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Welcome Address of the German Research Platform for Zoonoses

Dear colleagues,

We are delighted to welcome you to the **National Symposium on Zoonoses Research 2017** to celebrate with us 10 years of joint zoonoses research in Germany.

Over the past ten years, the research on zoonotic diseases in Germany has increased considerably, with the start of the nine research networks on zoonoses funded by the Federal Ministry of Education and Research (BMBF) in 2007 forming the basis for the later foundation of the German Research Platform for Zoonoses. The latter continues to thrive to optimize both interdisciplinary research networking and collaboration as well as to close the gap between research and practical application, forging links between representatives from all relevant sides, all within the meaning of the One Health concept. In regard to the latter, it is our great pleasure to have one of the main pioneers of this vital concept, Prof. Dr. Zinsstag, as one of our keynote speakers among us. To add to the diversity and internationality of your symposium, we're also proud to announce Prof. Dr. Mika Salminen from Finland and Prof. Dr. Keizo Tomonaga from Japan as well Prof. Dr. Simone Sommer as a German representative as part of this year's expert keynote speaker assembly.

This year's program structure will not only represent current exciting research results and projects; by introducing the different members of the new *Forschungsnetz Zoonotische Infektionskrankheiten* (funded by the BMBF), it will also allow an insight into the joint future of national zoonoses research in Germany. Furthermore, this insight will be completed by presentations of research consortia funded by other ministries. To ensure the event is accessible to everyone, some segments will be held in English and others in German.

The Young Scientists Breakfast will take place on Friday morning. Early-stage researchers are invited to take advantage of this relaxed

setting to talk with experienced colleagues about possible career paths and other topics.

We would like to take this opportunity to thank all those, both past and present, who in the last years have submitted abstracts, and prepared posters and presentations, some of whom have travelled a great distance to join us. All of you have made a positive contribution to the success of our symposia.

We hope you enjoy and benefit from the *National Symposium on Zoonoses Research 2017*.

Martin H. Groschup
(Greifswald, Germany)

Stephan Ludwig
(Münster, Germany)

Sebastian C. Semler
(Berlin, Germany)

Directors of the German Research Platform for Zoonoses

Welcome Note of the Federal Government

Grußwort der Bundesregierung zum

National Symposium on Zoonoses Research 2017
10 Jahre vernetzte Zoonosenforschung

Zoonotische Infektionskrankheiten verursachen weltweit bedeutende Gesundheitsprobleme. Um wirksame Behandlungs- und Präventionsansätze entwickeln zu können, müssen verschiedene wissenschaftliche Disziplinen, voran die Human- und Veterinärmedizin intensiv zusammenarbeiten. Damit dies gelingt, haben das Bundesministerium für Bildung und Forschung (BMBF), das Bundesministerium für Gesundheit (BMG) und das Bundesministerium für Ernährung und Landwirtschaft (BMEL) bereits im März 2006 die Forschungsvereinbarung zur Erforschung von Krankheiten geschlossen, die von Tieren auf Menschen übertragbar sind. Damit begann die gezielte Förderung der Zoonosenforschung in Deutschland.

Ziel der Bundesförderung war und ist es, die unterschiedlichen Forschungszweige zu vernetzen und Strukturen für die interdisziplinäre Zusammenarbeit aufzubauen, zu fördern, zu stärken und international sichtbar zu machen. Unter diesem Leitgedanken fand auch das erste Nationale Symposium für Zoonosenforschung im Jahr 2007 in Berlin statt. Es bildete den Auftakt des seither jährlich durchgeführten Symposiums, das in diesem Jahr sein zehnjähriges Jubiläum begeht und mittlerweile zu einem festen Termin für Zoonosenforscherinnen und -forscher in Deutschland geworden ist. Von dem Symposium profitiert insbesondere auch der wissenschaftliche Nachwuchs, der sich hier in einem komplexen Forschungsfeld orientieren kann.

Zehn Jahre vernetzter Forschung haben bereits viel bewegt. Dennoch entwickeln sich die Herausforderungen stetig weiter. Die Erkenntnis, dass die Gesundheit von Mensch und Tier sowie auch der Umwelt außerordentlich eng miteinander verwoben ist, fordert die Wissenschaft auf, alte Erkenntnisse neu zu bewerten und neue Wege zu gehen, um dem Anspruch des „One Health“-Konzepts gerecht zu werden. Für uns bedeutet dies, die Erforschung von

Infektionserregern, die bei Mensch und Tier vorkommen und die über die Umwelt oder Lebensmittel übertragen werden, auszurichten. Die Politik hat dies erkannt. Anfang 2016 wurde deshalb die Forschungsvereinbarung zu Zoonosen erneuert und auf das „One Health"-Konzept ausgerichtet. Als viertes Bundesministerium beteiligt sich seither auch das Bundesministerium der Verteidigung (BMVg) an der Vereinbarung. Dies unterstreicht den hohen Stellenwert der Zoonosenforschung für die Bundesregierung.

„One Health" ist eines der zentralen Themen der Globalen Gesundheit – besonders auch beim Kampf gegen antimikrobielle Resistenzen. In der Abschlusserklärung des G20-Treffens im Juli 2017 in Hamburg riefen die Staats- und Regierungschefs zur Bildung einer neuen, internationalen Plattform auf. Sie soll die Zusammenarbeit im Bereich Forschung und Entwicklung verbessern und gemeinsame Projekte zur Entwicklung von neuen Therapeutika und Diagnostika für Infektionskrankheiten fördern. Dabei stehen vor allem die von der WHO identifizierten Pathogene mit dem höchsten Forschungsbedarf für neue Antibiotika und die Tuberkulose im Fokus.
Das vom BMBF mit erheblichen Mitteln neu gegründete „Forschungsnetz zoonotische Infektionskrankheiten" wird einen wichtigen Beitrag zur Umsetzung des „One Health"-Ansatzes leisten. Es begann im Juli 2017 mit seiner Arbeit unter dem Motto „Forschung trifft Praxis". Die enge Verzahnung mit den öffentlichen Gesundheitsdiensten wird seine Forschungserfolge schnell und gezielt in die Anwendung bringen.

Wir wünschen damit allen Teilnehmerinnen und Teilnehmern ein spannendes Symposium, interessante Gespräche und zahlreiche neue Ideen für gemeinsame Projekte.

Federal Ministry of Education and Research (BMBF)

Federal Ministry of Health (BMG)

Federal Ministry of Food and Agriculture (BMEL)

Federal Ministry of Defense (BMVg)

Program

Thursday, October 12, 2017

08:00	**Registration (Poster Mounting)**
10:00 – 12:00	**Plenary Session I: Keynotes (Room Ballsaal)** Chair: *Stephan Ludwig*
10:00	**Opening Remarks: 10 Years of Interdisciplinary Zoonoses Research in Germany** Stephan Ludwig, Münster, Germany **Welcome Note of the Federal Government** Andrea Spelberg, BMBF, Berlin, Germany
10:30	**Keynote 1:** **Ecological and Genomic Drivers of Zoonotic Infections** Simone Sommer, Ulm, Germany
11:15	**Keynote 2:** **Global Health Security Agenda: Antimicrobials, Zoonoses und Biosafety/Biosecurity** Mika Salminen, Helsinki, Finland
12:00	*Lunch and Poster Viewing*

14:00 – 15:30 Session 1: Novel Methods, Diagnostics and NGS
(Room Zehlendorf)
Language: English
Chairs: *Martin H. Groschup and Claudia Kohl*

14:00 **Ancient viral DNA: a systematic feasibility study**
V. Schuenemann, **S. Calvignac-Spencer**

14:15 **Hybridization capture as a paleovirological tool for the detection of recent filoviral integrations in potential hosts' genomes**
A. Düx, J. Gogarten, L. Müller, F. Leendertz, S. Calvignac-Spencer

14:30 **Molecular typing of Listeria monocytogenes in foodstuffs to combat human listeriosis in Germany**
S. Lüth, S. Kleta, A. Flieger, S. Halbedel, R. Prager, R. Merle, T. Alter, S. Al Dahouk

14:45 **Advanced characterization of Cowpox virus infection in a human skin equivalent-based 3D model**
M. Neumann, R. Koban, F. Groeber, H. Walles, H. Ellerbrok

15:00 **A novel approach to tackle respiratory pathogens responsible for great ape population declines**
L.V. Patrono, **S. Calvignac-Spencer**, F.H. Leendertz

15:15 **Identification of biomarkers of zoonotic pathogens by ORFeome phage display**
M. Hust, **G. Moreira**

14:00 – 15:30 Session 2: Pathogen-cell interaction (Room Steglitz)
Language: English
Chairs: *Martin Beer and Jonas Fuchs*

14:00 **Phosphorylation of Tripartite motif containing 28 (TRIM28) during HPAIV infection occurs via the stress-activated protein kinase pathway p38-MSK1**
T. Krischuns, C. Nordhoff, S. Ludwig, L. Brunotte

14:15 **Endogenous Borna-like N elements in shrews**
D. Nobach, J. Müller, S. Herzog, M. Eickmann, C. Herden

14:30 **Infection of differentiated swine airway epithelial cells by influenza A viruses of different species origin**
D.-L. Shin, N-H. Wu, W. Yang, F. Meng, G. Herrler

14:45 **Comparative loss of function screens reveal common pathways required by Paramyxoviridae and Pneumoviridae**
K. Pfeffermann, D.E. Anderson, S.Y. Kim, B. Sawatsky, P. Duprex, M.A. Garcia-Blanco, V. von Messling

15:00 **The role of Salmonella Pathogenicity Island 2 (SPI2) in the course of neonatal non typhoidal Salmonella infections**
K. van Vorst, A. Dupont, K. Zhang, C. Pfarrer, U. Repnik, G. Griffiths, M. Hensel, P. Valentin-Weigand, M. Hornef, M. Fulde

15:15 **Hypoxia induces dormancy in *Coxiella burnetii***
F. Fischer, J. Schulze-Lührmann, I. Hayek, S. Wirtz, J. Jantsch, **A. Lührmann**

14:00 – 15:30 Session 3: Antimicrobial Use and Resistance (Room Ballsaal)
Language: English
Chairs: *Birgit Walther and Denise Rabold*

14:00 **MIC distributions for glyphosate in farm animal-associated Enterobacteriaceae determined by the broth microdulition method**
K. Bote, J. Poeppe, O. Makarova, U. Roesler

14:15 **Traceability of MRSA in the pork food chain and survival in meat products**
A. Fetsch, B. Ballhausen, S. Rose-Meierhöfer, D. Bade, G. Arendt, A.-C. Brück, M. Ebert

14:30 **ESBL-/AmpC-producing Enterobacteriaceae in broiler fattening farms after cleaning and disinfection – identification of critical control points**
K. Daehre, C. Robé, A. Blasse, A. Friese, U. Roesler

14:45 **Use of antimicrobial peptides for the reduction of multi-resistant pathogenic bacteria and prevention of biofilm formation in dairy processing**
S. Kersting, A.E.M. Schmidt, K. Rapsch, J. Assmann, A. Bethe, L.H. Wieler, C. Fidelak, T. Janßen, M. Tadros, E. Ehrentreich-Förster, M. von Nickisch-Rosenegk

15:00 **Colonisation of broiler chickens with ESBL-/AmpC- producing E. coli using a seeder-bird model and detection of in vivo transformants**
C. Robé, A. Blasse, K. Daehre, U. Roesler, S. Guenther

15:15 **Multispecies and clonal dissemination of OXA-48 type carbapenemase in**

Enterobacteriaceae from clinical samples of animals (2009-2016)
S. Pulss, I. Stamm, E. Prenger-Berninghoff, S. Göttig, I. Stolle, C. Heydel, T. Semmler, C. Ewers

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15:30 *Coffee Break and Poster Viewing*

16:00 – 17:30 **Plenary-Session II: Vielfältige Zoonosenforschung in Deutschland (Room Ballsaal)**
Language: German
Chairs: *Martin H. Groschup and Stephan Ludwig*

16:00 **Forschungs-Ergebnisse der Arbeiten aus BMEL-geförderten Zoonosenprojekten**

BMEL-Verbund: Culi-Mo: Monitoring of Mosquitoes in Germany
Helge Kampen

BMEL-Verbund: Culi-Fo: Research on Mosquitoes in Germany
Egbert Tannich, Hamburg, Germany

BMEL-Verbund: Tubercolosis
Karin Schwaiger, Munich, Germany

16:30 **Neuroborreliose-Surveillance bei Kindern und Erwachsenen; Infektionsprävalenzen und -inzidenzen in der Allgemeinbevölkerung (BMG-Förderung)**
Hendrik Wilking, Berlin, Germany

16:45 **Zoonosenforschung unter dem Dach des BMVg**
Sabine Sauer

17:00 **One Health-Initiative der Bundesinstitute**
Mathias Niedrig, Berlin, Germany

17:15 Discussion

17:30 – 19:30 **General Assembly German Research Platform for Zoonoses (Room: Ballsaal)**
Language: German
Chair*: Sebastian C. Semler*

20:00 ***Welcome Reception/Social Dinner*** **(Room: Ballsaal)**

20:45 **100.000 € für den Beweis der Existenz des Masernvirus - im Ernst?**
David Bardens, Hudiksvall, Sweden

Friday, October 13, 2017

07:30 – 09:00 **Young Scientists Breakfast**
(Room: Restaurant)

09:00 – 10:30 **Plenary Session III: Netzwerk Zoonotische Infektionskrankheiten**
(Room Steglitz)
Language: German
Chairs: *Martin H. Groschup, Stephan Ludwig, Sebastian C. Semler*

09:00 **Das Netzwerk Zoonotische Infektionskrankheiten**
Vorstellung des Netzwerks durch den Sprecher
Christian Drosten, Berlin, Germany

09:05 **Kurz-Vorstellung der einzelnen Verbünde (à 5 Min.)**

#1Health-PREVENT - One Health Interventions to Prevent Zoonotic Spread of Antimicrobial Multidrug-Resistant Bacterial Microorganisms
Robin Köck, Münster, Germany

The Zoonotic Bornavirus Consortium (ZooBoCo)
Martin Beer, Greifswald – Insel Riems, Germany

Verbund Campylobacter – Preventing and combating Campylobacter infections: On track towards a One Health approach
Stefan Bereswill, Berlin, Germany

The RAPID consortium – Risk Assessment in Pre-pandemic Respiratory Infectious Diseases
Christian Drosten, Berlin, Germany

Q-GAPS: Q fever – GermAn Interdisciplinary Program for reSearch
Anja Lührmann, Erlangen, Germany

Strengthening public health by understanding the epidemiology of rodent-borne diseases (RoBoPub)
Rainer Ulrich, Greifswald – Insel Riems, Germany

TBENAGER: The German Tick-Borne Encephalitis Consortium
Gerhard Dobler, München, Germany

10:00 **Diskussionsrunde mit allen Nachwuchs-gruppen (20 Minuten)**

Persistence of Toxoplasma gondii: Mode of action of candidate anti-protozoals and the role of physiological heterogeneity in zoonotic infections
Martin Blume, Berlin, Germany

Ecology of emerging arboviruses - ARBOSPREAD
Sandra Junglen, Berlin, Germany

Vector biology of Aedes albopictus and eco-bio-social drivers for effective vector prevention & control in cooler ecoregions (AECO)
Ruth Müller, Frankfurt, Germany

Development of novel tools to study the zoonotic vector biology of Ixodes ricinus using CRISPR/Cas and artificial tick feeding systems
Ard Nijhof, Berlin, Germany

RNA-VIRT: Emerging RNA viruses and their interaction with the human and animal host
Imke Steffen, Hannover, Germany

Immunological requirements for protective vaccination against zoonotic infections
Asisa Volz, Munich, Germany

10:20 **Gemeinsame Diskussion mit den Koordinatorinnen und Koordinatoren**

10:30 *Coffee Break and Poster Viewing*

11:00 – 12:30 **Session 4: Risk Assessment, Epidemiology and Modelling**
(Room Steglitz)
Language: English
Chairs: *Sandra Eßbauer and Reimar Johne*

11:00 **Bushmeat hunting and zoonotic transmission of Simian T-lymphotropic virus 1 in tropical West and Central Africa**
G. Schubert, A. Mossoun, S. Calvignac-Spencer, C. Akoua-Koffi, E. Couacy-Hymann, J.J. Muyembe-Tamfum, S. Karhemere, R.M. Wittig, F.H. Leendertz

11:15 **Characteristics profiles of cefotaxime-resistant *E. coli* from German livestock farms and potential association with farm factors**
K. Hille, M. Felski, I. Ruddat, J. Woyd, A. Schmid, A. Friese, J. Fischer, L. Falgenhauer, S. Hörmansdorfer, A. Käsbohrer, U. Rösler, L. Kreienbrock

11:30 **Hantavirus infections as a cause of fever of unknown origin in Kazakhstan**
N. Tukhanova, A. Shin, K. Abdiyeva, N. Turebekov, S. Frey, R. Yegemberdiyeva, L.

Yeraliyeva, Z. Shapieva, G. Fröschl, G. Zhumabaeva, A. Zhalmagambetova, S. Essbauer

11:45 **Lyme borreliosis in Germany: description of surveillance data 2013-2016**
J. Enkelmann, H. Wilking, M. Böhmer, V. Fingerle, C. Siffczyk, D. Werber, M. Littmann, S.-S. Merbecks, C. Helmeke, S. Schroeder, S. Hell, U. Schlotthauer, F. Burckhardt, A. Schielke

12:00 **Spatial Distribution and Risk Areas in Germany – Results of the National Database for Alveolar Echinococcosis**
J. Schmidberger, T. Gräter, T.F.E. Barth, W. Kratzer, B. Grüner

12:15 **Spread of human dirofilariasis in Europe**
R. Lühken, E. Tannich

11:00 – 12:30 Session 5: New and emerging zoonoses (Room Ballsaal)
Language: English
Chairs: *Martin Pfeffer and Rainer Ulrich*

11:00 **Pathogenesis and transmission of the novel highly pathogenic avian influenza H5N8 2016 virus in ferrets and mice**
D. Hoffmann, M. Naguib, T.C. Harder, C. Grund, M. Beer

11:15 **Experimental transmission of Zika virus by mosquitoes from Central Europe**
A. Heitmann, **S. Jansen**, R. Lühken, M. Leggewie, M. Badusche, B. Pluskota, N. Becker, O. Vapalahti, J. Schmidt- Chanasit, E. Tannich

11:30 **Hepeviruses in small mammals: RabbitHEV in two populations in and around Frankfurt/ Main, Germany**
R. Ryll, M. Eiden, E. Heuser, M. Weinhardt, M. Ziege, M.H. Groschup, R. Johne, R.G. Ulrich

11:45 **The impact of polymorphisms within the Ebola virus glycoprotein on host cell entry**
M. Hoffmann, L. Crone, E. Dietzel, J. Paijo, M. González Hernández, I. Nehlmeier, U. Kalinke, S. Becker, S. Pöhlmann

12:00 **Multiple detection of zoonotic variegated squirrel bornavirus 1 in different squirrel species**
K. Schlottau, B. Hoffmann, R.G. Ulrich, K. Franzke, M. Beer, D. Hoffmann

12:15 **Diverse novel orthobunyaviruses detected in sylvatic mosquitoes of the Panama Canal Zone**
M. Marklewitz, K. Hermanns, J. Schmid, G. Eibner, F. Zirkel, J.R. Loaiza, P. Trippner, S. Sommer, C. Drosten, S. Junglen

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11:00 – 12:30 Session 6: Public Health and Social Issues of Zoonoses Research (Room Zehlendorf)
Language: English
Chairs: *Uwe Rösler and Shari Fell*

11:00 **Comparison and optimization of methods for virus detection in frozen berries**
C. Bartsch, E.Trojnar, R. Johne

11:15 **Full genomes of new HEV and HPgV strains, Sudan**
T. Muzeniek, C. Kohl, M. Eldegail, A. Radonic, I. Mahmoud, A.A.Osman, A. Nitsche

11:30 **Zika virus infection is enhanced in the presence of dengue antibodies in human placenta explants**
K. Hermanns, C. Göhner, A. Kopp, A. Schmidt, W. Merz, U. Markert, C. Drosten, S. Junglen

11:45 **Interdisciplinary Communication in the Zoonoses Research Community in Germany**
A. Debski

12:00 **Haemolytic uraemic syndrome (HUS) outbreak caused by sorbitol-fermenting (SF) Shiga toxin-producing *Escherichia coli* (STEC) O157, Germany, December 2016 to May 2017**
S. Vygen-Bonnet, B. Rosner, H. Wilking, A. Fruth, R. Prager, A. Kossow, C. Lang, S. Simon, J. Seidel, M. Faber, A. Schielke, K. Michaelis, A. Holzer, R. Kamphausen, D. Kalhöfer, S. Thole, A. Mellmann, A. Flieger, K. Stark

12:15 **Evaluation of predisposing factors for a long-time colonization of pigs with livestock-associated MRSA via the airborne route in a newly established animal model**
K. Rosen, F. Ebner, A. Friese, S. Hartmann, U. Roesler

12:30 *Lunch and Poster Viewing*

14:30 – 16:00 Plenary Session IV: Keynotes (Room Ballsaal)
Chair: *Martin H. Groschup*

14:30 **Keynote 3:**
Bornavirus infection: a new model of evolution and coexistence of RNA viruses
Keizo Tomonaga, Kyoto, Japan

15:15 **Keynote 4:**
One Health – (Not) Just a Buzz-Word
Jakob Zinsstag, Basel, Switzerland

16:00 **Poster Awards**

16:20 **Farewell**

General Information

Date and Venue
October 12-13, 2017

Hotel Steglitz International
Albrechtstraße 2, 12165 Berlin
www.si-hotel.com

How to get there

The venue is accessible within 15 minutes from Berlin Tegel Airport and within 25 minutes from Berlin Schönefeld Airport. You can travel by car or by public transportation. A subway (U-Bahn) and a city railroad station (S-Bahn) are located in front of the hotel. The hotel features an indoor parking garage and offers special parking rates to hotel guests.

Public transportation:
From Berlin Tegel Airport: Take Bus 109 to "Zoologischer Garten". From there, take Subway U9 to "Rathaus Steglitz".

From Berlin Schönefeld Airport: Take Train S45 to "Schöneberg". From there, take Train S1 (direction "Wannsee") to "Rathaus Steglitz".

From Berlin Central Station: Take Train S5, 7 or 75 to "Zoologischer Garten". From there, take Subway U9 to "Rathaus Steglitz" or take Train RB14, S5, 7 or 75 from Berlin Ceentral station to "Friedrichstraße". From there take Train S1 to "Rathaus Steglitz".

Conference Languages
The official conference languages are English and German.

Steering Committee
Martin H. Groschup (Greifswald - Insel Riems)
Stephan Ludwig (Münster)
Sebastian C. Semler (Berlin)

Organization
Office of the German Research Platform for Zoonoses

Münster:
Stephan Ludwig
Friederike Jansen
Sebastian Sprengel

Greifswald - Insel Riems:
Martin H. Groschup
Nils Kley

Berlin:
Sebastian C. Semler
Ilia Semmler
Kerstin Splett

With kind support of
Stefan Fischer (Greifswald)
Juliane Gehrke (Berlin)
Björn Kaesz (Greifswald)
Patrick Wysocki (Greifswald)

Review Committee
Members of the Internal Advisory Board of the German Research Platform for Zoonoses in 2016-2017:

Anton Aebischer, Berlin, Germany
Martin Beer, Greifswald - Isle of Riems, Germany
Christian Drosten, Berlin, Germany
Sandra Eßbauer, Munich, Germany
Martin H. Groschup, Isle of Riems, Germany
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Jonas Schmidt-Chanasit, Hamburg, Germany
Karin Schwaiger, Munich, Germany
Sebastian C. Semler, Berlin, Germany
Rainer Ulrich, Riems, Germany
Birgit Walther, Berlin, Germany

Poster Award Committee
The poster awards are selected by the members of the Internal Advisory Board of the German Research Platform for Zoonoses.

Keynote Speakers
Mika Salminen, Helsinki, Finland
Simone Sommer, Ulm, Germany
Keizo Tomonaga, Kyoto, Japan
Jakob Zinsstag, Basel, Switzerland

Young Scientists Breakfast
The Young Scientists Breakfast is going to take place at the “Pavillon” room of the hotel on Friday, October 13, at 7:30 am.
The attending senior scientists are:

Stefanie Becker, Hannover, Germany
Robin Köck, Oldenburg, Germany
Reinhard Straubinger, Munich, Germany
Cornelia Silaghi, Isle of Riems, Germany
Asisa Volz, Munich, Germany

Lunch Set-up
Due to the capacity of the venue premises, lunch will be served in two consecutive shifts. Please exercise some patience while seating yourself accordingly.

Continuous Medical Education
The National Symposium on Zoonoses Research 2017 is registered for **6 CME points** of category A **per day** by the Berlin Chamber of Physicians (Ärztekammer Berlin). Please note that you will need one barcode label per day for the confirmation of participation.

Continuous Veterinary Education
The National Symposium on Zoonoses Research 2017 is registered for **12 hours (ATF-Stunden)** by the Federal Chamber of Veterinarians (Bundestierärztekammer). You will receive your certificate during the lunch break on the second day of the symposium.

Poster Presentations
Posters will be presented during both days of the conference. Poster presenters will obtain their poster number during registration process and are requested to refer to this booklet and the relevant bulletin on the blackboard to find the poster session and board number assigned to them. Please use the poster board with the designated number. Poster presenters are responsible to remove the posters at the end of the conference.

Oral Presentations
Oral presentations should be handed over on a common data carrier at the registration desk on Thursday, October 12, between 8.00 am and 1.00 pm. All session rooms will be equipped with a PC computer and a LCD projector. Apple computers are not available. Please make sure, that you use either a powerpoint or a pdf file format.

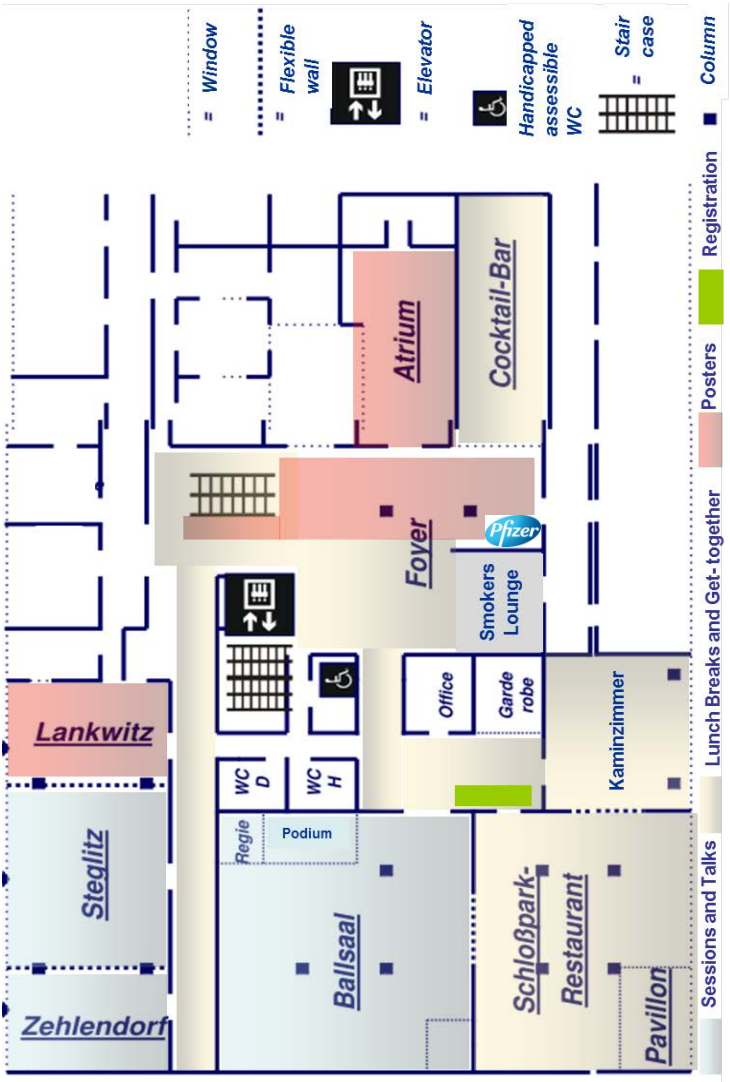
Internet Access
For internet access you are pleased to register at the registration desk. WLAN will be provided without charge.

Funding
The National Symposium on Zoonoses Research is funded by the Federal Ministry of Education and Research.

Sponsoring
The National Symposium on Zoonoses Research is kindly supported by:

Please feel free to take a look at the Pfizer booth in the foyer.

Floor Plan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Column
Registration
Posters
Lunch Breaks and Get-together
Sessions and Talks
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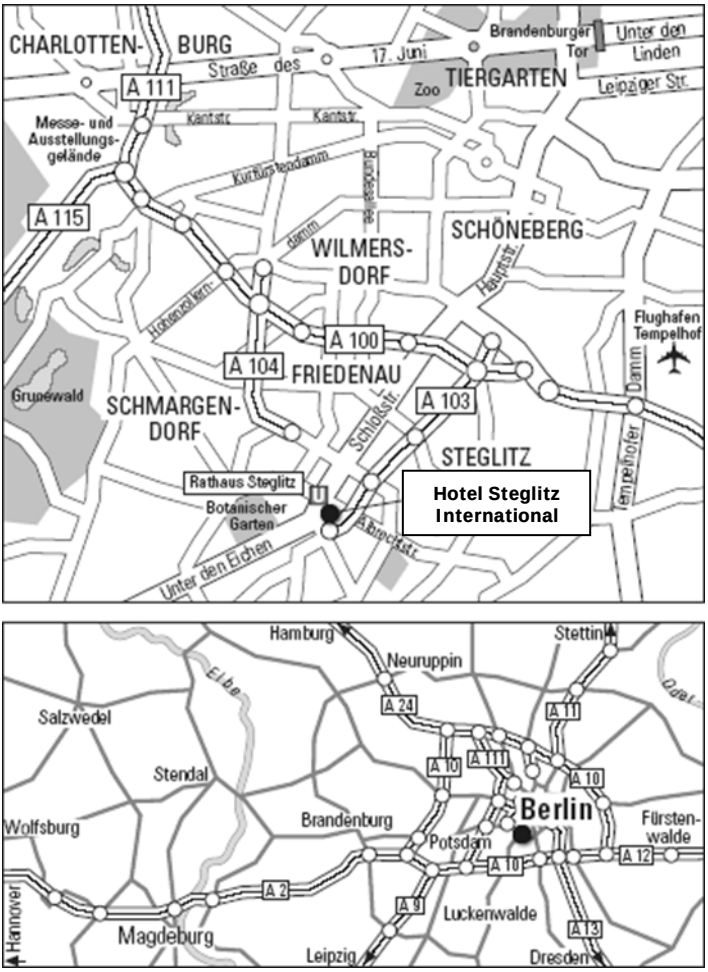
Zehlendorf
Ballsaal
Regie
Podium
WC D
WC H
Foyer
Atrium
Cocktail-Bar
Pfizer
Smokers Lounge
Office
Garde robe
Kaminzimmer
Schloßpark-Restaurant
Pavillon

Site Plan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Straße des 17. Juni
Brandenburger Tor
Unter den Linden
TIERGARTEN
Zoo
Leipziger Str.
A 111
Messe- und Ausstellungs-gelände
Kantstr.
Kantstr.
Kurfürstendamm
A 115
WILMERS-
DORF
SCHÖNEBERG
Hauptstr.
Flughafen Tempelhof
A 100
A 104
FRIEDENAU
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SCHMARGEN-
DORF
Schloßstr.
STEGLITZ
Rathaus Steglitz
Botanischer Garten
Hotel Steglitz International
Unter den Eichen
Albrechtstr.
Tempelhofer Damm
Hamburg
Stettin
Neuruppin
Elbe
Oder
Salzwedel
A 24
A 11
A 10
A 111
A 10
Stendal
Berlin
Wolfsburg
Brandenburg
Potsdam
Fürsten-walde
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Luckenwalde
A 13
Hannover
Magdeburg
Leipzig
Dresden

About the German Research Platform for Zoonoses

The German Research Platform for Zoonoses is a central information and service network, initiated and funded by the German Federal Ministry of Education and Research (BMBF) in 2009, for all working groups operating in Germany in the field of zoonoses research.

The objective of the platform and its currently over 700 members is to increase the exchange of professional experiences and knowledge at national and international levels and thus intensify research activities in the field of zoonoses research, promoting broad horizontal cross-linking of human and veterinary medicine as well as other scientific disciplines related to zoonotic disease research and public and veterinary health services. To develop and maintain sustainable and flexible solutions strengthening research, prevention and therapy of zoonotic infectious diseases in Germany, the Research Platform offers the following measures:

- Organization and realization of joint events that support interdisciplinary exchange and interaction.
- Encouragement of communication as well as national, European and international collaboration.
- Registration, harmonization and standardization of existing resources, including the setting up of both real and virtual specimen databases (i.e. the Database Internet Portal)
- Providing information about zoonotic infectious diseases for the general public
- Initiation and realization of innovative and interdisciplinary pilot projects of a cross-sectional nature
- Support and counseling for the design and implementation of zoonotic funding schemes
- Furtherance of junior scientists in the field of zoonosis research

Acting as a central service point that provides fact-oriented, transparent information relating to research on zoonoses both for politics and the general public, the German Research Platform aims to be the definite voice of German zoonosis research. Additionally,

the platform also promotes a continuous and intensive exchange of expertise between scientists from all over the world.

As part of these activities, the German Research Platform for Zoonoses organizes every year the National Symposium on Zoonoses Research with up to 350 participants.

Furthermore, scientific workshops, also for researchers at the beginning of their career, are organised, where specific topics are presented and discussed.

All researchers working on zoonoses in Germany are welcomed to join the German Research Platform for Zoonoses.

For further information please visit our website www.zoonosen.net.

Oral Presentations

Plenary Sessions

October 12, 2017
10:30 – 12:00
Room: Ballsaal
Chair: Stephan Ludwig

16:00 – 17:30
Room: Ballsaal
Chairs: Martin H. Groschup and Stephan Ludwig

and

October 13, 2017

9:00 – 10:30
Room: Ballsaal
Chairs: Martin H. Groschup, Stephan Ludwig and Sebastian C. Semler

14:30-16:00
Room: Ballsaal
Chair: Martin H. Groschup

Plenary Session I: Keynotes

Ecological and Genomic Drivers of Zoonotic Infections

S. Sommer[1], J. Schmid[1], A. Heni[1], S. Brändel[1], M. Tschapka[1], V. M. Corman[2], C. Drosten[2]

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Keywords: environmental change, virus infections, wildlife

Anthropogenic environmental change and loss of biodiversity has been shown to increase the infection prevalence in wildlife reservoirs and drive zoonotic diseases. However, despite recent advances in theoretical concepts and mathematical models, empirical data concerning the ecological and genomic drivers of infections in wild animal populations, especially from the tropics, remain scarce. We have studied small mammal and bat populations in three tropical landscapes in central Panama differing in their degree of human disturbance to test whether changes in species richness affect host population density, species ecology and virus prevalence. We furthermore investigated the effects of host adaptive (TLR, MHC) diversity on infection and resistance pattern to infer the impact of genomic constraints and reduced genetic diversity. Our study has revealed ecological and genomic mechanisms by which human-induced landscape change can have significant effects on infectious diseases.

Joint External Evaluation (JEE) in assessing the implementation of the International Health Regulations (2005) – a OneHealth approach to Global Health Security

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Keywords: IHR(2005), WHO, JEE

Background;
The Ebola epidemic of 2014-2015 in West Africa demonstrated that implementation of the International Health Regulations, IHR (2005) is severely lacking and that the current model of self-reporting by countries to the WHO often does not represent true operational capabilities. In 2015 WHO introduced a new model of peer-to-peer expert external evaluations, the Joint External Evaluation (JEE) and a score-based indicator data collection instrument (JEE Tool), for which voluntary implementation started in early 2016. The performance of the process and tools in accurately measuring IHR capacity and capability is not known, but would be important both for the acceptability of the process and for providing a basis for planning for improvement at country level.
Methods:
We postulated that, as the IHR core capacities which the JEE tool is intended to measure, represent a subset of essential public health functions, high JEE scores should correlate with a lower disease burden of communicable diseases and some other health outcomes, as well as indirectly with other indicators of social development. Similarly, developed economies should receive higher scores, if resources have been used for prevention. Scores for the 19 key technical areas in the JEE tool were collected from publicly available country reports and compared for their correlation with widely used publicly available socioeconomic, development, health service and health outcome indicators from the same countries.
Findings:
As expected, JEE scores varied substantially between countries. The JEE scores correlated strongly with major indicators of health and social development outcomes (i.e. life-expectancy, mortality rate and human development index). No correlation was found between JEE

scores and the level of official development assistance received by countries.

Interpretation: The JEE appears to adequately summarize the strength of a country's public health infrastructure. Our results also indicate that while there is a correlation between the economy of a country and the JEE scores, a fairly well-functioning health security system can still be achieved within a wide range of direct investments.

Plenary Session II: Vielfältige Zoonosenforschung in Deutschland

CuliMo – Mosquito monitoring in Germany

H. Kampen[1], N. Becker[2], M. Groschup[1], E. Kiel[3], S. Klimpel[4], A. Rose[5], J. Schmidt-Chanasit[6], E. Tannich[6], D. Walther[7]

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Keywords: Culicidae, Germany, monitoring

Prompted by the recent emergence of mosquito-borne diseases and invasive mosquitoes in southern Europe, monitoring activities were consolidated in Germany in 2015 in order to update data on the spatiotemporal distribution of mosquito species and circulating mosquito-borne pathogens. Mosquitoes were trapped in 2015 and 2016, each, at 148 sites in near-natural, rural and urban landscape structures within a given grid cell pattern. In addition, mosquitoes were passively collected by the citizen science project 'Mueckenatlas'. Using these approaches, some 120,000 mosquitoes belonging to 6 genera and 51 species were recorded, including several invasive species such as the Asian tiger mosquito. A considerable part of these as well as further specimens sampled in floodplains along rivers, where mosquitoes may come into contact with migratory birds serving as virus reservoirs and amplifiers, were subjected to a pathogen screening. Among 206,000 mosquitoes tested, 5 were found infected with Sindbis virus, 12 with Usutu virus and 17 with *Dirofilaria repens*. Furthermore, numerous mosquitoes showed PCR signals for yet uncharacterised mosquito flaviviruses and filarial worms.

The data collected in the framework of the CuliMo project, which will cover a third field season in 2017, are entered into the mosquito database CULBASE and are made available for preparing distribution maps and performing mosquito-borne disease risk analyses in the CuliFo project.

CuliFo – Mosquito Research in Germany

E. Tannich[1], N. Becker[2], F. Conraths[3], M. Groschup[3], H. Kampen[3], E. Kiel[4], S. Klimpel[5], J. Schmidt-Chanasit[1], D. Walther[6], R. Wieland[6]

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Keywords: Culicidae, Germany, research

In addition to CuliMo, a further research consortium has been established, designated CuliFo, that investigates specific questions relevant for risk assessment and control of mosquitoes and the transmission of mosquito-borne (mobo) viruses in Germany. Research includes (i) development of a data base on the genetic variability of German mosquito species, (ii) analysis of vector competences of abundant German mosquitoes for selected viruses, (iii) impact of autochthonous mobovirus infections in human and animals, (iv) risk assessment for establishment and/or further spread of new invasive mosquito species such as *Aedes albopictus* and *Aedes japonicus*, and (v) development of models and distribution maps predicting the risk for transmission of moboviruses in Germany. These models will include detailed microclimate and resting sites data. The CuliFo consortium entered into operation in 2016, one year after CuliMo had been initiated. First results indicate (i) high risk of permanent establishment of *Ae. albopictus* in Germany, (ii) vector competence of different German mosquito species for a number of non-endemic viruses, and (iii) widespread mobovirus circulation in Germany.

One Health-Initiative der Bundesinstitute (GOHI)

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According to the definition of the CDC, the One Health approach recognizes that the health of people is connected to the health of animals and the environment. The goal of One Health is to encourage the collaborative efforts of multiple disciplines – working locally, nationally, and globally – -to achieve the best health living conditions for people, animals, and our environment. Such an integrated approach is important because 6 out of every 10 infectious diseases in humans originate from animals.

To intensify the collaboration on projects of common One Health interest, the four German federal research institutes BfR, FLI, PEI, RKI, which are dedicated to different aspects of veterinary and human health, have started a concerted initiative for One Health in 2016. To foster close interactions between these institutes, eight PhD research projects were selected to investigate different aspects of zoonotic pathogens like Hepatitis E virus, *Toxoplasma gondii*, Ebolavirus, Mumps virus and - as part of Anti-microbial resistance (AMR) - *Klebsiella pneumoniae*.

The presentation will provide an overview on the current and planned activities of our German One Health Initiative (GOHI

Plenary Session III: Netzwerk Zoonotische Infektionskrankheiten

Das Netzwerk Zoonotische Infektionskrankheiten

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Mit Bekanntgabe vom 29. 1. 2016 hat das Bundesministerium für Bildung und Forschung (BMBF) zur Einreichung von Arbeitsvorschlägen für thematisch gruppierte Forschungsverbünde und Nachwuchsgruppen aufgerufen, die die Etablierung eines Nationalen Forschungsnetzes für Zoonotische Infektionserkrankungen ermöglichen sollen. Positive Förderentscheidungen wurden zu 7 Vorschlägen im Fördermodul „Verbünde“ und 6 Vorschlägen im Fördermodul „Nachwuchsgruppen“ getroffen. Hier wird kurz das Koordinationskonzept des Forschungsnetzes erläutert und in die Vorstellung der einzelnen Teilverbünde und Nachwuchsgruppen eingeführt.

#1Health-PREVENT - One Health Interventions to Prevent Zoonotic Spread of Antimicrobial Multidrug-Resistant Bacterial Microorganisms

R. Köck[1], A. Mellmann[1], K. Becker[1], S. van Alen[1], U. Kaspar[1], M. Boelhauve[2], A. Fetsch[3], B.-A. Tenhagen[3], B. Ballhausen[3], B. Walther[4], A. Lübke-Becker[5], W. Ziebuhr[6], S. Schoenfelder[6], A. T. Feßler[5], S. Schwarz[5], K. Kadlec[7], C. Cuny[4], W. Witte[4]

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Keywords: antibiotics, resistance, BMBF

Background and objectives: Multidrug-resistant microorganisms (MDRO) have emerged in livestock, humans and companion animals. This comprises a plethora of different bacteria including methicillin-resistant Staphylococcus aureus, extended-spectrum β-lactamase producing and/or carbapenemase-producing Enterobacteriaceae, colistin-, linezolid-, and daptomycin-resistant pathogens and commensals (e.g. enterobacteria, enterococci, coagulase-negative staphylococci).
Materials and methods: The consortium #1Health-PREVENT aims to perform epidemiological studies and interventional studies to prevent the zoonotic spread of various MDRO. The interventions focus on limiting direct transmission routes (e.g. barrier precautions, reducing environmental MDRO contamination, vaccination) as well as on primary preventive approaches (e.g. antibiotic stewardship). The interdisciplinary consortium comprises partners from veterinary and human medicine, public health and agriculture, which allows for establishing a research approach, truly tackling a novel One Health perspective.
Results and conclusion: The results of the consortium shall indicate concrete successful intervention strategies to limit zoonotic MDRO dissemination, which can be directly established as part of preventive bundles for humans (e.g. farmers, veterinarians), in veterinary clinics and livestock holdings. Moreover, #1Health-PREVENT will answer open epidemiological questions related to zoonotic antimicrobial resistance spread.

The Zoonotic Bornavirus Consortium (ZooBoCo)

M. Beer[1], R. G. Ulrich[2], B. Hoffmann[1], D. Höper[1], D. Hoffmann[1], C. Herden[3], H. Feldmann[4], M. Schwemmle[5], D. Rubbenstroth[5], D. Tappe[6], J. Schmidt-Chanasit[6], T. Homeier-Bachmann[7], H. Wilking[8], J. Rissland[9]

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Several deaths from infection by the novel squirrel bornavirus VSBV-1 were recently recorded in Germany. This unexpected incidence raises serious concerns about the zoonotic and pathogenic potential of VSBV-1 and related bornaviruses. The number of squirrels or humans currently infected with or exposed to potentially harmful bornaviruses is presently not known. It is also unclear whether only squirrels can transmit such viruses or whether a reservoir of other hosts exists that needs to be identified. Therefore, the “Zoonotic Bornavirus Consortium (ZooBoCo)” will provide urgently needed data for a better understanding of the zoonotic potential of these viruses, their distinctive features differentiating them from classical bornaviruses as well as their putative reservoirs and way of transmission. Furthermore, adequate intervention strategies will be explored. The consortium consists of members working in veterinary or human medicine, at universities, clinical research institutes and governmental institutions. Collaborations with international partners will allow performing risk assessment studies in non-human primates. The main goal of this „One health“-approach is to provide a solid basis for improved public health measures and guidelines helping to identify and handle pathogenic bornaviruses and their reservoir species. Finally, a diagnostic workflow building on metagenomics of encephalitis cases of unknown infectious etiology will be establishe

Verbund Campylobacter – Preventing and combating Campylobacter infections: On track towards a One Health approach

S. Bereswill[1], T. Alter[2] (On behalf of all consortium partners)

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The increasing number of disease cases in humans worldwide creates the need for the development of novel strategies for prevention, control and treatment of *Campylobacter* infections. In frame with the One Health concept, the goal of this project is to lower the incidence of human infections by a reduction of the *Campylobacter* burden in meat production. To achieve this, we are proposing several coordinated lines of research: in the first line, we will establish and implement intervention strategies along the poultry production chain in order to limit pathogen colonisation and spread in affected animals. Based on these data, a risk intervention model that simulates the impact of the specific interventions will be generated and tested in frame with the reduction of the *Campylobacter* load in chicken meat. These data are completed by integrative research activities focusing on the role of the environment for *Campylobacter* epidemiology by investigating farm emissions and by the study of pathogen survival strategies outside the host. In the second line, we will apply various *in vitro* assays and *in vivo* animal models to develop novel therapeutic approaches to combat *Campylobacter* infection and/or limit post-infectious sequelae in humans. The combined expertise of veterinary/ human medicine as well as environmental sciences and epidemiology in this One Health approach will enable us to tackle the multifaceted socioeconomic problems associated with *Campylobacter* infections.

The RAPID consortium – Risk Assessment in Pre-pandemic Respiratory Infectious Diseases

C. Drosten[1], D. Muth[1], A. von Brunn[2], S. Hippenstiel[3], T. Wolff[4], F. Weber[5], J. Ziebuhr[6], V. Thiel[7], A. Volz[8], G. Sutter[8], V. Herder[9], W. Baumgärtner[9], A. Osterhaus[9], U. Wernery[10], T. Meyer[11], S. Pöhlmann[12], P. Nagy[13], J. Juhasz[13]

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Zoonotic viruses that undergo onward transmission in humans via the respiratory route bear a high pandemic potential. Based on the MERS-coronavirus, a paradigmatic prepandemic virus, the RAPID consortium will collaboratively transform the latest methodology from basic respiratory virus research into a public health laboratory toolbox that provides assays to assess the pandemic potential of variant and novel respiratory agents. Hallmarks of pandemic potential will be identified by systems biology studies and advanced infection surrogate models. Specific for the MERS-CoV, we will implement an intervention that provides proof of the OneHealth approach by conducting a vaccine trial in camels (the zoonotic source of MERS-CoV), using a human vaccine that is scheduled to enter human phase I trials. Our program will integrate German public health and translational research structures into a One Health research resource that comprises the whole chain of emergence of MERS, one of the most severe threats to human health security today.

Q-GAPS: Q fever – GermAn Interdisciplinary Program for reSearch

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There are no efficient therapies available for chronic Q fever and for the Q fever fatigue syndrome caused by *Coxiella burnetii*. To develop novel treatments, it will be essential to increase our knowledge about *C. burnetii* pathogenesis. In addition, specific precautions are urgently needed to prevent *C. burnetii* spreading; however, we still do not fully understand the modes of transmission. In order to develop new diagnostic tools, establish risk indicators as well as a catalog of countermeasures for the public health service, the Q-GAPS network aims to address several unsolved questions concerning the epidemiology, pathogenesis, surveillance and control of *C. burnetii*. We will clarify the role of ticks as vector for *C. burnetii* transmission, and we will investigate antigen presentation during *C. burnetii* infection in order to set the basis for new vaccination strategies. By sequencing diverse isolates and correlating the genomes with their virulence, we will identify genes that are associated with distinct host species and/or pathogenic potential. The analysis of the function of these genes will allow us to identify candidate virulence markers of *C. burnetii*. Additionally, we will shed light on the development of the Q fever fatigue syndrome. All information gathered will be integrated into a catalog of countermeasures and an interactive database to help the human and veterinary health services to monitor *C. burnetii*, prevent bacterial spreading and control outbreaks.

Strengthening public health by understanding the epidemiology of rodent-borne diseases (RoBoPub)

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Hantaviruses and *Leptospira* spp. cause notifiable human diseases and are transmitted by persistently infected rodents. Frequent manifestations are unspecific, flu-like symptoms. Therefore, human infections with these pathogens are commonly under- or misdiagnosed resulting in a high level of underreporting. The emergence of hantavirus or *Leptospira* infections is complex and associated with individual pathogen properties like virulence and tenacity, but also environmental and climatic factors, as well as geographical distribution, reservoir association and population dynamics. Finally, susceptibility and behaviour of the human population strongly affects likelihood of infection. Notification of human disease cases is influenced by the awareness of physicians and diagnostic capabilities, risk perception and behaviour of the human population as well as severity of manifestation.

This interdisciplinary consortium aims to create the necessary knowledge of hantavirus and *Leptospira* epidemiology and to translate these findings into public health intervention measures. Within this OneHealth initiative the pathogen-, rodent reservoir-, environment-related aspects of pathogen transmission as well as human disease manifestation and detection, and social aspects of awareness and risk perception will be studied. The final aim is a thorough risk assessment with the generation of risk maps, early warning modules, risk management plans and public health recommendations.

TBENAGER: The German Tick-Borne Encephalitis Consortium

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Tick-borne encephalitis (TBE) is the most important viral CNS infection in Central Europe. Beside ticks since 2016 also milk-borne TBE has been reported in Germany. During the last years TBE is an emerging infection in Germany. So far, the factors involved in the increasing emergence of TBE are unclear. The natural transmission cycle is only poorly understood.
The aim of the TBENAGER Consortium is the interdisciplinary collaboration of various research groups and public health services in the field of TBE. The TBENAGER Consortium includes research groups working on the epidemiology and ecology of TBE. The different virus strains in Germany and neighbouring countries will be genetically and phenotypically characterized. Experimental infection techniques elucidate the role of the vectors and of the vertebrate hosts for the transmission cycle. Pathological and immunologic studies will provide a better understanding of the pathogenesis and individual risk factors for severe clinical outcome of disease. These data will be used for the development of new innovative TBE vaccines and treatment options.
A better understanding of the eco-epidemiology and pathogenesis of TBE will improve the surveillance for TBE and provide information on TBE for a targeted prevention in risk groups and for highly endemic areas. With these epidemiological data the public health services will be able to sustainably decrease the risk of TBE infection and therefore the number of human TBE cases in Germany.

Persistence of *Toxoplasma gondii*: Mode of action of candidate anti-protozoals and the role of physiological heterogeneity in zoonotic infections

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Keywords: Toxoplasma gondii, Metabolism, Persistence

Life-long persistence of pathogen subpopulations in their hosts is a hallmark of many zoonotic infections. *Toxoplasma gondii* is an Apicomplexan parasite that infects virtually all warm-blooded animals and causes life-long infections in up to 30% of humans globally and >50% in Germany. Chronically persisting parasite tissue cysts, termed bradyzoites, are key for pathogenesis as they underlie remission and transmission via undercooked meat products. However these stages cannot be targeted by available chemotherapies. My junior group identifies inhibitors of chronic Toxoplasma stages from chemical libraries and investigates their mode-of-actions using comprehensive LC/MS and GC/MS-based metabolomics. In a complementary approach the group will directly investigate the physiological basis of persistence in veterinary and clinical isolates. Although, *T. gondii* infections are widely clonal, tissue cysts are thought to be phenotypically highly heterogeneous. We will use fluorescent parasite reporter cell lines to monitor key physiological parameters such as cell division status and metabolic changes at single cell resolution and feed these data into computational model that allows us to understand the contribution of phenotypic heterogeneity to the persistence of *T. gondii*.
We use *T. gondii* as a model to deepen our molecular understanding of persistence and identify intervention strategies that will be applicable to other zoonotic diseases.

Ecology of emerging arboviruses - ARBOSPREAD

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Vector-borne infections account for about one fifth of all infectious diseases. Especially mosquito-borne viruses that often originate in tropical areas manage to spread to new hosts or new geographic regions with relative ease. However, our knowledge of arbovirus diversity, evolution, and geographic spread is mainly based on epidemic variants. To fill critical gaps in our knowledge on pre-epidemic viruses and viral emergence, we will characterize arbovirus diversity along tropical disturbance gradients to document and dissect microevolutionary processes in viral habitat expansion.
The overall aims of the group are: (i) to provide a comprehensive sample of mosquitoes from ecologically undisturbed and adjacent disturbed habitats in three main tropical rainforest ecosystem types occurring in Africa. We will characterise the true taxonomic diversity of arthropod-associated viruses including novel prototypic deep lineages still existing in natural ecosystems using state of the art methods; (ii) to identify the interplay between host community composition and viral emergence processes using mosquito-associated viruses as a multi-host and multi-pathogen system; and (iii) to analyse whether and how arbovirus emergence is driven by positive selection involving fitness gains of emergent strains using genetic and phenotypic viral population studies. Taken together we will obtain the first comprehensive assessment of viral microevolution during emergence in a real-world scenario.

Vector biology of *Aedes albopictus* and eco-bio-social drivers for effective vector prevention & control in cooler ecoregions (AECO)

R. Müller[1], M. Dhimal[2], D. Montag[3], U. Kuch[1], D.A. Groneberg[1]

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Aedes albopictus belongs to the world's most feared mosquito species with high social and medical importance because it transmits dengue and chikungunya viruses amongst others.
The eco-bio-social research plan of the BMBF funded junior research group AECO focus on (i) the vector biology of the highly invasive mosquito *A. albopictus* and (ii) eco-bio-social aspects influencing vector prevention & control practices along a climatic gradient in a dengue and chikungunya epidemic country (Nepal).
We analyse the potential association of cold hardiness and morphological and epigenetic plasticity in *A. albopictus* field-collected eggs in order to better understand the rapid adaptation of this originally tropical to subtropical species to cooler ecoregions (such as Germany). Physiological data will feed species distribution models with new phenological data and thereby advance the forecast of this invasive species in temperate regions under climate changes.
In addition, we assess the people´s knowledge and attitude on mosquito-borne diseases and their vector prevention and control practice at different altitudes. Based on the analysis of an eco-bio-social dataset, the main drivers for the practice and social acceptance of different preventive and control measures against mosquitoes will be assessed in order to support national one-health strategies to efficiently combat dengue and chikungunya illnesses in Nepal.

Development of novel tools to study the zoonotic vector biology of *Ixodes ricinus* using CRISPR/Cas and artificial tick feeding systems

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Ticks are obligate hematophagous parasites and prolific disease vectors. In Europe, the main tick species of both medical and veterinary importance is *Ixodes ricinus*, a widespread vector for a wide variety of zoonotic tick-borne pathogens including tick-borne encephalitis virus (TBE) and Lyme borreliosis. In this research group, we aim to contribute to the elucidation of the vector biology of *I. ricinus* by:

1) establishing CRISPR/Cas technology and a gene drive system in tick cell lines and ticks for functional genomic studies
2) continuing studies on the artificial feeding of *I. ricinus* and evaluating its suitability for *in vitro* infection models for various zoonotic pathogens

The studies performed within the framework of this project will form a major advancement for research on tick-host-pathogen interactions, spawning new paradigms to control zoonotic tick-borne diseases.

RNA-VIRT: Emerging RNA viruses and their interaction with the human and animal host

I. Steffen[1]

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An important feature of zoonotic viruses is the differential pathogenesis in different susceptible hosts. While some hosts develop symptomatic disease, others express only mild or no symptoms, despite their susceptibility to viral infection. A better understanding of the interaction of antigens, virulence factors and immunological reactions in animals and humans is crucial for a better detection and interpretation of host responses to viral infections. The proposed research projects will systematically compare molecular and immune-regulated host responses to flavivirus infections in different hosts.
The requirements for tick-borne and Japanese encephalitis virus replication in different hosts will be assessed using subgenomic replicon-based systems. Changes in cellular gene expression will be determined to identify host cell factors that are selectively induced or down-regulated in flavivirus infections of different hosts. In addition, humoral immune responses against tick-borne encephalitis virus will be compared between different host species based on antibody profiles against selected viral antigens and functional antibody tests for the measurement of antibody-dependent cellular and systemic immune responses.
The resulting data will be used to identify protective mechanisms in natural reservoir hosts that can be transferred to other hosts for therapeutic use.

Immunological requirements for protective vaccination against zoonotic infections

A. Volz[1]

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Zoonotic pathogens pose a major challenge to public health and necessitate the rapid development of new vaccines that confer protection in case of an emergency. This has been vividly demonstrated in the recent past with Severe Acute Respiratory Syndrome (SARS) or Middle East Respiratory Syndrome (MERS) coronaviruses as prominent examples of emerging infections causing severe disease and/or major epidemics. Thus the development of a novel general approach for innovative and rational design of vaccines is urgently needed. In that context the induction of pathogen specific protection is a major focus of modern vaccinology, as in addition to efficient inhibition of transmission this might specifically protect against specific forms of the disease outcome. An aspect of innovative vaccine development includes the identification of novel antigens that mediate rapid protection against pathogen specific disease manifestations. Utilizing Zika Virus (ZIKV) as a prototype zoonotic pathogen we will test ZIKV proteins in vivo for their ability to induce disease specific protection. For this, the Modified Vaccinia virus Ankara (MVA) serves as a highly versatile expression system to allow for stable synthesis of ZIKV proteins. The results from this project allow a more detailed understanding of protective mechanisms of immune responses elicited against pathogen specific diseases. This will be essential for rational design of vaccines.

Evening lecture

100.000 € for the proof of the existence of the measles virus - for real?

D.Bardens[1]

[1] Region Gävleborg, Sweden

Keywords: measles, vaccination

Measles is a highly contagious and dangerous infection, caused by the well-studied measles virus. Vaccination is a safe, cheap, and effective way to prevent infection.
However, many people choose to not vaccinate their kids and spread conspiracy theories about the disease. In 2012, a German biologist offered to pay out 100.000 € to the person who can prove that the measles are caused by a virus. David Bardens took the challenge and brought the case to Germany's highest court. In his entertaining talk he tells the story of what happened. [The talk will be held in German.]

Plenary Session IV: Keynotes

Bornavirus infection: a new model of evolution and coexistence of RNA viruses

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Keywords: Bornavirus, Endogenous viral elements, Evolution

Bornavirus research has entered a new era. Borna disease virus (BoDV) was identified as the etiological agent of Borna disease of horses and sheep in the early 20th century in Germany. Studies of BoDV has uncovered many unknown properties of animal-derived RNA viruses, while the lack of species diversity in the genus Bornavirus had long remained an enigma. However, in the past decade, our knowledge regarding the species diversity of bornavirus has dramatically increased by the discovery of new viruses that are clearly related to BoDV in many vertebrates, especially birds. Furthermore, in 2015, a zoonotic mammalian bornavirus, named variegated squirrel bornavirus, was identified in the brains of the patients died from fatal encephalitis. At present, there are at least 8 different species in the genus. In addition, in 2010, endogenous sequences highly homologous to current bornaviruses, called endogenous bornavirus-like (EBL) elements, were discovered in the genomes of many mammalian species, including humans. We have recently showed that the products from some EBL elements can affect the infection of exogenous bornavirus in cultured cells. These epoch-making discoveries have not only had great impact on the understanding of the interaction between host and bornavirus, but make this unique virus as a model of evolution and coexistence of RNA viruses. In this presentation, I will provide an overview of recent progress and implications for future directions of bornavirus research.

One Health – Not Just a Buzz-Word!

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Keywords: One Health, Zoonoses, Added value

The inextricable linkage of human and animal health has been increasingly recognized in the past decades. However, human and veterinary medicine are often working so much in separation that human and animal health is affected. We define One Health (OH) as the added value in terms of human and animal health benefits, financial and other resource savings and improved environmental services compared to the two medicines working in separation. An integrated assessment of human and animal health requires methods capable of assessing effects on the animal – human interface. For example livestock mass vaccination against brucellosis is not profitable for the public health sector alone but becomes largely profitable from a societal perspective including all involved sectors. Dog rabies control in Africa by mass vaccination of dogs becomes less costly than human post-exposure prophylaxis alone after ten years. Other examples are provided from health services, integrated antimicrobial resistance surveillance and joint laboratory infrastructure. Conceptually OH is embedded in broader ecosystem approaches to health which can also be called health in social-ecological systems or health in human-environment systems which is also important for non-communicable diseases. In this way human and animal health improvements will be developed while considering social dynamics and sustained ecosystem services.

Session 1: Novel Methods, Diagnostics and NGS

October 12, 2017
14:00 – 15:30

Room Ballsaal
Chairs: Martin H. Groschup and Claudia Kohl

Ancient viral DNA: a systematic feasibility study

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Keywords: virus, ancient DNA

Background and objectives: We most often characterize zoonotic events retrospectively through inferences derived from the genetic diversity of present-day pathogens. Ideally though, we would complement this approach with a direct exploration of pathogen genetic diversity in the past, using ancient DNA techniques. While ancient bacterial genomes have been determined from a variety of ancient samples, it remains unclear whether ancient viral DNA can be recovered from normal tissues/bones (i.e. neither mummified nor preserved at cold). Here we will present the framework of and some preliminary results generated by a pilot project currently funded by the National Research Platform for Zoonosis. This pilot project offers the first systematic investigation of the feasibility of ancient viral DNA retrieval.
Materials and methods: For this, we will make use of an exceptional set of nonhuman primate bones collected in a tropical environment over the last three decades. These samples will be analyzed with a combination of hybridization capture and next generation sequencing, using a specific capture tool targeting genomes/large genomic fragments from a number of dsDNA viruses and reverse-transcribing RNA and DNA viruses already known to infect the corresponding host species.
Results: (Not applicable)
Conclusion: This approach guarantees a convincing answer to the question of the feasibility of ancient viral DNA studies will be obtained.

Hybridization capture as a paleovirological tool for the detection of recent filoviral integrations in potential hosts' genomes

A. Düx[1], J. Gogarten[1,2], L. Müller[1], F. Leendertz[1], S. Calvignac-Spencer[1]

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Keywords: Paleovirology, Filovirus, Reservoir

Background and objectives: The discovery of ancient filovirus-like endogenous viral elements (EVEs) in mammalian genomes has unlocked an unexpected source of information in the search for filovirus reservoirs. We hypothesize that species with an ongoing long-term association with filoviruses may harbour recent EVEs in parts of the population. We test a novel bench approach to paleovirology using hybridization capture with RNA baits spanning the genomes of the five Ebolavirus species and the two viruses within the Marburgvirus species.
Materials and methods: To validate the method we used this approach on human DNA spiked with plasmids containing a 292 bp fragment of the Ebolavirus L gene. We were able to detect an equivalent of a single heterozygous integration in 1% of individuals. We then tested libraries built from pools of 30-90 individuals of susceptible or potential reservoir species (fruit and insectivorous bats, chimpanzees and humans from outbreak regions). Specimens were selected to span the species' geographic range and maximize genetic diversity in the pools.
Results: No filoviral EVEs were detected.
Conclusion: These results provide no additional evidence for the role of these species as Filovirus reservoirs. The method will expand the paleovirological toolkit, as it does not depend on sequence databases and can thus be used on any species and on a large number of individuals, allowing for the detection of recent EVEs that are rare variants within a population.

Molecular typing of *Listeria monocytogenes* in foodstuffs to combat human listeriosis in Germany

S. Lüth[1,2], S. Kleta[1], A. Flieger[3], S. Halbedel[3], R. Prager[3], R. Merle[2], T. Alter[2], S. Al Dahouk[1,4]

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Keywords: Listeria monocytogenes, molecular typing, WGS

Background and objectives: *Listeria monocytogenes* (Lm) is a zoonotic pathogen and the causative agent of listeriosis. Transmission mainly occurs via contaminated foodstuffs leading to severe illness in elderly, pregnant or immunocompromised people. A relatively high mortality rate renders listeriosis a major public health concern. For effective surveillance and disease control, extensive molecular tracing is indispensable. However, the analytical basis can be improved by using the potential of novel techniques. Therefore, pulsed-field gel electrophoresis (PFGE) and whole genome sequencing (WGS) as typing tools will be compared for various German Lm-field isolates in our project MolTypList5.
Materials and methods: A total of 458 food-isolates from ready-to-eat products (the most frequent origin of infection) and 54 isolates from production environments were selected from 1207 samples sent to the German National Reference Laboratory (NRL) for Lm in 2016 for molecular typing.
Results: The largest percentage of food-isolates originated from Baden-Württemberg (29%). Most frequent matrices were meat and fish products with 41% and 27%, respectively. More than 50% of the isolates belonged to serotype IIa. PFGE and WGS data revealed diverse Lm-strains in Germany and clusters based on epidemiologically linked food-isolates.

Advanced characterization of Cowpox virus infection in a human skin equivalent-based 3D model

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Keywords: 3D culture, Cowpox virus, transcriptomics

Background and objectives: Zoonotic Cowpox virus (CPXV) infections usually occur as localized skin lesions. 3D organoid models facilitate the examination of viral replication and virus-cell interactions in a more physiological context. Therefore human skin equivalents were established as an infection model to study CPXV infection via RNA-Seq-based transcriptome analysis.
Materials and methods: Differentiated human skin equivalents were generated from primary keratinocytes and fibroblasts and infected with CPXV. Viral replication was monitored via qPCR and IHC. RNA-Seq analysis of infected keratinocytes from the epidermal part of the equivalent or from 2D cultures was performed in biological triplicates.
Results: CPXV caused permissive infections in human skin equivalents. Increasing amounts of viral nucleic acids and viral particles were detected over time. The sequencing run generated reads, representing around 15.000 different human and all 229 viral genes. Comparison of different time points and cultivation setups revealed cell- or model-specific regulated genes and GO terms. For skin samples 4 % of host genes were significantly regulated due to infection. Changes in expression were confirmed via qPCR, IHC and Western Blotting.
Conclusion: Taken together infection experiments in a skin equivalent-based infection model combined with RNA-Seq transcriptome analysis have provided new insights in virus spread and regulation of cellular genes by zoonotic CPXV.

A novel approach to tackle respiratory pathogens responsible for great ape population declines

L. V. Patrono[1], S. Calvignac-Spencer[1], F. H. Leendertz[1]

[1]Project Group Epidemiology of Highly Pathogenic Microorganisms, Robert Koch Institute, Berlin, Germany.

Keywords: anthropozoonoses, great apes, hybridization capture

Background and objectives: Human respiratory pathogens have caused lethal outbreaks in endangered great ape communities across Africa, leading to population declines. Targeted genome capture and next generation sequencing pave the way to genomic insights on pathogen epidemiology, providing relevant information to reduce the risks of disease introduction to wildlife. The aim of this study is to develop a targeted genome capture strategy to characterize human-introduced viral and bacterial pathogens in a habituated chimpanzee community living in the Taï National Park.
Materials and Methods: Isolates of human metapneumovirus (HMPV), respiratory syncytial virus (HRSV) and *Streptococcus pneumoniae* were used to generate baits encompassing full viral genomes and pneumococcal specific genes. Lung tissue from individuals that died of respiratory disease (n=10) was transformed into Illumina libraries. Following a 48h bait-to-library hybridization, enriched libraries were sequenced on a MiSeq platform.
Results: Reads mapping to HMPV and HRSV were captured in 5/10 samples. In two samples, we reconstructed nearly the full HRSV genome. Where only few reads were captured, these mapped to different parts of the genome, providing little but valuable information on different genes. Reads mapping to *S. pneumoniae* genes were captured in 9/10 samples, with an average coverage of at least 20X.
Conclusion: The capture strategy developed herein is a promising tool to significantly expand our knowledge on human pathogens infecting great apes.

Identification of biomarkers of zoonotic pathogens by ORFeome phage display

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Keywords: biomarker, phage display, ticks

Background and objectives: The identification of biomarkers from pathogens is a prerequisite for the development of vaccines and diagnostic assays. Ticks are an important vector for disease, e.g. lime disease and tick born encephalitis. A vaccine against ticks could omit the infections with several pathogens.
Materials and methods: In our approach, we are using ORFeome phage display to select immunogenic proteins from zoonotic pathogens and ticks. The phage display libraries are constructed from whole genomes these pathogens, respectively from cDNA from ticks (Ixodes scapularis).
Results: We will present our ORFeome phage display technology and show the identification of novel immunogenic proteins of zoonotic pathogens. Humans which are often in contact with ticks develop an increased immune answer to tick bites resulting in drop down or death of the tick before feeding and potential transmission of pathogens. Using blood of these donors, we identified an immunogenic metalloprotease (MP1) from Ixodes scapularis. MP1 is a zinc metalloprotease, has a fibrinolytic activity and is involved in blocking haemostasis which is essential for blood feeding.
Conclusion: ORFeome phage display is a powerful tool to identify novel immunogenic proteins of pathogens. The metalloprotease1 of Ixodes scapularis is a candidate for further vaccinations studies to prevent tick borne diseases

Session 2: Pathogen-cell interaction

October 12, 2017
14:00 – 15:30

Room Zehlendorf
Chairs: Martin Beer and Jan Schinköthe

Phosphorylation of Tripartite motif containing 28 (TRIM28) during HPAIV infection occurs via the stress-activated protein kinase pathway p38-MSK1

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Keywords: HPAIV, TRIM28, MAPK

Background and objectives: Zoonotic transmission of highly pathogenic avian influenza A viruses (HPAIV) is accompanied by excessive cytokine production mediated by incompletely understood mechanisms. Phosphoproteome analysis revealed that the transcriptional co-repressor TRIM28 is selectively phosphorylated during HPAIV infection, indicating a potential role in cytokine dysregulation.
Materials and methods: In this study, diverse stress stimuli and kinase inhibitors were used to identify responsible kinases for TRIM28 phosphorylation. In addition, siRNA-mediated knockdown established a biological role of TRIM28 during IAV infection.
Results: In response to cellular stresses TRIM28 phosphorylation at Serine 473 is mediated by the kinases Chk2 and MK2. In contrast to this described pathways, our results demonstrate for the first time that during HPAIV infection TRIM28 phosphorylation is mediated by the kinases p38 and MSK1, which are both important mediators in the cytokine expression following activation of RIG-I and TLRs. In line with this, we can demonstrate that phosphorylation of TRIM28 occurs in response to different immune stimuli, indicating a similar mechanism during HPAIV infection. Most importantly, downregulation of TRIM28 reduces viral replication suggesting a proviral function.
Conclusion: Our data suggest that TRIM28 is an undescribed transcription factor downstream of p38 and MSK1 that may contribute to the excessive cytokine expression during HPAIV infection.

Endogenous Borna-like N elements in shrews

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[1]Infection Biology Unit, German Primate Center, Göttingen, Germany

Keywords: Bornaviridae, Endogenous borna-like elements, Shrews

Background and objectives: Endogenous viral elements have been demonstrated in various vertebrate and invertebrate genomes, mostly derived from retroviruses due to their replication strategy. Intriguingly, Bornaviridae-like sequences, termed endogenous borna-like N (EBLN), are found in vertebrate genomes as well. While integration in some mammalian species, like primates, has been phylogenetically dated back to millions of years ago, in some shrew species (Crocidura leucodon) the integration seems to be a more recent event associated with an individual Borna Disease Virus-1 (BoDV-1) infection. EBLN may play a role in antiviral immunity, but their function in shrews is not yet fully understood.
Materials and methods: A population of shrews (n=26) were tested for BoDV-1 infection and EBLNs by RT-PCR and PCR. For DNA-isolation ear punch tissue was used. As the shrews were part of a breeding colony, the pedigree and family relationships of the individuals were partly known.
Results: 3/7 naturally infected shrews show an EBLN. Interestingly, 1/19 non-infected shrew also harbours an EBLN without any relatives sharing this characteristic.
Conclusion: Besides BoDV-1 infection-derived EBLN, there is evidence for inheritance of unprecedented EBLNs not readily detected via published PCR protocols. A thorough analysis of this issue should be part of future studies.

Infection of differentiated swine airway epithelial cells by influenza A viruses of different species origin

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Keywords: Air-liquid interface, Influenza A viruses

Background and objectives: Pigs play an important role in the interspecies transmission of influenza A viruses (IAV). Therefore, it is necessary to further study the infection of the swine airway epithelium by IAV from different origins.
Materials and methods: An air-liquid interface culture system for well-differentiated porcine airway epithelial cells was applied to analyze the respiratory epithelium for virus-induced detrimental effects. Swine ALI cultures were infected via the apical membrane domain by IAVs comprising swine, human, and avian IAVs of different subtypes (H1N1, H3N2 and H9N2).
Results: Virus was continuously released from the apical surface up to eight days post-infection in all IAV infection groups. The amount of infectious virus released into the supernatant was highest in the case of swine IAV. Interestingly, there was a large variation in the infection efficiency among the different avian viruses. Though the differentiated swine epithelial cells underwent a dramatic loss of cilia, they retained the barrier function.
Conclusion: Air-liquid interface cultures of differentiated swine airway epithelial cells are a valuable culture system to analyse the interspecies transmission of influenza viruses.

Comparative loss of function screens reveal common pathways required by Paramyxoviridae and Pneumoviridae

K. Pfeffermann[1], D. E. Anderson[2], S. Y. Kim[3], B. Sawatsky[1], P. Duprex[4], M. A. Garcia-Blanco[2,3,6] , V. von Messling[1,2,5]

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Keywords: Paramyxoviridae, ABCE1, genome-scale siRNA screen

Background & objectives: Paramyxo- and pneumoviruses include many pathogens with great relevance for human and animal health, however host factors involved in their life cycle remain elusive. To identify common cellular host factors involved in the Paramyxo- and Pneumviridae life cycle as a basis for new insights in the virus biology and the development of rationally designed therapeutics.
Materials & methods: Comparative genome-wide siRNA screens with measles, mumps and respiratory syncytial viruses were conducted in A549 cells. One of the top hits, ABCE1, was chosen for characterization and its role in viral replication and translation was analyzed by viral growth kinetics and Western blot analysis. Its impact on viral and global cellular translation was elucidated using metabolic or catalyzed fluorescent protein labelling.
Results: Comparative analysis of the three screens yielded 42 common proviral host factors. In addition, proteasomal degradation, RNA processing and translation were top pathways required by all three viruses. Characterization of ABCE1 revealed that ABCE1 knockdown strongly inhibited the production of MeV proteins, while only modestly affecting global protein synthesis.
Conclusion: This study highlights the importance of ABCE1 as Paramyxo- and Pneumoviridae translation factor. The data set will be an invaluable resource for the development of broad-spectrum antivirals not only against the three viruses included but also against related emerging pathogens

The role of Salmonella Pathogenicity Island 2 (SPI2) in the course of neonatal non-typhoidal Salmonella infections

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Keywords: Salmonella, effector proteins, neonatal infection

Background and objectives: Non-typhoidal *Salmonella* (NTS) are among the most prevalent causes of infectious diarrheal disease in humans and pigs, but also contribute to invasive infections in human infants. Their pathogenicity is conferred by horizontally acquired chromosomal regions, called Salmonella pathogenicity islands (SPIs), encoding sets of effector proteins, which are delivered into the host cell via specific type-three secretion systems. Several *in vitro* studies identified SPI2 as a requirement for the establishment of a Salmonella containing vacuole (SCV), an intracellular compartment allowing survival and replication inside the host cell.
Materials and methods: We used our newly established mouse model to clarify the role of SPI2 in establishment and progression of systemic NTS infections in the neonate host.
Results: Oral infection with wildtype and SPI2-deficient NTS resulted in similar bacterial loads of the gastrointestinal tract, but re-isolation rates of mutants from systemic organs were significantly decreased. In contrast to the general understanding of SPI2 as prerequisite for SCV formation *in vitro*, mutants established and maintained SCVs and even grow to high numbers without harming their host cell.
Conclusion: By evaluating 15 isogenic SPI2 effector deficient *Salmonella* strains, we demonstrate that SifA significantly contributes to the SPI2-dependent phenotype. Its lack seems to prevent transmigration of enterocytes and, finally, systemic spread.
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Hypoxia induces dormancy in *Coxiella burnetii*

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Keywords: Coxiella burnetii, dormancy, hypoxia

Background and objectives: *Coxiella burnetii* is the causative agent of Q fever, which might result in an acute or chronic disease. Chronic Q fever develops years after infection and mainly manifests as endocarditis. How *C. burnetii* persist in patients is unknown. As tissue oxygen levels are low at sites of infection we investigate the influence of hypoxia on *C. burnetii* replication in macrophages (MΦ), their primary target cells.
Materials and methods: Murine bone marrow-derived MΦ were infected with *C. burnetii* under normoxic or hypoxic conditions.
Results: Under normoxic conditions *C. burnetii* replicates in MΦ. Exposure to 0.5% oxygen stabilized hypoxia-inducible factor 1α (HIF1α) and abolished *C. burnetii* replication in MΦ. Hypoxic-induced inhibition of *C. burnetii* replication was neither due to enhanced bactericidal MΦ activity nor linked to an altered intracellular trafficking. Our results rather indicate that *C. burnetii* enters a dormancy period under hypoxic conditions, which was dependent on HIF1α stabilization. HIF1α leads to reduced and delayed activation of STAT3. While constitutive active STAT3 rescued hypoxia-mediated impairment of *C. burnetii* replication, ablation of STAT3-signalling results in impaired *C. burnetii* replication under normoxia.
Conclusion: Hypoxia-induced stabilization of HIF1α impedes activation of STAT3 in MΦ, which in turn results in dormancy in *C. burnetii*. Therefore, hypoxic areas could provide a niche for *C. burnetii* persistence.

Session 3: Antimicrobial Use and Resistance

October 12, 2017
14:00 – 15:30

Room Steglitz
Chairs: Birgit Walther and Denise Rabold

MIC distributions for glyphosate in farm animal-associated Enterobacteriaceae determined by the broth microdilution method

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Keywords: MIC, glyphosate, Enterobacteriaceae

Background and objectives: Glyphosate is currently the most widely used herbicide in the world. Glyphosate acts on enolpyruvylshikimate-3-phosphate synthase (EPSPS), which is also present in bacteria. Concerns have been raised about the potential effects of glyphosate in animal feed on bacteria in farm animals. Here, we investigated the current levels of resistance to glyphosate (as an active ingredient and as a part of a complete herbicide formulation) in diverse isolates of *Escherichia coli* and *Salmonella* spp.
Materials and methods: We determined minimal inhibitory concentrations (MIC) of glyphosate and Roundup LB plus in Müller-Hinton I medium using a broth microdilution method for 120 *Salmonella* spp. and 238 *E. coli* isolates.
Results: The distribution of MIC values for *Salmonella* spp. was 40-80 mg/ml for both glyphosate and Roundup, and 5-10 mg/ml for glyphosate and 20-40 mg/ml for formulation for *E. coli*. Among the isolates with a MIC higher than the 95%-percentile the pathogenic *E.coli* were the dominant subgroup. Phylogenetic relationship, antibiotic resistance profile and geographical location of sample isolation did not have a large effect on glyphosate sensitivity in both species. Whereas, as well as host animal species and time point of isolation seem to have some slight effects on glyphosate MIC.
Conclusion: These results demonstrate that although *Salmonella* spp are more resistant to glyphosate than *E. coli*, the MIC distribution within the species is small, suggesting the absence of a highly resistant subpopulation in farm animal-associated Enterobacteriaceae.

Traceability of MRSA in the pork food chain and survival in meat products

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Keywords: MRSA, food chain, meat products

Background and objectives: Food of animal origin may be a source of Livestock-associated Methicillin-resistant *Staphylococcus aureus* (LA-MRSA) in humans. This project aimed to trace MRSA along the pork food chain and to test its survival in the final product.
Materials and methods: Three MRSA positive fatting pig farms (farm 1-3) and 9-10 animals/farm were included in the study. Animals harboring MRSA at farm level were re-sampled at slaughter and material was collected from the same animals. Minced meat and three different types of meat products (sausages) were manufactured and tested for the presence of MRSA throughout the shelf life. Further samples were taken from the slaughterhouse environment. All MRSA were *spa*- and SCC*mec* typed.
Results: Overall, MRSA were detected until the end of the shelf life in 4/9 samples of spreadable and 1/9 samples of firm raw sausages. Tracing of identical LA-MRSA types (t034, *mec*V) was successful from the farm to the final product as follows: detection at slaughter, but not in minced meat or any of the sausages (farm 1); detection at slaughter and in the final product (spreadable raw sausage) (farm 2); detection in minced meat and in the final product (spreadable and firm raw sausage) but not at slaughter (farm 3). Also, typical human-associated MRSA (t007) were found in the final meat products and in the environment.
Conclusion: Survival of MRSA in raw meat products needs to be further investigated from a consumer health risk perspective.

ESBL-/AmpC-producing Enterobacteriaceae in broiler fattening farms after cleaning and disinfection – identification of critical control points

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Keywords: broiler, ESBL, cleaning and disinfection

Background and objectives: Broiler farms are known as reservoirs for ESBL-/AmpC-producing Enterobacteriaceae. The reduction of these bacteria should be achieved by cleaning and disinfection procedures (C&D) between different fattening periods. Aiming on the elucidation of possible niches for the survival of ESBL-/AmpC-producing Enterobacteriaceae and the identification of critical control points (CCP) we investigated five broiler farms before and after C&D.
Material and methods: Five ESBL-/AmpC-positive broiler farms were identified by an initial screening of the current fattening flocks. Following, these farms were investigated intensively by taking gauze swabs and boot swabs after C&D from both the inside and the outside of the farms. ESBL-/AmpC-producing Enterobacteriaceae were isolated and examined for their bacterial species, phylogroup and resistance genes. Additionally, all samples were investigated for Enterococci as an indicator for faecal contamination.
Results: In our study, both investigated microorganisms survived the C&D in 4/5 farms. Thereby, ESBL-/AmpC- producers were only detected in samples that simultaneously were positive for Enterococci. Isolates detected at the initial screening and after C&D of the same farm showed equal molecular characteristics which will be further investigated by PFGE in this ongoing study.
Conclusion: ESBL-/AmpC-producing Enterobacteriaceae can survive C&D and therefore pose a risk for the colonisation of consecutively fattening flocks.

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Use of antimicrobial peptides for the reduction of multi-resistant pathogenic bacteria and prevention of biofilm formation in dairy processing

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Keywords: Antimicrobial peptides, dairy processing, antimicrobial surface coatings

Background and objectives: Bacterial adhesion on surfaces in milk production and processing environments leads to increased opportunity for microbial contamination of dairy products. In biofilms survival of multi-resistant pathogens is promoted and bacteria are partly protected from cleaning or disinfection procedures. New control and prevention strategies for biofilms are required. Antimicrobial peptides (AMPs) are novel therapeutic agents with a broad bactericidal activity against gram-positive and gram-negative bacteria.

Materials and methods: In a joint project (FKZ 2817700514) effective and suitable immobilization strategies of AMPs onto surfaces preventing biofilm formation by reducing the microorganism load upon contact were developed and optimized. Several natural and modified AMPs were employed and the antimicrobial mode of action was tested using isolates of mastitis-causing pathogens. Experiments for cytotoxicity in a eukaryotic cell model indicate the biocompatible properties of the AMPs used. Optimized antimicrobial surface coating were adjusted to lowest necessary peptide amount and relevant surfaces in milk production: Steal, rubber and silicone.

Results: Biocidal surfaces on the basis of AMPs that reduce microbial load successfully were generated.

Conclusion: Bacteria have great difficulties to acquire resistance against short-chained peptides. Therefore the approach may offer an alternative to the use of antibiotics and consequently may assist to reduce the spread of zoonotic diseases between animals and humans.

Colonisation of broiler chickens with ESBL-/ AmpC- producing E. coli using a seeder- bird model and detection of in vivo transformants

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Keywords: ESBL, AmpC, broiler

Background and objectives: Colonisation of broiler with ESBL- and AmpC- producing Enterobacteriaceae is well known, resulting in the possibility of transfer to humans either by a close contact to broiler flocks or through contaminated retail meat. To examine potential intervention strategies regarding hygiene- and management measures a broiler colonisation model was established.
Materials and methods: Broiler were conventionally housed in to identify the oral colonisation dose and to establish a seeder- bird-colonisation model close to real farming conditions. ESBL-/ AmpC-negative day- old broiler were orally co-infected on the third day of life with one ESBL- and one AmpC- producing *E. coli* strain. Colonisation success was proven by cloacal swabs over a period of 14 to 35 days and a final section. By using different selective media it was possible to verify in vivo plasmid transfer between the two strains.
Results: An oral infection dose of 10^2 cfu per animal is sufficient to colonise broiler 24 h *post infection* up to the end of the trial. A relation of 1:5 infected (seeder) to susceptible animals is adequate to colonise the complete flock. In addition transformants carrying both resistant plasmids were detectable after 72 h *p.i.*
Conclusion: Given the low infection dose and seeder numbers the spread of ESBL-/ AmpC- producers in conventional farms is not surprising. We will test hygiene- and management interventions using this model as a possible approach for reduction.

Multispecies and clonal dissemination of OXA-48 type carbapenemase in Enterobacteriaceae from clinical samples of animals (2009-2016)

S. Pulss[1], I. Stamm[2], E. Prenger-Berninghoff[1], S. Göttig[3], I. Stolle[1,4], C. Heydel[1], T. Semmler[5], C. Ewers[1]

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Keywords: OXA-48, companion animal, carbapenem

Background and objectives: One of the most urgent areas of antimicrobial drug resistance is the increasing spread of carbapenem resistance in gram negative bacteria. We report the occurrence of carbapenemases in Enterobacteriaceae from clinical specimens of animals over an 8-year period.

Materials and methods: *Escherichia coli* (Ec), *Klebsiella* spp. and *Enterobacter* spp. isolated from diagnostic samples from 2009 to 2016, mainly obtained from companion animals, horses, cattle and swine, were screened for carbapenem resistance using meropenem-containing media. Carbapenemase (CP) genes were determined by PCR and sequence analysis. CP-positive isolates were screened for ESBL and AmpC-type β-lactamase genes and phenotypic resistance using VITEK2. MLST, PFGE and whole genome sequencing was performed to determine the clonal relatedness.

Results: Among 21,569 isolates, OXA-48 could be identified as the sole carbapenemase type in 137 (0.64%) strains. K. pneumoniae (Kp) was the predominant carbapenemase producer (n=86 isolates), followed by Ec (n=22), E. cloacae (Ecl) (n=28), and K. oxytoca (n=1). OXA-48 was only found in non-livestock animals, i.e. in dogs (120/3182; 3.8%), cats (13/792; 1.6%), guinea pig (1/43; 2.3%), rat (1/23; 4.3%), mouse (1/180; 0.6%), and one rabbit (1/144; 0.7%).

Session 4: Risk Assessment, Epidemiology and Modelling

October 13, 2017
11:00 – 12:30

Room Steglitz
Chairs: Sandra Eßbauer and Reimar Johne

Bushmeat hunting and zoonotic transmission of Simian T-lymphotropic virus 1 in tropical West and Central Africa

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Keywords: bushmeat; non-human primates; zoonotic pathogen transmission

Background and objectives: Simian T-lymphotropic virus 1 (STLV-1) enters human populations through contact with non-human primate (NHP) bushmeat. We tested whether differences in the extent of contact to STLV-1 infected NHP foster regional differences in prevalence of human HTLV-1.
Materials and methods: Using serological and PCR assays, we screened humans and NHP at two sub-Saharan African sites where subsistence hunting was expected to be less (Taï region, Côte d"Ivoire, CIV) or more developed (Bandundu region, Democratic Republic of the Congo, DRC).
Results: Only 0.7% of human participants were infected with HTLV-1 in CIV (N=574), and 1.3% of humans in DRC (N=302). Two Ivorian human virus sequences were closely related to simian counterparts, indicating ongoing zoonotic transmission. Multivariate analysis of human demographic parameters and behavior confirmed participants from CIV to be less often exposed to NHP than participants from DRC through direct contact, e.g. butchering. At the same time, numbers of STLV-1 infected NHP were higher at CIV (39%, N=111) than at DRC (21%, N=39).
Conclusions: We conclude that a similar ultimate risk of zoonotic STLV-1 transmission - the product of prevalence in local NHP and human rates of contact to NHP carcasses - contributes to the observed comparable rates of HTLV-1 infection in humans in CIV and DRC. Young adult men and mature women were most likely exposed to NHP at both sites. The identification of such high risk groups of NHP exposition may guide future prevention efforts of zoonotic disease spread.

Characteristics profiles of cefotaxime-resistant *E. coli* from German livestock farms and potential association with farm factors

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Keywords: ESBL/AmpC; broiler; cattle; fattening pigs

Background and objectives: To better understand the epidemiology of ESBL/AmpC-producing *E. coli* we investigated characteristics profiles of CTX-resistant E. coli strains from German livestock farms and the association with management factors of these farms.
Materials and methods: In a cross-sectional investigation on the prevalence of cefotaxime-resistant *E. coli* samples from 194 livestock farms in Germany were collected. During farm visits, data on farm management were recorded by questionnaires. CTX-resistant *E. coli* were isolated from samples of 150 farms, and characterised further. For 469 isolates the ESBL-genes and the phylogroup were determined. Additionally, the phenotypic antimicrobial resistance was tested. This information was used to define different profiles characterising the isolates. Multivariate analyses using the distance-based permutation test were performed to investigate dependencies between characteristics profiles and conditions observed in the farms (e.g. farm size, hygiene factors or antimicrobial use).
Results: No co-occurrence of ESBL-genes in one isolate was observed. The Frequency of phylogroups was as follows: A (55%), B1 (35%), D (17%) and B2 (3%). The most frequent resistance profile was resistance to cefepine alone (27%).Results show, that the characteristics profiles of isolates from broiler farms differ substantially from the characteristics profiles of isolates from other

farm types. Results on characteristics profiles and association analyses will be presented.

Hantavirus infections as a cause of fever of unknown origin in Kazakhstan

N. Tukhanova[1,2], A. Shin[1,2], K. Abdiyeva[2,3], N. Turebekov[2,3], S. Frey[4], R. Yegemberdiyeva[2], L. Yeraliyeva[2], Z. Shapieva[5], G. Fröschl[3], G. Zhumabaeva[1], A. Zhalmagambetova[1], S. Essbauer[4]

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Keywords: hantavirus, fever of unknown origin, serological method

Background and objectives: Fever of unknown origin (FUO) can be caused by broad variability of zoonotic infectious agents. Studying zoonotic diseases in Kazakhstan is of great interest as the vastness of the territory harbors many natural foci for zoonotic infections. Aim of this study was to explore the seroprevalence of hantaviruses in patients' sera suffering on FUO in Kazakhstan.
Materials and methods: The study was set up in 2015-2016 among patients with FUO in the two selected regions (Almaty, Kyzylorda) in 13 hospitals. Blood sampling was performed on the day of hospitalization and after 10-14 days. Paired sera were stored in aliquots at -20°C. For antibody investigation, commercial ELISA kits were used.
Results: In total 375 and 423 blood samples were investigated from Almaty and Kyzylorda region. 5 and 12 % (median 8.8%) of 2nd sera from the 9 hospitals in Almaty region contained hantavirus IgG. 2-22% (median 13,3%) of sera from the 4 hospitals in Kyzylorda region contained hantavirus IgG. Work on differentiation of acute and former infections is in progress.
Conclusion: Our study shows for the first time that hantavirus infections might be present in Kazakhstan. The data obtained also indicate that the diagnostics of hantaviruses among people with FUO is important. So far data on hantaviruses in rodents in Kazakhstan are sparse. Further studies on rodent are planned in order to characterize the circulating virus strains in the two pilot regions.

Lyme borreliosis in Germany: description of surveillance data 2013-2016

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Keywords: Lyme borreliosis, surveillance, epidemiology

Background and objectives: Lyme borreliosis (LB) is an important tick-borne disease. In 9/16 German federal states, notification of erythema migrans (EM), neuroborreliosis (NB) and lyme arthritis (LA) is mandatory. We describe surveillance data in order to allow estimation of disease burden, detect trends and to identify risk groups and high-incidence regions.

Materials and methods: We used cases notified in the 9 states and confirmed by the local health offices, 2013-2016, to calculate incidence by time, place and person.

Results: In total, we observed 45,181 cases. Disease onset peaked yearly in July/August. Incidence decreased from 41/100,000 in 2013 to 26/100,000 in 2015, then increased again to 38/100,000 in 2016 with marked variations on district level (range 0.5-138/100,000).

Median age of cases was 54 years with peaks in boys 5-9 years (36/100,000) and women 50-69 years (56/100,000). Of those with known manifestation 42,312/44,564 (95%) had EM only, 5% had severe forms; 1,256/44,564 (2.8%) NB, 1,019/44,564 (2.2%) LA. Overall 54% were female, but more men had NB (56%) and LA

(52%), p<0,001. 1,589/38,097 (4.3%) were hospitalized; 778/1,158 (68%) of NB and 78/877 (8.9%) of LA.
Conclusion: LB is frequent, with a high proportion of NB-cases requiring hospitalization. Health authorities should promote prevention strategies among the general population (tick-bite-protection, prompt tick removal and early medical consultation) with communication tailored to risk groups and raise awareness among physicians to facilitate early diagnosis and treatment.

Distribution and Risk Areas in Germany – Results of the National Database for Alveolar Echinococcosis

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Keywords: Alveolar Echinococcosis, Echinococcus multilocularis, Spatial Distribution

Background and objectives: Alveolar echinococcosis (AE) caused by the parasite *Echinococcus multilocularis* is a rare disease in Germany. The aim of this analysis was the detection of high risk areas and the determination of prevalence of AE in Germany.
Materials and methods: EpiInfo ™ was used for cartographic representation and visualization of the penetration position. The Moran's I geoanalysis by clusters and risk areas was performed with GeoDa™. SAS Version 9.2 was used for statistical analysis.
Results: The analysis of the AE cases (N= 523) revealed a concentration of diseases in Baden-Württemberg and Bavaria. Analysis based on Moran's I gave a positive spatial autocorrelation for Germany ($I = 0.208815$, $Z = 32.6175$, $p<0.001$). The prevalence for Germany in the period 1992-2016 resulted in 0.64 cases per 100,000 inhabitants. For Baden-Württemberg, the estimated prevalence was 2.18 cases and in Bavaria 1.48 cases per 100,000 inhabitants. The analysis on spatial autocorrelation and possible risk areas shows the direct neighborhood of "high-high" risk areas in the southeastern regions of Baden-Württemberg ($I = 0.188514$, $Z = 11.3197$, $p <0.001$) and in the southwestern part of Bavaria ($I = 0.176953$, $Z = 13.5144$, $p <0.001$).
Conclusion: Striking are clusters of illnesses in the area of the Swabian Alb uplands, the Alps and the foothills of the Alps. By informing the population in "high-high" risk areas, AE cases can possibly be prevented or diagnosed at an early stage.

Spread of human dirofilariasis in Europe

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Keywords: Dirofilaria, dirofilariasis, spread

Background and objectives: Dirofilariasis is considered an emerging infectious zoonosis in Europe. Humans are aberrant hosts, but infections can cause subcutaneous nodules and even cases of meningoencephalitis were observed. Recent reports indicated a noticeable spread of the disease from the Mediterranean to Eastern and Central Europe.
Materials and methods: In order to understand the spread of the disease over the past decades, the correlation between the length of the potential annual transmission periods and reports of human cases were analysed to determine the threshold of days differentiating between areas with and without risk of *Dirofilaria* transmission to humans.
Results: Due to rising temperatures in Europe, the annual periods of time suitable for *Dirofilaria* transmission became longer for many regions in Eastern and Central Europe resulting in a higher risk of human infections, which clearly corresponds to the increased number of published human case reports.
Conclusion: The spread of dirofilariasis in Europe is directly linked to an increase of the annual time windows allowing extrinsic incubation. While the spatial extent of the area under risk did not change significantly until the end of the 20th century, the disease showed a tremendous spread within the past two decades. Due to the clear temperature dependence of the parasite, rising temperatures in course of the climate change will probably result in a further spread of human dirofilariasis in Europe.

Session 5: New and Reemerging Zoonoses

October 13, 2017
11:00 – 12:30

Ballroom
Chairs: Martin Pfeffer and Rainer Ulrich

Pathogenesis and transmission of the novel highly pathogenic avian influenza H5N8 2016 virus in ferrets and mice

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Keywords: HPAIV, ferret model, virulence

Background and objectives: The incursion of new reassortants of HPAI H5N8 resulted in epidemic outbreaks among wild birds and poultry throughout Europe. In this study, the potential of cross-species infection was evaluated in mammalian animal models.
Materials and methods: The virulence and pathotype of A/tufted duck/Germany/AR8444/2016 H5N8 was assessed by experimental inoculation of ferrets and Balb/c mice. Viral titers in nasal washes or organ samples were determined. Sero-responses were measured by ELISA (NP and H5 proteins) and HI assay.
Results: The H5N8 virus replicated in mouse lungs and spread systemically to the brain without prior adaptation. An infectious dose of 10^2 $TCID_{50}$/animal resulted in a survival rate of 40%, while animals receiving higher dosages had to be euthanized until 9 DPI. Therefore, HPAIV H5N8 replicate in mice and demonstrated a high-pathogenicity phenotype.
Ferrets were i.n. inoculated with 10^6 $TCID_{50}$/animal with naive ferrets served as transmission sentinels by direct contact. No respiratory symptoms and only minor changes in body weight and body temperature were detected. From a single nasal wash sample virus was re-isolated, and moderate viral titers were determined from organ samples. The contact ferrets did not seroconvert.
Conclusion: Clade 2.3.4.4 H5N8 virus exhibited a mildly virulent phenotype in ferrets and was not transmissible, consistent with lack of reported human cases, while it demonstrated high-pathogenicity in mice.

Experimental transmission of Zika virus by mosquitoes from Central Europe

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Keywords: Zika virus, Aedes albopictus, vectorial capacity

Background and objectives: In 2015, Zika virus (ZIKV) emerged in Columbia and Brazil and spread rapidly across the American continent and the Caribbean, causing an epidemic with notable numbers of associated clinical cases of microcephaly and Guillain–Barré syndrome. Mosquitoes of the species *Aedes aegypti* (*Ae. aegypti*) and *Aedes albopictus* (*Ae. albopictus*) are considered the primary and secondary vectors of ZIKV. Due to the spread of *Ae. albopictus* in Europe, it is of interest to assess the vectorial capacity. Furthermore, the question remains whether other mosquito species. play a role in transmission of ZIKV. In addition, to assess the risk of possible spread to regions with temperate climate such as Central Europe, information is required on ZIKV vector competence under reduced temperature conditions (< 20°C).
Material and methods: This study aimed to evaluate the vector competence of central European mosquito species for ZIKV. Therefore, taxa of the *Culex pipiens* complex and the invasive species *Ae. albopictus* were collected in Germany and challenged with ZIKV at 18 °C or 27 °C.
Results: None of the *Culex* taxa showed vector competence for ZIKV. In contrast, saliva of *Ae. albopictus* was positive for infectious virus particles, but only at elevated temperature of 27°C. Of note, none of the tested *Aedes* populations were susceptible to ZIKV at 18°C.
Conclusion: These results may limit the spread of ZIKV in central Europe to short summer periods with high temperatures.

Hepeviruses in small mammals: RabbitHEV in two populations in and around Frankfurt/ Main, Germany

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Keywords: rabbitHEV, zoonotic, Europe

Background and objectives: Rabbit hepatitis E virus (RabbitHEV) has a high zoonotic potential and belongs to the human pathogenic HEV-genotype 3 (HEV-3). It was reported for the first time in rabbits from China, USA, France, Italy and Germany, but recently also in patients from France.
Materials and methods: About 70 rabbits (*Oryctolagus cuniculus*), trapped in the city of and around Frankfurt/Main were investigated by specific HEV3-RT-qPCR, conventional RT-PCR and commercial antibody ELISA.
Results: In 25 of 72 animals HEV-specific antibodies were detected, but only 13 animals positive in the RT-qPCR and on the other hand all of them plus 5 were RNA-positive in conventional RT-PCR. Only seven animals were antibody and HEV-RNA-positive at the same time. Phylogenetic analysis revealed a subclade of novel sequences from Germany as sister-clade to already known rabbitHEV-sequences. Showing a higher similarity to Chinese sequences compared to European ones.
Conclusion: Identification of rabbitHEV in two populations in and close to a human high-density area indicates a potential health threat.

The impact of polymorphisms within the Ebola virus glycoprotein on host cell entry

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Keywords: Ebola virus, Glycoprotein, Polymorphisms

Background and Objectives: Ebola virus (EBOV) infection can cause a severe disease, Ebola virus disease (EVD), with high case-fatality rates. Host cell entry is facilitated by the EBOV glycoprotein (GP). Due to the large scale of the EVD outbreak in West Africa in 2013-2016 it was questioned if the host cell interactions of the responsible EBOV strain differed from those of other ebolaviruses.
Materials and Methods: Using rhabdoviral vectors, virus-like particles and authentic EBOV, we compared host cell entry driven by the GP of the virus responsible for the West African EVD epidemic (EBOV2014-GP) and the virus circulating in Zaire in 1976 (EBOV1976-GP).
Results: We observed that in relation to EBOV1976-GP, EBOV2014-GP mediated entry into cells of nonhuman primate origin as well as primary human macrophages and dendritic cells with reduced efficiency. By generation of mutant GPs we further showed, that single amino acid polymorphisms, present in the receptor binding domain (RBD) and internal fusion loop (IFL), affect host cell entry. While the RBD polymorphism had only a moderate effect, switching the polymorphic residue within the IFL between both GPs strongly affected entry efficiency. Moreover, the latter polymorphism was found to be responsible for the aforementioned reduced entry capacity.

Multiple detection of zoonotic variegated squirrel bornavirus 1 in different squirrel species

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Keywords: variegated squirrel 1 borna virus, encephalitis, animal trial

Background and objectives: A recently discovered novel putative zoonotic bornavirus: variegated squirrel bornavirus 1 (VSBV-1), caused fatal encephalitides in three squirrel breeders and a zookeeper.
Materials and methods: We screened more than 750 squirrels of 18 different species (oral swab samples and in parts fecal pool samples) using the published VSBV-1 specific RT-qPCR. We co-cultivated infected primary squirrel cells with a permanent cell line and isolated infectious virus from these passaged cells. The virus isolate was used for electron microscopy and for intracranial infection of neonatal rats.
Results: The screening revealed a positivity rate of 3.5%, including squirrels of the subfamilies *Sciurinae* (1.5%) and *Callosciurinae* (8.5%). Phylogenetic analysis of the 28 VSBV-1 sequences revealed a holding-related clustering of the sequences, independent from the squirrel species. Electron microscopy exhibited the typical structure of a bornavirus. After a few weeks, two of four rats were VSBV-1 genome positive and virus re-isolation from rat brain material was successful.
Conclusion: In conclusion, the non-invasive sampling methods seems to be suitable for rapid screening of squirrels and revealed further infected animals of different squirrel species, representing a threat for humans handling squirrels. In addition, we were able to isolate the virus and passage it in rats, which is a precondition for further animal trials.

Diverse novel orthobunyaviruses detected in sylvatic mosquitoes of the Panama Canal Zone

M. Marklewitz[1,2], K. Hermanns[1], J. Schmid[2,3], G. Eibner[2,3], F. Zirkel[1,4], J. R. Loaiza[2,5], P. Trippner[1], S. Sommer[3], C. Drosten[1,4], S. Junglen[1,4]

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Keywords: arbovirus, bunyavirus, mosquito

Background and objectives: Members of the genus *Orthobunyavirus* (order *Bunyavirales*, family *Peribunyaviridae*) can cause severe diseases in humans & animals, such as *La Crosse* & *Schmallenberg orthobunyavirus*. Little is known about the diversity of orthobunyaviruses in sylvatic amplification cycles. Here we tested for orthobunyavirus infections in sylvatic Central American mosquitoes.
Materials and methods: Mosquitoes (n=13.807) collected in the Panama Canal Zone in 2013-14 were tested for orthobunyaviruses by generic RT-PCR. Insect & vertebrate cells were used for virus isolation and growth kinetics. Full genome sequencing was performed by NGS & RACE-PCR.
Results: Five diverse new orthobunyaviruses and a novel strain of *Gamboa orthobunyavirus* were detected by RT-PCR. The viruses showed identities of 54-79% in the palm motif of the viral polymerase to known orthobunyaviruses suggesting the identification of five new species. A virus named *Gigante orthobunyavirus* (GIGV) was isolated from *Culex Melanoconion sp.* in C6/36 & VeroE6/7 cells. Proteins of GIGV showed maximal identities of 26-70% to known viruses. Phylogenetic analyses suggest that GIGV may belong to a new serogroup. Human & bird cells were susceptible to GIGV infection and replication peaked in rodent cells suggesting that rodents may play a role in the amplification cycle. Rodent serum samples collected simultaneously are being tested for GIGV infections.
Conclusion: Our data show that sylvatic mosquitoes are infected with a high genetic diversity of previously unknown orthobunyaviruses

Session 6: Public Health and Social Issues of Zoonoses Research

October 13, 2017
11:00 – 12:30

Room Zehlendorf
Chairs: Uwe Rösler and Shari Fell

Comparison and optimization of methods for virus detection in frozen berries

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Keywords: detection method, viruses, frozen berries

Background & objectives: Frozen Berries have been repeatedly identified as vehicles for transmission of human-pathogenic viruses, such as noroviruses (NoV) and hepatitis A virus. In addition, zoonotic viruses like rotavirus and hepatitis E virus have been identified in this type of food. However, detection of viruses in berries is difficult due to the presence of high amounts of PCR inhibitors. In this study, available virus detection methods were compared and further optimized.
Materials & methods: Different virus extraction methods in combination with real-time RT-PCR were compared using frozen strawberries artificially contaminated with NoV and the recovery rates (RRs) were calculated.
Results: Testing different batches of strawberries, highly variable RRs indicating the presence of different amounts of PCR inhibitors were recorded. Out of 5 published protocols, the method according to ISO/TS15216 revealed the best RRs. Further improvement of RRs from 2.83±2.92% to 15.28±9.73% was achieved by an additional RNA purification step using Sephacryl-based columns. The optimized protocol also showed better NoV detection rates by testing strawberries previously involved in a gastroenteritis outbreak.
Conclusion: Inclusion of an additional RNA purification step can increase the NoV detection rates using the ISO method in frozen berries leading to significantly higher detection rates

Full genomes of new HEV and HPgV strains, Sudan

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Keywords: Hepatitis E Virus, Human Pegivirus, Hemorrhagic Fever

Backgound and objectives: In 2014 an outbreak of hemorrhagic fever was reported from different states of Sudan (South Darfur, West Kordofan, South Kordofan). The National Public Health Institute, Khartoum forwarded 28 sera samples to the RKI.
Material and methods: To elucidate possible yet unrecognized infections, all samples were subjected to metagenomic deep sequencing on an Illumina MiSeq. Further, qPCR and ELISA were utilized to investigate the new strains in the sample set.
Results: By sequencing, two new virus strains were discovered. The full genome of a Hepatitis E virus (HEV_Sudan_2014) and the full genome of a Human Pegivirus (HPgV_Sudan_2014) could be assembled, annotated and phylogenetically analyzed (RdRP, ORF1, ORF2, ORF3). ELISA analysis for HEV resulted in high IgM and IgG titers for all samples collected in South Darfur region.
Conclusion: Phylogenetically, all HEV_Sudan_2014 ORFs clustered as the closest relative to strains from Chad. However, the phylogenetic reconstruction of ORF3 resulted in different clusters and the higher divergence of this ORF was also confirmed by the heatmap of distances. The strain HPgV_Sudan_2014 is closely related to strains from South Africa and East Africa. The results of the analysis will be discussed in context of outbreak investigations.

Zika virus infection is enhanced in the presence of dengue antibodies in human placenta explants

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Keywords: zika virus (ZIKV), placenta, antibody dependent enhancement (ADE)

Background and objectives: The current ZIKV outbreak is associated with severe neurorological disorders (e.g. microcephaly) in neonates. The mode of transmission from an infected mother to the fetus is unclear but it was shown that ZIKV can infect placenta cells. Preexisting antibodies against the phylogenetically related flavivirus dengue virus (DENV) are cross-reactive with ZIKV and can enhance ZIKV infection *in vitro* and lead to a more severe pathogenesis in mice. Our aim was to study the infection and replication of ZIKV at the placenta barrier in the presence and absence of dengue antibodies.

Materials and methods: Explants from amnion, decidua and villi of term human placenta were infected with ZIKV alone or with ZIKV preincubated with human sera against DENV, and control sera (e.g. against Chikungunya virus). Viral RNA was extracted from the supernatant and virus replication was measured by quantitative real-time PCR.

Results: ZIKV replicated in all types of explant tissues and reached a plateau 4-8 dpi. Preincubation of ZIKV with DENV antibodies enhanced infection rates of the explants. Additionally the preincubation with DENV antibodies led to a faster replication and significantly higher numbers of genome copies in comparison to ZIKV alone or to ZIKV preincubated with control sera.

Conclusion: Pregnant women having DENV antibodies are likely to have higher chances of maternal-fetal ZIKV transmission through the placenta.

Interdisciplinary Communication in the Zoonoses Research Community in Germany

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Keywords: interdisciplinary communication, zoonoses research community, expert interviews

Background and objectives: There are several structures and frame conditions in Germany, allowing the implementation of a One Health approach. However there is still a strong need for direct communication among the stakeholders ranging from research to application.
Materials and methods: To determine their communicational structures, needs and conditions ten experts, working in the field of zoonoses, were interviewed by using a standardized questionnaire in 2015. The experts answered the questions by using a sample project, which could be working in a research cluster, managing a crisis or organizing an event in the field of zoonoses.
Results: Despite the interviewee's different positions, projects and tasks – 'science' was the most linking area among them. The areas of 'human' and 'animal health', as well as 'laboratory' constituted a key role. In the chosen sample projects 'data management' was a recurring topic, especially the harmonization of data between human and animal medicine. The general satisfaction with the communicational situation in the sample projects was surprisingly high. If measures for optimizing the communication in the sample project were implemented, 90% of the interviewees considered these measures as (rather) successful.
Conclusion: The questionnaire developed in this study may be applied in further investigations with a larger target group for evaluating and comparing the existing communicational structures, needs and conditions in the field of zoonoses.

Haemolytic uraemic syndrome (HUS) outbreak caused by sorbitol-fermenting (SF) Shiga toxin-producing Escherichia coli (STEC) O157, Germany, December 2016 to May 2017

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Keywords: haemolytic uraemic syndrome (HUS); Shiga toxin-producing E. coli - STEC; outbreak

Background and objectives: In 2/2017, sorbitol-fermenting (SF) *stx1* negative, *stx2* positive, *eae* positive *E. coli* (STEC) O157:H- was detected in 4 patients with haemolytic-uremic syndrome (HUS). In order to control the outbreak, we initiated investigations to identify the causal source of infection.

Materials and methods: Cases were defined as confirmed outbreak cases if they had HUS or STEC enteritis from 12/2016 on, were residing in Germany and had laboratory confirmation via WGS or PFGE. We conducted explorative interviews and a case-control study to identify food items as possible infection vehicles. We estimated matched odds ratios (OR) for consumed food items and supermarkets using the Mantel-Haenszel test. Food safety authorities performed sampling of food and trace-back investigations.

Results: Molecular typing showed close relatedness between isolates from 14 confirmed cases. Mean age was 8.5 years (range: 1–36),

50% were male, 13 developed HUS; one patient died. We included 9 cases and 35 controls in the analysis. Cases had eaten mixed minced meat more frequently than controls (p=0.015), and were likely to having bought it at supermarket chain X (OR: 14.1; 95% CI: 1.2–174.9).
No cases were detected outside Germany. Food safety investigations are ongoing.
Conclusion: This is the largest outbreak of the highly virulent SF STEC O157 in Germany since 2002. As of May 2017, further cases may occur as the source appears to remain active. Food safety investigations are continuing to identify the definite cause of the outbreak.

Evaluation of predisposing factors for a long-time colonization of pigs with livestock-associated MRSA via the airborne route in a newly established animal model

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Keywords: MRSA, aerosol, pig

Background and objectives: Methicillin-resistant *Staphylococcus aureus* (MRSA) is a concern in healthcare systems. The occurrence of livestock-associated MRSA is a common phenomenon in pigs and a spread via an airborne route was assumed.
The aim of our ongoing study is to identify the dose for a successful airborne colonization of pigs with MRSA ST398 and to evaluate the impact of predisposing factors on MRSA colonization.
Materials and methods: For this, we established an experimental aerogen infection model for pigs using an aerosol chamber and we following investigated the impact of a decreased systemic immunity on the La-MRSA infection rate.
Results: All animals were colonised after the exposure with a dosage of 10^4 cfu/m^3), followed by a dropping number of colonized animals until day 14 after infection. In contrast, all animals infected with 10^6 cfu/m^3 were MRSA-positive for the entire screening period (21 days).
Conclusion: These data indicate that an airborne route of MRSA exists. Moreover, our current data show that a general immunosuppression of piglets does not significantly influence a La-MRSA colonization of weaned piglets.

Poster Presentations

Poster Session Risk assessment, epidemiology and modelling

R01

Circulation of Influenza A viruses in Ghana: The human-animal interface

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Keywords: Avian Influenza Virus, poultry, animal-handlers

Background and objectives: Aquatic birds are the reservoir of avian influenza viruses and serve as a constant but low-risk infectious source to domestic poultry. Avian influenza viruses are low pathogenic (LP) and high pathogenic (HP) in nature. LP types presents with mild to no clinical manifestations and some can evolve into HPs, causing severe illness and death in susceptible species. Avian influenza virus can mutate and show increased transmissibility and pathogenicity in humans. Regular surveillance in susceptible species is necessary for the identification of newly introduced viruses, to gain knowledge of viral reassortments that could be of public health concern, and to evaluate their pandemic potential. Information on LPAI in Ghana is poor. We plan to develop tools to evaluate circulating influenza strains for their public health importance. We investigated if LPAI circulates in poultry and poultry farmers in Ghana.
Materials and methods: 1,200 tracheal and cloacal swabs each were collected from chickens in 65 farms in the Ashanti Region. 159 throat swabs were also collected from farmers. Swabs were analysed for Influenza A by PCR.
Results: 9.4% animal pools tested positive. Of these 58.1% and 41.9% were sampled during the rainy and dry season respectively. 83.9% of positive pools were cloacal samples. 16.7% of human throat sample pools tested positive.
Conclusion: Influenza A viruses circulate in domestic poultry and poultry farmers in the Ashanti Region, raising issues for public health concern.

R02

Modelling spatial risk of Usutu disease in Europe: a comparison between a species distribution model and an epidemic model

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Keywords: MaxEnt, SIR, vector-borne disease

Background and objectives: Dealing with vector borne diseases, risk mapping is essential, especially under changing climate. This can be achieved by species distribution models (SDMs) and also by epidemic models (EMs). To provide information on model choosing, a widely used SDM, MaxEnt, and an EM, namely Susceptible-Infected-Removed model (SIR), were compared in this study. An emerging mosquito borne zoonotic disease, caused by Usutu virus (USUV), was used as a case study.
Materials and methods: SDM and EM model were performed within a rectangular study area across Europe, with a time span of 2003-2016. The EM, mainly driven by daily mean temperature, was adapted to calculate the basic reproduction number within each raster cell. To run MaxEnt, bio-climatic variables were derived from daily temperature and precipitation values, from the same database as used in the SIR model. Occurrence records of USUV were derived from field (BNI) and literature data. The risk maps from the two models were compared afterwards.
Results: Both models show similar patterns near the eastern border of France, in the Pannonian Basin as well as northern Italy. While the SDM correctly predicts the occurrences in the Netherlands and western Germany, the EM highlights additional areas along the Mediterranean coast and across the Iberian Peninsula.
Conclusion: The SDM matches the observed occurrence more closely than the EM. However, the EM has the advantage of being able to reflect temporal dynamics.

R03

Abundance of *Ixodes ricinus* and the prevalence of Lyme Borrelia pathogenic to humans

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Keywords: Ixodes ricinus, Lyme disease, theoretical risk of exposure

Background and objectives: Lyme disease (LD) is a zoonotic, bacterial infection in humans caused by spirochetes of the *Borrelia burgdorferi* sensu lato complex, transmitted by *Ixodes ricinus* ticks. This study focusses on LD prevention by examining the distributional patterns of questing Ixodes ricinus.
Materials and methods: From 2010-2012 the survey was performed at three study sites in Berlin (Gatow, Tegel, Wannsee). Additionally, the prevalence of genospecies pathogenic to humans was determined in questing ticks. This prevalence of infection in conjunction with tick densities results in a theoretical exposure risk for people.
Results: When comparing the tick activity between study sites for 2010-2012, Tegel appeared to be dominated by nymphal ticks and Gatow by adult ticks. The highest prevalence of *Borrelia* sp. in questing ticks was detected at the Wannsee study site, where it exceeded the European average. The results illustrate that the activity of questing ticks and their prevalence of pathogenic borreliae varies tremendously.
Conclusion: Keeping this in mind, risk maps for borreliae-infected ticks seem to be of little use. Risk assessment for people should be based on the theoretical risk of exposure, determined locally. The methods developed in this study may be applied in further investigations. These future investigations may provide a basis for targeted landscaping in the sense of the One Health initiative.

R04

Alaria spp. *mesocercariae* in wild boar (*Sus scrofa*, Linnaeus, 1758) in the Berlin-Brandenburg region

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Keywords: Alaria mesocercariae, wild boar, Berlin-Brandenburg

Background and objectives: *Alaria* spp. are parasitic trematodes of carnivores. Its life cycle consists on seven developmental stages with two intermediate and one final host. The life cycle can be extended at the mesocercarial stage by the addition of a paratenic host such as omnivorous mammals. In Europe, *Alaria mesocercariae* have been increasingly found in game meat, particularly in the wild boar (*Sus scrofa*). Even though there are no reports of infected humans by *Alaria alata mesocercariae*, presently its zoonotic potential cannot be excluded. The objective of this study was to assess the presence of *Alaria* spp. mesocercariae in wild boars from the Berlin-Brandenburg region.

Materials and methods: From January to December 2016, samples from 75 wild boar carcasses obtained from hunted animals in the Berlin-Brandenburg region were tested using the well/established Alaria Migration Technique (AMT). The isolated mesocercariae from positive samples (muscle and adipose tissue) were morphologically identified as *Alaria* spp.

Results: Most samples originated from Brandenburg (70) and all positive samples (8) come from this area, specifically from Oranienburg (5), Storkow (2), and Zeuthen (1). Only adult animals tested positive for AMT. No positive samples were detected in Berlin (5). The prevalence of *Alaria* Spp. Mesocercariae in our study population was 8/75 (10.66%).

Conclusion: Our study highlights the presence of *Alaria* spp. *mesosercariae* in wild boars from the Berlin-Brandenburg region, since its zoonotic potential cannot be discarded, consumption of

undercooked wild boar meat could constitute a health risk for humans.

R05

Towards a monitoring of *Francisella tularensis* in Microtus arvalis in Europe

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Keywords: Francisella tularensis, APHAEA, common vole

Background and objectives: *Francisella tularensis* is a zoonotic, gram-negative bacterium that causes tularemia in humans. *Francisella tularensis* can infect a broad range of organisms including small mammals like hares and common voles (*Microtus arvalis*). This rodent species is involved in transmission in a waterborne cycle, especially during years with massive population outbreaks. Within the project “harmonized Approaches in monitoring wildlife Population Health, and Ecology and Abundance” (APHAEA) harmonized methods for abundance estimation of rodents and for diagnosis of rodent-borne pathogens in Europe have been defined.
Materials and methods: In this study common voles and other small mammals were collected according to the APHAEA-standard protocol in Germany and the Czech Republic. These and additional small mammals from France and Switzerland were investigated by a standard Francisella tularensis real-time polymerase chain reaction (PCR) assay and isolation.
Results: *Francisella tularensis* DNA was detected by tul4-qPCR in common voles from Switzerland and in field voles and a bank vole from one of our trapping sites in Germany. Typing revealed *Francisella tularensis* subspecies holarctica. The positive animals were all collected during autumn.

Conclusion: We confirm the importance of rodents as host of this zoonotic bacterium. In addition, we could also show that Francisella tularensis can be detected in certain close range hotspots.

R06

A research agenda for vector-borne disease risk in Europe: spatio-temporal hotspots and potential economic burden

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Keywords: vector-borne diseases, cost-of-illness, burden of disease

Background and objectives: Zoonoses like Dengue, Chikungunya, West-Nile, Zika, Rift Valley and Crimean Congo haemorrhagic fever are vector-borne highly symptomatic viral infections causing fever and in some cases persistent complaints. These diseases are an emerging global health risk. We aim to assess the potential health and economic burden of Zoonoses across Europe.
Materials and methods: The following research agenda for modelling zoonoses outbreaks will be applied: (0) selection of possibly important zoonoses in Europe by expert consultation, (A) bioclimatic envelope modelling to project climatically suitable areas at risk for the establishment of vectors and pathogens, (B) development of indicators of vulnerability and modelling the course of infections, (C) combination of A and B to analyse spatial and temporal hotspots and the associated health and economic burden.
Conclusion: Our structured and interdisciplinary research approach is promising to project potential health and economic burden of vector-borne diseases across Europe

Poster Session Pathogen-cell interaction

P01

Establishment of primary cell cultures from a microbat and study of their host cell factors for filovirus entry

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Keywords: Ebolavirus, microbat, virus entry

Background and objectives: The Ebola virus (EBOV) entry process involves host cell factors including Niemann-Pick C1 (NPC1), T cell immunoglobulin and mucin domain (TIM) and TAM receptor tyrosine kinases (Axl, Mer and Tyro3). The significance of these factors for EBOV entry has been determined in cell lines derived from humans, spillover hosts and potential reservoir hosts including fruit bats. Those factors potentiating EBOV entry into cells derived from microbats, another potential reservoir host, remain unstudied. In order to examine the capacity for EBOV entry into microbat cells, we derived primary cell cultures from numerous organs from *M. condylurus*, which has been shown as a susceptible species to experimental infection in a prior study. Primary cell cultures from different organs were generated for the receptor expression characterization, as various bat cell types have been shown to take up EBOV-GP with different efficiencies.
Materials and methods: Receptor expression was characterized with use of immunofluorescence in an attempt to correlate it with cell permissiveness to EBOV infection. Susceptibility of primary cells to EBOV infection was determined by the presence of CPE and growth kinetics.
Results: We could show that the microbat cells are positive for various receptors and EBOV is replicating successfully.
Conclusions: Overall, our findings elucidate the relationships between host cell receptor expression and EBOV replication efficiency in vitro in a potential EBOV reservoir species.

P02

Zoonotic virus infections in cell lines from bank voles and common voles

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Keywords: NETs, meningitis, antimicrobial peptide

Background: Zoonotic infections and emerging diseases are of major interest for human health. As adequate cell culture and animal models are lacking, pathogenesis and adaption to the reservoir host is often poorly understood. For a better understanding of the association of zoonotic viruses to a putative animal host, we generated cell lines from bank voles (*Myodes glareolus*, lung) and common voles (*Microtus arvalis*, brain, kidney).
Materials and methods: Spontaneously immortalized cells were inoculated using different viruses and observed for cytopathic effects (cpe): Cowpox virus (CPXV), Encephalomyocarditis virus (EMCV) 1 and 2, Murine herpesvirus 68 (MHV-68), Rift Valley fever virus (RVFV), Schmallenberg virus (SBV), Sindbis virus (SINV), Tick-borne encephalitis virus (TBEV), Usutu virus (USUV), Vaccinia virus (VACV) and West Nile virus (WNV).
Results: CPXV, VACV, MHV-68 and RVFV replicated in all cell lines causing cytopathic effects. Lung and kidney cells were susceptible to SBV, SINV and EMCV1 with SBV showing start of cpe only after 6-7 days. Flavivirus infection was observed in kidney cells (WNV) of common voles and lung cells of bank voles (TBEV). Interestingly, several of the viruses are productively replicated in the rodent cells without inducing cytopathic effects. This may indicate an adaption of the virus to its reservoir.
Conclusion: These newly developed cell lines are a useful tool to study host factors for rodent associated viruses and could be a valuable system to mimic virus infection in natural host tissues.

P03

Comparison of eleven G. duodenalis isolates suggests Caco-2 monolayers as a model for asymptomatic Giardia sp. infections

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Keywords: insect-specific virus, host range, species barrier

Background and objectives: The protozoan *Giardia duodenalis* is responsible for more than 280 million cases of gastrointestinal complaints ('giardiasis') every year, worldwide. A proposed pathomechanism is the induction of epithelial barrier dysfunction by apoptosis or alterations of the tight junction complex, which increase epithelial permeability and may impact nutrient uptake and normal gut function, as well as infiltration of luminal antigens and invasion of pathogens. However, enigmatic to giardiasis is the range of medical conditions from severe chronic enteritis to complete asymptomatic courses. We investigated whether *G. duodenalis* alone can compromise epithelial barrier function in our Caco-2 in vitro monolayer setup.
Materials and methods: Measurements of trans-epithelial electric resistances (TEER) on *Giardia*-infected Caco-2 transwell-monolayers were used to indicate epithelial permeability and alterations of tight junctions were analyzed via immunofluorescence assays.
Results: No evidence for barrier compromising effects using 11 G. duodenalis isolates (4x A, 6x B, 1x E assemblage) were found. Contrarily, using various culturing conditions and infection doses indicated a highly consistent dose-dependent TEER-increase. Furthermore, proposed on tight junction proteins was not consistently found.
Conclusion: Our investigations suggest that *Giardia*, in our Caco-2 model, follows the course of an asymptomatic infection and may require additional, yet unknown factors to induce barrier dysfunction.

P04

Integrin αvβ3 ablation impairs Zika virus replication

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Keywords: Zika virus, cellular receptor, viral replication

Background and objectives: Recently, Zika virus (ZIKV, genus *Flavivirus*) became an important pathogen for public health. Like other related flaviviruses ZIKV is transmitted by mosquitoes from reservoir hosts to susceptible organisms. To cause viremia in these organisms the virus has to be able to enter host cells and replicate therein. Continuing our previous work with Yellow fever virus and West Nile virus, we studied the role of integrins as putative cellular receptors for ZIKV and their involvement in replication.
Materials and methods: Mouse embryonic fibroblasts (MEF) devoid of integrin subunits αv and β3 were infected with ZIKV. Internalization of virus as well as viral replication was studied using a ZIKV specific RT-qPCR and a negative-strand specific RT-PCR.
Results: Internalization assays show that ZIKV is able to enter the cells independently from the expression of investigated integrin subunits. In contrast to this, the generation of viral particles found in the cell culture supernatant is impaired in integrin αvβ3 deficient cells. Our studies also showed that the intermediate generation of negative stranded RNA is reduced in cells with lower replication.
Conclusion: Like other flaviviruses, ZIKV does not depend on αvβ3 integrins as cellular receptors to enter their host cells. However, the replication is impaired in αvβ3 integrin deficient cells as also shown for other viruses of this genus. This is probably due to the lower level of negative strand RNA synthesis.

P05

Chimeric mumps viruses expressing the fusion and hemagglutinin-neuraminidase protein of a bat-derived paramyxovirus are highly fusion active and neurovirulent

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Keywords: bat-derived paramyxovirus, mumps virus, fusion

Background and objectives: Mumps is a highly contagious childhood disease with usually mild symptoms. In rare events, mumps can result in complications like encephalitis or meningitis. So far, humans are the only known host of MuV. The detection of a bat-derived paramyxovirus (batMuV) with high phylogenetic relatedness to human mumps viruses (hMuV) raises the question if bats may serve as an intermediate or reservoir host. So far, the zoonotic potential of batMuV is unknown.
Materials and methods: To analyze the batMuV fusion (F) and hemagglutinin-neuraminidase (HN) glycoproteins in a viral context, recombinant MuVs (rMuVs) in which the ORFs for F and HN of a hMuV strain were replaced by the corresponding ORFs of batMuV were generated.
Results: Both batMuV glycoproteins were incorporated into chimeric rMuVs. rMuVs expressing batMuV F and HN were highly fusogenic. The growth kinetics of rMuVs expressing the bat-derived viral glycoproteins were similar to the backbone virus. Neutralizing antibodies directed against different hMuV strains inhibited infection mediated by chimeric rMuVs. The enzymatic activity of batMuV HN was similar to that of hMuV and could be inhibited by group IV anti-MuV antibodies. Replacement of the hMuV HN gene by the corresponding batMuV gene led to a slight reduction in neurovirulence of the highly neurovirulent backbone strain.

Conclusion: We report the generation of rMuVs expressing F and HN of batMuV, providing a tool for further examination of this novel virus.

P06

NS segment reassortment can increase growth kinetics and pathogenicity of the 2009 pandemic H1N1 influenza A virus in vitro and in vivo

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Keywords: H1N1pdm09, reassortment, pathogenicity

Background and objective: The 2009 pandemic influenza A virus (IAV) H1N1 strain (H1N1pdm09) circulating in humans and swine together with other human and avian IAVs raises the concern that possible reassortment between H1N1pdm09 and these IAVs would occur increasing the pathogenicity in different susceptible hosts.
Materials/methods: we explored the impact of NS segment reassortment between H1N1pdm09 strain A/Giessen/06/09 (Gi-wt) and human IAVs H1N1 (A/Puerto Rico/8/34, (Gi-NS-PR8)); and H3N2 (A/Victoria/3/75, (Gi-NS-Vict)), and avian IAVs of H5-, H7- and H9-subtypes.
Results: a significant increase in the growth kinetics was noticed in vitro with Gi-NS-PR8 in A549 (human lung) and NPTr (porcine tracheal) cells, and increased HA mRNA transcription in differentiated THP-1 (human macrophage-like) cells, whereas, Gi-NS-Vict showed reduced replication efficiency compared to Gi-wt. All other Gi-NS-reassortants showed similar replication efficiencies to Gi-wt. The ex vivo tracheal organ cultures (TOCs) of turkeys demonstrated increased replication of Gi-NS-PR8 compared to Gi-wt. Also, Gi-NS-PR8 induced a markedly higher expression of cytokines, chemokines

and ISGs in A549 cells, THP-1-derived macrophages (dHTP) and TOCs. In vivo, Gi-NS-PR8 induced an earlier onset of mortality than Gi-wt in mice, whereas, six-week-old chickens were found to be resistant to both viruses.
Conclusion: NS-reassortment of H1N1pdm09 may lead to increased virulence with variable impact on different host species.

P07

Inhibition of subtilisin kexin isoenzyme-1 (SKI-1) and signal peptide peptidase (SPP) interferes with Ebola virus glycoprotein-driven host cell entry

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Keywords: influenza viruses, well-differentiated epithelium, air-liquid interface

Background and objectives: Ebola (EBOV) and Lassa virus (LASV) harbor a single viral glycoprotein in their envelopes for entry into target cells. Both GPs are synthesized as inactive precursors and depend on priming by host cell proteases for activation. EBOV can be primed by the cysteine proteases cathepsin B/L however other proteases might be involved upon virion uptake. Here, we analyzed the role of SKI-1 and SPP in EBOV-GP and LASV-GPC priming.
Materials and methods: We employed rhabdoviral vectors to analyze LASV-GPC- and EBOV-GP-driven entry and its inhibition by the SKI-1 inhibitor PF-429242 and the SPP inhibitor (Z-LL)2 ketone.
Results: As expected, the incubation of cells producing LASV-GPC-bearing VSV particles with SKI-1 inhibitor markedly reduced particle infectivity, while the effect on infectivity of EBOV-GP-bearing particles was modest. Entry driven by both LASV-GPC and EBOV-GP was efficiently reduced if inhibitor was added prior to infection. Blockage of LASV-GPC-dependent entry was rescued by addition of cholesterol, indicating cholesterol metabolism interference accounting for the block of LASV-GPC-driven entry by SKI-1 inhibitor. In contrast, no rescue was observed for EBOV-GP. Finally, the SPP inhibitor blocked EBOV-GP- but not LASV-GPC-dependent entry, and time-of-addition experiments revealed that entry was inhibited at an early stage.
Conclusion: Our results suggest that EBOV-GP maturation in infected cells is independent of SKI-1 and SPP activity while EBOV-GP-driven host cell entry depends at least partially on these proteases.

Poster Session Immune response and Vaccines

I01

Neutrophil function in response to zoonotic pathogens in wild and domestic animals of Costa Rica

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Keywords: neutrophils, wild animals, Costa Rica

Background and objectives: The first line of defence against infectious diseases in human as well as animals is mediated by neutrophils, which are well known as multitasking cells. They can mediate antimicrobial activity by different strategies depending on the pathogen they encounter. Besides phagocytosis, a key strategy against extracellular pathogens is the formation of neutrophil extracellular traps (NETs). Those NETs mainly consist of DNA that is decorated with antimicrobial components and mediates entrapment and killing of various pathogens.
The goal of this study is to compare differences of neutrophil function from wild and domestic animals of Costa Rica. This could lead to a better understanding how neutrophils of various animals react in response to zoonotic pathogens.
Materials, methods and results: Based on a systemic literature search, we analyzed the differences of neutrophils derived from various animals, focusing on domestic animals and wild animals in Costa Rica. Experimentally, neutrophils of wild animals of Costa Rica were characterized after a DIFF Quick staining of blood smears by microscopy analysis. Furthermore, the formation of NETs in dogs and wild animals was compared in response to relevant zoonotic pathogens of Costa Rica like Trypanosoma cruzi. The analysis was conducted by immunofluorescence microscopy.
Conclusion: In conclusion, the characterization of neutrophils in various animals and humans may be helpful to understand the antimicrobial capacity and overall role of neutrophils against zoonotic pathogens.

I02

Establishment of bone marrow-derived dendritic cells from microbats for the study of Ebola virus infection

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Keywords: Microbat, Immunology, Ebola virus

Background and objectives: It has been established with in vivo models that dendritic cells (DCs) are early targets of Ebola virus (Ebov) infection. Following Ebov entry, DC maturation is impaired, albeit high-titer replication is supported. The interactions between Ebov and DC have been characterised with use of human, mouse or non-human primate DCs, however the response of reservoir-host DCs to Ebov infection remains unstudied.
Materials and methods: We have developed a culture system for bone marrow-derived dendritic cells (BMDCs) from *Mops condylurus* (Moco), an Angolan Free-tailed Bat, suspected to be involved in spill-over during the 2014 West African outbreak. BM cells harvested from Moco were cultured with recombinant equine GM-CSF and IL-4. Their morphology and susceptibility to Ebov infection was further investigated.
Results: Following 5 days of culture, BMDCs appeared as semi-adherent clusters and after 11-12 days they exhibited prominent DC-like veiled morphology. The permissiveness of BMDC to Ebov-Zaire was also assessed to determine the suitability of these cultures for studying viral-reservoir-host DC interactions.
Conclusions: Equine GM-CSF and IL-4 supplementation of bat BM cultures generated cells with DC morphology and DC marker expression. Upon Ebov-Zaire infection of the DC cultures poor viral replication ensued, which differs from high Ebov replication evident in DCs from other species. The interaction between bat DC and Ebov warrants further investigation.

I03

Bourbon Virus: An unusual Thogoto-Orthomyxovirus

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Keywords: innate immunity, virus-host interaction, tick-borne zoonosis

Background and objectives: Thogotoviruses (THOV) are tick-transmitted Orthomyxoviruses that cause disease in livestock. A recent fatal human case of Bourbon virus (BRBV) infection in the US (2014) suggests a zoonotic potential. C57BL/6 mice succumb to THOV infection; however, no disease is detectable in BRBV infected mice. In this project we aim to develop a mouse model for BRBV infection and to elucidate the cause of viral restriction in C57BL/6 mice.
Material and methods: IFNAR/IL28R-/-, STAT1-/- or wt C57BL/6 mice were infected with BRBV. The virulence, organ tropism and LD50 were evaluated under BSL3 conditions.
Results: In contrast to C57BL/6 mice, mice deficient for the type I and III interferon receptor got progressively ill after BRBV infection. Within the first eight days the mice lost up to 18% bodyweight but fully recovered at day 12 even after high dose infections (105 PFU). Strikingly STAT1-/- mice died within 3 days after low dose infections (1 PFU).
Conclusion: We successfully established an in vivo model for BRBV infection in mice deficient in critical components of the interferon system. Our data indicate that an interferon-stimulated gene downstream of STAT1 is restricting BRBV and it stands to reason that the US patient might have had a defect in his innate immunity. Our future studies intend to clarify the discrepancy in disease progression between the STAT1-/- and IFNAR/IL28R-/- mice and to identify the gene product responsible for BRBV restriction.

I04

Identification and characterization of bacterial immunoactive factors of *Enterococcus faecium*

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Keywords: Enterococcus faecium SF68, NF-κB, intestinal epithelial cell line

Background and objectives: *Enterococcus faecium* SF68 (NCIMB 10415) is a probiotic bacterium shown to improve symptoms of intestinal inflammation in human and animal studies. In a prior animal feeding trial we observed significant reductions in immune-associated gene expression in intestinal tissues and associated lymphoid organs in post-weaning piglets.
Materials and methods: To identify and characterize possible immunomodulatory factors, cell-free, whole bacterial lysates of commensal and pathogenic *Enterococcus* strains were screened using an NF-κB reporter derivative of the porcine intestinal epithelial cell line IPEC-J2 and the human intestinal epithelial cell line Caco-2.
Result: All E. faecium isolates showed inhibitory effects on NF-κB activation, compared to untreated cells. In contrast, lysates prepared in the same manner and concentrations from *E. avium*, *E. gallinarum* and *E. casseliflavus* isolates did not show similar effects. Cytotoxicity and cell viability assays showed no measurable host cell cytotoxicity. Treated cells no longer responded to TLR2, TLR1/TLR2, or TLR4-ligands, although the cells remained capable of NF-κB activation in response to TLR5- and TLR3-ligands. Assays using proteinase K or heat treatment indicate the factor(s) present in E. faecium SF68 are proteinaceous in nature.
Conclusion: The results indicate E. faecium strains produce an immunosuppressive factor inhibitory for NF-κB activation in porcine and human intestinal epithelial cell lines.

I05

Novel monoclonal antibodies and alpaca derived single domain antibody fragments (VHH) against Rift Valley fever virus

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Keywords: antibodies, PRNT, RVFV

Background and objectives:
Rift Valley fever virus (RVFV) is a mosquito borne virus, which is responsible for large outbreaks affecting human and a wide range of vertebrate hosts, especially ruminants throughout Africa and the Arabian Peninsula.
There are no drugs approved to date for the therapy of RVF in humans. One frequently discussed promising option is the application of neutralizing antibodies against RVFV infection.
Materials and methods:
For generation of monoclonal antibodies BALB/c mice were immunized with the vaccine strain MP-12. Supernatants from generated hybridoma cells were screened by a newly established indirect immunofluorescence assay (IIFA). Neutralizing activities of reactive mabs were assessed using a serum neutralization test (SNT). Furthermore a new protocol was established to generate single-domain antibody fragments against RVFV by immunization of Alpacas with MP-12 strain. Specific VHH were identified by phage display, followed by IIFA and subsequent assessment for neutralizing activity.
Results:
One specific neutralizing mab could be identified, which displayed synergistic effects in combination with recently generated mab Gn3 . Moreover 5 of 7 IIFA positive VHH showed neutralizing activities.
Conclusion:
Antibodies with neutralizing activity against RVF were generated and will be further evaluated for in-vivo studies.

I06

Lactobacillus johnsonii ameliorates intestinal, extra-intestinal and systemic pro-inflammatory immune responses following murine *Campylobacter jejuni* infection

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Keywords: Campylobacter jejuni, bacterial in vivo competition, bacteria-host interaction

Background and objectives: *Campylobacter jejuni* infections are progressively increasing worldwide. Probiotic treatment might open novel therapeutic or even prophylactic approaches to combat campylobacteriosis.
Materials and methods: In the present study secondary abiotic mice were generated by broad-spectrum antibiotic treatment and perorally reassociated with a commensal murine *Lactobacillus johnsonii* strain either 14 days before (i.e. prophylactic regimen) or 7 days after (i.e. therapeutic regimen) peroral C. jejuni strain 81-176 infection.
Results: Following peroral reassociation both *C. jejuni* and *L. johnsonii* were able to stably colonize the murine intestinal tract. Neither therapeutic nor prophylactic *L. johnsonii* application, however, could decrease intestinal C. jejuni burdens. Notably, *C. jejuni* induced colonic apoptosis could be ameliorated by prophylactic *L. johnsonii* treatment, whereas co-administration of *L. johnsonii* impacted adaptive (i.e. T and B lymphocytes, regulatory T cells), but not innate (i.e. macrophages and monocytes) immune cell responses in the intestinal tract. Strikingly, *C. jejuni* induced intestinal, extra-intestinal and systemic secretion of pro-inflammatory mediators (such as IL-6, MCP-1, TNF and nitric oxide) could be alleviated by peroral *L. johnsonii c*hallenge.
Conclusion: Immunomodulatory probiotic species might offer valuable strategies for prophylaxis and / or treatment of *C. jejuni* induced intestinal, extra-intestinal as well as systemic pro-inflammatory immune responses in vivo.

I07

When a virus uses another entrance - Immune mechanisms involved in innate anti- Lyssaviruses immune response in nasal cavity of European bats

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Keywords: lyssavirus, bat, interferon

Background and objectives: Lyssaviruses are neurotropic viruses causing fatal encephalitis of central nerve system, rabies. In Europe, bats act as reservoir hosts for two specific Lyssaviruses, European Bat Lyssavirus (EBLV-1 and -2). Although cases of rabies in bats caused by EBLV 1 or 2 are described, there are no reports about epidemics in bats and Lyssavirus specific antibody titers in European bats were only rarely detected. This indicates that innate immune pathways might be responsible for the observed resistance in bats
Materials and methods: The interferon Type I and III family of two European bats species E.serotinus and M.myotis were cloned and sequenced. Using established cell lines from nasal epithelium (MmNep) nervus olfactorius (MmNol), M. myotis brain (MmBr) the IFN responses along the aerosol infection route by investigation of IFN signaling pathways, induction of IFNs and interferon stimulated genes (ISGs) and anti-viral effects in correlation to the expressions of viral receptors was analyzed in-vitro..

Results: The sequenced Type I IFN α, β, δ, ε, κ, ω and τ and Type III IFN λ of both bat species are structurally and functionally typical IFN's. Interestingly, a gradual decreased susceptibility along the aerosol route combined with an increased IFN response was measured.
Conclusion: Bats as reservoir host for EBLV 'provide' a peripheral replication site but laos block effectively the spread of Lyssaviruses to central nerve system indicating a specific co-evolutionary relation.
Conclusion: Immunomodulatory probiotic species might offer valuable strategies for prophylaxis and / or treatment of C. jejuni induced

intestinal, extra-intestinal as well as systemic pro-inflammatory immune responses in vivo.

I08

Comparative analysis of immunogenicity of Rift Valley fever virus glycoprotein Gc recombinantly expressed in different expression systems

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Keywords: Rift Valley fever virus, Leishmania tarentolae, protein expression

Background and objectives: Rift Valley fever virus (RVFV) is a mosquito borne virus, which is responsible for large outbreaks affecting human and a wide range of vertebrate hosts, especially ruminants throughout Africa and the Arabian Peninsula.
There are no drugs approved to date for the therapy of RVF in humans. One frequently discussed promising option is the application of neutralizing antibodies against RVFV infection.
Materials and methods: For generation of monoclonal antibodies BALB/c mice were immunized with the vaccine strain MP-12. Supernatants from generated hybridoma cells were screened by a newly established indirect immunofluorescence assay (IIFA). Neutralizing activities of reactive mabs were assessed using a serum neutralization test (SNT).
Furthermore a new protocol was established to generate single-domain antibody fragments against RVFV by immunization of Alpacas with MP-12 strain. Specific VHH were identified by phage display, followed by IIFA and subsequent assessment for neutralizing activity.
Results: One specific neutralizing mab could be identified, which displayed synergistic effects in combination with recently generated mab Gn3 .
Moreover 5 of 7 IIFA positive VHH showed neutralizing activities.
Conclusion: Antibodies with neutralizing activity against RVF were generated and will be further evaluated for in-vivo studies.

I09

Epidemic versus reservoir-associated SARS-CoV: Papain-like protease of the epidemic strain has an increased interferon antagonistic activity

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Keywords: SARS-related Bat-Coronavirus, Papain-like protease, Interferon antagonist

Background and objectives: SARS-Coronavirus (SARS-CoV), a zoonotic agent hosted by *Rhinolophus* sp. bats, caused an epidemic with sustained human-to-human transmission. Although there is a plethora of sequence information available, little is known about functional differences between SARS-CoV and its bat-borne pendants. The papain-like protease (PLP) plays a central role in the viral replication cycle of SARS-CoV. PLP cleaves the viral polyprotein, deubiquitinates viral and cellular proteins and antagonizes the interferon (IFN) response.
Materials and Methods: To study the functional diversity we compared SARS-CoV PLP (SA-PLP) with bat-CoV PLP (bt-PLP) in in-vitro assays. To compare anti-IFN functions in a full replicating virus we constructed a chimeric virus and exchanged SA-PLP with bt-PLP.
Results: We found that all SA-PLP functions were conserved in bt-PLP in human cells in in vitro assays. Bt-PLP perfectly integrated into the genomic background of the recombinant SARS-CoV and supported viral growth to wild-type levels in IFN deficient cells. In IFN competent cells, we found that SA-PLP has an additional anti-IFN function that does not depend on protease activity. Compared to the chimeric virus, this resulted in an elevated robustness towards IFN.
Conclusions: Overall our data suggest that the reservoir contains a considerable degree of functional diversity that interferes with highly conserved host functions. In the case of host transition these pre-existing variations may form the basis for virus evolution.

I10

Bat influenza virus chimeras as basis for the development of a new type of vaccine backbone for livestock vaccination

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Keywords: bat influenza, vaccination, modified live vaccine

Background and objectives: Effective and safe influenza A viruses (IAV) live vaccines for animals do not exist with very few exceptions. This is mainly due to the risk of reassortment events between vaccine and wildtype strains, and partly because of the pathogenicity associated with live vaccine strains especially in very young animals. However, we recently succeeded to generate chimeric viruses containing the bat influenza A-like H17N10 backbone, and the glycoproteins hemagglutinin and neuraminidase from prototypic IAVs. These chimeric viruses do not reassort with ordinary IAVs.
Materials and methods: We passaged the chimeric viruses in eggs and day old chicks to adapt the viruses to the avian system. Chimeric vaccine viruses were subsequently tested for safety in chicken and in addition challenge experiments with HPAIVs were performed.
Results: By passaging the LP chimeric viruses in eggs and chicken the replicative potential was clearly improved as demonstrated by a reduced mean death time in eggs. These vaccine prototypes induced no clinical signs in neither adult nor day old chicks. Protection levels in adult chicken against a HP H5N1 or a HP H7N1 challenge achieved an efficiency of around 70%.
Conclusion: Bat influenza based modified live vaccines are able to induce protective immune responses in chicken. However, further adaptation to the avian system will likely improve the replicative potential and the corresponding immunological responses.

I11

Identifying cat transmission-relevant rodent reservoirs for virulent *Toxoplasma gondii* strains by analyzing IRGs-mediated resistance mechanisms

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Keywords: Toxoplasma gondii, non-model rodents, vole Interferon-γ, innate immunity

Background and objectives: One *Mus musculus* resistance mechanism to infection with virulent type I *T. gondii* strains relies on the polymorphic IFN-γ-induced Immunity-Related GTPase *Irgb2-b1*. However, cats prey more on other rodent species, such as *Myodes* spp., *Microtus* spp. and *Apodemus* spp., which also show higher *T. gondii* seroprevalences compared to *Mus* spp. We aim at assessing whether specific *Irg* sequences confer resistance to type I *T. gondii* infection in these rodents. The results will help defining ecologically important intermediate hosts for virulent parasite transmission to cats.
Materials and methods: Having access to a large collection of tissue samples across Germany from *Myodes* spp., *Microtus* spp. and *Apodemus* spp., we amplify *Irg* sequences identified in these rodents' genomes and express them in lab mice-derived fibroblasts susceptible to infection with type I *T. gondii* strains. Following infection we observe whether the introduced sequences lead to parasite death via disruption of the PVM.
Results: First results indicate substantial IRG amino acid diversity between and also within these species. As reference we have cloned *Irgb2-b1*-like cDNAs of available rodent cell lines, i.e. BVK168 *Myodes glareolus*, FMN-R *Microtus arvalis* and AAL-R *Apodemus agrarius,* after induction with either our custom-produced recombinant vole IFN-γ (MgIFN-γ) or mouse IFN-γ, respectively.
Conclusion: We confirmed the hypothesized *Irg* diversity in these rodent species

I12

Neuraminidase-expressing vesicular stomatitis virus replicons induce subtype-specific protection against influenza A virus

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Keywords: bat influenza, vaccination, modified live vaccine

Background and objectives: Effective and safe influenza A viruses (IAV) live vaccines for animals do not exist with very few exceptions. This is mainly due to the risk of reassortment events between vaccine and wildtype strains, and partly because of the pathogenicity associated with live vaccine strains especially in very young animals. However, we recently succeeded to generate chimeric viruses containing the bat influenza A-like H17N10 backbone, and the glycoproteins hemagglutinin and neuraminidase from prototypic IAVs. These chimeric viruses do not reassort with ordinary IAVs.
Materials and methods: We passaged the chimeric viruses in eggs and day old chicks to adapt the viruses to the avian system. Chimeric vaccine viruses were subsequently tested for safety in chicken and in addition challenge experiments with HPAIVs were performed.
Results: By passaging the LP chimeric viruses in eggs and chicken the replicative potential was clearly improved as demonstrated by a reduced mean death time in eggs. These vaccine prototypes induced no clinical signs in neither adult nor day old chicks. Protection levels in adult chicken against a HP H5N1 or a HP H7N1 challenge achieved an efficiency of around 70%.
Conclusion: Bat influenza based modified live vaccines are able to induce protective immune responses in chicken. However, further adaptation to the avian system will likely improve the replicative potential and the corresponding immunological responses.

Poster Session Antimicrobial use and resistance

A01

Reduction of *Escherichia coli* cell numbers by bacteriocins

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Keywords: bacteriocins, Escherichia coli

Background and objectives: Food can be contaminated with *Escherichia (E.) coli* along the food chain as a result of hygienic deficiencies. The use of bacteriocins provides a possible way to reduce the number of *E. coli* in food. Bacteriocins are proteins that are produced by almost all bacteria to lyse strains of the same and closely related species.
Materials and methods: The efficacy of bacteriocins to reduce the number of E. coli was analyzed at 4 °C in medium. Therefore, the indicator strain *E. coli* DH5α was mixed with bacteriocin concentrate and *E. coli* cell numbers were determined after different timepoints.
Results: Initial experiments showed a 6 log reduction of *E. coli* cell numbers after 15 min at 4°C with an initial cell number of 107 to 108 CFU/ml. After bacteriocin treatment, *E. coli* cell numbers remained constant with 102 to 103 CFU/ml for up to 24 h at 4°C.
Conclusion: Thus, bacteriocins could be an effective agent for decontamination of food under cooling temperatures.

A02

AMP-coated silicone membranes display high antimicrobial activity

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Keywords: Mastitis, AMP, antimicrobial surfaces

Background and objectives: Besides the clinical burden of affected animals, mastitis causes financial losses in the dairy industry worldwide, due to decreased milk production, medication or even culling of diseased animals. By reducing carry-over of bacteria from one cow to another during milking process, the rate of new infections can be declined. Thus, the aim of this study was to establish a coating strategy for antimicrobial peptides (AMPs) on teat rubbers to generate a surface with antimicrobial activity and thereby prevent bacterial adhesion.
Materials and methods: Silicone surfaces were modified by silanization with APTES or allylamine polymerization, followed by coating with two different AMPs, OH-CATH30 or BMAP-27, using the hetero bifunctional linker NHS-PEG-Mal. Antimicrobial activity against clinical mastitis isolates, one multidrug-resistant (MDR) *E. coli* and one methicillin-resistant *S. aureus* (MRSA), was investigated using a modified protocol based on ISO22196.
Results: OH-CATH30 coated on NHS-PEG-Mal-associated surfaces showed no remaining living bacteria, in contrast to $1{,}3 \times 10^8$ CFU/mL in the untreated control. For immobilized BMAP-27, we detected a microbial load of $4{,}1 \times 10^5$ CFU/mL. Antimicrobial activity of the surfaces was higher against MDR *E. coli* than MRSA.
Conclusion: We successfully established a method for coating of silicone membranes with AMPs, resulting in surfaces with detectable antimicrobial activity against two different mastitis pathogens

A03

Identification of extended-spectrum β-lactamase (ESBL)-producing Escherichia coli isolates in diseased food-producing animals from GERM-Vet 2008-2015

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Keywords: MRSA β-lactams, $bla_{CTX-M-1}$, multidrug-resistance

Background and objectives: Extended-spectrum β-lactamase (ESBL)-producing bacteria represent risks for public health. The aim of this study was to investigate ESBL-producing *Escherichia coli* isolates originating from diseased food-producing animals.
Materials and methods: A total of 7,810 *E. coli* collected from diseased cattle (n= 3,188), pigs (n= 1,834) or poultry (n= 2,788) in the GERM-Vet (2008-2015) were subjected to antimicrobial susceptibility testing and screened for ESBL phenotype. ESBL genes were investigated by PCR/sequencing. ESBL-producing isolates were further characterized by phylotyping.
Results: ESBL-producers were identified in 396 bovine, 95 swine and 24 avian isolates. The isolates were distributed among phylogenetic groups A (57.5%), D (23.7%), B1 (17.9%) and B2 (0.9%). The most common ESBL genes detected were: blaCTX-M-1 (68.1%), blaCTX-M-15 (15.0%) and blaCTX-M-14 (11.8%). Additional resistance to non-β-lactam antibiotics (especially sulphonamides/trimethoprim and tetracyclines) was seen in 488/515 of ESBL-producing isolates (94.8%) and 451/515 (87.6%) isolates were multidrug-resistant (resistant to at least three classes of antimicrobial agents).
Conclusion: The dominant ESBL gene among *E. coli* isolates from diseased food-producing animals over time is blaCTX-M-1. Additional resistance to non-β-lactam antibiotics may play an important role in the persistence and dissemination of ESBL genes.

A04

Multidrug-resistant extended-spectrum β-lactamase-producing Escherichia coli isolates originating from fresh vegetables

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Keywords: ESBL, multidrug-resistance plasmid, vegetables

Background and objectives: Extended-spectrum β-lactamase (ESBL)-producing *Escherichia coli* isolates from vegetables play an important role in foodborne outbreaks. The aim of this study was the characterization of ESBL-producing *E. coli* from vegetables.

Materials and methods: ESBL-producing *E. coli* were found in salad (2/202) and sprouts (5/43). They were investigated by antimicrobial susceptibility testing (AST), XbaI-PFGE, MLST and phylotyping. ESBL genes were detected by PCR/sequencing. Transformants carrying ESBL genes were investigated by AST, S1-nuclease PFGE, replicon typing, conjugation and tested for co-located antimicrobial resistance genes. Plasmid sequencing of one $bla_{\text{CTX-M-15}}$- and $bla_{\text{CTX-M-125}}$-carrying plasmids was performed using a HiSeq 2500 system.

Results: No clonal relationship was seen among the seven *E. coli*. They carried single plasmid-borne ESBL genes, $bla_{\text{CTX-M-14}}$ (n=2), $bla_{\text{CTX-M-15}}$ (n=3), $bla_{\text{CTX-M-65}}$ and $bla_{\text{CTX-M-125}}$ (each n=1). The isolates and 6/7 ESBL gene-carrying plasmids (70-245 kb) displayed multidrug-resistance. Sequencing revealed the ESBL gene in close location to the plasmid-mediated quinolone resistance gene *qnrS1* and $bla_{\text{TEM-1}}$ (*qnrS1*-IS*2*-Δ*tnpA*-$bla_{\text{CTX-M-15}}$-IS*Ecp1*-Δ*tnpA*-*tnpR*-$bla_{\text{TEM-1}}$) or the fosfomycin resistance gene *fosA3* ($bla_{\text{CTX-M-125}}$-ΔIS*903*-*fosA3*-*orf1*-*orf2*). All plasmids were conjugative, except an IncFIA-FIB plasmid.

Conclusion: Multidrug-resistance was detected among ESBL-producing *E. coli* and their ESBL gene-carrying plasmids.

A05

Novel extended-spectrum β-lactamase blaCTX-M-146 gene detected in a bovine Escherichia coli isolate

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Keywords: β-lactams, ESBL diversity, ESBL gene expression

Background and objectives: Frequency and diversity of extended-spectrum β-lactamases (ESBLs) have increasingly been reported. The aim of this study was to describe the novel ESBL gene variant *bla*$_{CTX-M-146}$.

Materials and methods: A putative ESBL-producing *E. coli* isolate, collected from a calf in the GE*RM*-Vet, was submitted to ESBL phenotypic confirmatory tests, ESBL gene investigations by PCR/sequencing, transformation and hybridization. A fragment containing the *bla*$_{CTX-M-146}$ gene and its flanking regions was amplified by PCR, cloned and transformed.

Results: The ESBL phenotype of the original *E. coli* isolate was confirmed by cefepime +/- clavulanic acid test. A novel ESBL gene, designated *bla*$_{CTX-M-146}$, was detected. The amino acid sequence of CTX-M-146 revealed 99% identity with CTX-M-1, a single amino acid exchange (K234R) was found. Transfer experiments were unsuccessful and hybridization experiments confirmed a chromosomal location of *bla*$_{CTX-M-146}$. The original isolate displayed lower cefotaxime, ceftazidime or cefepime minimal inhibitory concentrations (MICs) than the clone harbouring *bla*$_{CTX-M-146}$: <0.25 vs 6 mg/L, <0.5 vs 12 mg/L or 2 vs >16 mg/L, respectively. The ESBL phenotype of the clone was confirmed by cefotaxime, ceftazidime and cefepime +/- clavulanic acid tests.

Conclusion: Functionality of the novel ESBL gene *bla*$_{CTX-M-146}$ was confirmed by cloning. However, different MICs of the original isolate and the clone suggest a higher expression when plas

A06

Colistin resistance gene mcr-2 in pathogenic Escherichia coli from swine

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Keywords: colistin, mcr-2, swine

Background and objectives: In 2016, a new plasmid-borne colistin resistance gene, *mcr-2*, was reported in porcine *Escherichia coli* in Belgium soon after the discovery of the paradigm gene *mcr-1*. While *mcr-1* has disseminated globally, *mcr-2* has not been reported since the original publication. Therefore, we examined the presence of *mcr-2* in *E. coli* from pigs.
Materials and methods: Faecal *E. coli* (n=7,220) isolated from piglets in 1999 to 2015 and tested positive for at least one virulence gene typically associated with disease in swine were tested for *mcr-2* by PCR. Samples originated from 16 European countries, mostly (86%) from Germany. MICs were determined by broth microdilution and VITEK2. Clonal relatedness was tested by PFGE; resistance, virulence, MLST and plasmid data were examined by WGS.
Results: The *mcr-2* gene was detected in 14 (0.37%) isolates obtained from Belgium (n=11; 3 different farms), Germany (2), and Spain (1). Predicted pathotypes were enterotoxigenic *E. coli* (ETEC; n=12), enteropathogenic *E. coli* (EPEC; n=1), and non-typeable (n=1). Six STs and nine pulsotypes (A-I) were identified; clonal strains were present in two of the Belgium farms. MICs of colistin ranged between 0.5 and 4 mg/L. Mcr-2 was predominantly encoded on transferable IncX4 plasmids with >99% sequence similarity to reference plasmid pKP37-BE. In one isolate (ST29; EPEC), *mcr-2* was located on the chromosome but revealed the same flanking region (IS*Ec69*) as the plasmid variants.
Conclusion: Mcr-2-mediated colistin resistance in *E. coli* occurs at very low frequency in pig holdings. Its potential threat to livestock breeding and public health requires specific monitoring in the future.

A07

Impact of the horizontal gene transfer in *Klebsiella* spp. for the spread of antimicrobial resistances

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Keywords:, Antimicrobial resistance, Whole Genome Sequencing, Mobile Genetic Elements

Background and objectives: As antimicrobials are used both in the veterinary and human medicine, the resulting selective pressure may promote the establishment of resistance genes in bacteria. These bacteria may form a reservoir for the transfer of resistances to human pathogens or the human microbiota via direct contact or food handling. In this study, klebsiellae from animal species were analyzed regarding to their impact for spreading resistance genes to human pathogens.
Materials and methods: Isolates were pheno- and genotypically characterized. Antimicrobial susceptibility testing was performed using broth microdilution following CLSI guidelines and EUCAST epidemiological cut-off values. WGS and bioinformatical analysis were performed to reveal the genome composition. Filter-mating studies were used to determine the transferability of the plasmids.
Results: We show that a broad spectrum of *Klebsiella* spp. isolates from animals exhibit limited phenotypic resistances against antimicrobials, while only a few are multidrug-resistant. Filter-mating revealed that most of the resistance genes are located on plasmids that are transmissible to other Enterobacteriaceae. Analysis of the plasmid genome composition gives further insight into the transferability.
Conclusion: Antibiotic selective pressure contributes to the transfer of acquired resistance genes. In this context, animals may represent an important reservoir for the spread of resistance genes to human pathogens.

A08

MRSA and MRSP among employees and the environment of a small animal clinic

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Keywords: zoonotic pathogen, staphylococci, multiresistance

Background and objectives: The aim of the study was to investigate methicillin-resistant *Staphylococcus (S.) aureus* (MRSA) and methicillin-resistant *S. pseudintermedius* (MRSP) among employees of a small animal clinic and the clinic environment.
Materials and methods: Swabs from 96 employees and 73 from the clinic environment were investigated. CAMHB+6.5% NaCl was used for enrichment before plating on MH agar+2% NaCl+0.25 mg/L oxacillin. The species was determined using MALDI-TOF MS. The isolates were subjected to mecA-PCR, macrorestriction analysis, and antimicrobial susceptibility testing.
Results: MRSA were present in five employees and six environmental samples, MRSP in two employees and three environmental samples. All isolates harboured mecA. Susceptibility testing revealed that all but one isolates were multiresistant. All isolates were resistant to β-lactams and fluoroquinolones. All but one isolates were resistant to macrolides and lincosamides. A single MRSA was resistant to gentamicin. All MRSP were resistant to trimethoprim/sulfamethoxazole and resistant or intermediate to gentamicin. One isolate was also resistant to tetracycline. Macrorestriction analysis revealed three main SmaI patterns for MRSA and two main SmaI patterns for MRSP.
Conclusion: The finding of indistinguishable MRSA and MRSP among employees and in the environment of a small animal clinic suggests the possibility of transfer of these bacteria between humans, animals, and the clinic environment.

A09

Investigation of potential risk factors for the occurrence of Escherichia coli isolates from German fattening pig farms harbouring the mcr-1 colistin resistance gene

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Keywords: antimicrobial resistance, cross sectional investigation

Background and objectives: Soon after the finding of the plasmid-mediated colistin resistance gene mcr-1 in November 2015, working groups all over the world confirmed the presence of this gene in their strain collections. However, to date no analysis of factors associated with the occurrence of the mcr-1 gene in livestock has been published.
Materials and methods: Within the scope of a cross-sectional investigation on fattening pig farms conducted in 2011 and 2012, 48 fattening farms in Germany were investigated. Primary cultures of boot swabs and collective faecal samples were stored at 80°C and currently screened for the presence of the mcr-1 gene. Using logistic regression models the association between occurrence of mcr-1 and farm information were investigated.
Results: *E. coli* carrying the mcr-1 gene were isolated from 26 out of 216 mixed bacterial cultures (12.0%) originating from 12 out of 48 farms (25.0%). Farms and stables with low numbers of pigs were associated with a lower number of mcr-1-positive samples. The change of housing during fattening was associated with a higher number of mcr-1-positive samples. We found no statistically significant association between antimicrobial use and the occurrence of the mcr-1 gene.
Conclusion: Our results indicate that the transmission between pigs or their direct environment is crucial for the occurrence of resistant bacteria. Therefore, small animal groups, prevention of contact between animal groups and thorough application of hygiene measures should be recommended.

A10

Development of a high-throughput method for identification of Methicillin-susceptible *Staphylococcus aureus* (MSSA) progeny from epidemic MRSA

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Keywords: MRSA, resistance, large serine recombinases (ccr)

Background and objectives: Since accumulation of resistance in bacteria is promoted by use of antimicrobials, alternative ideas to combat antibiotic resistance are urgently needed. *Staphylococcus aureus*, especially Methicillin-resistant variants (MRSA), are a frequent cause of infectious diseases in humans and animals. Methicillin-resistance is mediated by the mecA gene, which is carried by the Staphylococcal Cassette Chromosome mec (SCCmec). Site-specific genomic excision/integration of SCCmec is catalysed by cassette chromosome recombinases (ccrA/ccrB or ccrC). Recent research indicates that the ccr activity is tightly regulated and influenced by growth conditions.
Materials and methods: MRSA_ST398_spa type t12359 was used as initial strain to set up a high-throughput screening procedure based on concurrent subcultures. To confirm candidate colonies as Methicillin-susceptible “offspring” generated during different growth scenarios, antimicrobial susceptibility testing, PCR proving the lack of mecA and subsequent whole genome sequencing was employed.
Results: Pheno- and genotypic analysis confirmed the ability of our method to identify methicillin susceptible colonies with a screening rate of approximately 800cfu/experiment/person/day.
Conclusion: We have set-up a procedure to identify growth conditions affecting the activity of the ccr genes in MRSA with respect to loss of mecA respectively the SCCmec element, a promising method for studying SCCmec integration/excision kinetics in further projects.

A11

Tracking the effects of high-Zinc oxide diets in porcine intestinal E. coli populations

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Keywords: Antimicrobial resistance, Escherichia coli (E. coli), zinc

Background and objectives: As an alternative to antimicrobial growth promotors, zinc oxide is currently used in large amounts in the pig production sector. However, effects of high-zinc diets on bacterial populations are not fully understood yet. Former piglet feeding trails indicated an increase of genetic diversity among intestinal *E. coli*; furthermore the proportion of *E. coli* expressing multi-drug resistant (MDR) phenotypes was enhanced. The aim of this study is to analyse representatively selected *E.coli* from the high-zinc-feeding piglets and the control group, respectively.
Materials and methods: Whole genome sequencing (WGS) data of 179 *E. coli* isolates ("high zinc": n=99; "control" n=80) were screened *in silico* for the occurrence of antimicrobial resistance- and virulence associated genes and genetic determinants using an in-house developed BLAST based screening tool and Chi-square test with an Holm-Bonferroni correction.
Results: First results showed significant differences (p: < 0.01) for *E. coli* subsets representative for both feeding groups with respect to the sulfonamide resistance gene *sul*1 (high zinc: 27.3%; control: 3.8%). Concerning virulence determinants, differences were detected e.g. for the *Salmonella* iron transporter (*sit*ABCD) (high zinc: 29.3%; control: 2.5 %) and the urease gene cluster *ure*ABCDEFG (high zinc: 19.2%; control: 0 %).
Conclusion: First results indicate the influence of high-zinc diets on genetic determinants associated with antibiotic resistance and oxidative stress tolerance in intestinal *E. coli.*

A12

Novel multiresistance integrative and conjugative element ICE*Pmu2* from a German bovine *Pasteurella multocida* isolate

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Keywords: zoonotic pathogen, Pasteurellaceae, multiresistance

Background and objectives: The aim of the study was to identify the antimicrobial resistance genes in a multiresistant *Pasteurella multocida* isolate and to detect their location on mobile genetic elements.
Materials and methods: A single *P. multocida* isolate from 2010 (among 375 collected in 2004-2010) was resistant to tilmicosin with a minimal inhibitory concentration (MIC) of ≥256mg/L. It was also resistant to chloramphenicol streptomycin, spectinomycin and tetracycline. Whole genome sequencing was performed to identify the resistance genes and to analyse their genetic environment.
Results: Whole genome analysis revealed the presence of an integrative and conjugative element (ICE). Similarities were seen in comparison to the first described ICE in *P. multocida* ICEPmu1. One resistance gene region was located in the same position as seen in ICEPmu1. The genes sul2 (sulfonamide resistance), catA3 (chloramphenicol resistance), strA and strB (streptomycin resistance) were located in the same orientation. No macrolide resistance gene was identified in ICEPmu2. In contrast, ICEPmu2 conferred also tetracycline resistance. The gene tet(Y) was for the first time identified in *P. multocida* and located downstream of the resistance gene cluster sul2-catA3-strA-strB.
Conclusion: The identification of an ICE conferring multiresistance in an isolate from Germany is alarming. The novel ICE, designated ICEPmu2, is to the best of our knowledge the first ICE identified in a German isolate.

A13

Used Daily Dose vs. Defined Daily Dose – Advantages and disadvantages of different dosage assumptions for the monitoring of antimicrobial usage in livestock

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Keywords: antibiotic use, monitoring, Defined Daily Dose

Background and objectives:Antimicrobial resistance is a serious threat for public health globally. Tackling this problem requires amongst others valid data, which as a consequence requires harmonized monitoring of antibiotic use and a benchmarking system on farm level.

Up to date there is no harmonized monitoring of antibiotic use and system for assessment of these data Europe-wide, which may facilitate a direct comparison between European countries. Most of the monitoring systems are based on sales data. Therefore, to assess the number of animals treated, overall assumptions about the defined daily doses and also about the weights of the treated animals have to be made.

Materials and methods: The VetCAb-database maintains detailed information about the number of animals treated, the treatment duration, application route and also the indication. So the calculation of the used daily dose for every treatment is possible.

In this evaluation we calculated the treatment frequency for each farm on the basis of UDD (Used Daily Dose) and in contrast the treatment frequency based on the DDD (Defined Daily Dose) published by ESVAC in April 2016 for pigs, cattle and poultry.

Results and conclusion: Results showed that there are strong differences between both outcomes, which may have serious implications for the benchmarking of farms. Furthermore, it reflects that the calculation procedure also has an impact on the comparison between populations which needs further reflection.

A14

Endolysin HY-133 - a putative novel antimicrobial candidate against livestock associated methicillin resistant *Staphylococcus aureus*

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Keywords: LA-MRSA, Phages, Endolysin

Background and objectives: Livestock-associated methicillin-resistant *Staphylococcus aureus* (LA-MRSA) significantly colonize and infect humans. The recombinant endolysin HY-133 is active against *S. aureus,* but its activity against LA-MRSA has not been specifically determined yet. Here, the *in vitro* activity of HY-133 was studied against a large collection of LA-MRSA.
Materials and methods: The activity of HY-133 (Hyglos GmbH, Bernried) was analyzed against 100 LA-MRSA comprising CC398 and other clonal complexes either possessing the methicillin resistance genes *mecA* or *mecC*. *In vitro* activity was analyzed via broth microdilution according to CLSI guidelines. Killing kinetics of HY-133 were analyzed in time-kill-curves using a subset of four LA-MRSA strains.
Results: HY-133 showed similar activities towards *mecA* and *mecC* carrying LA-MRSA. MIC_{50} and MBC_{50} values were at 0.25 µg/mL for both groups, MIC and MBC ranges appeared wider for *mecC* strains (0.12-4 µg/mL vs. 0.06-1 µg/mL). In time-kill-curves, all concentrations of HY-133 led to a rapid decrease in growth reaching bactericidity with a 16-fold MIC. At bactericidal concentrations, regrowth of LA-MRSA was observed after prolonged incubation times.
Conclusion: High activity of HY-133 was shown for a large collection of different LA-MRSA with MIC/MBC values similar to those against other *S. aureus* lineages. Killing kinetics of HY-133 showed a fast mode of action. The regrowth phenomenon warrants further investigation.

A15

Investigation of Methicillin-resistant *Staphylococcus aureus* and Extended-spectrum beta-lactamase-producing Enterobacteriaceae in pigs

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Keywords: antibiotic-resistant bacteria, epidemiology

Background and objectives: Antibiotic resistance is a major public health concern. Extended-spectrum ß-lactamase (ESBL)-producing E. coli strains and livestock-associated *Staphylococcus aureus* (LA-MRSA) are widely distributed among pigs. They seem to play a more important role in conventional farms compared to organic farms.
Materials and methods: A literature search was performed in PubMed and Web of Science. The keywords pig(s), MRSA, ESBL, antibiotic resistance, Germany, organic farming and livestock were used.
Results: Two cross-sectional studies had investigated the occurrence of MRSA and ESBL in livestock in conventional as well as organic farms in Mecklenburg-Western Pomerania. There were MRSA-positive pig farms in all tested districts, the organic farms tested MRSA-negative. Most conventional pig farms and all organic farms tested positive for ESBL. Mostly fattening pigs as well as suckling and weaned piglets were affected by MRSA.
Conclusion: The cohort studies conducted in the past did not monitor more than one fattening period. Therefore, conventional and organic pig farms will be examined for the presence of resistance marker genes in a longitudinal study over a period of twelve months. The burden of ESBL-producing Enterobacteriaceae as well as methicillin-resistant S. aureus in pig farms in Mecklenburg-Western Pomerania will be determined. In conjunction a mobile PCR based approach for quantification of resistance marker genes will be developed. According to the results, a categorization scheme and intervention strategies will be developed.

A16

Comparisons of AmpC-beta-lactamase CMY-2 encoding plasmids from *Escherichia coli* from humans, livestock and food in Germany

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Keywords: antibiotic resistance, AmpC, NGS

Background and objectives: The most frequent plasmidic encoded AmpC enzyme is CMY-2. It is produced by ca. 1% and ca. 30% of the third-generation cephalosporin-resistant *Escherichia coli* isolated from humans and poultry, respectively. In this study we are comparing plasmids from human, meat products and livestock animals from Germany, by whole-genome sequencing.

Materials and Methods: Genomic DNA of 168 CMY-2 positive *E. coli* from different sources (humans n=51, healthy broilers n=51, chicken meat n=56, turkey meat n=7, diseased pigs/chickens n=5,) was extracted and sequenced using the Illumina® MiSeq platform. Phylogenetic markers, such as plasmid multi-locus sequence type and plasmid replicon types were identified, as well as mobile genetic elements and whole plasmid comparisons were conducted.

Results: Plasmids carrying bla_{CMY-2} were identified in 145 of the 168 sequenced isolates; in the rest of the isolates bla_{CMY-2} was integrated in the chromosome. Plasmids of the replicon type IncI1 (n=64) and IncK (n=76) were the most prevalent ones and among these, identical plasmid sequences (nucleotide identity 96-99%) present in all habitats, were observed. Further four IncA/C plasmids were identified.

Conclusions: The results showed highly related plasmids from different reservoirs. This indicates a possible plasmid-mediated spread and zoonotic potential of *bla*$_{CMY-2}$ carrying plasmids across the *E. coli* host populations.

A17

Induction of resistance to glyphosate, a common herbicide, in *E. coli in vitro*

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Keywords: glyphosate, resistance induction, Enterobacteriaceae

Background and objectives: Residues of glyphosate, the most used herbicide in the world, are commonly found in the environment and food supply chain. Recently, its effects on microorganisms and antibiotic resistance have been recognised, raising concerns about the effects of glyphosate in animal feed on microbiome. The objective of this study was to investigate the ability of glyphosate to induce resistance *in vitro* in *Escherichia coli* isolated from farm animals.
Materials and methods: We used two strains of *E. coli* (with and without ESBL resistance markers). After initial determination of the minimal inhibitory concentration (MIC), we passaged them daily in gradually increasing concentrations of glyphosate alone and as a part of formulation (Roundup LB plus). To assess the stability of resistance, we determined the final MIC after the stability passage (in the absence of glyphosate).
Results: Resistance induction response for Roundup was similar for ESBL and non-ESBL *E. coli* strains, with early extinctions of bacterial populations at 2x MIC. The ESBL strain was also unable to grow at concentrations >MIC, whereas the non-ESBL strain readily adapted to growth at 2-4x MIC of glyphosate.
Conclusion: Our results demonstrate that there are differences between glyphosate alone and as a part of herbicide formulation and individual bacterial strains in the ability to induce resistance. Overall, although it is not easy to induce resistance to glyphosate, it is nonetheless possible.

A18

A survey on the competencies and learning outcome of undergraduate veterinary students in three German faculties attending an elective course on prudent antibiotic use

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Keywords: antibiotic stewardship, resistance, E-Learning

Background and objectives: Antibiotic resistance is considered as an increasingly serious threat to global public health. Therefore, we developed “VetMAB” as a certified interactive E-Learning-tool for veterinarians and veterinary students focusing on responsible handling and prudent use of antibiotics in livestock.
Materials and methods: During April and July 2017, VetMAB is held as a compulsory elective course in veterinary faculties at three German universities in Munich, Gießen and Berlin. Enrolled 4th-year-students are obliged to attend two appointments at the beginning and end of the course and, in the meantime, work self-dependent through two E-Learning modules (a basic and an animal specific module about bovine respiratory infections). Moreover, the students are requested to anonymously fill in a questionnaire capturing the students´ *status quo* about prudent use of antibiotics at the beginning and the end of the course. Statistical analysis will enable to evaluate the learning process during the course and draw comparisons between undergraduate students of three veterinary faculties in Germany.
Results and conclusion: In total, 92 4th-year-students enrolled in the course, 24 in Munich, 40 in Gießen, and 28 in Berlin. Preliminary analysis suggests a good basic knowledge about antibiotic use and resistance.

A19

Molecular characterisation of MRSA CC3286 from primates

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Keywords: spa typing, macrorestriction analysis, DNA microarray

Background and objectives: A novel MRSA ST3268-SCC*mec*V clone was detected in primates shipped to the Washington National Primate Research Center in 2015. The aim of this study was to characterize further isolates of the same clonal complex obtained in the primate center.
Materials and methods: In total, nine MRSA CC3296 SCC*mec*V from *Macaca* (*M.*) *mulatta*, *M. fascicularis* and *M. nemestrina* were selected for characterization by DNA microarray, macrorestriction analysis, antimicrobial susceptibility testing, *spa* and SCC*mec* typing.
Results: Four isolates from *M. mulatta* were *spa* type t15469, and five isolates with *spa* type t13638 were from two *M. fascicularis* and three *M. nemestrina*. Seven isolates had indistinguishable PFGE profiles. Two *spa* type t13638 isolates had related PFGE patterns. All isolates were resistant to tetracycline [*tet*(K)], fluoroquinolones, penicillin [*blaZ*, *blaI*, *blaR*] and oxacillin [*mecA*]. The five *spa* type t13638 isolates were resistant to kanamycin and resistant/intermediate to gentamicin [*aacA-aphD*]. The *qacC* gene, encoding quaternary ammonium resistance, was found in three t13638 isolates, resulting in a reduced susceptibility to benzalkonium chloride. All isolates were positive for capsule type 5 alleles, the accessory gene regulator *argVI*, the *hlgA* locus, different enterotoxin genes and the SCC*mec* associated genes *ugpQ* and *ccrC* .

Conclusion: MRSA CC3268 isolates could be subdivided into two *spa* types that were associated with different host species.

A20

Eat organic? Antimicrobial resistance in *E. coli* from conventional and organic dairy cows and broilers

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Keywords: microarray antimicrobial resistance, organic, animal production

Background and objectives: Antimicrobial resistance (AMR) in bacteria from food producing animals contributes to the burden of AMR in humans although the extent of the contribution cannot be quantified easily. Public opinion believes that organic farming may limit AMR in food producing animals based on the restrictions imposed on antimicrobial use in these farming systems. It was the objective of this study to analyse whether this public believe proofs robust against scientific challenge.
Materials and methods: Samples of bulk tank milk and of broiler flocks were collected in the framework of the national zoonosis monitoring. *E. coli* were isolated from the samples by the regional laboratories and submitted to the National Reference Laboratory for Antimicrobial Resistance (NRL-AR). Isolates were tested for their susceptibility to 14 antimicrobials according to the methods described in Commission Implementing Decision 2013/652/EU.
Results: AMR was significantly higher in *E. coli* from broilers than from dairy cows and within the respective animal species in *E. coli* from conventional compared to organic farms. The highest proportion of fully susceptible isolates was found in bulk tank milk from organic dairy herds, followed by BTM from conventional cows, boot swab samples from organic and from conventional broilers.
Conclusion: Sometimes, public believe agrees with scientific results. Further research needs to determine the factors that contribute to the difference.

A21

Identification of mecB-encoded methicillin resistance in *Macrococcus caseolyticus* from food samples

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Keywords: Macrococcus caseolyticus, mecB, food samples

Background and objectives: Macrococci have been considered as colonizers of animal skin or recovered from food. Homologs of the mecA/mecC genes coding for methicillin resistance in *Staphylococcus aureus* (MRSA) have been described for *Macrococcus caseolyticus*, designated mecB and mecD. This study was aimed to identify macrococci from food products and to determine their susceptibilities to betalactams.
Materials and methods: Food products of Thai and Chinese origin purchased in Canada were screened for macrococci. Strains were identified using MALDI-TOF mass spectrometry (MS) and 16S rRNA gene sequencing. Disk diffusion antimicrobial susceptibility testing was performed according to CLSI guidelines. Betalactam-resistant strains were tested for the presence of the mecB gene by PCR and its genetic location was determined.
Results: Overall, ten macrococci were recovered from food samples including clams (n=2), frog legs (n=7) and a soft shell turtle (n=1). MALDI-TOF MS and sequencing confirmed the species affiliation *M. caseolyticus*. Overall, 9/10 isolates were phenotypically resistant against cefoxitin and penicillin. All cefoxitin-resistant isolates carried the mecB gene, which was always plasmid-encoded.
Conclusion: This study revealed that mecB-encoded plasmid-borne methicillin-resistant *M. caseolyticus* isolates may occur in high prevalence in specific food samples. This is alarming since they might represent a reservoir for inter-species dissemination of resistance genes.

A22

Monitoring antibiotic usage in animals: Do existing systems provide comparable data?

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Keywords: antimicrobial usage, monitoring systems, variables

Background and objectives: Monitoring of antimicrobial use (AMU) in animals is important to understand the development of antimicrobial resistance (AMR). Monitoring systems exist in many countries, but there is no global consensus on the documentation of AMU, thus hampering comparability of data. The main objective of this investigation is to analyse different AMU monitoring systems, the variables used and whether they facilitate integration with AMR data.
Materials and methods: Selected AMU monitoring systems are analysed and compared. Differences in data collection, analysis and documentation are described.
Results: Most countries collect overall sales data on antibiotics. Some do also provide usage data on the level of farms and animal species, using different variables. AMU data should be related to the respective animal population, but the choice of the denominator also differs. Additionally, some systems aim at benchmarking farmers or veterinarians.
Conclusion: It is not recommended to use sales data to evaluate AMU if the purpose is to analyse the impact on AMR. In this regard, most existing AMU monitoring systems are not sufficient yet. Moreover, the differences in systems collecting more detailed information do not allow for direct comparison. An integrated One Health approach to monitor AMU is needed. Despite initiatives of several international organisations to support the development of AMU data collection, existing AMU monitoring systems in animals lack harmonisation.

Poster Session Novel methods, diagnostics and NGS

D01

Virulence profiling of animal pathogenic *Escherichia coli* by multiplex real-time PCR

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Keywords: E. coli, animals, real-time PCR

Background and objectives: Enteropathogenic *E. coli* are the main cause of diarrhoea (ETEC, EPEC, STEC) and edema disease (EDEC) in piglets and calves. Sole serotype identification does not allow a reliable conclusion on pathogenicity. Thus, determination of virulence factors is required. In this study a multiplex real-time PCR was developed for virulence marker detection and consequently for the establishment of virulence profiles.
Materials and methods: In a first step, oligonucleotides were designed for single identification of the following virulence factors: K88, K99, 987p, F41, F107, LT/ST, Stx1/2 and intimin. Interaction and specificity analyses were performed. In a second step, two 4plex PCR systems were created by using specific reporter dyes (FAM, VIC, ROX, Cy5).
Results: The 4plex I system covers K88, 987p, F41, and K99, while 4plex II detects F107, Stx1/2, intimin, and LT/ST. Both were subjected to inhouse validation checking specificity, ruggedness, accuracy, LOD, and stability. Technical validation was conducted on various devices: Agilent Mx3005P, Agilent AriaDx, Roche LightCycler® 480 II, Roche LightCycler® 480 Cobas, Applied Biosystems 7500, BioRad CFX 96. All virulence factors could be successfully detected and differentiated by both systems in their respective channels.
Conclusion: This diagnostic test allows the rapid and sensitive detection of the most important virulence factors of animal pathogenic *E. coli* and might therefore contribute to reducing the risk of spreading and improving the recovery chances of infected animals.
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D02

Clinical comparison, standardization and optimization of Zika virus molecular detection

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Keywords: Arbovirus, diagnostic

Background and objectives: To examine the diagnostic performance of real-time reverse transcription-polymerase chain reaction (RT-PCR) assays for Zika virus detection.
Methods: We compared seven published real-time RT–PCR assays and two new assays that we have developed. To determine the analytical sensitivity of each assay, we constructed a synthetic universal control RNA (uncRNA) containing all of the assays' target regions on one RNA strand and spiked human blood or urine with known quantities of two Zika virus strains. Viral loads in 33 samples from Zika virus-infected patients were determined.
Results: Oligonucleotides of the published real-time RT–PCR assays, showed up to 10 potential mismatches, compared with 0 to 4 mismatches for the new assays. The 95% lower detection limit of the seven most sensitive assays ranged from 2.1-12.1 uncRNA copies/reaction. The mean viral loads in samples from Zika virus-infected patients were 5×104 RNA copies/mL of blood and 2×104 RNA copies/mL of urine.
Conclusion: We provide reagents and updated protocols for Zika virus detection suitable for the current outbreak strains. Some published assays might be unsuitable for Zika virus detection, due to the limited sensitivity and potential incompatibility with some strains. Viral concentrations in the clinical samples were close to the technical

detection limit, suggesting that the use of insensitive assays will cause false-negative results.

D03

Outbreak of canine brucellosis in a Brazilian kennel

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Keywords: Brucella canis, dogs, zoonoses

Background and objectives: Canine brucellosis may cause reproductive failure in dogs leading to economic losses in commercial kennels. *Brucella canis* is a zoonotic pathogen that can be transmitted to humans by close contact to the domestic animals. We will describe an outbreak of canine brucellosis in a Brazilian kennel using classical microbiological methods and whole genome sequencing.
Materials and methods: 24 Gram-negative, coccoid rod-shaped bacteria were isolated from 17 dogs of a kennel in São Paulo, sampled in 2014. Classical microbiological tests (CO2 requirement, H2S production, urea hydrolysis, agglutination with monospecific sera (anti-A, anti-M, and anti-R), dye sensitivity (basic fuchsin and thionine), and phage lysis (F1, F25, Tb, BK2, Iz, Wb, R/C)) were conducted to identify and sub-differentiate the bacteria. Mass spectra were acquired with a Microflex LT (Bruker Daltonik). Whole genome sequencing was carried out using MiSeq (Illumina).
Results: MALDI-TOF MS analysis assigned all isolates to the genus *Brucella*. The phenotypic data clearly identified *Brucella canis* with only slight differences in phage typing profiles. The genomic data proved a single outbreak strain exhibiting only four single-nucleotide polymorphisms (SNPs) in three strains.
Conclusion: An outbreak of canine brucellosis is defined by homologous *Brucella canis* strains isolated from individual dogs of the same kennel. Clonal brucellae can be identified using next generation sequencing.

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D04

Development and Validation of Real-Time RT-PCR based Yellow Fever surveillance in the Americas

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Keywords: Yellow fever virus, PCR, diagnostic

Background and objectives: The yellow fever virus (YFV) is one of the most important human arboviral pathogens. In 2016, Brazil has reported the beginning of the country's largest sylvatic YFV outbreak in decades. Large-scale vaccination campaigns reaching 19 million Brazilians have been launched in response to this outbreak. Since rare vaccine adverse events (VAE) following YFV vaccination have been reported from several countries including Brazil, surveillance of YFV VAE is required to monitor the vaccine's safety. Discrimination between YFV vaccine strains and wild-type (wt) strains is essential in regions of endemic transmission. We developed two new real-time RT-PCR assays for this purpose.

Materials and methods: The assays target genomic regions that are highly diverse between YFV vaccine strains and wt strains from the Americas. The assays were examined regarding their sensitivity, specificity and clinical performance.

Results: The limits of detection (95% CI) were below ten copies per reaction. YFV vaccine and American wt strain were specifically detected and discriminated in clinical matrixes spiked with viral particles and in clinical specimens. A panel of 41 arboviruses was tested negative.

Conclusion: The new assays enable YFV detection with diagnostic sensitivity. The feature to specifically discriminate between vaccine and American wt strains is highly beneficial for the surveillance of YFV VAE in the Americas.

D05

Comparison of canine distemper virus strains infecting aquatic and terrestrial mammals

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Keywords: morbillivirus, seals, reverse genetics

Background and objectives: Canine distemper virus (CDV) is an animal morbillivirus with a worldwide geographical distribution which causes epizootic disease outbreaks in multiple wildlife species. Comparatively few whole genome sequences have been reported for unpassaged CDV-infected wildlife strains. In this project, we have sourced a large collection of CDV-infected tissues representing both aquatic and terrestrial mammals. This includes tissues from European domestic and wildlife species and CDV-infected Phoca species including original samples from epidemics in Lake Baikal seals in 1988 and in Caspian Sea seals in 2000. The primary aim of this project is to obtain a better mechanistic understanding of the molecular determinants underlying CDV cross-species infections.
Materials and methods: CDV-positive frozen tissue samples were processed and prepared for deep sequencing and analysis of viral genomes.
Results: Phylogenetic analysis showed that a CDV strain derived from a novel lineage most closely related to American I, was responsible for the large outbreak in the Caspian Sea. Additional genetic variation was identified in European wildlife and Lake Baikal CDV strains. Based on the resulting consensus CDV sequences, new reverse genetics systems have been generated for dog, raccoon and Caspian Sea wild-type CDV strains.
Conclusion: New reverse genetics systems for wild-type CDV strains will facilitate analysis of unique mutations identified in infected wildlife species.

Poster Session New and re-emerging zoonoses

N01

Application of a luciferase immunoprecipitation system (LIPS) to investigate the host spectrum of bovine hepaciviruses

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Keywords: BovHepV, serology, host tropism

Background and objectives: Hepaciviruses have been detected in numerous animal species including horses, where infections with so-called “non-primate hepaciviruses (NPHV)” are described. Recently, we identified a novel HCV-like virus in bovine serum samples (bovine hepacivirus, BovHepV). It was shown, that the virus is able to establish persistent infection in cattle. However, further studies investigating the host tropism and a zoonotic potential of bovine hepaciviruses are still pending.
Materials and methods: To address this issue, we developed serological tests and broadly reactive RT-PCR assays to examine serum samples of different animal species and humans.
Results: We found that about 30% of investigated bovine serum samples reacted serologically positive in a LIPS assay based on BovHepV NS3 helicase domain. In addition, sporadic antibody positive reactions were observed by analyzing pig and horse sera. However, 200 serum samples from human blood donors did not elicit any serological reactivity and no BovHepV RNA was detected in sera from patients with liver disease of unknown etiology.
Conclusion: These results do not imply zoonotic transmission of BovHepV. If positive LIPS results in animal species other than cattle are attributable to cross-reactive antibodies induced by infections with NPHV or additional hepaciviruses remains to be elucidated.

N02

Development of serological screening assays for the detection of antibodies against the most relevant zoonotic viruses in *Eidolon helvum* and *Rousettus aegyptiacus* fruit bats

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Keywords: fruit bats, zoonotic disease, screening assay

Background and objectives: Fruit bats have recently attracted increased interest, as they may act as reservoir species for a variety of highly pathogenic viruses. However, the analysis of their role in certain disease outbreaks is often hampered by the lack of specific diagnostic tools. Therefore, we have developed ELISA assays for the specific detection of IgM and IgG antibodies against the most relevant zoonotic viruses in *Eidolon helvum* and *Rousettus aegyptiacus* fruit bats.
Materials and methods: Bats were immunized with recombinant viral proteins (glycoproteins of Hendra virus, Nipah virus and Lagos Bat Lyssavirus, nucleoproteins of Ebola virus and SARS-Coronavirus) to generate positive control sera for serological assays. Besides relevant negative controls, these antigens are used as a basis for ELISA assays. As a conjugate, we used our recently developed monoclonal antibodies specifically raised against IgM or IgG antibodies from *Eidolon helvum* or *Rousettus aegyptiacus*.
Results: Using these newly developed sera and specific conjugates, we are now able to reliably detect past infections with these zoonotic viruses in free ranging bats of *Eidolon helvum* or *Rousettus aegyptiacus* bats.
Conclusion: These newly generated tools will considerably improve the diagnostic situation for fruit bat samples by enabling the determination of a humoral immune response in animals that may have been in contact with infectious agents.

N03

Development and evaluation of an in vitro skin infection model for the zoophilic dermatophyte *Trichophyton benhamiae*

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Keywords: infection model, emerging dermatophyte, Trichophyton benhamiae

Background and objectives: Pets represent a major source of infection for humans with zoophilic dermatophytes such as *Trichophyton benhamiae*. The resulting dermatophytoses are often associated with highly inflammatory skin reactions requiring long lasting treatment.
Materials and methods: Initially, optimal skin explant culture conditions for infection studies were determined. Skin morphology was monitored histologically (HE, Ki 67) during the course of culture (14d). For the *in vitro* skin infection with *T. benhamiae*, a new fungal medium was developed.
Results: Histological evaluation confirmed maintained integrity of skin explants. The new fungal medium allowed for the induction and isolation of arthro- and chlamydospores as a basis for *in vitro* infection. Initial infection experiments showed skin invasion by dermatophyte hyphae.
Conclusion: The model is suitable to study the dermatophyte skin infection process and enables investigations on the underlying pathomechanisms.

N04

Novel marsupial hepatitis A virus corroborates frequent host shifts during hepatovirus evolution

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Keywords: Hepatitis A virus, Marsupialia, evolution

Background and objectives: The *Picornaviridae* genus *Hepatovirus*, represented by Hepatitis A virus (HAV) was thought to be restricted to primate hosts. Recent studies showed that small mammals harbor diverse hepatoviruses. To investigate hepatovirus evolution, we analyzed marsupials, which represent one of the evolutionary oldest mammalian lineage.
Materials and methods: We collected 118 wild marsupials (75 liver and 58 blood samples) representing 5 species in northeastern Brazil. Samples were screened for hepatovirus infection using taxon-specific PCR and competitive ELISA.
Results: One liver sample contained hepatovirus nucleic acid (1.3%). Phylogenetic analysis based on full viral genome placed this virus in basal relationship to a rodent hepatovirus recently found in Mexico. This lineage was closely related to human HAV, and thus not compatible with ancient co-speciation of hepatoviruses and their hosts. Overall seroprevalence was 20.7%, indicating frequent exposure of marsupials to hepatoviruses.
Conclusion: Our data corroborate frequent host shifts during hepatovirus evolution. Since marsupials are commonly consumed as bushmeat in Latin America, the zoonotic potential of the identified HAV-related virus may merit further investigation. However, the genetic relation of the marsupial and human hepatoviruses may imply limited potential of a new zoonotic introduction into humans due to HAV-induced herd immunity. Since *Marsupialia* only occur in the Americas and Australia, the question if related viruses exist in other marsupial species requires further invesitgation.

N05

Neotropical Bats that Co-habit with Humans Function as Dead-End Hosts for Dengue Virus

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Keywords: Dengue, bats, virus reservoirs

Background and objective: Several studies have shown Dengue nucleic acids and/or antibodies present in Neotropical bats, suggesting that they may be susceptible to DENV infection. Here we aim to elucidate the role of house-roosting bats in the DENV transmission cycle.
Materials and methods: Bats were sampled in households located in high and low dengue incidence regions during rainy and dry seasons in Costa Rica. We captured 318 bats from 12 different species in 29 households.
Results: Necropsies were performed and histopathology studies from all organs showed no significant findings of infection. Sera were analysed by PRNT90 for a seroprevalence of 21.2% and by RT-PCR for 8.8% positive bats. From positive bats, some intestine samples were DENV RNA+ for the same dengue serotype detected in blood. Virus isolation from positive samples was unsuccessful. Viral load analyses in sera by qRT-PCR showed low virus concentrations. Mosquitoes were collected using EVS-CO2 traps and analysed for DENV and feeding preferences. Only 3 mosquitoes were found DENV+ and none was positive for bat cytochrome b. Our results suggest an accidental presence of DENV in bats probably caused from oral ingestion of infected mosquitoes. Phylogenetic analyses suggest also a spillover event from humans to bats.

Conclusion: We conclude that bats in these urban environments do not sustain DENV amplification, they do not have a role as reservoirs, but function as epidemiological dead end hosts for this virus.

N06

Hantavirus screening of insectivores from Germany

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Keywords: Hantavirus, Shrew, RT-PCR, Seewis virus

Background and objectives: Recently many novel hantaviruses have been discovered in shrews, moles and bats. Shrew-borne Seewis virus (SWSV) and Asikkala virus (ASIV) were detected in several European countries. This pilot study was focusing on the molecular detection of insectivore-associated hantaviruses in Germany.
Materials and methods: A total of 700 shrews of three species were collected during a monitoring study at four regions in Germany. Additional 213 shrews of three species were collected at further sites by collaborators. Lung tissue RNA was used for hantavirus RT-PCR analysis targeting the S, M and L segments.
Results: SWSV RNA was detected in 42 of 700 (6.0 %) shrews from monitoring areas and in 5 of 213 shrews (2.3 %) from non-monitoring sites. The majority of affected shrews were *Sorex araneus*, but also a few *S. minutus*, and *S. coronatus* were found to be SWSV RNA-positive. Phylogenetic analysis indicated a clustering of the novel sequences with the previously published SWSV sequences. ASIV RNA was detected in 2 out of 54 (3.7%) pygmy shrews. No RNA was detected in any of the 27 greater white-toothed shrews.
Conclusion: This study confirms the continuing abundance of SWSV in common shrews and rare spillover infections to other shrew species. Future studies will be dedicated to understand the potential influence of changes in shrew populations on the prevalence and molecular evolution of insectivore-associated hantaviruses and to evaluate their zoonotic potential.

N07

Tick-borne encephalitis – an emerging viral infection in Germany

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Keywords: Tick-borne encephalitis, epidemiology, Germany

Background and objectives: Tick-borne encephalitis (TBE) is the most important tick-borne viral disease of humans in Europe and Asia. In Germany between 200 and 450 human cases are registered every year. Human cases mainly occur in Southern Germany, but sporadically also in other areas in Germany and are also imported from other endemic areas of Germany and Europe.
Materials and methods: Human TBE cases, reported in 2016 and 2017 were followed and ticks were collected in Lingen/Ems, Cuxhaven, Battaune, Schleching, Schwandorf, Traunstein or Tübingen. Ticks were identified to species level, stage and sex and following homogenization and nucleic acid extraction tested for TBE virus by a real-time RT-PCR (Schwaiger & Casinotti). For phylogenetic analysis the E genes of the virus isolates were sequenced and analyzed.
Results: Several thousand ticks were collected and tested. In Battaune, Saxony and Schwandorf, Schleching, and Traunstein (all Federal State of Bavaria) new virus strains were detected. The phylogenetic analysis provide data that TBE virus is spreading over long distances and short distances by different mechanisms.
Conclusions: TBE is emerging in new areas and also in new locations in known endemic areas in Southern Germany. There is a clear tendency of emergence and northward and westward spreading of human TBE cases in Germany. Genetic analyses of the causing TBE virus strains will help to uncover the mechanisms of spread of TBE viruses through Germany and Europe.

N08

Almaty region as a melting pot for the new tick-borne Rickettsiae species in Kazakhstan

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Keywords: Rickettsiae, Kazakhstan, tick

Background and objectives: About 65 years ago, tick-borne rickettsioses (TBR) were described in Kazakhstan. Since 1995 an increase of cases diagnosed by complement fixation test has been observed, but data on *Rickettsia* (R.) species circulating in Kazakhstan are sparse. We investigated the prevalence and *Rickettsia* species circulating in ticks in (i) a region with no known human rickettsioses (Almaty) and (ii) a region with the highest rickettsioses incidence (Kyzlyorda) in Kazakhstan.
Materials and methods: 2341 ticks were collected by flagging, sorted in pools, homogenized and DNA was extracted. After screening with real-time PCR (*gltA),* positive samples were further investigated by multi-locus sequence typing (MLST) targeting seven fragments. After sequencing BLAST comparisons and phylogenetic trees were done.
Results: Three *Dermacentor* species and one *Ixodes*, *Hyalomma*, *Haemaphysalis* and *Rhipicephalus* species were identified. The *Rickettsia* minimum infection rate (MIR) was high in Kyzylorda (-88%) and Almaty (-20%). Five Rickettsiae were identified: in *Dermacentor* species *R. raoultii* and for the first time *R. slovaca* in Kazakhstan. Further 2 new *Candidatus R. yenbekshikazakhensis* and *Candidatus R. tekelensis* and one new genotype *R. talgarensis* were found.

Conclusion: Two of the found *Rickettsia* are known to be pathogenic (*R. raoultii, R. slovaca).* The data indicate that *Rickettsia* in ticks and TBR should be further investigated in Kazakhstan to estimate the clinical impact.

N09

The epidemiological influence of tick-borne encephalitis in the southern part of Kazakhstan

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Keywords: Kazakhstan, Tick-borne encephalitis virus, Ixodes persulcatus

Background and objectives: One of the most severe arboviral infections in Kazakhstan is tick-borne encephalitis (TBE). TBE is a serious health problem in Kazakhstan with up to 50 cases per year. Therefore, we examined the prevalence of antibodies against TBE virus (TBEV) in humans suffering on fever of unknown origin (FUO) and the presence of TBEV in ticks from six districts in Almaty (AO) and Kyzylorda oblast (KO).
Materials and methods: In the six districts of AO and KO 2341 ticks were collected. Ticks were sorted in pools and homogenized. Extracted RNA was screened for the presence of TBEV. Products of a conventional E-Gen RT-PCR were sequenced and a phylogenetic analysis was carried out. Paired sera (day 0 and 10-14 after hospitalization) from 795 patients with FUO from 13 hospitals in AO and KO were investigated for the presence of IgG/IgM against TBEV.
Results: The Minimum Infection Rate (MIR) of TBEV in three districts of AO are between 1.1% and 4.4% whereas in the districts of KO none of the tested pools were TBEV positive. Sequencing results show that the TBEV belongs to the Siberian subtype. TBEV IgG antibodies were detected in 23 (2.9%) out of 795 sera samples.
Conclusion: In this study, we present new data on the circulating TBEV subtype in ticks in AO. For the first time, a serological study was carried out to rule out the role of TBEV as a cause of FUO in Kazakhstan. Our results will help to improve TBEV surveillance and prevention of this infection in Kazakhstan.

N10

Isolation and characterization of a novel paramyxovirus related to both murine and human respiroviruses 1

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Keywords: novel respirovirus, zoonotic potential

Background: Emerging infectious diseases pose threats to human and animal health, and novel infectious agents therefore need particular attention concerning their unknown zoonotic potential. Here, we describe a novel paramyxovirus that is phylogenetically related to both human respirovirus 1 (former human parainfluenza virus 1) and its murine counterpart, murine respirovirus (former Sendai virus).
Materials and methods: The novel virus was isolated on porcine thyroid cells from sample materials of a grizzled giant squirrel from Sri Lanka, which died during quarantine in a German zoo. Electron microscopy was used for further analysis, and the whole-genome sequence of the isolate could be obtained by next-generation sequencing (Illumina MiSeq) and de novo assembly.
Results: Pathologic examination of the animal revealed an acute hemorrhagic-necrotizing pneumonia with high-grade diffuse alveolar damage and a middle-grade lymphoplasmacellular dominated enteritis. Electron microscopy demonstrated paramyxovirus-like particles. The newly discovered virus isolate shows an overall sequence identity of 71 % with known murine respiroviruses, and 68 % with human respiroviruses 1. Phylogenetic analysis suggests a novel branch between murine and human respiroviruses.
Conclusion: As the virus has similarity to both murine and human respiroviruses, a zoonotic potential cannot be excluded and has to be further evaluated. A screening for the prevalence in squirrels is currently initiated.

N11

Diversity of viruses in bats from the Daintree Rainforest

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Keywords: Virome, Rainforest, bats

Background and objectives: Despite all differing possibilities of virus emergence, sixty per cent of emerging viruses have a zoonotic origin, thus highlighting spill-over events from animals to humans as a major threat to public health. Consequently, it has become evident that surveillance of viruses prevalent in wildlife is of particular importance. Hereby it is important to focus on apparently healthy animals in their natural habitat to maximize discovery potential and understand environmental disease factors, biology and ecology that are further influencing the risk of virus spreading and shedding.
Materials and methods: In this collaborative pilot study the Tropical North Queensland (TNQ) rainforest has been chosen because it is the oldest rainforest in the world and comprises a unique diversity of bats. We investigate how the biodiversity of bats, the second largest order of mammals, influences the evolution of highly pathogenic viruses and vice-versa. To assess the viral diversity we are using metagenomic sequencing, serology and virus isolation.
Results: Metagenomic analysis of oral swabs and urine samples yielded more than 60 novel virus strains from different families (e.g. Paramyxovirus, Adenovirus, Flavivirus, Poxvirus, Retrovirus).
Conclusion: Study layout and sampling methods provide the expected results. This study can be seen as a starting point to further investigate biodiversity and virus-host coevolution in these remote habitats.

N12

High variability and distinct geographic clustering of Hantavirus sequences from Banana pipistrelle bats (*Neoromicia nanus*) in Côte d'Ivoire

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Keywords: Hantavirus, Neoromicia nanus, Côte d'Ivoire

Background and objectives: Hantaviruses are emerging zoonotic viruses usually carried by rodents showing host-specific association. Recently they were also found in insectivores and bats. In Africa, all bat hantaviruses detected up to now were only found in single animals. Here we conducted a screening of bats from Côte d´Ivoire (CI) to invesitgate the ecology and evolution of indigenous hantaviruses in chiropteran hosts.

Methods: 397 bats of 23 species were captured in 2013-2016 in 15 villages across CI to collect tissue and blood samples. Lung samples were screened for hantavirus RNA using PCR. In addition, we established an in-solution capture assay enabling targeted next generation sequencing. To examine the genetic relationship of the virus strains found here, phylogenetic analysis was performed.

Results: 39/167 insectivorous bats (*Neoromicia nanus*) tested PCR-positive for hantaviruses in Taï region (Western-CI). Using an in-solution capture approach, we were able to obtain large fragments of the L-segment. Phylogenetic analysis showed that L-segment sequences were related to bat-borne Mouyassué virus (73-96% nucleotide identity) and small-scale geographic clustering was observed.

Conclusion: Hantavirus sequences have been detected in several individuals of local pipistrelles which confirms *N. nanus* as a natural host. The sequence differences indicate that at least one of the newly found strains may form a distinct species within the genus *Orthohantavirus*.

N13

The European polecat, *Mustela putorius*, as a host for ticks but not for tick-borne pathogens?

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Keywords: hexagonus, polecat, tick-borne pathogens

Background and objectives: The European polecat occurs almost in all Europe. Even though it is widespread species, there is not much data about its role in the circulation of tick-borne pathogens (TBP). The aim of this study was to investigate whether polecats pose an epizootiological and epidemiological threat in Germany.
Materials and methods: In total spleen samples from 118 polecats collected in different regions in Germany were tested by real time and conventional PCR methods to detect such pathogens as *Anaplasma phagocytophilum*, *Babesia* spp., *Bartonella* spp., *Candidatus* Neoehrlichia mikurensis and *Hepatozoon* spp. Additionally, randomly selected engorged *Ixodes hexagonus* ticks (100 females and 100 nymphs) collected from polecats were tested for: *A. phagocytophilum*, *Babesia* spp., *Bartonella* spp., *Borrelia* spp., *C*. Neoehrlichia mikurensis and *Rickettsia* spp.
Results: The polecats were infected only with two pathogens, *A. phagocytophilum* (n = 5; 4.3%) and *C*. Neoehrlichia mikurensis (n = 1; 0.9%). And there was one case of co-infection. *Ixodes hexagonus* ticks were positive only for one pathogen – *Bartonella* spp. with MIR (minimum infection rate) 0.5%.
Conclusion: The European polecats are not main reservoirs and do not play an important role in spreading tick-borne pathogens. They maintain the circulation of *I. hexagonus* ticks which, from the medical point of view, are not significant vectors of TBD.

N14

Encephalitis of unknown origin in horses from Brazil

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Keywords: S. aureus, sodium hypochlorite

Background and objectives: Tissue-based diagnostic investigation of equine encephalitis from Brazil.
Materials and methods: Paraffin-embedded CNS from horses with neurological signs were investigated with IHC for rabies virus, Borna disease virus, EHV-1, *Listeria* sp., *Toxoplasma gondii*, *Neospora* sp., *Sarcocystis neurona*, double-stranded RNA, and for astroglial (GFAP) and microglial activation (Iba1). RNA isolation was modified to minimize cross-linking, and nucleic acid quality was verified with equid-GAPDH PCRs. Pan-PCRs were carried out for viruses and coccidia.
Results: Out of 35 CNS samples, 5 displayed non-inflammatory lesions, while 27 presented lymphocytic and 3 granulomatous infiltration. This was associated with microglial activation (mild in 20/35, moderate in 9/35, and severe in 5/35 horses), and astroglial activation (mild in 9/35 brains, moderate in 22/35 and severe in 4/35). A GAPDH 64bp or 170bp product was amplified in 26 or 23 samples, respectively. No specific agent was determined with pan-PCRs lengthening ≥ 200bp. Rabies virus antigen and RNA were demonstrated in 1/35 and S. neurona schizonts in 4/35 brains. Arbovirus RNA (flavivirus, alphavirus) were detected in 2/35 samples, and EHV-1 DNA in 3/35 brains.
Conclusion: The inflammatory lesions and glial reaction pattern point to an infectious etiology. For FFPE material, special conditions were employed which allowed the first identification of flaviviruses in a

horse from Brazil. This study raises awareness to new emergent and re-emergent diseases in Brazil.

N15

Low Zika virus (ZIKV) prevalence and no interference of pregnancy and malaria on the performance of a ZIKV ELISA in Nigeria, 2016

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Keywords: virus, Pregnancy, Nigeria

Background and objectives: Zika virus (ZIKV) was first isolated in Africa in 1948 but since then human infections have been detected only sporadically. In 2015, the first introduction of ZIKV into South America caused an explosive outbreak throughout the continent. Although usually asymptomatic or a benign disease there are now several lines of evidence supporting a causal relationship between ZIKV infection during pregnancy and congenital defects and malformations including microcephaly. Nigeria has a high birth rate but little is known about ZIKV prevalence especially among pregnant women.

Materials and methods: In a prospective study, we included 405 serum samples of individuals presenting at Jos University Teaching Hospital in 2016. All of them were female and 74% of them were pregnant. All samples were analysed for ZIKV antibodies using the Euroimmun ZIKV Anti-NS1-IgM/IgG-ELISA and microscopy for *Plasmodium* species was done for 274 patients.

Results: Overall we found a ZIKV IgM seroprevalence of 5% and a ZIKV IgG seroprevalence of 3%. In pregnant women, 4% were positive for ZIKV IgM and 3% for ZIKV IgG, whereas in non-pregnant women 9% and 4% were positive for IgM and IgG respectively. Rates did not differ among pregnant women with a positive

Plasmodium microscopy result compared to those with a negative result.
Conclusion: Our data indicate a low ZIKV seroprevalence in Nigeria and no potential interference with pregnancy or malaria with the Euroimmun ZIKV ELISA.

N16

High seroprevalence against Zika virus in Salvador, north-eastern Brazil limits the potential for further outbreaks

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Keywords: Zika virus, microcephaly, serology

Background and objectives: During 2015-2016 Brazil notified more Zika virus (ZIKV) cases than any other country, yet population exposure remains unknown. Serological studies on ZIKV have been hampered by cross-reactive immune responses against heterologous viruses in affected populations.

Materials and methods: We conducted serosurveys for ZIKV, Dengue (DENV) and Chikungunya (CHIKV) virus using ELISA and plaque-reduction neutralization tests in 910 individuals sampled during 2013-2016, including HIV-infected patients, and microcephaly and non-microcephaly pregnancies, tuberculosis patients, and university staff

in Salvador city, Brazil. Individuals were georeferenced and sociodemographic indicators were compared between ZIKV-positive and -negative areas, and areas with and without microcephaly cases. Epidemiological key parameters were modelled in a Bayesian framework.
Results: ZIKV seroprevalence increased rapidly during 2013-2016, reaching 63.3% by 2016 comparable to DENV (75.7%) and higher than CHIKV (7.4%) Of 19 microcephaly pregnancies, 94.7% showed ZIKV-IgG antibodies, compared to 69.3% of 257 non-microcephaly pregnancies (p=0.017). Analyses of sociodemographic data revealed higher than average ZIKV prevalence in low socio-economic status (SES) areas. High seroprevalence, combined with case data dynamics allowed estimates of the basic reproduction number R0 of 2.1 (CI, 1.8-2.5) at outbreak onset and an effective reproductive number Reff <1 in subsequent years.
Conclusion: Our data corroborate ZIKV-associated congenital disease, an association of low SES and ZIKV infection and suggest population immunity caused cessation of the outbreak. Similar studies from other areas in Latin America will be required to determine the fate of the ZIKV outbreak.

N17

Novel anthropozoonotic transmission of human coronavirus OC43 to habituated great apes

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Keywords: human coronavirus OC43, chimpanzee, anthropozoonosis

Background and objectives: Pandemic human respiratory viruses have caused lethal outbreaks in several great ape populations, with pneumoviruses being repeatedly identified as the causative agents. Between late December 2016 and early January 2017 several individuals of a habituated chimpanzee community living in the Taï National Park, Ivory Coast, presented mild respiratory symptoms. Our study non-invasively investigated a potential infectious agent implied in this outbreak.
Materials and methods: Collection of faeces from symptomatic and asymptomatic individuals was routinely performed as part of the health monitoring programme of the Taï Chimpanzee Project. Total nucleic acids were extracted from 79 samples collected between November 2016 and February 2017. PCR screening included major respiratory viruses such as human pneumoviruses, influenza A and B, coronaviruses, enteroviruses, parainfluenza and adenoviruses.
Results: The PCR screening, coupled with sequencing, revealed the presence of a human coronavirus OC43 in 20/79 samples belonging to 11 different individuals. Positive samples matched the symptomatic individuals and the time frame of clinical presentation.
Conclusion: We report the first identification of a human coronavirus OC43 in habituated great apes. Our results provide additional evidence supporting the high likelihood of pathogen exchange among sympatric primates, highlighting the importance of further improving existing preventive strategies.

N18

Diversity of ticks and tick-borne pathogens from areas at the Black Sea in Bulgaria

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Keywords: tick-borne pathogens, Ixodes spp., Rhipicephalus sanguineus

Background and objectives: To date, there are only a few studies concerning ticks and tick-borne pathogens from Bulgaria especially at the Black Sea region. This is why this study is aiming for investigating ticks morphologically and for tick-borne pathogens in this area. Material and methods: In 2013/2014 ticks were collected at six different sites in Bulgaria. After species identification, ticks were tested in pools for *Anaplasma phagocytophilum*, *Babesia* spp., *Borrelia* spp., *Rickettsia* spp. and *Candidatus Neoehrlichia mikurensis* via conventional and real-time PCR.
Results: Altogether, 1541 ticks belonging to 12 species were collected. Apart from 181 specimens collected from pets and livestock, all other ticks were questing. Dominating tick genera were Ixodes and Rhipicephalus. Very high prevalences were achieved for *Rickettsia* spp. (90.77%), followed by *A. phagocytophilum* (6.78%), *Borrelia burgdorferi* s. l. (1.96%), *Babesia* spp. (0.08%) and *Candidatus Neoehrlichia mikurensis* (0.08%). Only *Rickettsia* spp. and *A. phagocytophilum* were found in ticks collected from animals.
Conclusion: This study presents high tick diversity in Bulgaria especially in areas near the Black Sea. This study shows very high prevalences for *Rickettsia* spp. in ticks from Bulgaria and moderate to low prevalences for all other pathogens. One should take into account that tick bites in this area could lead to *Rickettsia* infection in humans and mammals.

N19

Swarm incursions of multiple reassortants of highly pathogenic avian H5 influenza strains during winter 2016/17 in Germany

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Keywords: highly pathogenic avian influenza, multiple reassortants, zoonotic potential

Background: Highly pathogenic avian influenza viruses (HPAIV) H5 of Asian origin (goose/Guangdong/96) evolved into various phylogenetic clades, subtypes and numerous genotypes. Some of these lineages contain strains with enhanced zoonotic properties causing fatal human infections. Starting in November 2016, HPAIV H5 was prevalent in Germany, causing high mortality in wild and domestic birds.
Methods: A selection of HPAIV-H5 positive swab samples and/or tissues from dead wild birds, poultry and zoo birds were sequenced and analysed.
Results: More than 1200 cases of HPAIV H5 infected dead wild birds of at least 68 species were detected across Germany. In addition to wild birds more than 90 poultry holdings and 15 zoos were affected. The outbreaks were caused by HPAIV of subtype H5N8 and H5N5. The viruses were closely related to viruses detected at the Chinese-Mongolian and Russian-Mongolian border earlier in summer 2016. All sequenced German viruses contained reassorted genomes compared to their ancestors and therefore represent different genotypes. At least four different genotypes were identified.
Conclusions: HPAI H5N8 and H5N5 detected in birds in Germany 2016/2017 represent a swarm of several distinct reassortants of H5 viruses, indicating multiple independent entries of HPAIV into Germany.

N20

Discovery of Bluetongue virus, Tibet orbivirus and a novel rhabdovirus in mosquitoes from Nepal

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Keywords: bluetongue virus, rhabdovirus, mosquito

Background and objectives: Arthropod-borne viruses are one of the important etiological agents to cause diseases in humans and animals. Investigation of mosquito-borne viruses initiated in Nepal led to the discovery of novel viruses. The objective of the study was to characterize newly identified viruses.
Materials and methods: Virus isolation was done in C6/36 cells. Whole genomes were sequenced by NGS. Growth analyses were performed in insect and vertebrate cells.
Results: We isolated 3 BTV (bluetongue virus) strains, 1 TIBOV (Tibet orbivirus) strain, and 16 strains of Moaean rhabdovirus (proposed) in C6/36 cells. Full genome sequences of these viruses revealed typical genomic organization of orbiviruses and rhabdoviruses. BTV-Nepal was related to two different genotypes of BTV. TIBOV-Nepal showed closest relationship to TIBOV isolated in China. Phylogenetic analysis of Moaean virus indicated it belongs to a putative new genus. Growth kinetics demonstrated that viruses replicated well in *Aedes* cells C6/36 and U4.4, but showed different growth in *Culicoides sonorensis* and *Anopheles stephensi* cells. BTV-Nepal and TIBOV-Nepal replicated in various livestock cells. Moaean virus did not replicate in the tested vertebrate cells.
Conclusion: The detection of BTV in mosquitoes raises the question whether BTV can be transmitted by mosquitoes. Vector competence studies are ongoing. Further investigation is needed to determine if Moaean virus can infect vertebrates.

N21

CCHFV infection risk in Cameroon

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Keywords: Crimean-Congo hemorrhagic fever virus; multiplex RT-qPCR; Cameroon

Background and objectives: Crimean-Congo hemorrhagic fever (CCHF) is a fatal tick-borne viral infection of humans. The CCHF virus circulates in a tick-vertebrate-tick cycle. Following infection animals do not show clinical symptoms but may develop viremia for up to two weeks. Antibodies can be detected for several years. Therefore, the screening of ruminants is a good indicator for CCHFV presence in a country. However, virus genome detection is the ultimate proof of current CCHFV circulation. CCHFV is present in Africa, Asia and Europe, but most studies are outdated. In Cameroon no investigation on CCHFV was done so far, even though CCHFV is known to circulate in neighbouring countries.

Materials and methods: Approximately 1000 bovine serum samples were tested for CCHFV-specific antibodies by using two different ELISAs and/or IFA. Additionally 109 Hyalomma ticks, which were collected from infested cattle in a high prevalence area in Northern Cameroon, were tested for CCHFV genome using a novel multiplex one-step RT-qPCR. This highly sensitive and specific RT-qPCR enables to detect all six known CCHFV clades.

Results: An overall CCHFV-specific antibody prevalence of 74% was detected. The PCR investigation resulted in 7 CCHFV genome positive H. truncatum ticks. These CCHFV sequences clustered together with others form the African clade.

Conclusion: This first proof of CCHFV circulating in Cameroon shows that there is an infection risk for the human population, especially for risk groups.

N22

Hantaviruses in the natural host and in "spillover"-infected animals

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Keywords: Cameroon hantaviruses, evolutionary lineage, spillover-infection

Background and objectives: The *Puumala virus* (PUUV), one of the most important hantaviruses in Europe with the bank vole (*Myodes glareolus*) being its reservoir, causes a mild to moderate form of Hemorrhagic fever with renal syndrome in humans. The target cells of hantaviruses are poorly characterized in the natural reservoir and in „spillover"-infected animals.

Materials and methods: During spring and autumn of 2015 and 2016 101 bank voles, 73 yellow-necked mice *(Apodemus flavicollis)* and 23 wood mice *(Apodemus sylvaticus)* were trapped in the district Osnabrück (OS). The animal carcasses were dissected and tested for hantavirus-RNA by reverse transcription-PCR (RT-PCR), using lung tissue, and for serum antibodies by indirect IgG-ELISA. Furthermore, a *cytochrome b*-PCR and sequence-based classification of evolutionary lineages was done for all bank voles.

Results: The PUUV seroprevalence ranged between 11 and 48% in bank voles, between 6 and 20% in yellow-necked mice and between 0 and 12% in wood mice. PUUV-RNA was detected exclusively in bank voles; the RNA prevalence ranged between 0 and 44%. Single „spillover"-infections in yellow-necked mice and wood mice were indicated by exclusive detection of PUUV-reactive antibodies. According to the *cytochrome b* sequences all bank voles belong to the Western evolutionary lineage.

Conclusion: The results show the continuous presence of PUUV in the bank vole populations with a higher prevalence in spring than in autumn.

N23

Detection of *Arcobacter* spp. along the intestinal tract of broiler chickens

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Keywords: Arcobacter spp., chicken slaughterhouse

Background and objectives: Arcobacter spp. is regularly detected in chicken meat. The intestinal content of the chickens is assumed as the source of contamination during the slaughtering process. However, data on the occurrence and quantitative load of *Arcobacter* spp. along the complete intestinal tract of chicken are still scarce. Therefore, we examined the intestinal content of the duodenum, jejunum, caeca and colon of broiler chickens individually to test for the presence and the concentration of *Arcobacter* spp.
Materials and methods: Intestinal tracts of 25 broiler chickens from 5 different flocks were collected and intestinal content of the duodenum, jejunum, caeca and colon was extracted. Of each sample, 1 g was examined for the presence and quantitative load of *Arcobacter* spp. by selective enrichment. Suspected Arcobacter colonies were verified by mPCR and rpoB sequencing.
Results: In 44% (11/25) of the duodenal, 64% (16/25) of the jejunal, 8% (2/25) of the caecal and 92% (23/25) of the colonic samples *A. butzleri* were detected. The highest *Arcobacter* load was determined in the colonic content (23 MPN/g), followed by duodenum and jejunum (0.023 – 0.23 MPN/g), respectively.
Conclusion: Our data support the hypothesis that the intestinal tract has to be considered a source of entry for *Arcobacter* spp. into the poultry slaughterhouse, thereby enabling contamination and cross-contamination of the chicken meat. In contrast to *Campylobacter*, the highest numbers of *Arcobacter* spp. were detected in the colon, whilst the caeca showed the lowest *Arcobacter* concentration. We were also able to detect *Arcobacter* spp. in the small intestine, however, with lower bacterial numbers compared to colon. Therefore we recommend to use colonic content when testing for the presence of *Arcobacter* spp. in chicken at slaughter.

N24

First molecular biological analysis of Tick-borne encephalitis virus in Akmola Oblast and East Kazakhstan Oblast

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Keywords: Tick-borne encephalitis virus, Dermacentor marginatus; Ixodes persulcatus

Background and objectives: Tick-borne encephalitis (TBE) is endemic in Eurasia and is one of the most dangerous neuroviral infections in humans. In Kazakhstan (KZ), 216 TBE infections were reported from 2011-2016. We focused on two regions of KZ, the endemic region Eastern Kazakhstan Oblast (EKO) and a region not endemic for TBE, Akmola Oblast (AO). Our goal was to determine the minimum infection rate (MIR) of TBEV in ticks, to assign the TBEV subtype and to perform a first phylogenetic analysis.
Materials and methods: In 2016, 1522 ticks from 26 districts were collected in EKO and 437 ticks from one district in AO. Ticks were sorted into 485 and 90 pools, respectively. After homogenization, tick pools were tested by TBEV real-time RT-PCR. Subsequently a E-gene RT-PCR, sequencing and molecular analyses were carried out.
Results: TBEV RNA was detected in 5 pools of EKO (MIR 0.5%) and 2 pools of AO (MIR 0.5%). TBEV was found in *Ixodes persulcatus* and in AO also in *Dermacentor marginatus*. TBEV from both oblasts belong to the Siberian subtype. Phylogenetic analysis showed that the new TBEV strains from EKO are closely related to strains from China and Western Siberia. TBEV from AO are related to strains from Western Siberia.
Conclusion: In this study, we show first molecular biological data on TBEV in a previously known endemic region in EKO. AO, which was

not endemic, we detect TBEV for the first time in ticks. Our data will directly contribute to improving the KZ health system

N25

Screening of zoo animals for markers of hepatitis E virus infection

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Keywords: hepatitis E virus, zoo animals, rat HEV

Background and objectives: The number of notified hepatitis E cases is steeply increasing during the last years. The zoonotic hepatitis E virus (HEV) genotype 3 (HEV-3) is the main cause of these human diseases. Pigs and wild boars represent the major reservoirs of HEV-3. Additionally, several HEV-related viruses have been identified in other animal species, e.g. the rat HEV in wild rats, with unknown zoonotic potential. Here, several zoo animals representing a wide range of animal species and pest rats were investigated for markers (antibodies and/or viral RNA) of HEV infection.
Materials and methods: Sera were analysed for HEV-specific antibodies by commercial ELISA. The HEV genome was detected by real-time RT-PCR and a broadly reacting RT-PCR followed by sequencing of the PCR products.
Results: HEV-specific antibodies were detected in 35/503 (7%) animals and 19/82 (23.2%) species. Primates had a low seroprevalence, whereas suids and carnivores showed the highest antibody detection rates. No HEV-3 genome was detected in the sera, but one Syrian brown bear (Ursus arctos syriacus) was positive for rat HEV RNA. The sequence was most closely related to rat HEV strains from wild rats (*Rattus norvegicus*) from the same area.
Conclusion: HEV-specific antibodies can be detected in a wide range of animal species, thus posing a potential risk of infection for humans. Further investigations in carnivores are needed to estimate the significance of rat HEV.

N26

Zoonotic viruses detected in free-living animals in the Czech Republic

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Keywords: emerging zoonoses, free-living animals, Czech Republic

Background and objectives: From a large number of zoonotic viruses presented in Europe, we selected those that are well described in Europe, can cause diseases in humans with a scale of different symptoms and were also detected in the Czech Republic. We aimed at the detection of hantaviruses in the free-living rodents, flaviviruses in the free-living waterfowl and hepatitis E virus in wild boars and other game animals. These viruses belong to the so-called emerging viruses, and in some countries, they are monitored over a long period.

Materials and methods: For hantavirus detection, we used molecular screening method based on L segment. For the detection of specific antibodies against flaviviruses, we used plaque-reduction neutralization test and for specific antibodies against hepatitis E virus, we used commercially available ELISA test.

Results: We detected all selected viruses in free-living animals in the Czech Republic. From 500 rodents, one field vole and more than 30 common voles were positive for Tula virus. From 146 common coots, 9 had specific reaction with Usutu virus and 2 with West Nile virus. Out of 1 000 ungulates, 53 had specific reaction with West Nile virus. From 300 wild boars, 31 of them were seropositive for hepatitis E virus.

Conclusion: Presented results indicate that the studied viruses circulate in nature in selected areas and they represent a possible health threat to the human population.

N27

Seroprevalence of West Nile virus (WNV) in horses in selected locations of the Republic of Serbia

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Keywords: West Nile virus, horses, Serbia

Background and objectives: The aim of this study was to assess the seroprevalence of WNV in horses in selected locations of the Republic of Serbia as possible indicators of critical areas for human infections and to compare the diagnostic value of different tests.
Materials and methods: Horse blood sera (n=303) were collected from 10 different towns in Serbia in 2011-2013. For the determination of specific anti-WNV antibodies two different methods were used: 1. IIFT -"in house" test and commercial test Anti-West Nile virus IIFT (IgG) (Euroimmun, Germany) 2. ELISA- "in house" test and commercial competitive ELISA test ID Screen® West Nile Multi-species (IDVet, France). Diagnostic tests were statistically compared with McNemar ($\chi 2$ test) using Microsoft Excell 2010 and SPSS Statistics 20.
Results: The study revealed seropositivity in all 10 locations in 108/303 sera (35,6%) by "in house" IIFT, 75/234 sera (32,1%) by commercial IIFT, 81/301 sera (26,9%) by "in house" ELISA and 57/301 sera (19,9%) by commercial ELISA. There were no statistically significant differences between results of "in house" and commercial tests in both methods (IFAT $p<0.383$, ELISA $p=0.022$).
Conclusion: Horses were seropositive in all locations for WNV in the period from 2011-2013 in which also human epidemics occurred. There were no statistically significant differences between used "in house" and commercial test. IIFT is more sensitive and less expensive method than ELISA for WNV diagnosis.

N28

New Hepatitis E Virus Genotypes in Rodents, China

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Keywords: Hepatitis E virus, Genotype, Rodent

Background and objectives: Hepatitis E virus (HEV) is the prototype of the family Hepeviridae and cause the most common acute viral hepatitis globally. HEV variants have been detected in a variety of mammalian species, and some of them have been confirmed to have zoonotic potential. This study has investigated the wider host range of HEV within the species Orthohepevirus C, which is derived from the orders Rodentia, Soricomorpha and Carnivora.
Materials and methods: We tested 278 wild small mammal specimens from seven species in China for HEV RNA.
Results: Highly diversified and divergent HEV variants were detected in 60 (35.9%) of 167 Apodemus chevrieri, 6 (10.90%) of 55 Eothenomys melanogaster, and 1 (7.7%) of 13 Eothenomys chinensis. Four representative full genome sequences were amplified: RdHEVAc14 and RdHEVAc86 from A. chevrieri, and RdHEVEm40 and RdHEVEm67 from E. melanogaster. Pairwise amino acid distance based taxonomy indicated that RdHEVAc (>0.28) and RdHEVEm (>0.40) differed from other sequences. Quantitative RT-PCR and histopathology demonstrated that these rodent HEV variants have high hepatic tropism with liver inflammation.
Conclusion: This study supports data for the epidemiology and genetic diversity of HEV in Chinese rodents and gives new insights into the origin, evolution, and host range of HEV. With the framework and guideline provided by the latest consensus proposals for assigning new HEV variants, we provide convincing evidence that two new HEV genotypes can be designated within the species Orthohepevirus C, namely HEV-C3 and HEV-C4.

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N29

Characterization of *Arcobacter* spp. isolated from retail seafood in Germany

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Keywords: Arcobacter species, seafood, characterization

Background and objectives: *Arcobacter* species are considered emerging zoonotic pathogens which could provoke human gastroenteritis. However, information about the prevalence of *Arcobacter* in seafood products is still scarce. This study was aimed to investigate the prevalence of *Arcobacter* spp. in retail seafood like shellfish, shrimps and cephalopods in Germany, with further characterization of the isolates.
Materials and methods: *Arcobacter* spp. were recovered and isolated by cultural methods. By mPCR and rpoB sequencing *Arcobacter* was verified at genus and species level. All isolated strains were characterized by ERIC-PCR. Furthermore, the occurrence of 10 putative *Arcobacter* virulence genes were detected.
Results: Arcobacter spp. were isolated from 55 out of 318 seafood samples. Among all 62 isolates, 55% were *A. butzleri*, followed by 13% of both *A. cryaerophilus* and *A. venerupis*, 11% *A. aquimarinus*. *A. skirrowii* and *A. thereius* were only detected once, respectively. Three isolates could not be determined to species level. The ERIC-PCR results showed its capacity in *Arcobacter* spp. genotyping. The occurrence of virulence gene seemed to be higher in *A. butzleri* compared to the other species.
Conclusion: In this study, the prevalence of *Arcobacter* in retail seafood was 17%, coinciding with other studies. We suggest that *A. butzleri* possess the highest risk of human infection with *Arcobacter* due to its highest occurrence of putative virulence genes as well as highest prevalence among seafood.

N30

Sindbis virus – a wildbird associated zoonotic arbovirus also circulating in Central Europe?

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Keywords: Sindbis virus, wild birds, zoonosis

Background: Sindbis virus (SINV) (Togaviridae, genus Alphavirus) is an arbovirus that causes clinical symptoms including arthritis, rash and fever during acute human infections in Fennoscandia. Its transmission cycle involves mosquito species as vectors as well as wild birds as natural reservoir hosts. In Europe SINV outbreaks are largely restricted to Northern Europe. In Germany the first isolation of SINV was found in 2009 in mosquito species in the Upper Rhine valley and one year later in a hooded crow found injured in the city centre of Berlin.

Material and methods: Setting up a German nation-wide wild bird surveillance network for zoonotic arthropod-borne virus infections allows the monitoring of migratory and resident birds. RNAs were isolated from over 1.900 wild bird cruor samples (2014-2016) and subsequently analyzed using a SINV specific quantitative RT-PCR. Only one of the samples was positive for SINV, belonging to a common wood pigeon, found with neurological symptoms in the city of Giessen in 2016.

Results: Phylogenetic analysis of the complete SINV genome (cell culture supernatant), as determined by next generation sequencing (NGS), revealed a similarity of this virus to the virus found in the Upper Rhine valley but a difference to the one from the hooded crow.

Conclusion: SINV monitoring of wild birds and mosquitoes in Central Europe is needed to reveal its true prevalence and the SINV associated human infection risk and indigenous disease burden.

Poster Session Public Health and Social issues of Zoonoses Research

H01

Added value via crowdsourcing? An innovative exchange forum for science & public health

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Keywords: Crowdsourcing, exchange forum, public health

Background and objectives: In Germany no direct communication channel exists to share experiences and questions amongst members of the Public Health and Veterinary Services (PHS, PVS) and scientists. The aim is to present stakeholder feedback on a planned exchange forum with crowdsourcing character to strengthen exchange between PHS and PVS representatives and scientists working with zoonotic diseases.
Materials and methods: Crowdsourcing aims to increase the collective intelligence of participants. Access to the forum requires a voluntary monthly contribution of at least 20 minutes (alternative: quarterly fee). Members can post or comment on questions, indicate training and research needs, present best practice examples, scientific results and project ideas or form working groups. Network promotors recruit external experts, if further input is needed. A search database provides easy access to contents and is thus a valuable tool to inform research and training. A moderator maintains the forum and database. Innovative media are explored to address information or training needs. A needs assessment measures the need for and added value of the forum in a qualitative and quantitative manner.
Results: Feedback from various stakeholders consulted as part of the needs assessment is presented to encourage further feedback.
Conclusion: Besides strengthening information exchange and networking, the forum can lead to improved translation of science and more targeted research and training.

Poster Session Free Topics

F01

First detection of *Ehrlichia* sp. HF in *Ixodes apronophorus*, a neglected tick species in Romania

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Key words: Ixodes apronophorus, Romania, Ehrlichia species HF

Background and objectives: *Ixodes (I.) apronophorus* Schulze, 1924 (the "marsh tick") is primarily considered to be associated with rodents. Its role as vector of pathogens is unknown. In parts of Romania it is increasingly found as ectoparasite on dogs, foxes and hares.
Materials and methods: *Ixodes apronophorus* was sampled from dogs, foxes and hares in Arad, Caras-Severin, Dambovita, Galati, Mehedinti, Timiş counties and in Bucharest. Ticks were identified and individually homogenized analyzed for contained pathogens. The ticks were tested for *Borrelia* spp., *Rickettsia* spp. *Francisella tularensis*, *Mycoplasma* spp., *Coxiella* spp., *Neoehrlichia* spp., *Ehrlichia* spp., *Babesia* spp., *Theileria* spp. and *Hepatozoon* spp. using molecular detection techniques.
Results: All *I. apronophorus* were negative for *Borrelia* spp., *Rickettsia* spp., *Francisella tularensis*, *Mycoplasma* spp., *Coxiella* spp., *Neoehrlichia* spp., *Babesia* spp., Theileria spp., and Hepatozoon spp. However, for the first time *Ehrlichia* sp. HF in *I. apronophorus*, occurring in 16% (3/19) of the investigated ticks was found.
Conclusions: This is the first record of *I. apronophorus* in the sampled seven Romanian counties. *Ehrlichia* sp. HF, which was originally isolated in Japan and later also found in France was found for the first report in I. apronophorus and in Romania. Although

Ehrlichia sp. HF has not been found in humans it is a recognized pathogen in dogs. Its role as human pathogen has to be further studied.

F02

Susceptibility of avian influenza viruses to neuraminidase inhibitors

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Keywords: drug resistance, antiviral, surveillance

Background and objectives: Influenza type A viruses (FluA) occur naturally among wild aquatic birds and lead sporadically to outbreaks in poultry farms. Due to their pandemic potential, transmissions of such viruses to humans are an ongoing cause of concern. Neuraminidase inhibitors (NAI) are globally used for prophylaxis and treatment of influenza infections. The study focused on the susceptibility of FluA to the four NAI oseltamivir, zanamivir, peramivir, and laninamivir currently available commercially.
Materials and methods: Susceptibility of FluA to NAI was tested by using a validated fluorometric NA inhibition assay. FluA were selected to represent NA group-1 (NA-subtype N1, N4, N5, N8), and group-2 (NA-subtype N2, N3, N6, N7, N9) viruses as well as viruses of low and high pathogenicity (hemagglutinin-subtypes H5 and H7).
Results: All tested FluA showed *in vitro* susceptibility to the NAI tested. NAs belonging to group-1 were more sensitive to zanamivir than to oseltamivir, whereas group-2 NAs were more sensitive to oseltamivir than to zanamivir. Laninamivir inhibited FluA more efficiently than oseltamivir and zanamivir and even oseltamivir-resistant viruses. Peramivir was the most potent *in vitro* inhibitor of all tested FluA.
Conclusion: Although the tested NAs were sensitive to NAI the potential evolution of antiviral-resistant FluA should be closely monitored. This is even more important in the case of human infections with viruses from animal sources.

F03

Landscaping of cytomegaloviruses from wild African ape populations indicates high species-specificity

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Key words: Cytomegalovirus, great ape, species specificity

Background and objectives: Cytomegaloviruses (CMVs) are DNA viruses that are regarded as being highly species specific, with CMV genome sequence relationships reflecting the evolutionary relationship of their host. A number of CMV variants have been identified in samples isolated from free-ranging and captive great apes. However, the prevalence and genetic diversity of CMV in wild great apes has not been studied in depth. The present study uses non-invasive PCR-based analysis of stool samples to study the prevalence and strain variation of CMV in wild great ape populations.
Materials and methods: Stool samples were collected from 4 chimpanzee and 4 gorilla subspecies and bonobos in African national parks. Samples were analysed with generic PCR targeting glycoprotein B gene, a conserved region of the CMV genome. PCR products were directly sequenced and subjected to BLAST and phylogenetic analysis.
Results: Great apes from eastern, central and western Africa tested positive for CMV. CMV DNA presence was 6.15% (16/260) in chimpanzees, 57.14 % (20/35) in bonobos, and 14.6% (52/356) in gorillas. Sequences indicated high species specificity, with chimpanzee CMV, bonobo CMV and gorilla CMV being present in chimpanzees, bonobos and gorillas, respectively.
Conclusion: This study, using an innovative non-invasive method of analysis, represents the first large scale analysis of CMV distribution across diverse wild African ape populations and reports for the first time CMV in bonobos.

F04

Characterization of a new flavivirus isolated from sandflies in Panama

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Keywords: flavivirus, sandfly, sandfly-borne virus

Background and objectives: The genus *Flavivirus* contains viruses pathogenic for human and animals. Flaviviruses are mainly transmitted by mosquitoes and ticks. Two flaviviruses have been isolated from sandflies, Ecuador Paraiso Escondido virus (EPEV) and Saboya virus (SABV). The aim of this study was to test sandflies from Panama for infections with flaviviruses.
Materials and methods: In total 13.826 sandflies were collected in rainforest fragments in the area of the Panama canal. 1.778 pools were screened for flaviviruses using generic RT-PCR. Virus isolation was done in mosquito (C6/36), sandfly (LL5, PP9) and primate (Vero E6) cells. Genome sequencing was performed by PCR using generic primers.
Result: A novel flavivirus was detected in 22 pools of *Phlebotomus panamensis* sandflies by PCR. So far, 6.2 kb of the genome was sequenced. Closest similarity with 68% was found to the polyprotein of EPEV suggesting the detection of a previously unknown sandfly-associated flavivirus. The virus was isolated from four pools in C6/36 and LL5 cells.
Conclusion: Our data show that flaviviruses are more common in sandflies than previously thought. Further studies characterizing the virus are currently ongoing.

F05

A comparative analysis of subsampling methods to estimate the number of specimens and species in large mosquito samples

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Keywords: mosquito surveillance, large mosquito samples, subsampling

Background and objectives: Mosquito surveillance programs are conducted to assess the spatial-temporal distribution of vectors and associated pathogens. Thereby, single samples can consist of several thousand mosquito specimens making the sample analysis time- and money-consuming process, e.g. preventing short-termed decision for species-specific control measures. Therefore, the objective of this study was the evaluation of different techniques to accurately subsample large mosquito samples.
Materials and methods: In total, 23 samples comprising between 400 and 5000 mosquito specimens were analyzed using five different estimation methods: subsampling based on the area, volume and mass of the sample, automatized counting of specimens with a computer software and random selection of 200 specimens for species identification.
Results: Area-based sorting of 20% of the total sample resulted in an error of 10% for the number of specimens and non-detection of 20% species. Weight- and volume-based subsampling showed similar error rates. The computer software approach was suitable to estimate the total number of specimens only, whereas species detection based on 200 random selected specimens inadequately reflected the number of species.
Conclusion: Under the assumption that an error of 10% is acceptable and a shorter processing time, area-based subsampling of 20% of the total sample is the most appropriate method to estimate the number of specimens and species in large mosquito samples.

F06

Phylogenetic analysis of Cetacean morbilliviruses

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Keywords: morbillivirus, cetaceans, phylogeny

Background and objectives: In recent decades, outbreaks of cetacean morbillivirus (CeMV) in dolphins and whales have caused several mass mortality events (MME). This virus was initially recognized in 1990, when dolphin morbillivirus (DMV), a strain of CeMV, led to a MME in striped dolphins from the Mediterranean Sea. Furthermore, it has been documented to cross the species barrier, causing an epidemic in Mediterranean monk seals in 1997. Several other outbreaks and sporadic whale strandings due to CeMV have also been reported. The aim of this study was to identify mutations in CeMV leading to species adaptation. We compared full-length wild-type European CeMV strains from different species and geographical locations circulating at different time points. Moreover, laboratory strains were also compared to their wild-type counterparts.
Materials and methods: CeMV positive frozen tissue and cell culture samples were processed and prepared for deep sequencing. Full-length genomes were constructed and analyzed.
Results: Phylogenetic analyses indicate that DMV and porpoise morbillivirus (PMV) are in different clades. Low genetic diversity is presented within each strain (DMV: 96.1-99.9%; PMV: 99.2-99.5%) with DMV strains divergence based on geographical location, independently of their host species. Several unique mutations were identified in both DMV and PMV Vero-adapted strains.
Conclusion: DMV appears to infect a wide range of cetacean host species with minimal genetic adaptation.

F07

Bat Astrovirus Diversity: Insights into Virus Evolution Deduced from Sequence Information of the RNA-dependent RNA polymerase (RdRp) Gene

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Keywords: Astroviruses, Bats, Evolution

Background and objectives: Astroviruses (AstV) are abundant at high prevalences in many different mammalian and avian species. In infected individuals, symptoms range from diarrhoea to hepatitis, nephritis, respiratory syndrome and encephalitis, depending on the AstV strain. Recombination and spill-over events may have occurred during AstV evolution. However, little is known about the actual biological characteristics, pathology and evolutional history of bat AstV. Here, we present novel insights into AstV evolution deduced from bat AstV sequence information and sequence data from other host species.
Materials and methods: Bat-derived faecal and urine samples were obtained from three sampling sites in Germany (Würzburg, Wooster-Teerofen, Münster). Hemi-nested RT-PCR amplifying a 400 nt fragment encoding for the RNA-dependent RNA polymerase (RdRp) was performed followed by Sanger sequencing. Molecular evolutionary analyses were performed, including AstV variants derived from public databases using Geneious and MEGA software.
Results: Bat AstVs show a high variant diversity, forming distinct evolutionary lineages. Interestingly, several bat AstV variants are more similar to avian or other mammalian AstV variants than to known bat AstVs.
Conclusion: The evolutionary history of bat astroviruses indicates a rather fast radiation process within bats. A potential virus origin in bats followed by several spill-over events to other animal groups needs to be evaluated further.

F08

Development of a vaccination strategy against Bacillus anthracis and related pathogens in great apes

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Keywords: Bacillus anthracis, vaccination, wildlife conservation

Background and objectives: Since its discovery in Tai National Park (TNP), Ivory Coast, the bacterium *Bacillus cereus* biovar *anthracis* has continued to cause high mortality among chimpanzees in this tropical rainforest. This pathogen represents a serious threat to the survival of endangered wild great ape populations. First efforts to vaccinate a wild chimpanzee population in TNP using a blowpipe had to be discontinued in 2013, because the animals became afraid of the researchers following them. Hence, it is desirable to develop oral immunization procedures using food baits as an alternative vaccine administration method.
Materials and methods: Oral vaccination was tested in a habituated group of sooty mangabeys in TNP using the Sterne 34F2 vaccine. 10 times and 100 times parenteral dosage was tested; each dosage was administered to five mangabeys. Ten individuals served as control group, immunized by blowpipe or hand injection during anaesthesia. To evaluate the immunological response ELISA and Western Blot were used to detect antibodies against the Protective Antigen (PA) in blood and urine samples that were collected before and up to eight weeks after the vaccination.
Results: Oral vaccination did not trigger an anti-PA immune response and only 20% of the control group developed antibodies.
Conclusion: Neither oral nor injected vaccination succeeded. To see if the chimpanzee vaccinations in 2012 and 2013 have been successful, urine samples of these years are now being tested.

F09

Novel Double-Attenuated Influenza A Live Vaccines in Swine

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Keywords: swine influenza viruses, pandemic influenza, live attenuated vaccine

Background and objectives: Pigs are commercially relevant livestock and frequently infected with influenza. This disease is of great economic relevance and bears high zoonotic risks. To reduce disease burden and occlude virus reservoirs, effective vaccination has remained a key issue. However, the conventional inactivated vaccines often provide insufficient levels of protection. Therefore, we investigated the potential of a double-attenuated mutant, carrying a non-physiological, strictly elastase-dependent HA cleavage site and a C-terminally truncated NS1 protein, to serve as live vaccine in swine.
Materials and methods: By reverse genetics, we generated a double-attenuated mutant of the strain A/Bayern/74/2009 (H1N1v) and challenged prime-boost immunized pigs with different influenza strains.
Results: In-vitro, the double-attenuated mutant strain showed a strictly elastase-dependent growth. In pigs, prime-boost immunized animals developed neither clinical symptoms nor nasal virus shedding if challenged with the homologous wild-type. Furthermore, we observed considerably reduced clinical signs and no shed virus in nasal swabs after homo-subtypic infection with another unrelated H1N1 strain.
Conclusion: Overall, strong protection elicited by the elastase HA cleavage site/NS1 mutant against the same HA/NA subtype suggests broad tolerance against antigenic drift.

F10

Genetic variability of *Rickettsia africae* in Adama, Oromia Region, Ethiopia

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Keywords: swine influenza viruses, pandemic influenza, live attenuated vaccine

Background and objectives: *Rickettsia* are obligate intracellular Gram-negative bacteria that are transmitted by vectors. Tick-borne spotted fever group (SFG) rickettsiae are transmitted by ticks that parasitize domestic animals, especially cattle. These infections are considered as emerging infections in humans in various African countries.
Materials and methods: 291 ixodid ticks (245 *Amblyomma cohaerens*, 21 *Amblyomma variegatum*, 18 *Rhipicephalus pulchellus*, and 7 *Rhipicephalus decoloratus*) were collected from slaughtered cattle in Adama, Oromia, Ethiopia in 2007 (145 specimens) and 2016 (146 specimens). All ticks were individually screened for rickettsiae by a panRickettsia-real-time-PCR.
Results: A total of 32 (22.1%) samples (14 *A. cohaerens*, 17 *A. variegatum* and one *R. pulchellus*) were found positive for rickettsiae. All positive samples were further investigated using PCRs targeting the 23S-5S intergenic spacer, ompA and ompB genes. The sequence analysis revealed the circulation of three different clades of *Rickettsia africae* in the Oromia region. No association of the different *R. africae* variants to respective tick species was found.
Conclusion: Three different clades of *Rickettsia africae* are thus present in the Oromia region. Humans are at risk of acquiring rickettsial infections. Further studies are needed to investigate possible differences in the human-pathogenetic potential of these genetic variants.

F11

Decontamination of infrastructure and laboratory equipment in high-containment facilities by peracetic acid fumigation

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Keywords: peracetic acid, decontamination, infrastructure

Background and objectives: Decontamination of high-containment infrastructure is a challenging procedure especially in research facilities where highly contagious viruses are handled. In this study, we adapted a peracetic-acid-based room decontamination method to effectively decontaminate certain microorganisms.
Materials and methods: Laboratory equipment was placed in a supply airlock (14m³) and decontaminated with a dry-fog generator that releases an ultrafine mist composed of 1.3% peracetic acid (PAA) and 6.6% H2O2 (by volume) in water. Germ carriers (GCs) with ≥106 Geobacillus stearothermophilus spores, either commercially available or self designed, as well as GCs with an air-dried preparation of 104.5-7.0 infectious units of porcine enterovirus or murine norovirus were placed inside the airlock and laboratory equipment during each run.
Results: PAA dry fogging resulted in the absence of growth of most commercially available spores as well in a 6-log level reduction of most virus GCs depending on the location and the accessibility of PAA dry fog. Self- designed GCs were more resistant to PAA dry fog than commercially GCs. No damage to the equipment and the surfaces was seen after nine runs.
Conclusion: PAA dry fogging is a robust method for inactivation of viruses and bacterial spores. It is not advisable to rely only on the widely used commercial spore carriers as biological indicators due to false negative results. Parallel testing of microorganisms that are actually handled in the facility or their surrogates should be considered.

F12

Mycobacterium genavense infections in captive birds

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Keywords: Mycobacterium genavense, avian, co-infections

Background and objectives: *Mycobacterium genavense* (Mg) is a relevant pathogen for young, old, pregnant and immunocompromised people (YOPIs). In birds only a few studies exist suggesting that Mg is the primary cause of mycobacteriosis in psittaciform and passeriform pet birds, but there is still little information on this zoonosis.
Materials and methods: In this first study on the prevalence of Mg in naturally infected, captive bird populations, real-time PCR examinations were conducted in 134 individual passeriform and psittaciform birds to determine the prevalence in five different aviaries. Ante mortem examinations of faeces and cloacal swabs were compared with post mortem examinations of tissue samples to evaluate the reliability of ante mortem diagnostics. Birds were examined for viral infections, such as circovirus and polyomavirus, to indicate their immune status. Clinical signs from 83 birds were retrospectively evaluated.
Results: A varying detection prevalence of Mg infections in the flocks ranging from 10% to 91% based on post mortem testing was revealed. False negative ante-mortem results occurred in 64%. Viral co-infections and unspecific clinical signs were common.
Conclusion: It has to be assumed that Mg infections are more widespread in captive bird populations than anticipated. Viral infections might be an important risk factor. There is an urgent need for improvement of ante mortem diagnostics to evaluate the zoonotic risk originating from pet birds.

F13

The largest *Campylobacter coli* outbreak in Germany, associated with mincemeat consumption, May 2016

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Keywords: Nontuberculous mycobacteria, zoo animals, transmission

Background and objectives: Outbreaks of *Campylobacter coli* are rare in Germany and usually involve <5 cases. On 01/06/2016, the public health office of Märkisch-Oderland reported 11 *C. coli* cases. We aim to identify the source of the outbreak.
Materials and methods: Cases were residents in Märkisch-Oderland with symptoms' onset between 23/05-27/05/2016 and with positive C. coli-culture or epidemiologically linked symptomatic persons. Controls were selected from the same area using random digit dialing. We interviewed cases and controls with a structured questionnaire on food consumption and calculated odds ratios (OR) with 95% confidence intervals (95%CI). The local food safety authorities collected food and environmental samples. The National Reference Center subtyped human isolates by pulsed-field gel electrophoreses (PFGE).
Results: Of 15 identified cases (33% female, median age: 51; range: 4-69; 4 hospitalised), 12 had identical PFGE pattern. Cases were more likely to consume pork mincemeat (9/10 cases vs 6/14 controls; OR=12, 95%CI=1.2-122) and to consume it raw (9/10 cases vs 0/14 controls; OR=∞, 95%CI=16-∞). Seven cases, but no controls bought meat at the local butcher. Pork and environmental samples in the butchery tested negative.

Conclusion: This was the largest *Campylobacter coli* outbreak documented in Germany since 2001. Epidemiological evidence suggested raw mincemeat from a local butcher as the most likely vehicle of infection. Improvements of food safety regulations are still required to reduce contamination of pork with *C. coli*.

F14

Vector competence of European mosquitos for zoonotic *Dirofilaria* spp.

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Keywords: Dirofilaria spp., Aedes spp., larval development

Background and objectives: Transmission of the zoonotic mosquito-borne filarial nematodes *Dirofilaria immitis* and *D. repens* depends on the presence of competent mosquito species and temperatures suitable for the development of infectious filarial stages. This project investigated the development of these two *Dirofilaria* species under standardised experimental laboratory conditions in indigenous (*Aedes vexans*, *Ae. geniculatus*) and invasive (*Ae. albopictus*, *Ae. japonicus*) *Aedes* species.
Materials and methods: Mosquitoes were allowed to feed through artificial membranes on microfilariaemic blood. Blood-fed mosquitoes were incubated under realistic fluctuating summer temperature regimes (21°C and 27°C on average; range 12-29°C and 17.5-35°C) and dissected at pre-set time points under a microscope to observe filarial developmental stages. Additionally, real-time PCRs were carried out to assess infection rates.
Results: Around 500 individual blood-fed mosquitos were investigated. Results show vector competence for laboratory strains of *Ae. vexans* and *Ae. geniculatus* and field-collected *Ae. japonicus*, whereas laboratory strains of *Ae. japonicus* and *Ae. albopictus* were refractory to development of *D. immitis* and *D. repens,* respectively.
Conclusion: The results indicate the potential for endemic transmission of *Dirofilaria* spp. under Central European summer climate and emphasise the importance of using local vector populations when aiming to make risk assessments.

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