Fredrik Svensson, David Brown, Abhimanyu Singh, Susanne Schröder, Carmen Gil, Sanaa Botros, Naglaa El- Lakkany, Sayed Hassan Seif el-Din, Louis Maes, Irene Garcia Salado, Sarah Hendrickx, Koen Augustyns, Maria de Nazaré Correia Soeiro, Herman Verheij, Leonie Hussaarts, Kristina Orrling

Fraunhofer Institute for Molecular Biology & Applied Ecology – ScreeningPort, Hamburg, Germany

E-mail: Sheraz.Gul@ime.fraunhofer.de

Screening using miniaturised microtiter plate formats remains the most widely utilised methodology for identifying novel chemical starting points that are capable of modulating target function in a meaningful, biologically relevant manner. The initial screening Hits are optimised using multiple criteria including structure activity relationships, selectivity, physicochemical properties and liability that could result in a Lead compound series and eventually a clinical Candidate molecule. The compounds identified from this process are expected to possess appropriate physico-chemical properties, *in-vitro* off target liabilities, *in-vitro* toxicity and *in-vitro* ADME and safety profiles. This presentation will discuss the strategies that have been adopted by two EU funded translational neglected disease drug discovery projects:

NMTRYPI: The NMTrypI (New Medicines for Trypanosomatidic Infections) project aimed at obtaining new candidate drugs against Trypanosomatidic infections with appropriate efficiency from the lead phase to the final preclinical phase.

PDE4NPD: Phosphodiesterases (PDEs) have been studied extensively as drug targets, and various lead series against trypanosomes have already been identified and optimized in a previous project. The PDE4NPD consortium was established to further progress these compounds and to re-iterate the hit-finding process in order to identify new inhibitors for a broader range of neglected tropical diseases.

These projects have received funding from the European Union's Seventh Framework Programme for research, technological development and demonstration under grant agreement no. 603240 (NMTrypI - New Medicines for Trypanosomatidic Infections) and grant agreement no. 602666 PDE4NPD).

Aldehyde dehydrogenase, a potential drug target in Schistosoma

S. Häberlein[1], A. Blohm[1], P. Mäder[2], T. Quack[1], G. Lochnit[3], K. Becker[4], S. Rahlfs[4], M. Schlitzer[2], C.G. Grevelding[1]

[1]Institute of Parasitology, BFS, Justus Liebig University Giessen; [2]Institute of Pharmaceutical Chemistry, Philipps University Marburg; [3] Institute of Biochemistry, Justus Liebig University Giessen; [4]Biochemistry and Molecular Biology, Interdisciplinary Research Center, Justus Liebig University Giessen

E-mail: simone.haeberlein@vetmed.uni-giessen.de

Schistosomiasis is one of the most important tropical infectious diseases worldwide and caused by schistosome parasites. After decades of medication using praziquantel, there is upcoming fear of emerging resistance. In addition, no effective vaccine is available. Therefore, there is an urgent need to explore alternative treatment strategies.

To this end we searched for potential target molecules in the tegument of adult schistosomes. A proteomic approach indicated the presence of an aldehyde dehydrogenase (SmALDH) within the tegument. ALDHs are involved in metabolizing alcohol in humans, and Disulfiram is used as a drug inhibiting ALDH activity to treat alcoholism. In adult schistosomes, Disulfiram showed severe effects on egg production and tegument integrity *in vitro* as detected by light and confocal microscopy. Derivatives of Disulfiram were synthesized of which several induced severe phenotypes ranging from reduced gut peristalsis to the death of worms. An enzymatic activity assay indicated a concentration-dependent decrease of schistosomal ALDH activity with increasing Disulfiram concentration. *In situ* hybridization revealed SmALDH gene expression in the tegument and other tissues. Furthermore, we succeeded in expressing enzymatically active SmALDH. The recombinant protein is currently used for biochemical characterization studies and to screen the synthesized derivatives for their SmALDH-targeting potential.

Our findings suggest that ALDH is an interesting target molecule and may lead to promising alternatives not only for the control of schistosomiasis, but potentially also infections by other trematodes expressing ALDH-orthologs.

EIF-5A controls apoptotic damage of myocytes in experimental cerebral malaria

Annette Kaiser[1], Kirsten Heiß[2], Ann-Kristin Mueller[2], James Thujon Njuguna[3]

[1]Medical Research Centre, University Duisburg-Essen, Germany; [2]Centre for Infectious Diseases, Parasitology Unit, University Hospital Heidelberg, Germany; [3]Astel Diagnostics, Kampala, Uganda

E-mail: kaiser@microbiology-bonn.de

A determination of Troponin I and creatin kinase in whole-blood samples from a cohort of 100 small infants in the age of 2-5 years from Uganda with complicated *falciparum* malaria suggested the prevalence of impaired and damaged myocytes. It was recently reported that complicated malaria coincides with hypoxia in children. The obtained clinical data prompted us to further elucidate the underlying regulatory mechanisms of myocyte damage in a rodent model simulating cerebral malaria. Cerebral malaria remains the most common clinical presentation and might induce myocyte apoptosis by hypoxia. Eukaryotic initiation factor 5A (eIF-5A) is involved in hypoxia induced factor (HIF-1α) expression. EIF-5A is a small, acidic protein posttranslationally modified by hypusination involving catalysis of the two enzymes deoxyhypusine synthase and deoxyhypusine hydroxylase (DOHH) [1]. Treatment with GC7 [2], an inhibitor of deoxyhypusine synthase (DHS), the first step in hypusine biosynthesis in murine malaria decreased proinflammatory and proapoptotic myocardial caspase-1 activity in serum. Moreover, administration of GC7 reduced the release of cytochrome C from damaged mitochondria of myocytes. Lactate concentrations were reduced after inhibitor treatment due to a decreased glycolysis. In sum, administration of GC7 protected the infected mice from myocyte damage driven by hypoxia in cerebral malaria. This was furthermore supported by a reduced parasitemia and an extended survival rate of the treated animals. In sum we demonstrate involvement of EIF-5A in mitochondrial, apoptotic signaling pathways in a translational approach.

[1] Kaiser A, Gottwald A, Maier W, Seitz HM (2003) Targeting enzymes involved in spermidine metabolism of parasitic protozoa--a possible new strategy for anti-parasitic treatment. Parasitol Res. 91(6):508-516

[2] Lee YB, Park MH, Folk JE. (1995) Diamine and triamine analogs and derivatives as inhibitors of deoxyhypusine synthase: synthesis and biological activity. J Med Chem. 4;38(16):3053-3061

Developing a High Content Screening Assay to identify invasion and proliferation inhibitors of *Toxoplasma gondii*

Eric R. Kalkman[1], Susan Baillie[1], Matthew Gow[1], Paul Wyatt[2], Ian Gilbert[2], David Gray[2], Rich Elliott[3], Val Millar[4] and Markus Meissner[1]

[1]Wellcome Centre for Molecular Parasitology, Institute of Infection, Immunity and Inflammation, University of Glasgow, UK; [2]Drug Discovery Unit, Dundee, UK; [3]Bill & Melinda Gates Foundation; [4]Target Discovery Institute, Nuffield Department of Medicine, University of Oxford, UK

E-mail: Eric.Kalkman@glasgow.ac.uk

Toxoplasma gondii is a protozoan parasite and is one of the most common human parasite infections in the world. It can cause severe disease or death in immunocompromised patients, as well as severe congenital defects in prenatally infected infants [1]. Therefore understanding and seeking to control the mechanism(s) of invasion and parasite replication is crucial to the development of new means to combat *Toxoplasma*.

Due to their amenability to High Throughput assays, *in vitro* models are invaluable for studying mechanisms of host invasion, parasite conversion, and replication. We previously developed an siRNA High Content Screening (HCS) assay to investigate host kinome involvement in early stage *T. gondii* infection of HeLa cells (unpublished).

Based on this screen we developed a small molecule HCS assay to investigate the parasite infection of human foreskin fibroblast cells and subsequent parasite replication.

Toxoplasma can only be cultured and replicated in host cells. Human foreskin fibroblasts (HFF) are commonly used for this [2, 3]. HFFs with their ease of use were therefore also chosen as the *in vitro* host cells for the assay.

HFF cells are seeded in 384 TC microplates on top of a pre-dispensed compound library and after a number of hours wildtype GFP-expressing *Toxoplasma* parasites are added on top of the adhered HFF cells. The parasites are left to invade and replicate within the HFF cells overnight after which the microplates are fixed and stained with Hoechst 33342 to stain the host nuclei. The infected HFF cells are then imaged using an IN Cell 2000 HCS microscope and analysed using an automated analysis protocol written in IN Cell Investigator software.

We will be screening the Global Health Chemical Diversity Library, made available by the Gates Foundation and designed by the Drug Discovery Unit to interact with a diverse range of biological targets.

[1] McFarland et al. 2016. Antimicrob Agents Chemother 60, 7017–7034
[2] Rosowski et al. 2011. J. Exp. Med. 208, 195-212
[3] Niedelman et al., 2013. Infect. and Immun. 81, 4341-4349

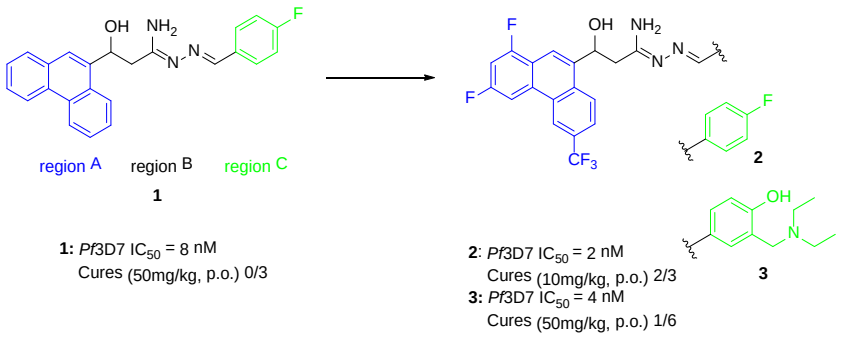
3-Hydroxy-*N'*-arylidenepropanehydrazonamides: Novel analogues of arylamino alcohols cure *Plasmodium berghei*-infected mice after peroral administration

T. C. Knaab[1], J. Held[2], M. Leven[1], B. Lungerich[1], S. Meister[3], C. Fischli[4], U. Lehmann[4], E. A. Winzeler[3], S. Wittlin[4], B. Mordmüller[2], T. Kurz[1]

[1]Institute of Pharmaceutical and Medicinal Chemistry, Heinrich-Heine-Universität Düsseldorf, Germany; [2]Institute of Tropical Medicine, Eberhard Karls Universität Tübingen, Germany; [3]Department of Pediatrics, School of Medicine, University of California, USA.; [4]Swiss Tropical and Public Health Institute, Switzerland

E-mail: tanja.knaab@hhu.de

The protozoan parasite *Plasmodium falciparum* caused approximately 212 new malaria cases in 2015 and 429 000 deaths globally [1]. Extensive efforts in prophylaxis and treatment steadily decreased incidence and prevalence from 2010 to 2015. Currently, artemisinin-based combination therapies (ACTs) are recommended by the WHO for the treatment of uncomplicated malaria infection in regions where chloroquine-resistant strains exist [2]. Unfortunately, emerging resistance to artemisinin and its derivatives are reported. This circumstance emphasises the urgent need for new antimalarial drugs. Previously, our group identified 3-hydroxy-*N*'-arylidenhydazonamides as potent *in vitro* antimalarial agents against the *P. falciparum* strains 3D7 and Dd2. The lead structure **1** shows promising *in vitro* activity in the nanomolar range [3]. Structure activity relationship studies revealed that the introduction of halogen substituents in region A provided new analogues with improved activity. However, the combination of the recently established halogen substitution and the 4-fluoro-phenyl (**2**) caused a low water solubility. Therefore, optimisation of region C was carried out by mannich type reactions, which led to compound **3**. These new compounds showed excellent antimalarial activity and cured *P. berghei*-infected mice in the standard Peters test.



[1] World Health Organization. World Malaria Report 2016
[2] Guidelines for the treatment of malaria, 3rd edition, 2015; World Health Organization
[3] Leven, M. et al., J. Med. Chem. 2014, 57, 7971−7976

Clinvet South Africa and Morocco: two contract research organisations offering unique opportunities for drug development and research

M. Madder[1], J. Fourie[1], T. Schetters[1]

Clinvet International, South Africa

E-mail: maxime.madder@clinglobal.com

Clinvet is a contract research organisation (CRO) based in Bloemfontein, South Africa and established in 1999. It specializes in conducting laboratory and field-based clinical trials to evaluate the safety, metabolism and efficacy of veterinary health products. A group of affiliated companies enables Clinvet to provide a wide range of services in support of laboratory and clinical trials performed on veterinary health products. With a wealth of experience, Clinvet has for many years been able to support leading veterinary health pharmaceutical in ground-breaking research and development programs. Conducting pivotal trials required for the registration of health products worldwide also forms part of Clinvet's portfolio.

With the new facilities based in Mohammedia, Morocco, which will be officially opened April 2017, Clinvet significantly increases its capacity to perform laboratory and field-based trials both on companion and production animals.

This presentation gives an overview of the opportunities Clinvet South Africa and Morocco offers for the scientific and pharmaceutical world.

First name	Last name	Institution	E-mail
José Maria	Alunda	Universidad Complutense de Madrid	jmalunda@vet.ucm.es
Sabine	Bachmaier	LMU München	sabine.bachmaier@lrz.uni-muenchen.de
Amir Reza	Bagheri	University of Bayreuth	amir.bagheri@uni-bayreuth.de
Michael	Boshart	LMU München	boshart@lmu.de
Collette	Britton	University of Glasgow	Collette.Britton@glasgow.ac.uk
Theodora	Calogeropoulou	National Hellenic Research Foundation Athens, Greece	tcalog@eie.gr
Anita	Cohen	Aix-Marseille Université	Anita.COHEN@univ-amu.fr
Maria	Costi	University of Modena and Reggio Emilia	mariapaola.costi@unimore.it
Manu	De Rycker	Drug Discovery Unit, University of Dundee	m.derycker@dundee.ac.uk
Koen	Dechering	TropIQ Health Sciences	k.dechering@tropiq.nl
Christian	Epe	Boehringer Ingelheim Animal Health, Duluth	christian.epe@merial.com
Martin	Folger	Boehringer Ingelheim Vetmedica GmbH	Martin.folger@boehringer-ingelheim.com
Reinhard	Forberger	Boehringer Ingelheim Animal Health GmbH	reinhard.forberger@boehringer-ingelheim.com
Julien	Furrer	University of Bern	julien.furrer@dcb.unibe.ch
Michael	Gaßel	Boehringer Ingelheim Veterinary Research Center GmbH & Co. KG	michael.gassel@boehringer-ingelheim.com
Jacob	Golenser	The Hebrew university of Jerusalem	Golener@md.huji.ac.il
Christoph	Grevelding	Justus Liebig University Giessen	Christoph.Grevelding@vetmed.uni-giessen.de
Sheraz	Gul	Fraunhofer IME-SP	sheraz.gul@ime.fraunhofer.de
Jurgen	Haanstra	Vrije Universiteit Amsterdam	j.r.haanstra@vu.nl
Helmut	Haas	*helmin*Guard	hhaas@helminguard.de
Simone	Häberlein	Justus Liebig University Giessen	Simone.Haeberlein@vetmed.uni-giessen.de

First name	Last name	Institution	E-mail
Kristina	Haeussler	Justus Liebig University Giessen	Kristina.Haeussler@ernaehrung.uni-giessen.de
John	Harrington	Boehringer Ingelheim Animal Health, Duluth	john.harrington@Merial.com
Andrew	Hemphill	University of Bern	andrew.hemphill@vetsuisse.unibe.ch
Thomas	Ilg	Bayer Animal Health GmbH, Leverkusen	thomas.ilg@bayer.com
Annette	Kaiser	Medical Research Centre, Bonn	Kaiser@microbiology-bonn.de
Eric	Kalkman	Wellcome Centre for Molecular Parasitology, University of Glasgow	Eric.Kalkman@glasgow.ac.uk
Ronald	Kaminsky	Para.C-Consulting for Parasitology and Drug Discovery	Para.C@gmx.de
Asaad	Khalid	Jazan University, Saudi Arabia	drasaad@gmail.com
Tanja Corinna	Knaab	Heinrich-Heine-Universität Düsseldorf	Tanja.Knaab@hhu.de
Friederike	Knapp-Lawitzke	Freie Universtität Berlin	Friederike.knapp-lawitzke@fu-berlin.de
Oliver	Koch	Technische Universität Dortmund	oliver.koch@tu-dortmund.de
Christoph	Krull	Freie Universtität Berlin	Christoph.Krull@fu-berlin.de
Rita	Kwoczalla	Bayer Animal Health GmbH	rita.kwoczalla@bayer.com
Gehan	Labib Elenain	Abu Dhabi University, UAE	gehan.elenain@adu.ac.ae
Loïc	Le Hir de Fallois	Boehringer Ingelheim Animal Health, Duluth	Loic.LeHir@Merial.com
Pasquale	Linciano	University of Modena and Reggio Emilia	p.linciano@unimore.it
Maxime	Madder	ClinGlobal, Oosterhout, NL	Maxime.madder@clinglobal.com
Marie	Miglianico	TropIQ Health Sciences	m.miglianico@tropiq.nl
Daria	Monaldi	University of Freiburg	daria.monaldi@pharmazie.uni-freiburg.de
Jeremy	Mottram	University of York	jeremy.mottram@york.ac.uk
Sandra	Noack	Boehringer Ingelheim Animal Health GmbH	sandra.noack@boehringer-ingelheim.com

First name	Last name	Institution	E-mail
Alec	O'Keeffe	London School of Hygiene and Tropical Medicine	Alec.o'keeffe@lshtm.ac.uk
Jan	Perner	Czech Academy of Sciences, České Budějovice	perner@paru.cas.cz
Alejandra	Peter	University of Marburg	Peteral@students.uni-marburg.de
Hedvika	Primasová	University of Bern	hedvika.primasova@dcb.unibe.ch
Georg	Rennar	University of Marburg	georg.rennar@pharmazie.uni-marburg.de
Heinz	Sager	Elanco Animal Health, Basel	heinz.sager@elanco.com
Eva-Maria	Schäfer	University of Marburg	schaef62@staff
Annette	Schlosser	Bernhard Nocht Institute for Tropical Medicine, Hamburg	annettebeatrix@gmail.com
Sandra	Schorderet-Weber	Neuchâtel, Switzerland	sandra.schorderet@hotmail.com
Frank	Seeber	Robert Koch-Institut, Berlin	SeeberF@rki.de
Paul	Selzer	Boehringer Ingelheim Animal Health GmbH	paul.selzer@boehringer-ingelheim.com
Ngoc	Thanh Truong	Bayer Animal Health GmbH	
Fiona	Tomley	University of London	ftomley@rvc.ac.uk
Georg	von Samson-Himmelstjerna	Freie Universtität Berlin	gvsamson@fu-berlin.de

We would like to thank the sponsors of the 18th Drug Design & Development Seminar of the German Society for Parasitology (DGP) for their support:

Boehringer Ingelheim Animal Health GmbH
Binger Straße 173
55216 Ingelheim am Rhein
Germany
www.boehringer-ingelheim.de

Jung-Stiftung für Wissenschaft und Forschung
Elbchaussee 215
22605 Hamburg
Germany
www.jung-stiftung.de

Merck KGaA
Frankfurter Straße 250
64293 Darmstadt
Germany
www.merck.de

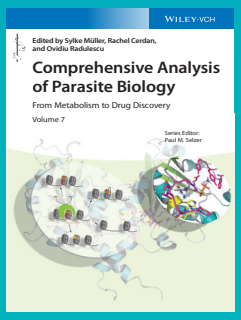
Looking for the **right resources?**

- Update your library today

20% Discount

– Order now to receive your discount*

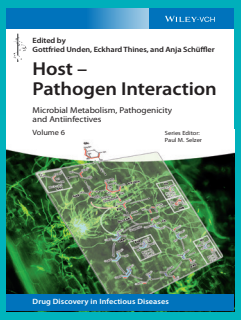
*Use promotion code **DDDS7** to receive your discount!



Comprehensive Analysis of Parasite Biology
From Metabolism to Drug Discovery

Sylke Müller (Editor),
Rachel Cerdan (Editor),
Ovidiu Radulescu (Editor),
Paul M. Selzer (Series Editor)

ISBN: 978-3-527-33904-4
576 pages • October 2016

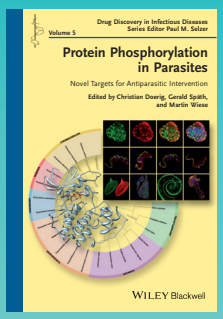


Host - Pathogen Interaction
Microbial Metabolism, Pathogenicity and Antiinfectives

Gottfried Unden (Editor),
Eckhard Thines (Editor),
Anja Schuffler (Editor),
Paul M. Selzer (Series Editor)

ISBN: 978-3-527-33745-3
240 pages • September 2016

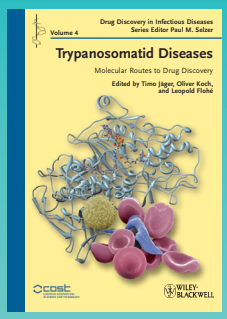
Also Available Online



Protein Phosphorylation in Parasites
Novel Targets for Antiparasitic Intervention

Christian Doerig (Editor),
Gerald Spaeth (Editor),
Martin Wiese (Editor),
Paul M. Selzer (Series Editor)

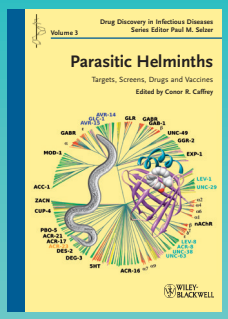
ISBN: 978-3-527-33235-9
456 pages • January 2014



Trypanosomatid Diseases
Molecular Routes to Drug Discovery

Timo Jager (Editor),
Oliver Koch (Editor),
Leopold Flohe (Editor),
Paul M. Selzer (Series Editor)

ISBN: 978-3-527-33255-7
576 pages • May 2013



Parasitic Helminths
Targets, Screens, Drugs and Vaccines

Conor R. Caffrey (Editor),
Paul M. Selzer (Series Editor)

ISBN: 978-3-527-33059-1
540 pages • August 2012

Go to **www.wiley.com** to find out more

27106

WILEY-VCH

WILEY