

FRIEDRICH-LOEFFLER-INSTITUT

FLI

Bundesforschungsinstitut für Tiergesundheit
Federal Research Institute for Animal Health



Junior Scientist Symposium 2023

15th to 17th November 2023

Jena

Impressum

Friedrich-Loeffler-Institut, Jena, Germany

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Jasmin Deutschendorf, Jonny Müller, Lena-Sophie Paul, Maike Richter,
Marwa Bassiouny, Mauricio Andino Molina & Selina Fuchs**

Jena, 2023

Welcome note

Dear young scientists,

It is a good tradition to have an annual Junior Scientist Symposium at the Friedrich-Loeffler-Institute organized by young colleagues from within our institute. This year the team from the institutes in Jena is in charge of hosting this scientific meeting of doctoral students and postdocs from the FLI.

This is the first time for me to welcome the young FLI scientist to the Junior Scientist Symposium. For some of you it will be the first time to speak about your scientific work to a large audience - and I can imagine, how excited and nervous you might be. At least, I remember “my first time” even after 30 years. A dear colleague of mine postulated that we agree to give presentations at conferences just to feel the deep relief when the talk or discussion is over. However, I hope that beyond all the work and tension during preparation of your contribution to this meeting you could feel the spirit and deep satisfaction, which comes with exploring the “terra incognita”, the unknown, the new frontiers of knowledge associated with your work. Without doubt, I am sure that you all can be proud of your achievements.

In addition to contributions from your young peers, the Jena organizers put together a really exciting program part featuring excellent guest speakers, who represent a diversity of scientific fields. You will hear about highly relevant topics like One Health or Emerging Pathogens. But while in a scientific career it is tempting to become an expert knowing more and more about less and less until in the end you know almost everything about nothing, some of the speakers will remind you about the world beyond the immediate field of microbiology or e.g. animal husbandry. I wish you an open mind to follow their ideas and insights.

Scientific meetings live from discussion and exchange of ideas - and analogous to your lab or computer work, this requires training and exercise. And just as for any sports, the earlier you learn the better you will be. Thus, I would like to nudge you to take an active part in the meeting beyond your own presentation. Listen, challenge ideas, bring forward suggestions: this will make the conference a living organism, will establish bonds between you and your peers and will be a seed of new ideas for your work.

For the meeting, in the “Lichtstadt” Jena, which is a well-fitting nickname for the city, I wish you and the organizers a good time in an inspiring, relaxed and collegial environment. A special thanks goes to the team in Jena for their efforts to organize and host the conference.

Prof. Dr. Christa Kühn
President of Friedrich-Loeffler-Institut

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General Information

Dates & Venues

Please note that we have **two different venues** for this year's Junior Scientist Symposium!

November 15th, 2023:

Friedrich-Loeffler-Institut, Naumburger Str. 96A, 07743, Jena.

November 16th-17th, 2023:

Förderverein Bären Lobeda e.V., Marktstraße 26, 07747 Jena.

Conference Language

The official conference language is English.

Organization Team

Christine Thomas, Felix Tetzlaff, Jonny Müller, Lena-Sophie Paul, Maike Richter, Marwa Bassiouny, Mauricio Andino Molina & Selina Fuchs.

Contact

JSS.Organization@fli.de

Social Events

Wednesday 15th (18:15 - 21:15)

*All participants of the meeting, are invited for **Bowling & Pizza Dinner Night** at "Bowling Roma" located in Löbstedter Str. 111, 07749 Jena. Please note that you will have to pay for the drinks at the location, as they are not included within the conference fee.*

Thursday 16th (21:30)

*Then, all participants are again invited after our **Social Dinner** for a **Movie Night** at "Schillerkino" located in Helmboldstraße 1, 07749 Jena. The feature will be a*

classic: “Back to the Future”. Please note that you will have to pay for the drinks or food at the location, as they are not included within the conference fee.

Social Dinner

*Our **Social Dinner** will take place **Thursday 16th** between 18:30 - 20:00 at the venue “Förderverein Bären Lobeda e.V.”; Please note that the dinner and drinks are included within the conference fee.*

Farewell Lunch

*A **Take-Away Lunch Pack** will be conveniently available at the venue location at noon of **Friday 17th**. This meal is included within the conference fee.*

Special Guests

- **Dr. Marco Körner**, “Science communication is a craft, and a craft can be learned.”
- **Dr. Anne Hartebrodt**, “Understanding systems biology: from genes to networks”
- **Prof. Dr. Stefan Schwarz**, “Transfer, co-selection and persistence of antimicrobial resistance genes among bacteria”
- **Dr. Marta Bonilla**, “Investigation of factors influencing the antimicrobial activity of porcine neutrophils during bacterial infections.”
- **Prof. Dr. Sascha Knauf**, “Fighting Neglected Tropical Diseases under the One Health Approach.”
- **Dr. Wutzler - Friedrich-Schiller-Universität Jena**, “Careers for doctoral graduates in science, industry and society.”

Posters’ Sessions

Each participant has the opportunity to present his/her poster during the meeting. Please stay near to your poster and be ready to present it during your assigned poster session. Additionally, the abstract corresponding to each poster is published within this abstract volume of the meeting.

Please hang your poster **Thursday 16th** morning around **08:30** at your designated poster stand. You find the information for your poster stand on page **26** of this Abstract Band.

Scientific Posters' Raffle

As a member of the *FLI Junior Scientific Community*, you are encouraged to attend this year's "Scientific Poster Session" where you will be introduced to a diverse range of topics; bear in mind you are free to choose which topics and posters interest you the most. After every poster presentation, the FLI Junior Scientist will award you with a collectable token to this card. In other poster sessions you are very welcome to visit other posters to discuss. *Those eagers for knowledge who collect the most of tokens will be awarded with a little surprise.*

Election of the doctoral student representative

The election of the doctoral student representative will take place at the end of the Junior Scientist Symposium. The *elected doctoral student representative* will represent the interests of all doctoral students at the FLI, and can be contacted if questions or problems occur during the course of the PhD time/program. The candidates will be shortly presented at the symposium.

Funding

The Junior Scientist Symposium is supported by the FLI Sponsors Association (FLI-SA). We would like to thank the FLI-SA for their financial support.

FLI Affiliations

- Institute for Bacterial Infections and Zoonoses (IBIZ)
- Institute of Epidemiology (IfE)
- Institute of Immunology (IfI)
- Institute for Infectious Disease Medicine (IMED)
- Institute for International Animal Health/One Health (IITG)
- Institute for Molecular Pathogenesis (IMP)
- Institute of Molecular Virology and Cell Biology (IMVZ)
- Institute of Livestock Genetics (ING)
- Institute for New and Novel Animal Disease Pathogens (INNT)
- Institute of Animal Nutrition (ITE)
- Institute for Animal Welfare and Animal Husbandry (ITT)
- Institute for Viral Diagnostics (IVD)
- Experimental Animal Husbandry and Biosafety Department (ATB)

List of participants

JSS 2023

Name & Surname	Institute	Mail	Participation Format	Number
Mauricio Andino Molina	IBIZ	mauricio.molina@fli.de	Poster	1
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Viktoria Korff	ATB	viktoria.korff@fli.de	Poster	11
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Jasmin Deutschendorf	IMP	jasmin.deutschendorf@fli.de	Poster	48
Joaquin Neumann	IITG/OH	joaquin.neumannheise@fli.de	Poster	50
Margret Vonholdt-Wenker	ITT	margret.vonholdt-wenker@fli.de	Talk	1
Ebere Roseann Agusi	External	roseebere8@gmail.com	Talk	2
Christine Thomas	IBIZ	christine.thomas@fli.de	Talk	3

Name & Surname	Institute	Mail	Participation Format	Number
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Constantin Lorenz	IVD	constantin.lorenz@fli.de	Talk	7
Johanna Wiethoff	IITG/OH	johannapauline.wiethoff@fli.de	Talk	8
Lea Lenhard	IFI	lea.lenhard@fli.de	Talk	9
Elena zu Klampen	ING	elena.zuklampen@fli.de	Talk	10
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Miles Winter	ITE	miles.winter@fli.de	Talk	16
Erik Bannert	ITE	erik.bannert@fli.de	Talk	17
Patrick Gutjahr	IMED	patrick.gutjahr@fli.de	Talk	18

Program

Wednesday, November 15th, 2023

Venue: *Conference Room, Institute of Bacterial Infections and Zoonoses, Jena*

From 13:30	Arrival and Registration
14:00 - 14:15	Welcome and Opening Prof. Dr. Heinrich Neubauer
14:15 - 15:00	Keynote Lecture 1 Dr. Marco Körner - Friedrich-Schiller-Universität Jena “Science communication is a craft, and a craft can be learned.”
15:00 - 15:15	Press
15:15 - 16:15	Guided tour at FLI Jena Prof. Dr. Christian Menge
16:15 - 17:00	Keynote Lecture 2 Dr. Anne Hartebrodt - FAU Erlangen/Bionets “Understanding systems biology: from genes to networks”
17:00 - 17:15	Coffee break
17:15 - 18:15	Talks I <u>Margret Vonholdt-Wenker (17:15 - 17:30)</u> <i>Evaluating and comparing sustainability aspects of animal husbandry systems in Germany: the web application InKalkTier makes it possible</i> <u>Ebere Roseann Agusi (17:30 - 17:45)</u> <i>Investigation of reverse spill-over transmission at the at high risk human-animal interfaces during COVID-19 pandemic in Nigeria</i> <u>Christine Thomas (17:45 - 18:00)</u> <i>Implementation, Validation, and Application of a Bioinformatics Framework for Analyzing Oxford Nanopore Technologies Genome Sequencing Data from Zoonotic Bacterial Pathogens</i> <u>Christoph Gerloff (18:00 - 18:15)</u> <i>Automatic detection of disease using Neural Networks on Audio-Data</i>
18:15 - 21:15	Dinner and Bowling

Thursday, November 16th, 2023

Venue: *Förderverein Bären Lobeda e.V., Jena*

08:30 - 09:00	Welcome, Information, Poster hanging
09:00 - 09:45	Keynote Lecture 3 Prof. Dr. Stefan Schwarz - Freie Universität Berlin “Transfer, co-selection and persistence of antimicrobial resistance genes among bacteria”
09:45 - 10:45	Talks II <u>Maike Richter (09:45 - 10:00)</u> <i>Determining antimicrobial resistance prevalence in Escherichia coli from fattening pigs by sampling animal transports at a slaughterhouse</i> <u>Christin Hennig (10:00 - 10:15)</u> <i>Novel swine influenza reassortant viruses: Risk assessment of their zoonotic potential</i> <u>Constantin Lorenz (10:15 - 10:30)</u> <i>Developing Novel Live Attenuated Vaccines for Foot-and-Mouth Disease</i> <u>Johanna Wiethoff (10:30 - 10:45)</u> <i>Cross-sectional study of zoonotic pathogen prevalence in cattle in Zanzibar, Tanzania</i>
10:45 - 11:00	Coffee break
11:00 - 11:45	Keynote Lecture 4 Dr. Marta Bonilla - Stiftung Tierärztliche Hochschule Hannover “Investigation of factors influencing the antimicrobial activity of porcine neutrophils during bacterial infections.”
11:45 - 12:45	Poster Session I
12:45 - 13:45	Lunch
13:45 - 14:45	Talks III <u>Lea Lenhard (13:45 - 14:00)</u> <i>Comparative analysis of immune responses and permissiveness to Cedar virus infection using multispecies approaches</i> <u>Elena zu Klampen (14:00 - 14:15)</u> <i>Single Cell Sequencing reveals transcriptional response to TGFβ1 and MFG-E8 treatment in equine endometrial fibroblasts</i> <u>Gregor Neufeld (14:15 - 14:30)</u> <i>Dissecting the bovine peri-implantation embryo development through single cell sequencing</i> <u>Benedikt Litz (14:30 - 14:45)</u>

Leaderless FMDV is fully attenuated in vivo and unable to establish persistent infection

14:45 - 15:45 Poster Session II

15:45 - 16:15 Coffee break

16:15 - 17:00 **Keynote Lecture 5**
 Prof. Dr. Sascha Knauf - Friedrich-Loeffler-Institute
 “Fighting Neglected Tropical Diseases under the One Health Approach.”

17:00 - 18:00 **Talks IV**
Md Rasheduzzaman (18:00 - 18:15)
Establishment of a Methodological Pipeline for the Characterization of the Virome of Different Mosquito Species
Richard K uchler (18:15 - 18:30)
Establishing a Phosphoproteomics Workflow for the Systematic Analysis of Virus-Induced Phosphorylation
Sophie-Celine Weinert (18:30 - 18:45)
Isolation attempts of neurotropic rustrela virus
Miles Winter (18:45 - 19:00)
Processing feces for downstream analyses of different non-invasive biomarkers

18:30 - 20:00 Dinner

From 21:30 Movie Night at Schillerkino - “Back to the Future” (1985)

Friday, November 17th, 2023

Venue: *Förderverein Bären Lobeda e.V., Jena*

09:00 - 09:30	Welcome
09:30 - 10:00	Talks V <u>Erik Bannert (09:30 - 09:45)</u> <i>Interrelations between lameness and water intake behavior of lactating dairy cows</i> <u>Patrick Gutjahr (09:45 - 10:00)</u> <i>Mosquitoes and peatland rewetting: An inconvenient nuisance or potentially harmful?</i>
10:00 - 10:30	Coffee break, Poster ranking
10:30 - 11:00	Selection of the new PhD representation Poster/Speaker Price
11:00 - 12:00	Keynote Lecture 6 Dr. Wutzler - Friedrich-Schiller-Universität Jena “Careers for doctoral graduates in science, industry and society.”
From 12:00	Lunch packages and Farewell

Venues' Locations

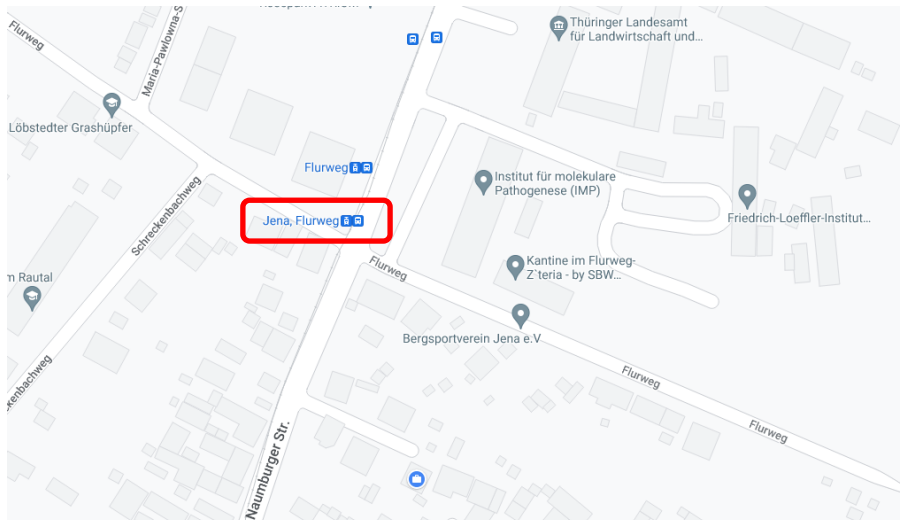
Friedrich-Loeffler-Institut

Institute for Bacterial Infections and Zoonoses (IBIZ)

Institute for Molecular Pathogenesis (IMP)

Naumburger Straße 96A

07743, Jena



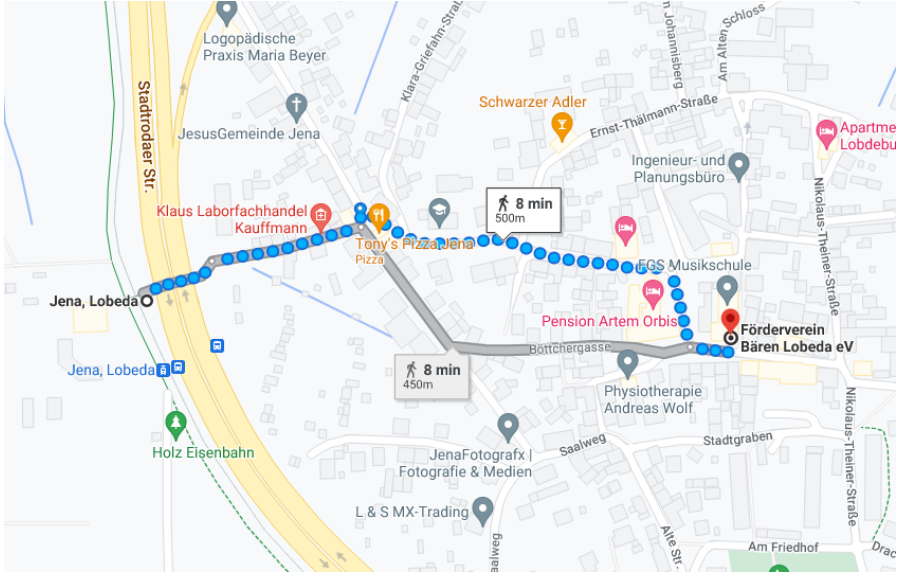
You can reach the FLI with the tram. By starting from the city center: take Tram Line 4 or 1 (direction 'Zwätzen') and leave at station 'Flurweg'. It is directly in front of the FLI.

Unfortunately, we currently have rail replacement traffic. Both Trams will only go to the stop 'Altenburger straÙe' - you then have to take the SEV bus to the station 'Flurweg' or you can walk (approx. 10 - 15 min).

For all guests who are not located in Jena: Please hold your ID-card ready. The security ward might check it.

Förderverein Bären Lobeda e.V.

Marktstraße 26,
07747 Jena.



You can reach our second Location 'Förderverein Bären Lobeda e.V.' with the Tram. By starting from the city center: take the Tram 4 (direction 'Lobeda-West') or Tram 5 (direction 'Lobeda-Ost'). Leave at station 'Jena-Lobeda'. From there you can reach the location in 5 minutes by foot.

Poster Sessions' Schedules

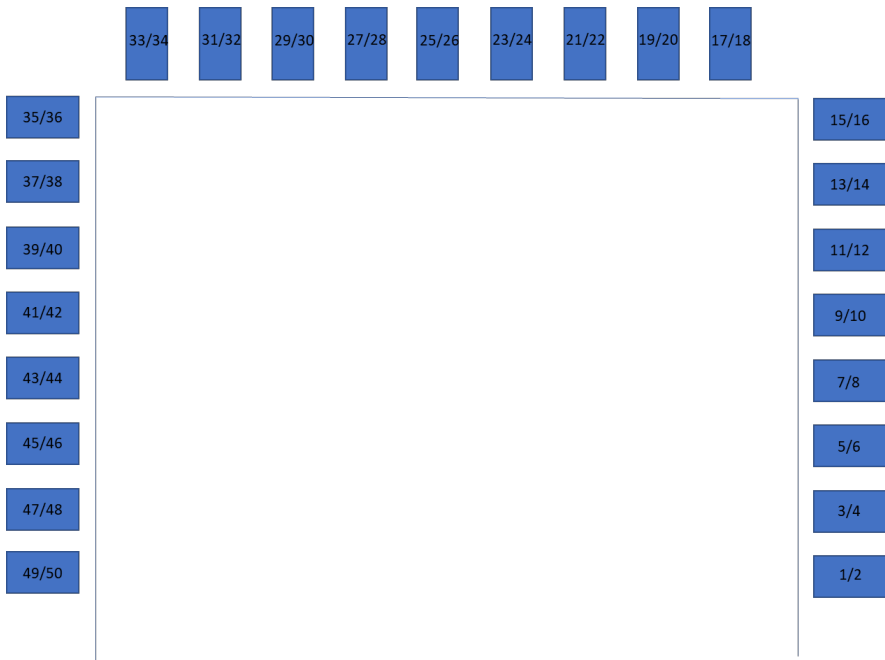
Poster Session I - November 16th, 11:45 - 12:45 **Fehler! Keine gültige Verknüpfung.**

Name	Number	Institute
Mauricio Andino Molina	1	IBIZ
Issam El-Debs	3	ATB
Martin Oettler	5	IfE
Charlie Fricke	7	IfI
Katerina Marcollova	9	ING
Viktoria Korff	11	ATB
Lars Teschke	13	IMVZ
Alexandra Blake	15	IMVZ
Franziska Schopf	17	INNT
Jana Kochmann	19	IfI
Jan Sluka	21	IMVZ
Sarah Jahn	23	IfE
Marie Weinberger	25	IfE
Lisa Bode	27	ITE
Markus Peschel	29	ITE
Judith Wedemeyer	31	IfE
Leonie Seemann	33	ITE
Luise Freier	35	IfI
Jule Brüssau	37	IfE
Deisy Johana Lancheros Buitrago	39	ING
Juan Pablo Fernández	41	ING
Annika Tietze	43	IITG
Benjamin Becker	45	IMP
Lena-Sophie Paul	47	IMP
Marwa Bassiouny	49	IBIZ

Poster Session II - November 16th, 14:45 - 15:45 **Fehler! Keine gültige Verknüpfung.**

Name	Number	Institute
Husnain Alvi	2	IfE
Juliane Lang	4	IMVZ
Anne Schwarzer	6	INNT
Kai Othmer	8	IFI
Anika Noldin	10	ITE
Mirette Eshak	12	IVD
Kristina Kissner	14	IMVZ
Katharina Padberg	16	ITE
Kira Wisnewski	18	IVD
Ole Pietsch	20	IVD
Christin Unruh	22	ITE
Felix Tetzlaff	24	IMP
Lea Boten	26	IMED
Susnato Das	28	ITT
Ronja Piesche	30	IVD
Josephine Friedrich	32	ATB
Isabel Suckau	34	IfE
Tamara Kozytska	36	IBIZ
Selina Fuchs	38	IBIZ
Nils Tadewaldt	40	IMED
Diana Palme	42	IMVZ
Tobias Britzke	44	ATB
Isabella Hrabal	46	INNT
Jasmin Deutschendorf	48	IMP
Joaquin Neumann	50	IITG/OH

Posters' Stands plan layout



Location: Förderverein Bären Lobeda e.V.

Date: 16th November, 2023

Set-up time: 08:30

Oral Presentations' Schedules

Talk Sessions I - November 15th, 17:15 - 18:15 **Fehler! Keine gültige Verknüpfung.**

Name	Institute
Margret Vonholdt-Wenker	ITT
Ebere Roseann Agusi	External
Christine Thomas	IBIZ
Christoph Gerloff	ITT

Talk Sessions II - November 16th, 09:45 - 10:45 **Fehler! Keine gültige Verknüpfung.**

Name	Institute
Maike Richter	IMP
Christin Hennig	IVD
Constantin Lorenz	IVD
Johanna Wiethoff	IITG/OH

Talk Sessions III - November 16th, 13:45 - 14:45 **Fehler! Keine gültige Verknüpfung.**

Name	Institute
Lea Lenhard	IFI
Elena zu Klampen	ING
Gregor Neufeld	ING
Benedikt Litz	IVD

Talk Sessions IV - November 16th, 17:00 - 18:00 **Fehler! Keine gültige Verknüpfung.**

Name	Institute
Md Rasheduzzaman	IMED
Richard Küchler	IMVZ
Sophie-Celine Weinert	IVD
Miles Winter	ITE

Talk Sessions V - November 17th, 09:30 - 10:00 **Fehler! Keine gültige Verknüpfung.**

Name	Institute
Erik Bannert	ITE
Patrick Gutjahr	IMED

Oral Presentations

Talk Session: I

Talk Nr. 1

Evaluating and comparing sustainability aspects of animal husbandry systems in Germany: the web application InKalkTier makes it possible

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In order to support the transition to sustainable livestock husbandry systems, a digital science-based information system that provides stakeholders with knowledge and insight into different aspects of sustainability is desirable. By comparing housing systems and visualizing their contribution to animal welfare, environmental emissions, and farm economy, actors can get informed about the main aspects of sustainability of current and future livestock housing systems to make informed decisions regarding future policies or investments. The InKalkTier (“Interaktives Kalkulations- und Informationssystem zu Tierwohl, Umweltwirkung und Ökonomie von zukunftsfähigen Tierhaltungsverfahren”) project creates such a comprehensive information system in the form of an online web-application, in which all relevant German animal housing systems can be evaluated and compared. A key aim is to assess animal welfare based on scientific knowledge, which requires transformation of qualitative data into quantitative data. To this end, we applied a method called semantic modelling that allows for a synthesis of scientific information and generates a weighted welfare score on a scale from 0 to 1 as output. The procedures of semantic modelling are based on logic and semantics of scientific information about technical elements of a housing system (e.g. floor type) and their effect on animal welfare. Accordingly, the input for the model consists of linguistically meaningful sections of scientific literature (i.e. quotes). Scientific literature on the three main production directions (i.e. fattening pigs, dairy cows, laying hens) was analyzed, which can be extended to other production directions in the future. The web-application will be launched end of 2023 (www.inkalktier.de).

Contact: Margret Vonholdt-Wenker (Margret.Vonholdt-Wenker@fli.de)

Talk Nr. 2

Investigation of reverse spill-over transmission at the at high risk human-animal interfaces during COVID-19 pandemic in Nigeria

E. Agusi¹, V. Allendorf², O. Asala¹, J. Schön³, I. Shittu¹, K. Dietze², D. Hoffmann³, A. Globig², C. Meseko¹

1 National Veterinary Research Institute Vom, Jos, Plateau State, Nigeria

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Human, domestic animals, as well as wildlife entanglement, can lead to zoonotic transmission events with known and unknown pathogens including sarbecoviruses. In late 2019, a new type of viral infection caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), most likely resulting from spill-over from animals, emerged and was globally documented as COVID-19 with a high rate of mortalities in humans.

To elucidate on the possible role of domestic animals and wildlife in the ecology and epidemiology of sarbecoviruses in Nigeria, and to analyze the possible circulation of other, undiscovered, but potentially zoonotic sarbecoviruses in animals, we tested 504 serum samples from dogs, rabbits, bats, and pangolins collected between December 2020 and April 2022. The samples were analyzed using an indirect multi-species enzyme-linked immunosorbent assay (ELISA) based on the receptor binding domain (RBD) of SARS-CoV and SARS-CoV -2, respectively. ELISA reactive sera were further analyzed by specific virus neutralization test and indirect immunofluorescence assay for confirmation of the presence of antibodies. In this study, we found SARS-CoV reactive antibodies in 16 (11.5%) dogs, 7 (2.97%) rabbits, 2 (7.7%) pangolins and SARS-CoV-2 reactive antibodies in 20 (13.4%) dogs, 6 (2.5%) rabbits and 2 (7.7%) pangolins, respectively. Interestingly, 2 (2.3%) bat samples were positive only for SARS-CoV RBD reactive antibodies. These serological findings of SARS-CoV and/or SARS-CoV-2 infections in both domestic animals and wildlife indicates exposure to sarbecoviruses and thus, reinforces the need for continuous surveillance for early detection of pathogens and intervention at the human-animal interface.

Contact: Ebere Agusi (roseebere8@gmail.com)

Talk Nr. 3

Implementation, validation, and application of a bioinformatics framework for analyzing Oxford Nanopore Technologies genome sequencing data from zoonotic bacterial pathogens

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Investigating bacterial genomes on nucleotide level requires whole-genome sequencing and bioinformatics analysis. Using bioinformatics tools often implies basic understanding of computer science. Therefore, automated bioinformatics pipelines enable researchers with little expertise in computer science to investigate their biological data sets, and ensure reproducibility. Regarding whole-genome sequencing, Oxford Nanopore Technologies (ONT) and IlluminaTM represent the main sequencing technologies in the field of bacterial genomics. Although ONT sequencing has a higher per-base error rate than Illumina sequencing, it has high potential to assemble bacterial genomes to closure due to producing significantly longer reads. For this work, a bioinformatics pipeline for analyzing ONT data is implemented, validated, and applied. The pipeline takes raw sequencing data and meta data as an input. The analysis includes basecalling, quality control, assembly, and polishing. Final results consist of high-quality genome assemblies, which can represent the basis for further analysis like genotyping or the detection of genetic markers. Since various bioinformatics tools are available for each step of the pipeline, benchmarking is performed during the implementation process. To achieve this, reference strains of bacterial pathogens are sequenced and the data is used to validate and optimize the best combination of tools to receive high quality bacterial genomes. Results show the utility of ONT sequencing in veterinary medicine for serotyping in *Salmonella* or genotyping of highly pathogenic zoonotic bacteria. Especially in plasmid reconstruction ONT sequencing has big advantages which is presented by a study on antimicrobial resistance in *Streptococcus uberis*.

Contact: Christine Thomas (Christine.Thomas@fli.de)

Talk Nr. 4

Automatic detection of disease using Neural Networks on Audio-Data

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Feather pecking and outbreaks of diseases continues to be the biggest problems in commercial turkey farming. Machine learning methods pose as a cost-effective technique to automatically detect agonistic feather-pecking and diseases. Computer vision is a rising field in Precision Life stock Farming. For this Video-data is often taken to be learned by an artificial intelligence model to then detect negative behavior in a flock of turkeys. Audio-Data on the other hand is rarely used even though the recording systems are relatively inexpensive and tend to be less vulnerable to dirt and dust in a barn. The Animals are also not hide from the audio recording. Therefore, this work seeks to investigate if there is a connection between vocalizations of turkeys in audio data and the presence of a disease or antagonistic pecking behavior. For this purpose, audio and video data have been recorded during turkey fattening. Spectrograms are generated from the audio data and the resulting data set is then used to train deep learning methods in order to detect the presence of feather pecking and health problems.

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Oral Presentations

Talk Session: II

Talk Nr. 5

Determining antimicrobial resistance prevalence in *Escherichia coli* from fattening pigs by sampling animal transports at a slaughterhouse

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Antimicrobial resistance surveillance data for livestock is aggregated at EU member state level and published with considerable delay, preventing timely and targeted interventions. Sampling animal transports at abattoirs with cross-regional catchment areas may allow up-to-date regional data collection with reasonable effort. This assumption was tested by sampling 1,022 vehicles carrying fattening pigs. One *E. coli* strain was isolated non-selectively per boot swab sample and its susceptibility to 14 antimicrobials determined using broth microdilution. Minimal inhibitory concentrations were obtained and isolates without (wild type) and with (non-wild type) phenotypically detectable acquired resistance were distinguished with ECOFFs from EUCAST.

Our results (n=992) show high (21.4% to 38.8%) non-wild type prevalence for antimicrobials grouped into category “D” (Prudence) from the EMA categorisation of antibiotics for prudent and responsible use in animals, which are intended for first-line treatment. Whereas antimicrobials, classified in categories “A” (Avoid), “B” (Restrict) and “C” (Caution), reveal only low (0.1% to 6.1%) prevalence. Regional analysis shows slight differences for some federal states and antibiotics.

Roughly one third (38%) of the isolates were completely susceptible, while approximately one quarter (24%) is resistant towards three or more antimicrobial classes. Selective screening of a subset (n=375) of our faecal samples for ESBL- and AmpC-producers unveils a prevalence of 86% in comparison to 2%, detected for the non-selective cultivation.

Resistance- and metadata will further serve to evaluate a ‘Lot Quality Assurance Sampling’ surveillance approach which classifies resistance prevalence as either above or below a threshold. This strategy requires less samples, thereby facilitating surveillance.

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Talk Nr. 6

Novel swine influenza reassortant viruses: Risk assessment of their zoonotic potential

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2 University Medical Center, Institute for Virology, Freiburg, Germany

Swine Influenza A viruses (swIAV) cause at global scale and seasonally independent respiratory disease invoking economic damage in the pig industry. Influenza A viruses are genetically highly flexible and may adapt rapidly by genetic drift/genetic shift to new (host) environments. Swine are known as the “mixing vessel” of IAV. Co-infections with IAVs of human, avian and swine origin in pigs can generate novel reassortant swIAVs, potentially bearing zoonotic and contingent pandemic potential.

Here we study the flow of swIAV across the swine-human interface in Germany. Nasal swab samples of pigs and of staff of swine holdings were collected to detect and characterize IAV, of swine as well as of human origin.

To date, in 122 of 214 swine holdings examined, an increased proportion of avian H1N1/H1N2 as well as pandemic H1N1 viruses has been documented. In contrast, no IAV could be found in 287 human samples obtained from the same holdings so far. However, during the time of this study a case of zoonotically transmitted swIAV in a man from Germany North Rhine-Westphalia confirms the topicality of the issue. Subsequently, by performing serological surveys to identify presence of cross-reactive antibodies in human and swine sera against circulating IAV and swIAV, possible gaps in the humoral immune response lines might be identified.

This study will give new insights into the frequency of zoonotic and reverse-zoonotic transmissions of swIAV and their impact at the swine-human interface

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Talk Nr. 7

Developing Novel Live Attenuated Vaccines for Foot-and-Mouth Disease

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Foot-and-mouth disease (FMD) is a highly contagious viral disease that affects cloven-hoofed animals. This disease has devastating consequences for animal health, welfare, and international trade. Existing inactivated vaccines for FMD work by preventing clinical disease and by a reduction of virus excretion. However, these vaccines also show several downsides, such as the need to administer them repeatedly via intramuscular injection and the inherent risks for the environment when inactivating large amounts of live virus. Their effectiveness is further limited by the presence of multiple serotypes, subtypes and ever-evolving antigenically divergent strains. In case of an outbreak all potentially infected animals - regardless of their vaccination status or infection state - are culled in order to quickly regain freedom from FMD transmission within affected regions. Live attenuated vaccines (LAV) are derived from virulent pathogens that have been modified to reduce their virulence while still eliciting a protective immune response. In our research, we consider different design possibilities that lead to virus attenuation, such as the deletion of virulence associated genes, codon deoptimization or chimeric viruses. Every vaccine candidate will be tested *in vitro* and *in vivo* using pig models to establish their efficacy and safety. These vaccines have the potential to provide long-term immunity, induce a robust immune response and confer protection against multiple serotypes. LAV, along with serological tests to differentiate vaccinated from infected animals, are an essential step in implementing “vaccinate-to-live” strategies and prevent the killing of non-infected animals.

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Talk Nr. 8

Cross-sectional study of zoonotic pathogen prevalence in cattle in Zanzibar, Tanzania

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Zanzibar, a semi-autonomous state within the United Republic of Tanzania, lies 50 km off the Tanzanian mainland in the Indian Ocean. Livestock is essential for its 2 million inhabitants, providing food, draught power and financial stability. A livestock census from 2017 identified chicken (7 million), cattle (90,000), and goats (48,000) as the primary livestock.

In December 2022, Zanzibar's Revolutionary Government launched its One Health Strategic Plan 2021-2025. It was followed by a multi-stakeholder workshop involving German and Tanzanian participants with the aim to create the road map for implementing the plan. The workshop highlighted Zanzibar's lack of case definitions and One Health laboratory capabilities to diagnose diseases impacting both humans and animals. Most diagnoses rely on clinical symptoms only.

To enhance the knowledge of zoonotic pathogens and associated health risks, this study focuses on selected pathogens in local cattle. The chosen pathogens - *Brucella* spp., *Coxiella burnetii*, Rift Valley Fever Virus, and *Leptospira* spp., - derive from a decision tree using workshop outcomes. Three districts with the highest cattle density on Unguja and Pemba Island were chosen as sample sites. A total of 731 blood samples from local and imported cattle were collected and analyzed using commercial ELISA kits. The results showed the highest seropositivity for Rift Valley Fever, followed by Q-Fever, Brucellosis, and Leptospirosis. The study may extend its methods to include PCR, serotyping, or sequencing analyses. Additionally, a semi-quantitative survey questionnaire aims to investigate the knowledge, attitudes, and practices of livestock keepers regarding the risk of zoonotic infections.

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Oral Presentations

Talk Session: III

Talk Nr. 9

Comparative analysis of immune responses and permissiveness to Cedar virus infection using multi-species approaches

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Nipah and Hendra virus are highly pathogenic zoonotic viruses that cause severe disease in humans and various livestock species. Recently numerous novel Henipaviruses with the potential of human spillover have been identified in Madagascar, South Korea and China. Cedar virus (CedPV) was first isolated from fruit bat urine samples in Australia. Later it was discovered to be non-pathogenic to humans and categorized as a BSL2 pathogen. Considering its high similarities to other highly pathogenic Henipaviruses, it provides the unique opportunity to study the genus under low biosafety conditions. It has been established that CedPV employs Ephrins A2, A5, B1 and B2 for its entry into human cells via attachment and fusion mediated by glycoprotein (G) and fusion protein (F). In order to elucidate the mechanism by which CedPV enters Roussetts cells, CHO-K1 cells expressing individual bat Ephrins have been generated and recombinant CedPV entry into each Ephrin expressing cells has been evaluated via immunofluorescence and high throughput microscopy. Our observations suggest that the expression of bat Ephrin B1 or B2 allows the entry of CedPV. This will be further validated in specific Ephrin knockdown cells generated by a lentiviral shRNA method. Furthermore, IFN signaling, proinflammatory gene expression and cellular stress responses upon CedPV infection will be assessed. In all, this project aims to elucidate the pathogenic potential of CedPV and uncover the mechanism how CedPV enters different host cells and manipulates innate immune responses.

Contact: Lea Lenhard (Lea.Lenhard@fli.de)

Talk Nr. 10

Single Cell Sequencing reveals transcriptional response to TGFB1 and MFGE8 treatment in equine endometrial fibroblasts

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Fibrotic degenerations of the endometrium in mares are a major cause of subfertility, especially in older animals. Although fibrotic changes can affect various organs of the body and often lead to incurable chronic diseases, the pathomechanisms are not yet fully understood. Fibrotic conditions are caused by an imbalance in stromal tissue architecture maintenance. Fibroblasts and their transition towards a more active and contractile phenotype called myofibroblast, play a central role in disease development. Transforming Growth Factor Beta 1 (TGFB1) is a prominent pro-fibrotic factor in this context. In arteriosclerosis MFGE8 seems to be a key player in the pathogenesis of fibrotic changes.

The effect of TGFB1 treatment on fibroblasts gene expression for individual genes can be studied by reverse transcriptase quantitative PCR. By this approach one will potentially overlook thousands of gene expression changes and ignore the fact that fibroblasts are a heterogeneous cell type being descended from different precursor cells. Innovative approaches such as Single Cell Sequencing address this problem and allow parallel investigation of the entire transcriptome and comparison of treatment effects on different fibroblast subpopulations. We have used this technology to study the effects of a TGFB1 and MFGE8 treatment on endometrial fibroblasts from four different mares cultivated in vitro on hydrogels. We found a tremendous effect of TGFB1 treatment on the various fibroblast cell population gene expression, while the effects of MFGE8 were more subtle.

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Talk Nr. 11

Dissecting the bovine peri-implantation embryo development through single cell sequencing

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The bovine peri-implantation period is important but relatively unknown in embryological development. Single-cell RNA sequencing (scRNAseq) allows us to determine the transcriptome of individual cells in high throughput and to reveal the heterogeneity of biological samples. This technology can help us gain more insight into this embryological time point. Therefore, scRNAseq was applied to day-17 bovine embryos obtained by flushing the uterus after slaughter of the animal. So far, three embryos derived from artificial insemination have been analyzed using scRNAseq (10X Genomics technology). Results show cell clusters from two major developmental layers identified by gene expression of marker genes: the cells originating from the inner cell mass (ICM) and the cells originating from the trophectoderm. The ICM-derived cells were assigned to different developmental cell types such as epiblast, mesoderm, and endoderm and were compared to a day-14-old human gastrulating embryo, showing striking similarities of this relatively conserved gastrulation process in mammals. The cells derived from trophectoderm could be assigned to two groups based on IFNT expression. The IFNT-positive group corresponds to trophoblast uninuclear cells, representing approx. 80% of the total trophoblast cell population. The IFNT-negative group comprises 1) trophoblast giant cells recognizable by the expression of specific marker genes such as pregnancy-associated glycoprotein and the family of prolactin genes. 2) one still unidentified cell population that expresses marker genes like e.g. WFDC2, and predicted transcription factors like e.g. NR3C1 or FOXO1. Analysis of cell-to-cell communication revealed a plethora of interactions between the different clusters of trophoblast cells.

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Talk Nr. 12

Leaderless FMDV is fully attenuated in vivo and unable to establish persistent infection

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3 Animal Health Laboratory, Foot-and-mouth disease reference laboratory, Virology JRU, ANSES, INRAE, ENVA, 13 Paris-Est University, Maisons-Alfort, France

Many ways of attenuation of foot-and-mouth disease virus (FMDV) have been explored. One successful method is the deletion of most of the coding sequence for the Leader proteinase Lpro. Lpro inhibits host cell mRNA translation and blocks the interferon response which promotes viral replication. Therefore, leaderless FMDV can replicate in vitro but is strongly attenuated in vivo. The ability of a leaderless variant of FMDV to establish persistent infection after simulated natural infection remains unexplored. We created a variant of FMDV O/FRA/1/2001 lacking Lpro and exposed cattle by intranasopharyngeal inoculation. The animals were observed for 35 days after exposure to determine if leaderless FMDV could establish a persistent infection in epithelia of the nasopharynx, similar to what is seen after wildtype FMDV infection. Despite its ability to replicate well in various cell lines, the leaderless virus was unable to cause an acute infection characterized by vesicular lesions and viral shedding nor establish persistent infection in pharyngeal tissues.

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Oral Presentations

Talk Session: IV

Talk Nr. 13

Establishment of a methodological pipeline for the characterization of the virome of different mosquito species

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Different mosquito species have different vector competence to pathogens. Previous studies showed the importance of mosquitoes as vectors of arthropod-borne viruses (arboviruses) and zoonotic disease-causing viruses like DENV, ZIKV, CHIKV. Also, mosquitoes have their own insect-specific viruses (ISVs) which may interact with arboviruses, possibly altering vector competence. To date, there are very few conclusive and systematic studies on how mosquito virome contribute to vector competence or disease transmission. Here, we are seeking to obtain the virome sequencing data of all available mosquito species from our in-house insectarium--namely *Aedes albopictus*, *Aedes aegypti*, *Aedes vexans*, *Culex pipiens pipiens* and a hybrid species (*Culex pipiens pipiens* × *Culex pipiens molestus*)--in that direction. The main idea of this study is to examine the mosquito virome using Oxford Nanopore Sequencing Technology, as it does not depend on biased prerequisite of preamplification or growing viruses in the cell culture. Nevertheless, established NGS methods may serve as a comparison. We are constructing a full pipeline to study the mosquito virome qualitatively and quantitatively. Therefore, the strategy will handle the problems from sample preparation to the virus identification in an integrative approach. The findings of this study will need to be validated in future transmission studies in the lab. The Bioinformatics Pipeline will be freely available as an executable, and extendable snakemake file.

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Talk Nr. 14

Establishing a Phosphoproteomics Workflow for the Systematic Analysis of Virus-Induced Phosphorylation

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Virus-host interactions are essential for the viral replication cycle, facilitating the cellular reprogramming of the host cell in favour of virus replication. The underlying mechanisms are highly complex including multiple targets and require a very tight spatial and temporal regulation. Many regulatory mechanisms in cells are implemented by phosphorylation and dephosphorylation of proteins. The precise control of kinases and phosphatases allows fast and reversible phosphorylation of various target proteins at defined phosphorylation sites. In infected cells, virus-encoded kinases are important players in this highly dynamic process. Systematic and detailed analysis of protein phosphorylation on a proteome-wide scale requires a dedicated approach for sample preparation and data analysis known as 'phosphoproteomics'. Here, we present a phosphoproteomics pipeline optimized for the analysis of virus-infected cells. As a proof of concept, we applied the workflow to compare the phosphoproteomes of cultured swine cells after infection with Pseudorabiesvirus (PrV), a herpesvirus of swine, and a kinase negative PrV-mutant. Technically, this strategy is applicable to any virus-infected cell culture of interest.

Contact: Richard Küchler (Richard.Kuechler@fli.de)

Talk Nr. 15

Isolation attempts of neurotropic rustrela virus

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Until recently, rubella virus (RuV; Rubivirus rubellae), exclusively infecting humans, was the only known virus of the genus Rubivirus (family: Matonaviridae). In 2020, two relatives of RuV were discovered in Africa and Europe: ruhugu virus (RuhV; Rubivirus ruteetense) found in oral swabs of presumably healthy bats, and rustrela virus (RusV; Rubivirus strelense) detected in brain tissue from diverse neurologically diseased mammals at several zoos in northern Germany. Recently, it was also published that the so-called staggering disease in cats is most likely caused by RusV. However, the zoonotic potential of those viruses remains unclear. Unfortunately, several attempts to isolate RusV, including inoculation of diverse cell lines with homogenized RusV-positive brain material or co-culturing cells derived from experimentally infected RusV-positive animals with a number of neuronal and non-neuronal cells were unsuccessful. Ongoing attempts for rescue and isolation of RusV include lipofection as well as electroporation of RusV RNA obtained from the above-mentioned brain material. Furthermore, it is aimed to infect cerebral organoids and brain slices of rodents with RusV. This will allow further characterization of viral tropism and molecular pathogenesis and provide important insights into this newly discovered neurotropic RNA virus and its potential for zoonotic transmission.

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Talk Nr. 16

Processing feces for downstream analyses of different non-invasive biomarkers

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Under the aspect of animal experiments and the increased need to reduce them, non-invasive biomarkers have gained elevated importance over the last years to monitor animal health. Noninvasively obtained matrices like feces have been shown to exhibit biomarkers indicating inflammatory processes.

Therefore, this project currently focuses on cow feces and the following biomarkers that are linked to inflammatory processes and thus potentially monitor animal health. Cortisol, a universal stress hormone with anti-inflammatory effects and Calprotectin, a protein secreted by neutrophilic granulocytes are both measured via ELISA assays. Cortisol is one of the main glucocorticoids secreted from the hypothalamic-pituitary-adrenal axis. Calprotectin detected in feces indicates the migration of neutrophils to the intestinal mucosa, which is a sign of intestinal inflammation. Lastly, miRNAs - small non-coding RNAs (20-22 nucleotides) - are involved in post-transcriptional gene regulation and are potentially linked to the specific immune response of cows to e.g. paratuberculosis (Johne's Disease).

Here, methods of sampling, processing and molecular downstream analyses of the cow feces are shown. The protein and RNA extractions are sensitive to many factors and thus different starting materials from fresh, to frozen and lyophilized feces are tested. The goal is to find the best way to extract these biomarkers to depict animal health. Therefore, the challenge between the best method on the one hand and the best practicality on the other hand is discussed with regard to the potential use of these biomarkers in common practice.

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Oral Presentations

Talk Session: V

Talk Nr. 17

Interrelations between lameness and water intake behavior of lactating dairy cows

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Lactating cows have a high water demand and their intake can rise, depending on ambient temperature and milking yield, up to 200 L per day. To reach this daily water intake 5 - 25 drinking bouts per day as single visits to a water trough are necessary. Lameness is a highly prevalent production disease in modern dairy production systems. Studies have shown a decreased dry matter intake and altered lying and standing behavior in lame versus sound cows. We therefore hypothesized, that lameness may influence the water intake behavior of lactating dairy cows.

To investigate this hypothesis data from an ongoing study, including 48 lactating German Holstein cows in a total confinement system (cubicles, slatted flooring), was used. Throughout 18 weeks the animals were fed with different feeding regimes according to GfE energy and nutrient supplies recommendations in 3 periods. Lameness scoring was conducted weekly (Flower and Weary, 2006). The daily drinking bout duration, water intake and activity were determined using the smaXtec Boli (smaXtec animal care GmbH, Graz, Austria) and the RIC weighing trough System (Insentec BV, Marknesse, The Netherlands).

For water intake, drinking bout duration and activity a significant interaction between lameness and period was observed. The clearest effect was observed regarding the drinking bout duration, which was significantly shorter in the lame cows (lame: 1064 sec; not lame: 1240 sec) in the period with the highest temperature-humidity-index (P1: 50,7; P2: 58,4; P3: 66,8).

Contact: Erik Bannert (Erik.Bannert@fli.de)

Talk Nr. 18

Mosquitoes and peatland rewetting: An inconvenient nuisance or potentially harmful?

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Recently, peatland ecosystems underwent a significant image change. For centuries, draining them to access the land for human development was seen as achievement of civilisation. Today however, rewetting peatlands is considered an effective method to mitigate climate change and biodiversity loss. Nevertheless, effective peatland management involves potential mosquito breeding sites yet little is known about the culicid fauna in these habitats. The spread of diseases like Dengue, Chikungunya or West-Nile-Fever to formerly unaffected parts of the world however indicates that wetland restoration combined with the influence of climate change on the spatial and temporal distribution of mosquitoes and mosquito-borne pathogens may present a health risk to the vicinity of such ecosystems.

To unravel the underlying ecological principles and environmental conditions of mosquitoes in peatlands, we monitor them in a partially rewetted fen and adjacent areas in North-eastern Germany. Preliminary results after the first of three seasons have revealed almost half of the known German mosquito inventory in a comparatively small area including vectors of aforementioned pathogens. To detect this diversity, we applied a combination of sampling methods beyond the scope of regular monitoring that enabled us to get rapid, in-depth insight into the ecology of the regional mosquito community. The data may hint towards potential shortcomings of previous monitoring efforts but also towards previously unknown ecological connections. Further ecological, genetic and virological analysis will enable us to connect these culicid species with their respective niches and habitats, identify their vector capacity and ultimately increase the safety of peatland restoration.

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Poster Presentations

Session: I

Poster Nr. 1

Clostridioides difficile in Honduras, Central America: an updated genomic and phenotypic characterization

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Clostridioides difficile is an anaerobic enteropathogen of noted clinical importance in hospital and community settings. Its ubiquitous presence in pets, animals, food products and the environment -together with its ability to form spores- favor its survival and dissemination. Some strains with possible zoonotic implications have been described over the years, and its dynamic is still under research. *C. difficile* poses a threat especially for elderly and immunosuppressed populations. In Central America, hypervirulent and multidrug-resistant genotypes have been previously reported. Thirty-one isolates from patients in two major hospitals in Tegucigalpa, Honduras were characterized using whole genome sequencing (WGS) and phenotypic antimicrobial susceptibility testing (AST). Two toxigenic PCR-ribotypes RT027 (ST1) and RT002 (ST8) were detected. All RT027/ST1 isolates (n=29) were found to be resistant to moxifloxacin, tetracycline, and linezolid, whereas RT002/ST8 isolates (n=2) were susceptible. This correlates with the presence of certain genetic elements associated with antimicrobial resistance in the analyzed strains. Worrisomely, core genome MLST cluster analysis shows a close relationship between the RT027/ST1 isolates, suggesting an ongoing outbreak of multidrug-resistant *C. difficile* in both hospitals with unknown sanitary and economic implications. This emphasizes the need for a One Health research approach to develop intervention measures for control and prevention.

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Poster Nr. 3

Establishment of mouse cerebral organoids as 3D infection model for neurotropic viruses

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Organoids are in great demand for research to further reduce animal experimentation. Mice are popular animal models to study neurotropic viruses such as pseudorabiesvirus (PrV) and to analyze the effects of viral proteins on neurovirulence and neuroinvasion in the brain. A suitable organoid model for equivalent fundamental investigations should assist in replacing animal testing. Organoids are self-organizing 3D aggregates generated either from embryonic or pluripotent stem cells that resemble the cytoarchitecture and cell types found *in vivo*. Although murine brain organoids mimic biochemical and physiological tissue development and function, the short cultivation time so far allows only the development of immature embryonic organoids. To generate mature organoids, the cultivation time was extended considerably by the implementation and improvement of distinct external conditions to secure adequate oxygen and nutrient supply. Once mature organoids are developed, they will be infected with wild-type and mutant PrV and morphological and functional characteristics after infection will be studied and compared to uninfected organoids. In addition, the infected murine organoids will be compared with brain samples from PrV-infected mice from previous animal trials. Different viral mutants will be studied in the organoid model and readouts will be identified to evaluate the impact of viral proteins on neurotropic properties. Organoids will be analyzed using conventional histopathology as well as modern imaging techniques such as light sheet microscopy and confocal imaging. Finally, the organoid model will be evaluated as a suitable replacement for animal experiments.

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Poster Nr. 5

Biosafety aspects of wood surfaces in livestock stable construction

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Wood is a traditional construction material in animal husbandry, but in recent years it has become less important compared to more modern materials. One reason for this could be the negative perception of the hygienic properties of wooden surfaces. Various guidelines demand a surface that is easy to clean and disinfect. Whether wood, as a porous material, meets these requirements is often questioned. To assess this, disinfectant tests were conducted on different construction timbers to evaluate the biosecurity of wood as a construction material.

The virucidal disinfectant tests were based on the national test guidelines for animal husbandry of the DVG and also current European standards. Five different types of wood carriers were contaminated with enveloped or non-enveloped viruses and subsequently treated with different disinfectants. In order to determine the effectiveness of the disinfectants, the experiments were subsequently evaluated quantitatively. Here, germ reduction by at least four decadal logarithmic levels was considered to be an effective disinfection.

The results show that an effective disinfection of the wooden surfaces is possible under laboratory conditions and that the concentrations required for this are below the recommended concentrations of the FLI list for surface disinfectants. For the tests with non-enveloped viruses, no significant differences were observed between the respective disinfectants on different wood surfaces, but significant differences were found for the enveloped viruses.

The use of wood as a traditional construction material in livestock buildings can contribute to a more sustainable economy and climate protection. However, aspects of animal hygiene and animal disease control must not be ignored. The presented investigations are intended to test the compatibility of wood with the aforementioned requirements.

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Poster Nr. 7

Early distinct immune responses against different SARS-CoV-2 variants in hACE2 transgenic mice

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Early immune responses to SARS-CoV-2 determine the progression and severity of COVID-19. Typically, immune events in humans are measured via blood or post-mortem tissue sampling, which does not fully reflect the early host response at the infection site. Assessing mucosal immunity in humans after natural infection is cumbersome, making it challenging to understand early immune responses to infection. To overcome these obstacles, we analyzed early immune responses to various SARS-CoV-2 variants in K18-hACE2 mice, which serve as an animal model for severe COVID-19. The mice were intranasally infected with the ancestral SARS-CoV-2, Beta or Delta virus variants, and tissue viral loads, clinical symptoms, and immune responses in the lungs and spleen were monitored over time. Infection led to differences in lung viral loads, inflammation, and immune cell accumulation between virus variants. Beta viral loads increased rapidly in the lungs, induced high levels of cytokines and chemokines, and increased pulmonary frequencies of monocytes and macrophages. One-week post-infection, lung CD4⁺ and CD8⁺ T cells upregulated activation, differentiation, or homing markers, such as CXCR3, CD103, CD69, and CD44. Virus-neutralizing antibodies and S-peptide-specific T cell responses developed regardless of the virus variant and were detected earliest 7 days post-infection. Depletion of lymphocytes led to increased viral burden in the respiratory organs. Our data unveil early immune responses to different SARS-CoV-2 variants, enrich knowledge about COVID-19 pathogenesis, and could help develop pan-variant countermeasures.

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Poster Nr. 9

Role of zinc in ejaculated boar sperm cells

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Although Antoni van Leeuwenhoek observed swishing sperm cells for the first time almost 400 years ago, a great deal of their intracellular ion regulation still remains veiled. Zinc has been implicated in playing an essential role in sperm physiological events.

The zinc ion (Zn²⁺) is generally highly abundant in male reproductive tissues. However, well-balanced and dose dependant zinc concentration seems essential for porcine sperm cells' proper functionality. Primarily is this role demonstrated in correct nucleus formation, adequate tail stiffness and correct mitochondrial membrane potential level for energy supply.

We observed by flow cytometry that addition of ZnCl₂ (0.01 mM to 2 mM) to zinc-free media readily results in a concentration-dependant Zn²⁺ uptake in viable sperm. Moreover, 1 mM ZnCl₂ was sufficient to raise free intracellular Zn²⁺ and thereby trigger acrosomal destabilisation in viable sperm cells. Mitochondrial membrane potential remained unaffected under these conditions. On the contrary, the removal of free intracellular zinc ions with the chelator TPEN resulted in a dose-dependent (10 nM to 1 μM) collapse of the mitochondrial transmembrane potential in viable sperm without any effect on acrosome integrity. Interestingly, addition of zinc did not restore the mitochondrial membrane potential, but resulted again in acrosome destabilization.

In conclusion, zinc has its undisputed role in physiological processes that are essential for sperm function.

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Poster Nr. 11

Why infect alphaherpesviruses only very specific regions in the brain?

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The human Herpes Simplex Virus 1 (HSV-1) and the swine pathogen Pseudorabies virus (PrV) are neurotropic alphaherpesviruses able to invade the central nervous system (CNS) which can lead to fatal encephalitis in humans and animals. Human Herpes simplex encephalitis (HSE) caused by HSV-1 virus infection is mainly restricted to the temporal lobe of the cerebral cortex. However, details of the neuronal invasion route are largely unknown.

A mouse model for HSE using a PrV mutant was established recently (Sehl et al., 2020). Mice infected with this PrV mutant showed striking analogies to HSE in humans including the characteristic viral distribution and inflammation in the temporal lobe making this model promising to uncover the specificity of the invasion route. To answer the question “How does the virus get there?” we use different approaches. Kinetic studies with labeled virus are used to target ports of entry of the virus into the CNS and to further track the invasion pathway. Stereotactic brain injection is done to follow neuronal projections and the susceptibility of different parts of the brain to viral infection. Animal experiments are complemented by infection of primary murine neuronal cells derived from different parts of the brain identifying the differences in susceptible versus refractory cells.

Preliminary results indicate that cerebellar neurons from CD1 mice appear to be less susceptible to PrV infection than cortical neurons, confirming virus invasion restricted to the cerebrum *in vivo*. Together, we aim to use these studies to identify the underlying mechanism for the strict tropism of alphaherpesviruses to the temporal lobe.

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Poster Nr. 13

Towards greater climate change resistance in crops: Identifying novel epigenetic regulators in plants

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Knowledge of epigenetic and epitranscriptomic regulation in non-model plants is limited. In order for agriculture to benefit from emerging epigenetic manipulation technologies such as targeted methylation, a better understanding of epigenetic regulation in crops plants is required. This can, for example, improve selective breeding approaches, hence generating plants with greater resistance to effects of climate change. To this end, we are applying an unbiased mass spectrometry approach to identify epigenetic regulators in 20 economically relevant species. After completion of the data set, an evolutionary comparison of the identified regulators should help us to decipher conserved epigenetic regulation across the plant phylum. We then will assemble the collected data in a publicly available data repository.

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Poster Nr. 15

Characterizing telomere-binding proteins in *Caenorhabditis elegans*

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Telomeres protect the ends of linear eukaryotic chromosomes in part by preventing inappropriate DNA damage responses, chromosome end fusions and progressive shortening of the genome. They are able to do so by engaging with telomere-binding proteins and other associated proteins which then carry out specialized functions. In humans, a protective protein structure associated with telomeres, termed Shelterin, has been described. Though some telomere-binding proteins have been described in *C. elegans*, no similar structure has been deduced for this species. With a combination of proteomic, molecular and computational approaches we aim to better characterize telomere-binding proteins in *C. elegans*.

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Poster Nr. 17

Comparative pathology of natural flavivirus infections in various wild and zoo bird species

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The National Reference Laboratory (NRL) for West Nile virus (WNV) infections at the FLI has documented the gradual expansion of this notifiable disease throughout Germany ever since its first introduction in 2018. Hundreds of samples, mainly from wild and zoo birds, but occasionally also from horses, have been submitted to the NRL by the veterinary federal state laboratories of Germany for confirmation of WNV infection and exclusion of the closely related Usutu virus (USUV). While horses and humans serve as dead-end hosts, wild and captive birds are often the reservoir hosts in the enzootic transmission cycles of both of these flaviviruses. Mosquitoes of the genus *Culex* spp. are the most common vectors.

A large number of the avian WNV cases submitted to the NRL were examined pathologically at the veterinary state laboratories or other facilities. Additionally, several USUV positive birds from the years 2021 and 2022 were necropsied directly at the FLI. We intend to combine the existing virologic and pathological findings with the results of future immunohistochemical testing of formalin-fixed paraffin-embedded (FFPE) tissue. In some cases, clinical reports of the diseased birds before death are available as well. This rich collection of data from natural flavivirus infections of a wide variety of bird species can provide further insight into viral tissue distribution, infection pathogenesis and species-dependent susceptibility. We thank the German veterinary federal state laboratories and our clinical cooperation partners for providing their FFPE samples, pathological findings and clinical reports, respectively.

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Poster Nr. 19

Evaluation of the protective capacities of vaccine-induced mucosal tissue resident lymphocytes in different vaccine concepts

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Mucosal vaccine delivery can induce lung resident memory lymphocytes which have been shown to contribute to protection against airborne pathogens such as Influenza. While systemic vaccine-induced specific lymphocytes can be recruited to the mucosal sites upon infection, local tissue resident lymphocytes might provide faster immune responses. However, it is not fully understood, which specific mucosal vaccine concept induces superior mucosal resident memory B cell (BRM) and T cell (TRM) compared to systemic vaccination.

Previous animal studies proved that using a Live Attenuated Vaccine (LAV) based on the One-to-Stop Codon modification is safe and protects against challenge with SARS-CoV-2. Here we characterize the quality and quantity of SARS-CoV-2 specific mucosal tissue resident T and B cells in K18-Human ACE2 mice and Syrian Gold Hamsters. We vaccinate these rodent models for SARS-CoV-2 with different experimental LAVs compared to non-replicating vaccine platforms, and different routes of vaccination.

We established protocols to monitor immune responses in both rodent models by Flow cytometry for SARS-CoV2 reactive T and B cells, SARS-CoV-2 specific IgG and IgA ELISA, as well as bulkRNA sequencing and scRNA sequencing. Further, we also established qPCR panels for early innate cytokine profiles in the Syrian Gold Hamster.

With these tools in place, we currently aim to fill substantial knowledge gaps about the underlying innate and adaptive immune mechanisms that establish mucosal immunity against SARS-CoV-2.

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Poster Nr. 21

Characterization of telomere binding proteins in *C. elegans*

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The aim of this study is to elucidate the role of proteins interacting with telomeric sequences in the model organism *C. elegans*. Telomeres, located at the end of chromosomes, are surrounded by specialized proteins that protect them from DNA damage and control the homeostasis of telomere length. In the nematode *C. elegans*, our current knowledge of the proteins that interact with the telomeric sequence is incomplete. In our previous study, DVE-1 and LIN-40 were identified as interactors with telomeric nucleotide sequences by quantitative mass spectrometry. Both proteins are involved in chromatin remodeling and display a lethal knock-out phenotype, indicating their essential role in *C. elegans* viability and fertility. To date, these proteins have not been described in the context of telomeres. Our goal is to gain a better understanding of their functions and interactions on telomeres by using state-of-the-art techniques such as quantitative mass spectrometry, next generation sequencing, and high-resolution confocal fluorescence microscopy.

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Poster Nr. 23

AI for Animal Health - Application of Computer Vision Methods for Monitoring in Livestock Farming

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Changes in the behavior and activity of animals can indicate discomfort or health issues. By identifying these changes early and taking countermeasures, both veterinary costs and the use of medications can be reduced or even entirely avoided. Activity behavior, including standing and lying phases, is often used as an indicator in cows. With the advancement of digitalization, various wearable sensors for activity measurement have been developed in different forms. Depending on the design, these sensors can be attached to the ear, collar, leg, or even ingested as a rumen bolus. These sensors can be uncomfortable or disruptive for the cow. Moreover, data collection is temporally dependent on battery life, and there is a possibility of loss or damage. Video surveillance is neither dependent on a battery nor does it disturb the cow in any way. Additionally, one camera can monitor multiple cows simultaneously. In this study AI for animal health, specifically through the application of computer vision methods in livestock farming, is using. By establishing reference values for the behavior and activity of healthy animals, the assessment of the health and welfare of the animals can be facilitated. To detect various behaviors and activity levels, Deep Learning techniques will be employed. These methods will analyze video footage to detect deviations from the norm and classify them. Ultimately, this will enable a largely autonomous and continuous monitoring of animal health and welfare

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Poster Nr. 25

Antibiotics minimization in broiler farming through hygiene measures and optimization of biosafety, animal health and management

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Effective biosecurity is considered the foundation of all disease control programs and therefore essential in animal production, including poultry farming. Biosecurity and proper hygiene management have been shown to have a positive impact on production numbers, use of antibiotics, and animal health. Therefore, we will investigate the impact of good biosecurity management on antibiotics minimization in broiler farming. For this purpose, conventional farms, as well as farms using slow-growing genetics, are first divided into two categories, those with relatively high and those with low use of antibiotics. Subsequently, stable- and management-specific parameters are recorded and evaluated, applying the so-called AI risk traffic light (University of Vechta) and the AI checklist (Friedrich-Loeffler-Institute), which were implemented to prevent the introduction and spread of avian influenza in poultry farms. Additionally, we will attempt to adapt these tools to the project and figure potential general improvements. This is followed by the implementation of measures to remedy the identified risk factors. A reinvestigation and evaluation will then be conducted to determine whether the implementation of the measures has been successful. Ultimately, the results and findings will be transferred into practice in the form of workshops, training sessions, lectures and the creation of training materials. The objectives of the project are to improve the biosecurity of broiler chicken farms, the overall hygiene and the animal health. But also, to minimize the use of antibiotics, transfer knowledge between broiler chicken farms with low and high antibiotic use, and optimize risk management based on knowledge transfer.

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Poster Nr. 27

Effects of per- and polyfluoroalkyl substances (PFAS) on animal health

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Per- and Polyfluoroalkyl substances (PFAS) are used in everyday objects mainly as surfactant or surface protection products. This group comprises more than 4700 different substances. However, their desired characteristics for industrial applications also result in environmental persistence and bioaccumulation. Numerous epidemiological studies and rodent experiments showed that PFAS have adverse effects on human and animal health such as e.g. reduced effectiveness of vaccinations, higher cholesterol levels, damage to the liver or increased risk of cancer. Farm animals may also be exposed to PFAS by contaminated environment (e.g. pastures), feed or water. Therefore, the aim of the project is to determine the effects of PFAS on health parameters of farm animals.

In vitro studies on the cytotoxicity on bovine peripheral blood mononuclear cells (PBMCs) as well as two feeding trials with dairy goats (N=10) and fattening sheep lambs (N=16) are carried out in cooperation with the German Federal Institute for Risk Assessment (BfR) and the Max Rubner-Institute (MRI). The dairy goats receive PFAS via contaminated hay. The lambs receive a defined mixture of PFAS standard substances with syrup to ensure voluntary oral intake. Health related parameters will be analyzed frequently in blood samples during the trial and in tissue samples obtained at the end of the trial.

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Poster Nr. 29

Preliminary data on the human-edible feed conversion efficiency (HEFCR) of lactating dairy cows with varying concentrate feed portions (CFP) in the ration

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The decreasing availability of agricultural area results in an increasing uncertainty of sufficient food supply for a growing world population. Due to these planetary boundaries, the feeding of human-edible (HE) biomass to livestock has to be discussed. Therefore, an experiment was conducted with dairy cows fed rations varying both in feed composition and CFP over several periods to investigate the conversion of HE feed components into milk. Within the periods, the cows were allocated into 4 groups according to a 2x2 factorial design. The CFP was set at 30% (C30) and 55% (C55). Concentrates consisted of HE components (wheat, barley, rye), by-products (rapeseed meal, beet pulp) and a premix. For the evaluation of the HEFCR data from the literature were used, which included 3 different scenarios for the assumption of the HE fraction of the feed components. To calculate HEFCR, HE output via milk was divided by HE input via the summed single components of the ration. The HE fraction of the rations ranged from 17.2% to 46.7%, and for the respective concentrates it ranged from 35% to 66%. Depending on the scenarios, the mean HEFCR of the C30-groups were 1.16, 0.81 and 0.59, in the corresponding C55-groups 0.80, 0.56 and 0.43, respectively (HEFCR >1 corresponds to a gain of food and HEFCR <1 to a loss). In conclusion, the CFP had a decisive influence on HEFCR and feeding of high CFP in conjunction with a high proportion of HE components should be avoided in future.

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Poster Nr. 31

Using metagenomic analyses to characterize the resistome in different animal species

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Antibiotic resistance is a well-known threat in human and veterinary medicine. Metagenomic sequencing can be used to analyze genes which are coding for antibiotic resistance in the microbiome of animals (resistome). This way, the resistance situation of a farm can be monitored and possible dissemination routes can be identified in order to derive adapted intervention measures in the future. In this project, metagenomic analyses are carried out in different animal species. For resistome analyses of horses as well as cattle, samples from different farms in Mecklenburg-Western Pomerania were collected and metagenomic sequencing was performed. Different tools and pipelines are used for the sequence analysis and the results are compared. The goal is to create a protocol with which metagenomic data from animals can be effectively evaluated.

In the further course of the project, these methods will be applied to study the resistome of a pig herd. In a longitudinal study, faecal samples were regularly taken at pen level in the pig herd over a period of one and a half years. Part of the animals are kept according to organic standards and have access to a free-range area, while the other part is kept according to conventional standards. Metagenomic sequencing will also be carried out and the methods developed for sequence analysis will be used to analyze differences in the resistome structure between conventional and organic husbandry as well as seasonal influences.

This contribution presents the methodological procedure and first results of the analysis of the horse and cattle data and gives an outlook on the resistome analysis of the pig herd.

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Poster Nr. 33

Effects of dietary L-carnitine supplementation on blood parameters of mid-lactating dairy cows during standardized lipopolysaccharide-induced inflammation

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Dairy cows are exposed to a variety of pathogens throughout their lives, and a well-functioning immune system is important to prevent performance losses and maintain cow health. L-carnitine, which is essential for the beta-oxidation of free fatty acids in the mitochondria, also provides energy for immune cells and could therefore have an impact on leukocytes and protein metabolism. To test this hypothesis, German Holstein cows were assigned to a control group (CON, n = 26) and an L-carnitine supplemented group (CAR, n = 27, 25 g rumen-protected L-carnitine/cow/d, with concentrate) and received an intravenous bolus injection of lipopolysaccharides (LPS, 0.5 µg/kg body weight, *E. coli*) on d 111 post-partum as a model of standardized systemic inflammation. Blood samples were collected from d 1 ante injection (ai) until d 14 post injection (pi), with frequent sampling through an indwelling venous catheter from 0.5 h pi to 12 h pi. The white blood count and clinical chemistry parameters were analyzed. All parameters of the white blood count responded significantly to LPS, whereas only few parameters were affected by L-carnitine supplementation. The mean eosinophil counts as well as the percentage of basophils were significantly higher in CAR than in CON over time. In addition, a significantly higher urea level was observed in CAR throughout the study, indicating changes in protein metabolism.

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Comparative immune competence analysis of three local chicken breeds

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Both genetics and husbandry management are fundamental to animal health. Due to the special requirements, this is a challenge especially in organic farming. It is often said, without any scientific evidence, that traditional or local livestock breeds are healthier and have a better immune system than high-performance lines. In contrast, little is known about immune competence of regional breeds. The aim of this project is to assess the suitability of three phylogenetically divergent regional chicken breeds (Altsteirer, Ramelsloher and Bielefelder Kennhuhn) for organic farming, where there is a particular risk of infection through contact with wild birds. For this reason, it is of utmost importance to study the general immunological competence as a basis for early and protective immunity. In this project we first aim to study these breeds with respect to animal health aspects. In the second part of project, we will also analyse their crosses with high-performing commercial lines. Therefore, we want to study the kinetics of the development of the juvenile immune system after hatching and the immune competence of naïve chickens by analyzing the blood and the lymphoid organs by flow cytometry. Since avian influenza viruses have a zoonotic potential, they pose a threat not only to poultry but also to humans. To assess the viral resistance of the chicken breeds we infect them with a modified avian influenza A virus, which serves as a model virus. We will analyze the clinic and the survival, the virus shedding and transmission, and humoral and cellular immunity.

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One gets it - the other doesn't. Comparison of outbreak and HPAI-free poultry farms in Lower Saxony including sociological aspects

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Avian influenza (AI) is a highly contagious viral infection that has among others natural reservoir hosts in wild waterfowl and can kill large numbers of farmed poultry. In addition to the outbreak years 2016/2017, in which more than 1.2 million animals had to be culled, Germany was severely affected by an AI epidemic in 2020/2021. The exact cause or the factors that led to the virus entering the herd often remain unclear and it seems possible that a combination of several factors is responsible for the entry. Therefore, biosecurity measures are considered the most important way for disease-free fattening and preventing the spread of AI.

Outbreak numbers over the past few months suggest that HPAI could become endemic in Central Europe. Although many flocks seem to have a similar risk of virus introduction due to homogeneous geographic location and husbandry farms are spared from an HPAI outbreak.

The aim of this project is to identify differences between outbreak farms and HPAI-free farms and thus to recommend measures that will form a starting point to protect poultry farms effectively from HPAI in the long term. To achieve this goal, the project will partly build on previous studies in Lower Saxony and will be divided into two phases.

In the first phase, a case-control study will be conducted using a "KAP" (Knowledge, Attitude, Practice) questionnaire to investigate knowledge, attitudes and implementation regarding biosecurity measures in HPAI-affected and non-affected poultry farms. In addition, the biosecurity measures of the participating farms will be evaluated using a checklist and weighted with regard to their potential effect.

Using a cross-sectional study, a "health belief model" will be applied in the second phase of the project, which should help to better understand the behavior of farmers regarding the implementation of biosecurity measures. The combination of the two sub-studies will help to identify barriers that hinder a consistent and effective implementation of biosecurity measures and thus create opportunities to counteract them.

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Poster Nr. 39

Use of 3D cell cultures devices for the in vitro culture of bovine embryos

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Assisted Reproduction Technologies (ART) have played a vital role in improving livestock production and preserving genetic diversity in various mammalian species for many years. Nevertheless, concerns have been raised about the association between ART, adverse pregnancy outcomes, imprinting disorders, and the timing of epigenetic reprogramming. Therefore, it is crucial to improve in vitro production of embryos techniques in terms of embryo quantity, time to pregnancy, likelihood of live birth and to ensure the health of next generations of in vitro fertilization offspring. This study aims to validate extracellular vesicles (EVs) collected from bovine oviduct epithelial cells (BOECs) cultured in an air-liquid interface as an alternative to the oviduct-on-a-chip. We will also compare the transcriptome and DNA methylation of in vivo, on-chip, and in vitro zygotes. BOECs will be isolated, pre-culture and will be transfer to Transwell® system. EV characterization will include nanoparticle tracking analysis, transmission electron microscopy, mass spectrometry and Western blot. Oocyte collection and in vitro maturation will involve the recovery of bovine oocytes through ovum pick up from Holstein heifers. Matured Cumulus-oocyte complexes will be randomly distributed between groups, and methylation patterns will be analyzed using fluorescent staining. Our research focuses on the utilization of novel cell culture devices for in vitro bovine culture and assessing the epigenetic effects of three-dimensional (3D) culture model systems. This will significantly improve our understanding of gamete interaction, fertilization, and early embryo development, by more accurately mimicking the in vivo environment during pregnancy establishment.

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Poster Nr. 41

Modifying the porcine genome for xeno-organ transplantation

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Xenotransplantation is a crescent alternative and solution for the increasing demand for organ transplantations. Due to the close similarity that human and pig organs share, domestic pigs have emerged as suitable candidates for organ donation. Nevertheless, the presence of xenoantigens limits the success of pig-to-human grafts. To address this issue, the generation of genetically edited pigs targeting xenoantigens and expressing human genes to achieve immunotolerance has been established.

One prominent method for generating these genetically modified pigs is Somatic Cell Nuclear Transfer (SCNT). This approach has demonstrated notable success in the production of triple-knockout pigs specifically engineered to minimize human IgG and IgM antibody binding to diverse pig tissues, thereby reducing the presence of xenoantigens. Nonetheless, this technique demands sophisticated equipment and a high level of expertise, making it labor-intensive and costly. Furthermore, its efficiency to generate knock out pigs is very low. Alternative methods to SCNT for introducing the CRISPR/Cas9 system into zygotes have gained popularity in recent years. Microinjection stands out as a technique that facilitates the intracytoplasmic delivery of ribonucleoprotein complexes or CRISPR-Cas9 plasmids into oocytes or zygotes.

On the other hand, electroporation causes the temporary disruption of the cell membrane by applying electrical pulses, thereby enabling the entry of ribonucleoproteins (RNPs) into the embryo. This study conducts a comparative analysis between microinjection and zygote electroporation methods, focusing on the assessment of mosaicism and mutation rates during the generation of GGTA1, CMAH and B4GALNT2 triple-knock out pig embryos.

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Poster Nr. 43

Whole-genome sequencing and development of an improved typification strategy for syphilis-causing *Treponema paraluisleporidarum* in European lagomorphs

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Treponema paraluisleporidarum is the causative agent of syphilis in hares and rabbits. For now, only a single genome of *Treponema paraluisleporidarum* strain Cuniculi A has been completely sequenced. The aim of this work is to do a whole genome sequencing of eight new strains of the bacterium. In an initial step, we identify different strains from genital swabs and tissue samples of wild hares from Finland by using a multilocus strain typing (MLST) method. The typing genes, tp0548 and tp0488, are Sanger sequenced as well as we introduce a new amplicon-based Oxford Nanopore Technology (ONT) sequencing system to optimize the MLST. From these sequences different haplotypes are identified and selected for whole genome sequencing. DNA target enrichment is applied using in-solution capture with RNA-baits. Subsequently, DNA libraries are prepared and sequenced on the Illumina MiSeq platform. Resulting reads will be downstream processed and mapped against the strain Cuniculus A reference genome. Subsequently, gaps in the draft genomes will be filled by Sanger sequencing or ONT sequencing. By obtaining the whole genome of different strains, the multilocus strain typing can be revised for optimization in lagomorph infecting *Treponema paraluisleporidarum*.

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Poster Nr. 45

Resilience of chicken towards *Salmonella*: using surrogate infection models to define a protective microbiome (ChiSaRe)

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In 2021 the European food safety authority (EFSA) reported 60,050 cases of Salmonellosis. This makes *S. enterica* the second most prevalent bacterium for zoonotic infections in humans. These infections are often associated with ingestion of chicken (*Gallus domesticus*) produce. Besides its ability to infect humans, *S. enterica* is a threat for its avian host as well. E.g. Serovars *S. enterica pullorum* and *S. enterica galliarum* are the causing agents of diseases, associated with mortality rates approaching 100 % in young chicken.

We intend to establish a protective microbiome in chicken, to prevent the proliferation and decrease pathogenicity of *S. enterica*. The first step is the identification of dominant and representative microbes of the gastro-intestinal tract (GIT) of *G. domesticus* from literature, to define a synthetic oligo-chicken microbiota (OCM).

This OCM will be subjected to competitive growth assays with different strains and mutants of *S. enterica* to identify promising phenotypes. The OCM will be applied on surrogate infection models, namely larvae of *Galleria mellonella* and organ-on-chip using cells of the avian GIT. The models will be infected with *S. enterica*, to investigate possible protective functions of the OCM. The underlying molecular factors will be characterized using omics approaches with a focus on (meta-)transcriptomics and proteomics especially.

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Poster Nr. 47

Capacity of the swine gut microbiome in phytate dephosphorylation and inositol catabolism

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Phytate, or inositol hexakisphosphate, is not only the major carbon source in soil, but also the main phosphorus storage molecule of plants and therefore part of the diet. Phytases are often added to animal food to raise the amount of phytate-derived phosphate absorbed by the host. Based on an *in-silico* survey, we estimated that approximately 27% of all bacterial species are able to utilize myo-inositol (MI), which is released from phytate upon complete dephosphorylation. The enzymatic activities of the gut microbiome involved in phytate dephosphorylation and inositol degradation, however, are largely unknown.

16S rRNA gene sequencing of nearly 300 commensals positive in MI utilization revealed a predominance of *Paenibacillaceae*, *Bacillaceae*, *Enterobacteriaceae* and *Planococcaceae*. Similar compositions were found in feces, colon as well as ileum and cecum of four swine. To then investigate how many and which commensal bacteria isolated by culturomics produce phytases, we performed a systematic bioinformatic survey of all bacterial species producing phytases. Analyses on all taxonomic levels revealed that the main classes of phytases are not ubiquitously present in bacteria. In a follow-up approach, we will derive the metagenomes and meta-transcriptomes of feces samples to delineate the influence of increasing amounts of phytate in the diet on the microbiota composition and activity.

This project combines different omics approaches to analyze the swine gut microbiome with a focus on the taxonomic distribution of phytases and MI utilization activities. The results are expected to improve our understanding of the phytate fate following ingestion and to pave the way for probiotic interventions to increase phosphate availability in the gut and to reduce eutrophication.

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Poster Nr. 49

ESKAPE Pathogens in Germany: An overview

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Antimicrobial resistance (AMR) represents a major global health threat and is increasing at alarming rates. The leading cause of nosocomial infections and AMR are the so called ESKAPE pathogens, i.e., *E. faecium*, *S. aureus*, *K. pneumoniae*, *A. baumannii*, *P. aeruginosa*, and *Enterobacter* species. ESKAPE pathogens have been isolated from humans, hospital environments, animals, and veterinary clinics, as well as from food and environmental samples in the majority of the Federal states of Germany. MDR isolates of ESKAPE pathogens harbouring several resistant determinants mainly mediating resistance to aminoglycosides, tetracyclines, cephalosporins, fosfomycin have been isolated from horses, dogs, cats, cattle, pigs, rabbits, poultry farms, milk samples, and wastewater in different localities in Germany. A typical feature of these pathogens is their rapid spread in communities and hospitals and their association with high mortalities. They are typically isolated from wound and skin infections as well as from respiratory and urogenital tracts of patients, particularly those hospitalized in intensive care units in large clinical centres.

Despite ESKAPE pathogens being intensively investigated in human medicine, they are still disregarded in veterinary wards and environmental health sectors, also due to limited financial and human resources. Large-scale genomic surveillance of strains from different resources and reservoirs is required to understand the mechanisms of resistance and strengthen the one health concept.

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Poster Presentations

Session: II

Poster Nr. 2

In-vivo study of the effect of ceftiofur free crystalline and ceftiofur (HCl) on different bacterial diseases in rabbits

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Bacterial infections have deleterious effect on health and production of livestock and result in huge economic losses of animal. These include *E. coli*, *S. aureus*, *Salmonella* spp. and *F. necrophorum*. After considering the problem the study was conducted for efficient antibacterial treatment. This study compared the effectiveness of the Ceftiofur free crystalline acid and Ceftiofur (HCl) against *E. coli*, *S. aureus*, *Salmonella* spp. and *F. necrophorum* with the help of C-reactive protein values for finding the prevalent undiagnosed infection and a prognostic tool to check success of therapy. 32 adult male rabbits which were managed in eight groups. After 192 hours CRP values were found maximum confirming that these bacteria had caused infection in rabbits. Groups A to D were treated with ceftiofur free crystalline acid and groups E to H were treated with Ceftiofur (HCl). Blood samples were collected at an interval of 24hrs for CRP test to determine the efficacy of both medicines. Ceftiofur free crystalline acid was significantly ($p < 0.05$) more effective against all the studied bacterial infections. C-reactive protein test was found effective to determine the underlying infections in animals.

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Poster Nr. 4

Pathobiology and variable genetic determinants for virulence, transmission and tissue tropism of H7N7 avian influenza virus in chickens, turkeys and different duck breeds

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High pathogenicity (HP) avian influenza viruses (AIV) cause severe mortality in chickens (*Gallus gallus domesticus*) and turkeys (*Meleagris gallopavo*), although turkeys are more sensitive than chickens. Little is known about the virulence determinants in both bird species, beyond the polybasic cleavage site (CS) in the hemagglutinin (HA). In 2015, HPAIV H7N7 and a low pathogenicity (LP) ancestor were isolated from the same chicken farm. The aim of this study was to investigate the pathobiology and the genetic determinants for virulence and transmission of these viruses in chickens, turkeys and ducks. Using reverse genetics different virus reassortants with an LP backbone carrying the polybasic CS (designated Lp_poly) and single or multiple HP segments were generated. In contrast to chickens and turkeys, LP and HP viruses were avirulent in ducks, where infection was restricted to the respiratory and digestive tracts with excretion at considerable levels. Interestingly, in turkeys and chickens, Lp_poly viruses carrying the HP NS or M gene segments were as virulent and transmissible as the parental HPAIV, respectively. All Lp_poly viruses were detected in the brain of chickens and ducks, although endothelial infection was exclusively found in chickens, where the HACS and to lesser extent NA are major determinants for this phenotype. Together, ducks are clinically resistant to this H7N7 virus but excreted high amounts of viruses. This study showed two major differences between chickens and turkeys: more genetic constellations to confer high virulence and transmission in turkeys than in chickens and neurotropism in turkeys vs. neuronal and endothelial tropism in chickens.

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Poster Nr. 6

Evaluation of an ELISA for distinguishing West Nile virus and Usutu virus infections in birds by using a mutated recombinant envelope protein

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The work package, part of the CuliFo3 project, focuses on zoonotic Flaviviruses: West Nile virus (WNV) and Usutu virus (USUV) which belong to the Japanese Encephalitis serocomplex. Mosquitoes function as vectors for both viruses and infect birds, which serve as reservoir hosts but also mammals. USUV is widespread across Germany, whereas WNV cases have occurred to date mainly in the eastern part of Germany.

Distinguishing USUV from WNV currently requires virus neutralization tests (VNTs) and commercial ELISA due to the high serological cross-reactivity. The project aims to discriminate these viruses in animals without the need for testing via VNTs by using a quadruple mutant E-protein (Equad) in an indirect ELISA. Sera of various bird species and reference horse sera are tested by this method.

The Equad-ELISA is based on recombinant E-proteins with four point mutations in the domain II of the fusion loop resulting in reduced cross-reactivity between closely related flaviviruses. Previous studies demonstrated that differentiation of WNV and USUV is specific in human samples, and is also possible in horse field sera. The project extends the application of Equad-ELISA in the veterinary field, by discriminating WNV from USUV-infected animals using horse and poultry sera. Equad-based competition ELISAs were used to further minimize cross-reactivity in horse, geese and duck sera.

The projects aim is to develop a rapid screening method for avian sera, eliminating the need for VNTs in the BSL3 laboratory. This will allow conclusions about the circulation of arbozoonotic pathogens in the vicinity of humans.

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Poster Nr. 8

The role of extracellular vesicles in the modulation of antiviral immunity during foot-and-mouth disease virus infection

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Foot-and-mouth disease virus (FMDV), the causative agent of foot-and-mouth disease in cloven-hooved animals is a highly contagious animal pathogen. Although the virus remains an ongoing threat to livestock globally, parts of the viral lifecycle remain incompletely understood. FMDV Leader Protease (Lpro) is a papain-like cysteine protease and deubiquitinase that acts as a potent virulence factor early after infection as it is the first protein to be translated off the viral genome. Lpro is vital to FMDV as it is involved in interference with interferon- and NFκB signalling thus allowing FMDV to overcome primary immune mechanisms and preventing cells from establishing an antiviral state. We were able to show that Lpro is localized to and secreted within extracellular vesicles (EVs), membrane-enclosed particles that are produced by all cell types. EVs have recently come into the spotlight as carriers of both protein- and nucleic acid cargo that play important roles in intercellular communication. We hypothesize that Lpro is secreted through EVs in order to reach other, non-transmissible cell types such as cells of the innate immune system in order to inhibit their antiviral functions. Within the scope of this PhD project we hope to further elucidate the mechanism of localization of Lpro to EVs as well as to characterize these EVs and their effect on potential target cells.

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Poster Nr. 10

Optimization of deoxynivalenol inactivation using sodium metabisulfite under hydrothermal conditions

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Deoxynivalenol (DON) is a mycotoxin produced by *Fusarium* species that grow on cereal crops. The contamination is not completely avoidable resulting in a need for inactivation strategies. Former studies demonstrated the inactivation of DON using sodium metabisulfite (SBS) by wet conservation (Paulick et al. 2015). However, this method cannot be applied to traded feed. Therefore, the present study aimed to titrate optimal conditions for the inactivation of DON under hydrothermal conditions which might be included into commercial feed production processes.

DON contaminated maize was mixed, with or without the addition of SBS, for 2 minutes prior to the addition of saturated steam for the adjustment of moisture and temperature in a stationary experimental conditioner. The addition of steam (0-3%), the conditioning time (10-600 seconds) and the dose of added SBS (0-10g/kg) were varied. DON concentrations were analyzed by HPLC (Oldenburg et al. 2007) with DAD after clean-up with immuno-affinity columns (DONPREP, R-Biopharm Rhône Ltd.).

DON concentrations were only reduced when SBS was added in the conditioning process. The optimal reduction to 4.06% in maize kernels and 6.37% in maize meal of the initial DON concentration, was reached with an addition of 3% saturated steam and 10g/kg SBS at a conditioning time of 10 seconds. Future investigations should validate the optimized conditions at different levels of DON contamination and in other matrices (e.g. wheat).

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Poster Nr. 12

Diving Deep into Fish Bornaviruses: Uncovering Hidden Diversity and Transcriptional Strategies through Comprehensive Data Mining

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Recently, we discovered two novel orthobornaviruses in colubrid and viperid snakes using an *in-silico* data mining approach. Here, we present the results of a screening of more than 100,000 nucleic acid sequence datasets of fish samples from the Sequence Read Archive (SRA) for potential bornaviral sequences. We discovered the potentially complete genomes of seven bornaviruses in datasets from osteichthyans and chondrichthyans. Four of these are likely to represent novel species within the genus *Cultervirus*, and we propose that one genome represents a novel genus within the family of *Bornaviridae*. Specifically, we identified sequences of Wùhàn sharp-belly bornavirus (WhSBV) in sequence data from the widely used grass carp liver and kidney cell lines L8824 and CIK, respectively. A complete genome of Murray-Darling carp bornavirus (MDCBV) was identified in sequence data from a goldfish (*Carassius auratus*). The newly discovered little skate bornavirus (LSBV), identified in the little skate (*Leucoraja erinacea*) dataset, contained a novel and unusual genomic architecture (N-Vp1-Vp2-X-P-G-M-L), as compared to other bornaviruses. Its genome is thought to encode two additional open reading frames (tentatively named Vp1 and Vp2), which appear to represent ancient duplications of the gene encoding for the viral glycoprotein (G). The datasets also provided insights into the possible transcriptional gradients of these bornaviruses and revealed previously unknown splicing mechanisms.

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Poster Nr. 14

Development of an efficient Vaccine against Porcine Reproductive and Respiratory Syndrome Virus

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Porcine Reproductive and Respiratory Syndrome Virus (PRRSV) is one of the leading pathogens to cause worldwide economic loss in pig farms. Existing commercial vaccines show only limited impact on the disease prevention and control due to highly variable circulating strains and the immunosuppressive nature of the virus. After PRRSV infection, pigs develop early but non-neutralizing antibodies which are thought to even enhance virus uptake. A neutralizing antibody response is detectable only at late times after infection probably due to the presence of immunodominant decoy epitopes and an efficient glycan shield of critical epitopes. Therefore, it is crucial to identify suitable targets eliciting efficient and cross-protective immunity.

The aim of this project, as part of the EU-funded Horizon REPRODIVAC consortium for the generation of new prevention tools against livestock diseases, is to develop a protective vaccine against PRRSV by identifying conserved antigenic regions which will be presented by using the Alphaherpesvirus Pseudorabies Virus (PrV) as a vector. The attenuated PrV strain Bartha (Ba) is widely used as a DIVA-suitable marker vaccine and has been successfully employed in the eradication of Aujeszky's disease in pigs, therefore making it applicable as a bivalent vaccine against PRRSV. Two different approaches are used for the generation of the PrV Ba-PRRSV recombinants, both of which include the insertion of full-length genes, parts encoding the ectodomains or antigenic peptide regions of PRRSV envelope proteins into non-essential gene loci of PrV Ba. Generation and in vitro characterization of the first PrV-PRRSV recombinants will be presented.

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Poster Nr. 16

Modeling Nitrogen excretion from dairy cattle to improve National Emission Inventories and on-farm assessment

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Excessive nitrogen emissions cause ecosystems to become eutrophic and nitrous oxide emissions to increase. They contribute to global warming, soil acidification and reduced biodiversity. The MoMiNe project (Modeling N excretion from dairy cattle to improve National Emission Inventories and on-farm assessment) was launched in order to develop tools to predict N excretion by cattle at the farm level as a precondition to adjust feeding and management practices for reducing N excretion. As a part of this project, feeding trials with dairy cows are performed at the experimental station Braunschweig aimed at induction of a high variability of N excretion as a basis for developing prediction models. Trial begins with an adaptation phase of 21 days, followed by a collection phase of 21 days. During the collection phase, samples of feces, urine, back fat thickness and blood are taken weekly. Rumen fluid is collected at the first and last sample week. The animals are fitted with numerous monitoring devices throughout the periods and are examined for methane emissions, rumination, animal wellbeing, monitoring of general health with health sheets, BCS, locomotion score, feed and water intake, live weight, milk yield and milk constituents. For each adaptation phase and for each collection phase a pool sample of the feed is processed. Statistical analyses are performed using R, SAS and Excel. The aim of the study is an adapted diet that meets the nitrogen requirements of the lactating dairy cow and still reduces nitrogen emissions without causing performance losses or reduced animal health

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Poster Nr. 18

Development of a Novel Live Attenuated Vaccine for Foot-and-Mouth Disease

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Foot-and-mouth disease (FMD) is an easily transmissible animal disease which poses a severe economic threat to farmers, particularly in developing countries. Current control measures rely on culling infected and nearby animals and vaccination with an inactivated vaccine. The inactivated vaccine has inherent shortcomings such as necessitating intramuscular injection, demanding a biannual re-administration, and perpetuating the risk of outbreaks linked to vaccine production facilities. Our research aims to address these challenges by developing a novel live attenuated vaccine. This innovative approach promises a safer, easily administered vaccine with durable immunity, along with the ability to distinguish between infected and vaccinated animals (DIVA). To achieve this, we are exploring various vaccine candidates, including chimeric viruses, codon-deoptimized viruses, and viruses with specific deletions. Our comprehensive study involves both in vitro experiments and in vivo studies conducted using pig models. By evaluating the safety and efficacy of these vaccine candidates, we aim to contribute to the development of a more effective and safe solution for controlling foot-and-mouth disease

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Poster Nr. 20

Detection of antibodies against Rotavirus A (RVA) infection in domestic pigeons, free-ranging feral pigeons (both *Columba livia*) and wild wood pigeons (*Columba palumbus*) in Germany

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In the period from 2021 to the present, there have again been many confirmed cases of domestic pigeons (*Columba livia* forma domestica) dying from infections with rotavirus (RVA) of genotype G18P[17]. The virus is member of the family *Reoviridae* and proven to be the causative agent of 'young pigeon disease' (YPD) which is characterized by its seasonal occurrence with high mortality rates of up to 50%. Mainly juvenile individuals are affected, showing predominantly intestinal manifestations and liver necrosis in association with inflammatory infiltrations.

During the annual racing season millions of pigeons come into contact with each other, resulting in the transmission of the pathogen.

Although YPD leads to immense losses, especially in pigeon husbandry, there is still little knowledge about the distribution within domestic pigeon populations as well as possible wild reservoirs. Therefore, the aim of this study was to further investigate the occurrence of RVA infections in domestic pigeons of different age groups, free-ranging feral pigeons and wild wood pigeons (*Columba palumbus*) by detection of RVA-reactive antibodies. This update on our RVA research will fill in missing gaps and contribute to a better understanding of the pathogen.

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Poster Nr. 22

The effects of lameness on methane production, intensity and yield in a dairy cow herd - preliminary results

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Studies simulating methane emissions in lame dairy cows suggest that lameness increases the global warming potential at the farm (Chen et. al.; 2016). To verify this hypothesis, a herd of dairy cows with varying degrees of spontaneously occurring lamenesses was analyzed in methane production, as part of an ongoing methane mitigation research project.

The trial consisted of a 48-head dairy herd, which was split in two groups, housed in 2 different pens in a uniform stable tract. It lasted 18 weeks and was divided into three periods. The cows were fed according to the recommendations for energy and nutrient supply of the GfE (2001) in a 2 x 2 factorial design. Wherein the first factor was the concentrate level, the second factor changed in every period and included different potentially methane reducing feeding regimes. Methane production of the cows was measured by a GreenFeed system (C-Lock Inc., Rapid City, SD, USA) in each pen. During the whole trial, the locomotion score was determined weekly according to Flower & Weary (2006). This resulted in 4 groups according to their locomotion score; healthy cows, slightly lame cows, lame cows and clearly lame cows.

Starting from a slight lameness, the cows begin to show comparatively higher methane emissions to healthy cows. Measurements show that the locomotion score has a significant impact on methane production ($p = 0,0169$), intensity ($p < 0,0001$) and yield ($p < 0,0001$).

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Role of $\gamma\delta$ T cells in protecting chickens against *Salmonella enterica* serovar Enteritidis infections

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Avian $\gamma\delta$ T lymphocytes are frequently found in the intestinal mucosa and are considered to be crucial in protecting chickens from *Salmonella* infection. To elucidate the role of $\gamma\delta$ T cells in the avian immune response, wild-type (WT) and $\gamma\delta$ T cell knockout (KO) chickens were orally infected with two different doses of *Salmonella Enteritidis*.

Up to 12 days post infection, the *Salmonella* load in liver and cecum, the absolute numbers of $\gamma\delta$ and $\alpha\beta$ T cells subsets in blood and transcription levels of different immune-related proteins in cecum of chickens were determined.

After high-dose *Salmonella* infection, KO chickens displayed a significantly higher bacterial load in the liver compared to WT animals. Immunohistochemical analysis revealed a more pronounced *Salmonella* invasion in cecum of KO chickens. Compared to WT animals, we found an expansion of monocytes in blood of KO chickens after high-dose *Salmonella* infection. Contrary to the detected increase in CD8 $\alpha\alpha$ ++ $\gamma\delta$ T cells in blood of WT chickens, an elevated number of CD8 $\alpha\alpha$ ++ $\alpha\beta$ T cells was observed in KO chickens after infection. Additionally, we identified an increase in CD25-expressing CD8 $\alpha\alpha$ ++ $\gamma\delta$ T cells in WT and CD8 $\alpha\alpha$ ++ $\alpha\beta$ T cells in KO chickens. In conclusion, our findings suggest that $\gamma\delta$ T cells play a critical role in early protection of KO chickens against *Salmonella Enteritidis* infection. However, later in the course of infection, immune functions typically fulfilled by CD8 $\alpha\alpha$ -expressing $\gamma\delta$ T cells might be compensated in KO chickens by an upregulation of their corresponding $\alpha\beta$ T cell counterparts.

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Poster Nr. 26

The power and promise of CRISPR/Cas9 genome editing for the identification of novel West Nile Virus host and vector factors

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Emerging and re-emerging arboviruses are a major global health threat. West Nile Virus, which is newly emerging in Germany, is a small, enveloped, positive-stranded RNA virus. It is a member of the Flaviviridae family and as such it is closely related to other arthropod-borne viruses like Japanese encephalitis virus or Yellow fever virus. WNV has a wide host range. In its enzootic life cycle, it circulates between mosquitoes and birds. But humans and other mammals can also be infected as dead-end hosts. In humans, it is the causative agent for West Nile fever and in rare cases encephalitis. So far, very few West Nile virus host/vector factors have been identified. To better understand the host/vector-virus dependencies, we aim to confirm suspected WNV host/vector factors via a gene specific Knockout-Rescue approach. The goal is to generate gene specific knockouts in human and insect cells using CRISPR/Cas9 and confirm the absence of target gene expression. Afterwards, the effect of the missing target gene expression on the life cycle of the West Nile Virus will be investigated. This approach will be complemented by protein expression using an expression construct. Here we present preliminary data on knock-out generation and protein overexpression and discuss challenges and pitfalls encountered so far.

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Poster Nr. 28

Connecting the dots with Tryptophan in laying hens

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Tryptophan is an essential amino acid in humans and non-human animals and needs to be supplied through diet. Majority of the ingested Tryptophan is metabolized in the periphery and the rest is transported to the brain. Tryptophan has been found to exert both immuno-stimulatory and -suppressive effects and its metabolism produces downstream metabolites like serotonin and kynurenine that serve as active neuromodulators in the brain. Altered concentrations of these metabolites in the brain have been found to be associated with altered behaviour (including fear responses, social behaviour among others) and cognitive abilities. Furthermore, the gut microbiota seems to play a pivotal role in the interaction between Tryptophan metabolites and behaviour by further affecting peripheral Tryptophan metabolism. The present study uses a dietary Tryptophan supplementation treatment in laying hens to explore:

- a) *The association between Tryptophan metabolism, behaviour and gut microbiota*
- b) *The effect on immune status*

The experiment will last from hatching until the rearing period using two feeding treatments with varying Tryptophan concentrations throughout: basal (0.2%) and surplus (0.5%). At repeated timepoints throughout the rearing period, behavioural data (fearfulness, sociality, locomotor activity), fecal samples (microbiota composition) and blood samples (concentrations of Tryptophan and its metabolites, quantification of immune cells) will be collected. At the end of the rearing period, learning performance and memory will be assessed, and central (brain) concentrations of Tryptophan and metabolites as well as cecal microbiota composition will be measured.

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From genotype to phenotype- diversity of HPAIV H5Nx genotypes and their pathogenicity in ducks (*Anas platyrhynchos* var. *domesticus*)

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Worldwide incidence of highly pathogenic avian influenza (HPAI) has gained considerable momentum since 2020, and it is now considered as panzootic with a growing number of different HPAIV genotypes. Anseriformes play an important role in the epidemiology of AIV and dispersal of HPAIV H5. For the currently circulating clade 2.3.4.4 HPAI H5 viruses, studies have shown that HPAIV infection may have minimal or no effects on duck health or behavior. However, little is known on genotype specific differences regarding pathogenicity in ducks and correlation to distribution. Here, we used the infection model of juvenile ducklings (*Anas platyrhynchos*) as a sensitive method to characterize virulence. Comparing 6 HPAIV H5N1 genotypes from Germany co-circulating in December 2021, we observed significant differences: Strikingly, two genotypes, that dominated the HPAIV H5N1 infection dynamic in 2021 and 2022 in Germany and Europe, were highly pathogenic for ducklings, inducing 100% mortality within two days. In contrast, other genotypes that were detected only sporadically, induced only mild signs of disease and no mortality. Host-parasite combinations in which virulence is low or tolerance is high are usually biased towards widespread pathogen dispersal. Our findings, in contrast, point to an infection dynamic in wild birds continuous spill-over and seeding infections mutually involving poultry and the huge reservoir of low pathogenicity AIV in wild birds.

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Poster Nr. 32

Anaphylactic reactions in vaccinated cattle after administration of Oxytetracyclin and Penicillin-Streptomycin

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In 2018, a new vaccine against bovine mastitis was licensed by the European Commission. The vaccine is produced from streptococcal biofilms and the active substance is Lipoteichoic acid (LTA). The European Pharmacovigilance database lists 113 fatal anaphylactic reactions when cows vaccinated with the new mastitis vaccine were treated with antibiotics. Most of the antibiotics that triggered these anaphylactic reactions are produced in *Streptomyces* spp. We hypothesize that vaccine-induced antibodies cross-react with a component, present in the involved veterinary antibiotics, triggering an anaphylactic shock.

Two veterinary antibiotics and one cell-culture additive containing Penicillin-Streptomycin (Pen-Strep) were tested by western blotting and incubated with serum of vaccinated cows. The veterinary medicines showed reactivity in contrast to the highly purified antibiotic.

Affinity purified vaccine specific antibodies of vaccinated cows showed cross-reactivity to Pen-Strep, while in contrast serum of non-vaccinated cows showed no reactivity.

The nature of the cross-recognized antigen has to be proven in further experiments. Additionally, we aim to establish the functional relevance of the observed western blot activity using a mast cell degranulation assay.

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Poster Nr. 34

Literature review on the impact of West Nile Virus

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Since its first discovery in 1937 in the West Nile region in Uganda, the Orthoflavivirus nilense, former known as West Nile Virus (WNV), spread around the world. Whether the epidemic episode in the United States (1999-2004) when WNV spread all over the US, resulting in about 57,000 disease cases and 2800 deaths, or the recent outbreaks since 2018 in Europe all the way North to Germany, there is yet no unified model to calculate the impact of an infection with WNV. The disease can reach forms from subclinical over West Nile Fever and West Nile Neuroinvasive Disease up to lethal cases.

Aim of the study is to use a literature review to obtain information on the impact of WNV as well as on the methods used to calculate the impact. We searched the databases Web of Science, Pubmed and Scopus using the keywords “West Nile Virus” and “economics” and found 79 fitting papers, mostly from the US and southern Europe.

The results showed, that the impact models are very different between working groups and countries. While some groups focused on costs in humans (USA), other concentrated on horses (Belgium) or the improvement of surveillance systems (Italy). Some studies showed only the costs of treatment in humans, while others used also productivity loss. The results will be used to develop an impact model for Germany.

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Poster Nr. 36

Comparison of *Acinetobacter* spp. antibiotic resistance results using CLSI and EUCAST breakpoints

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The prevalence of MDR *Acinetobacter* spp. has increased in recent years, resulting in the restriction of treatment of patients to certain critical antibiotics. One factor that affects the reliability of the results is the criteria used to interpret the results, e.g. CLSI or EUCAST. Resistance was determined in 123 isolates of *Acinetobacter* spp., using the MICRONAUT automated system and MICRONAUT-S MDR MRGN screening plates containing 12 antibiotic groups.

According to the results, one sample showed identical results for colistin, while the rest 122 strains were sensitive at increasing dose (I) according to CLSI and were sensitive (S) according to EUCAST; 50 samples showed identical results for ciprofloxacin, while 72 strains were S and one was I according to CLSI and 72 strains were I and one was resistant (R) according to EUCAST; 89 samples showed identical results for ceftazidim, while 34 strains were S according to CLSI and 32 were I and 2 were R according to EUCAST; 97 samples showed identical results for trimethoprim/sulfamethoxazol, while 25 strains were R and one was S according to CLSI and 26 were I according to EUCAST; 113 samples showed similar results to piperacillin/Tazobactam, while six were S, three were I and one was R according to CLSI, and six were I and four were R according to EUCAST. Evaluation of *Acinetobacter* spp. using CLSI and EUCAST yielded similar results for most antibiotics, with differences remaining for some compounds. Using both calculations can provide a comprehensive overview of the resistance profile of strains.

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Poster Nr. 38

Improved molecular surveillance and assessment of host adaptation and virulence of *Coxiella burnetii* in Europe

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Q fever, a worldwide occurring zoonosis. The bacteria display a broad host range and clinical manifestations of Q fever in ruminants are diverse and mainly associated with reproduction disorders and infertility in cattle, abortion in goats and asymptomatic infections or minor effects in sheep. Despite these differences, human Q fever outbreaks mostly originate from *C. burnetii* shedding by sheep and goats. The hypothesis of this project is, that differences in the zoonotic potential and clinical relevance of *C. burnetii* isolates correlate with differences in genomic traits. To increase the availability of isolates a biobank of *C. burnetii* containing materials will be created and isolation protocols from samples improved. To enrich *C. burnetii* concentration and limit sample contamination prior inoculation in cell culture or axenic media pre-treatment steps like physical sample treatment, such as filtration and ultrasound as well as chemical treatment with NaOH will be assessed. After inoculation post-treatment steps like the usage of antibiotics/antimycotics will be tested. Archived and new isolates will be sequenced for phylogenetic analysis and correlated with phenotypic characteristics originated from cell culture infection models. All data will be collated and read outs selected, which allow standardized discrimination of observed *C. burnetii* phenotypes. This information can be used to determine upcoming zoonotic threats posed by *C. burnetii* and provide a framework for assessing the risk and severity of future Q fever outbreaks.

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Poster Nr. 40

Molecular interactions during Flavivirus coinfections

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Multiple members of the genus Orthoflavivirus (formerly Flavivirus), many of which are arthropod-borne and transmitted by either mosquitoes or ticks, are of major concern for public health in countries across the world. Two members of the orthoflaviviruses, the West Nile virus and the Usutu virus co-circulate in Germany as well as other European countries. These viruses are transmitted by the same vectors, e.g. mosquitoes of the *Culex pipiens* species complex, and infect the same host species. In laboratory experiments it has both been shown that it is possible to infect mosquitoes with both viruses simultaneously as well as that co-infection can modulate their transmission. This shows the need to further understand the underlying molecular mechanisms and possible effects of co-infections on the hosts and vectors alike. Our goal is therefore to identify the mechanisms by which these viruses influence one another. Initially, the occurrence of co-replication or superinfection exclusion will be tested by using virus-specific antibodies. In order to differentiate between the two viruses, antibody cross-reactivity has to be ruled out beforehand. Additionally, reverse genetic systems like replicon or minigenome systems will be generated for both viruses and used with the intend of identifying the underlying mechanism(s) of co-replication or superinfection exclusion. Since equivalent proteins from different orthoflaviviruses can generally substitute for one another, crosstalk during co-infections is anticipated, which we aim to better characterize using these systems.

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Pathobiology and replication of a zoonotic H7N7 avian influenza virus in poultry, mammals and ex-vivo models

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Avian H7 influenza viruses (AIV) can evolve from a low pathogenicity (LP) to a high pathogenicity (HP) form in chickens, leading to severe systemic infections. In 2003, a HPAIV H7N7 outbreak in the Netherlands, Germany and Belgium resulted in the death and culling of ~30 million birds and 89 confirmed human infections. A poly-basic cleavage site (pCS) in the hemagglutinin (HA) protein is one of the main virulence determinants in HP AIV in chickens but less is known about the role of the viral polymerase segments in the transition of LP to HPAIV in chickens. Likewise, little is known about the role of pCS for adaptation of AIV in mammals.

Here, recombinant LP H7N7 viruses carrying a pCS (LP_pCS) with or without the polymerase segments from German HPAIV H7N7 were constructed and characterized. Chickens and mice were infected with recombinant viruses to assess virus virulence, and replication. In addition, ex vivo infected precision cut lung slices (PCLS) from mice were established as an alternate infection model. In chickens, we found that the pCS increased the virulence of LP H7N7, close to the level of HP H7N7, whereas the introduction of the HP H7N7 polymerase segments reduced the virulence of recombinant viruses compared to LP_pCS.

Interestingly neither the pCS nor the polymerase segments affected virulence of this H7N7 in mice or viral distribution in PCLS. These results indicate that virulence determinants of HPAIV H7N7 in mammals differ from those in poultry and highlight the feasibility of PCLS for studying the adaptation of HPAIV in mammals.

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Poster Nr. 44

A novel SARS-CoV-2 modified live vaccine with an optimized safety profile induces sterile immunity in syrian hamsters

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mRNA-based SARS-CoV-2 vaccines prevent from severe clinical outcomes in humans, but do not efficiently break transmission chains. Here we evaluated an attenuated live vaccine, based on the “one-to-stop (OTS)” genome recoding attenuation method in Syrian hamsters. We asked for the level of attenuation and protective potential, including the ability of inducing sterile immunity. For the attenuation-study, we inoculated Syrian hamsters intranasally with 103.6 TCID₅₀/animal of the live vaccine candidate OTS-228. Naive contact animals were screened for virus transmission. For protection, we immunized hamsters with OTS-228 and challenged them 21 dpi with homologous ancestral SARS-CoV-2, and heterologous Omicron BA.2 or BA.5. Attenuation and protection were evaluated through survival data, body weight, tissue virus RNA load, virus shedding, histopathology and virus antigen detection. OTS-228 vaccine candidate was fully clinically attenuated and led neither to pneumonia-related atelectasis nor SARS-CoV2 characteristic vascular lesions, peribronchial infiltrates or necrotizing bronchitis. Virus antigen was detected associated with slight expansion of the pulmonary interstitium by mainly macrophages. The vaccine virus was not transmitted to naïve contact animals. Vaccinated animals were protected from clinical disease and pneumonia-related atelectasis post challenge infection. OTS-228 vaccination also efficiently protected against replication of the challenge virus in the lungs, provided sterile immunity following homologous but not after heterologous challenge infection. The vaccine candidate OTS-228 meets all safety criteria for a SARS-CoV-2 live vaccine in the sensitive hamster model and has the efficacy to protect and induce strong immunity, that can even inhibit SARS-CoV-2 transmission.

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Vaccination approaches to inhibit autochthonous transmission of hepatitis E

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Hepatitis E virus is considered the most common cause of viral hepatitis worldwide. While genotypes 1 and 2 predominate in Africa and Asia and are associated with severe disease courses and death rates of 0.2-4%, and up to 25% in pregnancy, genotypes 3 and 4 are predominant in Europe. Unlike genotypes 1 and 2, they have zoonotic potential and usually cause only mild symptoms or subclinical, self-limiting courses of disease in healthy individuals. In immunocompromised patients, however, they can also lead to severe hepatitis with acute and chronic courses potentially involving other organ systems. Therapeutic options for hepatitis E are limited, so that hepatitis E disease in immunosuppressed patients is associated with a mortality rate of 20-27%.

In Germany, hepatitis E virus genotype 3 (HEV3) has the highest prevalence. It is responsible for 98% of HEV infections in the German population and leads to an estimated 417000 seroconversions per year. Domestic and wild boars are main reservoir hosts.

The VacInATE project addressed to the One Health aspect and to evaluate different vaccination strategies against HEV3 in pigs in order to prevent the transmission of the virus from infected pigs to humans and thus reduce the number of therapy-resistant clinical cases. For this purpose, vaccine candidates were generated and analysed in rabbits. The antibodies obtained were examined in vitro by ELISA, Western blot, immunofluorescence and serum neutralization assay. The most promising vaccines were finally applied in the porcine infection model. In addition, novel monoclonal antibodies were tested as alternative therapeutic option for treatment against HEV infection.

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Infectious potential and genome comparison of *Chlamydia avium* - a new player in Avian Chlamydiosis

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Until recently, *Chlamydia (C.) psittaci* was considered to be the only causative agent of chlamydiosis in birds. Using molecular diagnostic techniques, *C. avium* was described in 2014 as an additional chlamydial agent in pigeons and psittacines with different manifestations of the infection: While causing severe disease and death in psittacine birds, pigeons appeared to be less affected. To investigate these differences in host tropism and pathogenicity and to better characterize the new pathogen, we studied the growth and proliferation of four *C. avium* isolates in vitro and the pathogenic potential of twelve strains in an embryonated chicken egg model. Further, whole genome sequences were generated and compared with each other and with those of other avian *chlamydiae*.

In vitro, all four strains showed smaller inclusions, delayed proliferation and delayed formation of infectious particles compared to *C. psittaci*. In the egg model, even at a very high infection dose of 1×10^7 IFU per egg, *C. avium* isolates induced low mortality of 10-40 % or no mortality at all over an observation period of eight days, irrespective of whether they were obtained from sick parrots or healthy pigeons. In contrast, *C. psittaci* caused 100 % mortality after only five days.

A phylogenetic tree inferred from core SNP analysis presents with two distinct clusters, one comprising isolates from asymptomatic pigeons and the other mainly from psittacine birds with clinical manifestations. In comparison to other chlamydial species, *C. avium* lacks certain virulence-associated genes e.g. genes of the plasticity zone or some genes encoding polymorphic membrane proteins.

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Non-invasive sampling of wild ducks: Proof of concept for Avian Influenza

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Currently, highly pathogenic avian influenza virus (HPAIV) of subtype H5 goose Guangdong (gs/Gd) is circulating worldwide at unprecedented levels, causing epizootics in poultry and wild birds. Moreover, cases in terrestrial and marine carnivorous are reported at an alarming rate. The uncontrolled spread in captive and wild birds and the close contact between infected birds and mammals, including humans, poses the risk of the virus adapting to humans, which could lead to a significant pandemic potential. The current spatial spread of HPAI H5 gs/Gd has led to the introduction of HPAIV in Southern America where it infects a naïve host population. As a result, globally important biodiversity hotspots, such as the Galapagos Islands or Antarctica, face a conservation crisis.

Information from passive surveillance is limited and investigations in live birds are warranted to assess the extent of true prevalence. While active (sentinel-) surveillance in wild ducks is in demand but time and resource-intensive, the development of a simplified non-invasive surveillance strategy is needed. For this purpose, water samples that have come into contact with waterfowl and direct duck samples will be examined. We hypothesize that feeding water, with which wild birds have demonstrably been in contact, is just as suitable for detecting and phylogenetic characterizing influenza viruses as direct bird sampling. After proof of concept, the approach shall be tested in different field setting where outbreaks occur.

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