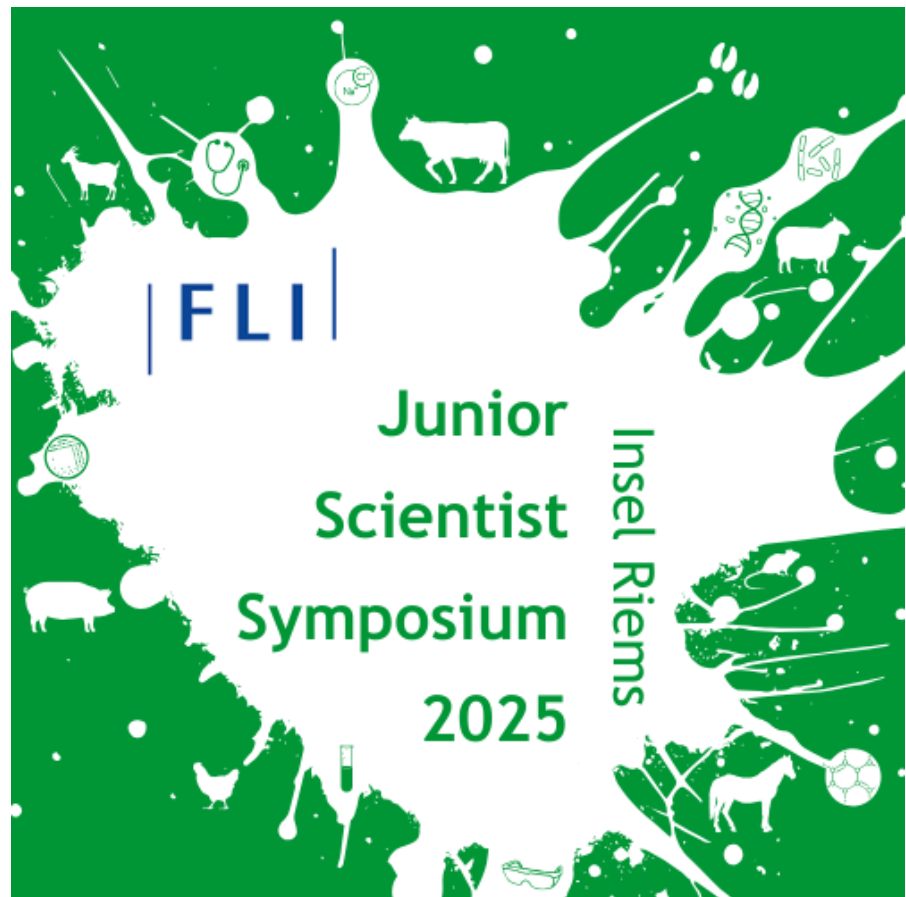




JUNIOR SCIENTIST SYMPOSIUM 2025

15-17 October 2025, Isle of Riems



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Welcome Note

Dear young scientists,

Why a young scientist symposium? I heard this question multiple times when talking to supervisors, doctoral students and others here at FLI. From the continuously decreasing participation in JSS, it seems that to some extent, there seems to be a “voting by feet”. And it is true: it takes a lot of effort, particularly from the (mostly few) organizers to set up such a meeting. They encounter delayed abstracts, very busy PIs, who are to be asked for key notes, and some reluctance by their peers to assist with all those small contributions, which create a pleasant and friendly workshop atmosphere. And still: all those of you, who attend this year seem to see the benefit of the JSS. It is a great opportunity to train for areas essential for a career, either in science, administration or industry and even veterinary practice. FLI wants to give you the chance to try yourself in many different ways. The JSS provide a “protected environment” for those of you, who double the plasma adrenalin concentration when standing at the lectern in front of an audience. You are among your peer: thus, you can be sure: nobody is perfect in the audience and very likely there is not a single person in the audience, who knows more about your PhD project than yourself. And you have the chance to discuss science really profoundly, ask questions - even if they may seem stupid to yourself. I can tell you from decades of conference participations: I hardly ever heard a “stupid question” from a doctoral student, but there are very few better ways to make yourself known in the community than to actively take part in discussions. And for all of this, the FLI JSS provides an excellent training ground. We want you to be successful, as scientist and also as mature person, educated to make your career path, wherever the future may carry you. And from own experience, I know that you might be extremely surprised to what horizons the future will carry you. Activities like the JSS will prepare you for this beyond the immediate presence of viral proteins, model equations or animal observation. I grant: we do live in times of some degree of uncertainty: but I can guarantee there is nothing better to prepare for the unknown than wide expertise, knowledge and skills. And if one of you realized that her/his vocation is conference management by organizing a JSS: well, this is not the expected outcome of a doctoral thesis, but

if you become a happy, self-conscious person I would also count it an FLI success story. FLI can and does provide opportunities with input of very substantial resources: it is up to you as young scientists to pick up this chance by participating in the JSS and the FLI graduate school. And to prove this way, that we set the right priorities in times of more and more budget constraints.

This year, the JSS is organized by the Riems team of doctoral students (a big thank-you for all those of you, who volunteered to make the JSS happen this year!): I hope those from outside get the chance to have at least a small view on the very impressive infrastructure here at the Island of Virus Hunters: access to Riems is not very easy for ordinary persons. For the JSS, I am looking forward to meeting you in Riems and learn from your presentations and discussions.

Prof. Dr. Christa Kühn

General Information

Organising Team & Contact

Nihal Telli, Alix Drobiniaik, Franziska Neffgen, Ender Menges, Laura Rzepa, Sarah Jahn & Jessica Junker

Contact: JSS.Organization@fli.de

Conference Language

The official conference language is English.

Dates and Venues

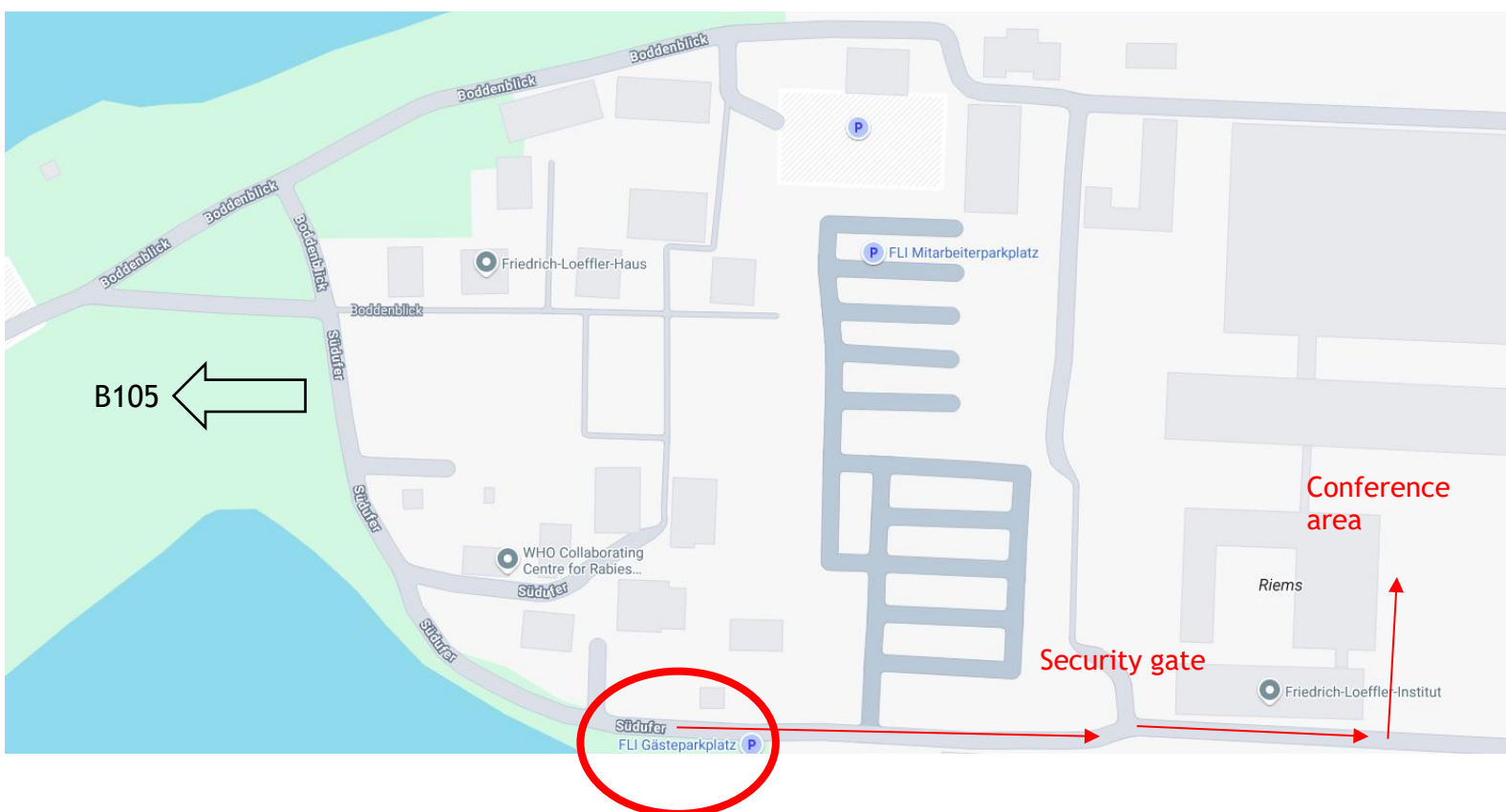
The Symposium will take place in the conference area of the Friedrich-Loeffler Institute on the Isle of Riems, Südufer 10, 17493 Greifswald-Insel Riems.

How to get there (from Greifswald)

By car:

B105 in the direction of Stralsund

Parking on the parking area for guests



By bus: Bus 127 to Riems Wendeschleife

Linie 127 Greifswald - Neuenkirchen - Riems - Stahlbrode

AVG mbH Betrieb Greifswald Land											
Verkehrstage	Montag - Freitag										
Fahrtnummer	1	3	5	7	11	9	13	17	35	37	21
Verkehrshinweise beachten			2				1 3 4 5	2	2		
Fußnoten	A	A	A	S	A F	S	S	S			S
Helmshagen..... ab	05:35										
Greifswald, ZOB (DB).....	05:45	07:15	08:12	09:42	12:07	12:17	13:17	13:17	13:17	13:17	
Greifswald, H.-Beimler-Str.....	05:51	07:23	:	:	:	:	:	:	:	:	
Greifswald, Bahnhof Süd (DB).....	05:54	07:26	:	:	:	:	:	:	:	:	
Greifswald, Ärztehaus.....	05:56	07:29	:	:	:	:	:	:	:	:	
Greifswald, Schönwalde I.....	05:58	07:30	:	:	:	:	:	:	:	:	
Greifswald, Schönwalde II.....	06:01	07:32	:	:	:	:	:	:	:	:	
Greifswald, Tolstoistraße.....	06:03	07:35	:	:	:	:	:	:	:	:	
Greifswald, Hellsinkirng.....	06:05	07:37	:	:	:	:	:	:	:	:	
Greifswald, Osttangente.....	:	:	:	:	:	:	:	:	:	:	
Greifswald, Ostseeviertel II.....	06:07	07:40	:	:	:	:	:	:	:	:	
Greifswald, Medigreif.....	:	:	:	:	:	:	:	:	:	:	13:30
Greifswald, Volksstadion.....	:	:	:	:	:	:	:	:	:	:	13:31
Greifswald, Ostseeviertel I.....	06:08	07:41	:	:	:	:	:	:	:	:	13:32
Greifswald, Ostseeviertel II.....	:	:	:	:	:	:	:	:	:	:	13:33
Greifswald, Hellsinkirng.....	:	:	:	:	:	:	:	:	:	:	13:35
Greifswald, Tolstoistraße.....	:	:	:	:	:	:	:	:	:	:	13:37
Greifswald, Volksstadion.....	06:09	07:42	:	:	:	:	:	:	:	:	
Greifswald, Am St.Georgsfeld.....	06:11	07:43	:	:	:	:	:	:	:	:	
Greifswald, An den Wurthen.....	06:12	07:45	:	:	:	:	:	:	:	:	
Greifswald, Schönwalde II.....	:	:	:	:	:	:	:	:	:	:	13:39
Greifswald, Schönwalde I.....	:	:	:	:	:	:	:	:	:	:	13:41
Greifswald, Ärztehaus.....	:	:	:	:	:	:	:	:	:	:	13:42
Greifswald, Lomonossowallee.....	:	:	:	:	:	:	:	:	:	:	13:44
Greifswald, ZOB (DB)..... an	:	:	:	:	:	:	:	:	:	:	13:56
Greifswald, ZOB (DB)..... ab	:	:	:	:	:	:	:	:	:	:	14:00
Greifswald, Bahnhofstraße.....	:	:	08:13	09:44	12:09	12:19	13:19	13:19	13:19	13:19	14:02
Greifswald, Platz der Freiheit.....	06:13	07:46	08:15	09:47	12:12	12:22	13:22	13:22	13:22	13:22	14:04
Greifswald, Steinbecker-Brücke.....	06:17	07:50	08:18	09:50	12:15	12:25	13:25	13:25	13:25	13:27	14:06
Neuenkirchen, Siedlung.....	:	:	:	09:52	12:18	12:28	13:28	13:28	13:28	13:28	14:10
Neuenkirchen, Gemverw.....	:	:	:	09:53	12:19	12:29	13:29	13:29	13:29	13:29	14:11
Neuenkirchen, Schule.....	:	:	:	:	:	12:30	13:30	13:30	:	:	14:12
Wampen..... an	:	:	:	:	:	:	:	:	:	:	14:18
Wampen..... ab	:	:	:	:	:	:	:	:	:	:	14:21
Neuenkirchen, Neubau.....	:	:	:	09:55	12:20	12:31	13:32	13:32	13:32	13:32	14:26
Neuenkirchen, EKZ.....	06:22	R 07:54	:	09:57	12:22	12:34	13:34	13:34	13:34	13:34	14:28
Mesekenhagen, Ausbau.....	:	R 07:57	:	10:02	12:27	:	:	:	13:39	13:39	:
Leist III.....	:	:	:	:	:	12:39	13:38	13:38	:	:	14:32
Leist I.....	:	:	:	:	:	12:41	13:40	13:40	:	:	14:34
Groß Karrendorf.....	:	:	:	:	:	12:44	13:43	13:43	:	:	14:37
Klein Karrendorf.....	:	:	:	:	:	12:46	13:45	13:45	:	:	14:39
Frätow.....	:	:	:	:	:	12:47	13:46	13:46	:	:	14:40
Frätow.....	:	:	:	:	:	12:48	13:47	13:47	:	:	14:41
Mesekenhagen, Schule.....	:	R 07:58	:	10:04	12:29	12:51	13:50	13:50	13:41	13:41	14:44
Mesekenhagen, Kulturhaus.....	:	R 07:59	:	10:05	12:30	12:52	13:51	13:51	13:42	13:42	14:45
Kowall.....	:	R 08:01	:	10:07	12:32	12:54	13:53	13:53	13:44	13:44	14:47
Gristow, Abzw.....	:	:	:	:	:	:	:	:	:	:	:
Gristow, Waldeslust.....	06:29	08:03	:	10:10	12:35	12:57	13:56	13:56	13:47	13:47	14:50
Gristow, Dorf.....	06:30	08:04	:	10:11	12:36	12:58	13:57	13:57	13:48	13:48	14:51
Riemserort.....	06:31	08:05	:	10:12	12:37	12:59	13:58	13:58	13:49	13:49	14:52
Riems, Yachthafen.....	06:32	08:06	:	10:13	12:38	13:00	13:59	13:59	13:50	13:50	14:53
Riems, Wendeschleife.....	06:33	08:07	:	10:14	12:39	13:01	14:00	14:00	13:51	13:51	14:54
Kirchdorf, Sportplatz.....	:	:	08:33	:	:	:	:	R 14:07	13:58	:	
Kirchdorf, B105.....	:	:	08:34	:	:	:	:	R 14:08	13:59	:	
Tremt, Hof Surbier.....	:	:	08:36	:	:	:	:	R 14:10	14:01	:	
Dömitzow, B105.....	:	:	08:37	:	:	:	:	R 14:11	14:02	:	
Reinberg, Stahlbroder Str.....	:	:	08:43	:	:	:	:	R 14:17	14:08	:	
Stahlbrode, Ausbau.....	:	:	08:45	:	:	:	:	R 14:19	14:10	:	
Stahlbrode, Zum Hafen.....	:	:	08:46	:	:	:	:	R 14:20	14:11	:	
Stahlbrode, Am Sund..... an	:	:	08:47	:	:	:	:	R 14:21	14:12	:	

A - verkehrt nicht am 24. und 31.12.

F - verkehrt nur in den Ferien

R - Bedarfshaltestelle Bedienung, nach tel. Voranm. 03834/81963 mind. 60 Min. v. Fahrtbeginn (IV

K - Kleinbus (eingeschränkte Platzanzahl)

Social Events and Dinner

Wednesday, 15/10/25

Dinner in the canteen of the FLI

This meal is included in the conference fee.

After the Dinner: Pub Quiz at the Studiclub Kiste e.V.

Makarenkostraße 49

17491 Greifswald

Organization Transport: everyone for themselves

You can bring your own food.

Thursday, 16/10/25

BBQ in the canteen

This meal is included in the conference fee.

Organization Transport: everyone for themselves

Farewell Lunch

A **Take-Away Lunch Package** will be conveniently available at the venue at noon on Friday 17th. This meal is included in the conference fee.

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.

Special Guests

- Prof. Dr. Christa Kühn - *President of the FLI*
- Dr. Christoph Staubach - *Friedrich Loeffler Institute, Institute of Epidemiology*
- Dr. Fee Zimmermann - *Helmholtz Institute for OneHealth*
- Prof. Dr. Lars Kaderali - *University of Greifswald*
- Dr. Christian Nawroth - *Research Institute for Farm Animal Biology*
- Dr. Alexandra Bahr - *Helmholtz Institute for OneHealth*

Talks

15min talk + 5min questions

Talks I, Wednesday, 15/10/25, 14:30-15:30

Nina Böttcher - *IMVZ*

"Cross-species analysis of Ebola virus and Sudan virus replication components identifies a novel polymerase-nucleoprotein interaction"

Susnato Das - *ITT*

"Influence of dietary Tryptophan supplementation on behaviour and gut microbial composition in pullets"

Gesa Krueger - *IfI*

"Deciphering responses to *Mycobacterium tuberculosis* and *Mycobacterium bovis* in a 3D model for bovine tuberculous granulomas"

Talks II, Wednesday, 15/10/25, 16:45-17:45

Assem Tara - *ING*

„Development of Highly Active Ligninase Genes for Future Ruminants with Reduced CO2 Footprint"

Nele Lechleiter - *IfE*

"Metagenomic analyses reveal seasonal dynamics and AMR presence in the gut microbiome of red deer (*Cervus elaphus*)"

FREE

Talks III, Thursday, 16/10/25, 11:00-11:45

Nils Tadewaldt - *IMED*

"Viral Interference During Flavivirus Coinfections"

Constantin Lorenz - *IVD*

"Too Weak to Protect, Too Virulent to Use: FMDV Vaccines with No Therapeutic Window"

Talks IV, Thursday, 16/10/25, 14:30-15:30

Laura Schmid - *IMVZ*

"Prefusion Stabilization of Rabies Virus Glycoprotein Impairs Viral Fusion and Enhances Safety"

Ender Menges - *IfE*

"PigVirScan: A Peptide Library for Sero-Epidemiological Surveillance of Swine Viruses"

Lina Spieß - *INNT*

"Identifying Risk Factors for Classical Scrapie in Icelandic Sheep: Results from a countrywide farm survey"

Talks V, Friday, 17/10/25, 9:15-10:15

Marie-Luise Eweleit - *IMED*

“West Nile virus in focus: Does host background influence the clinical outcome on cellular level?”

Lisa Hildebrand -*ITT*

“Keel bone fractures in low- and high-performing chicken genotypes”

Marine-Noel Klamke - *IMVZ*

“Characterization of Tacaribe virus matrix protein phosphorylation and its impact on protein functions”

“Fresh Brews and Fresh Views” (Poster Session)

This year, we would like to organise the poster session slightly differently.

As usual, your poster will be displayed during one of the poster sessions on Thursday. Rather than being required to stand next to your poster for the entire session, we encourage you to grab a tea or coffee and take a look at other posters in your session. Posters displayed close to each other cover similar topics, so this is a great opportunity to discuss your work and gain a fresh view on a related subject.

We have tried to categorise your posters. Please note that, as science is complex and often multi-directional, some posters could fit into more than one category.

Please hang your poster in your designated poster booth by **09:00 on Thursday**.

Odd numbers (e.g. 1, 3, 5...) will be presented in the early poster session (Thursday, 16 October 10:15 -11:00).

Even numbers (e.g. 2, 4, 6...) will be presented in the late poster session (Thursday, 16 October, 15:30 - 16:30).

Scientific Poster Feedback

As a member of the **FLI Junior Scientific Community**, you are encouraged to attend the **Scientific Poster Sessions**, where you will be introduced to a diverse range of topics. Keep in mind that you are free to choose which topics and posters interest you the most. This year, you have the opportunity not only to discuss any questions that arise during the sessions with the poster presenters, but also to leave a feedback note in the envelope attached to each poster. Whether you choose to provide positive feedback or suggestions for future improvements is entirely up to you. We look forward to your engagement and insights.

FLI Affiliations

- Institute for Bacterial Infections and Zoonoses (IBIZ)
- Institute of Epidemiology (IfE)
- Institute of Immunology (IfI)
- Institute of Infectology (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)

Participants

First Name	Institute	Participation Format	Number
Alina Anton	IITG/One Health	Poster	30
Stina Auerbach	IfE	Poster	16
Janine Benthin	ITT	Poster	13
Nina Böttcher	IMVZ	Talk	I
Sophia Brunnbauer	ITE	Poster	4
Susnato Das	ITT	Talk	II
Alix Drobinia	IMED	Poster	32
Marie Luise Eweleit	IMED	Talk	XI
Anna Fingerhut	ITE	Poster	3
Felix Fischer	ING	Poster	6
Nadine Heberl	ITT	Poster	12
Jonas Heck	IVD	Poster	
Janet Held	IfE	Poster	15
Maxi Hertel	ATB	Poster	29
Lisa Hildebrand	ITT	Talk	XII
Sarah Jahn	IfE	Poster	8
Jessica Junker	IfE	Poster	35
Nick Laurenz Kaiser	INNT	Poster	10
Kristina Kissner	IMVZ	Poster	24
Marine - Noel Klamke	IMVZ	Talk	XIII
Annemarie Knief	INNT	Poster	23
Marie Krebs	IVD	Poster	28
Gesa Krueger	IfI	Talk	III
Andrei Lazar	iFi	Poster	20
Nele Lechleiter	IfE	Talk	V
Constantin Lorenz	IVD	Talk	VII
Ender Menges	IfE	Talk	IX
Alaleh Mohammadi Aghbash	IMP	Poster	18
Franziska Neffgen	INNT	Poster	27
Joaquín Neumann-Heise	IITG/One Health	Poster	21
Cindy Nyanzi	ITT	Poster	11
Jessica Raabe	ITT	Poster	2
Laura Rzepa	IMVZ	Poster	26
Bidiepta Saha	IfI	Poster	37
Laura Schmid	IMVZ	Talk	VIII
Patricia Scholz	ITE	Poster	1
Louisa Schröter	IMP	Poster	38
Abhishek Sharma	IfI	Poster	19
Lina Spieß	INNT	Talk	X
Elisabeth Striese	IMED	Poster	33
Chi Mei Sun	ING	Poster	5
Nils Tadewaldt	IMED	Talk	VI
Aseem Tara	ING	Talk	IV
Nihal Telli	IMED	Poster	31

Filipe Tomaz	INNT	Poster	25
Christin Unruh	ITE	Poster	14
Josefine Wassermann	IfE	Poster	36
Judith Wedemeyer	IfE	Poster	34
Johanna Pauline Wiethoff	IITG/One Health	Poster	17
Deliah Tamsyn Winterfeld	IfE	Poster	9
Kira Wisnewski	IVD	Poster	22
Katharina Zavyalova	ITT	Poster	7

Program

Wednesday, 15/10/25

From 12:30	Arrival and Registration
13:30 - 13:45	Welcome and Opening Prof. Dr. Christa Kühn - <i>President of the FLI</i>
13:45 - 14:15	Keynote Lecture 1 Prof. Dr. Christa Kühn - <i>President of the FLI</i> "Bovine genome annotation: challenges and potential benefits"
14:30 - 15:30	Talks I <u>Nina Böttcher - IMVZ</u> "Cross-species analysis of Ebola virus and Sudan virus replication components identifies a novel polymerase-nucleoprotein interaction" <u>Susnato Das - ITT</u> "Influence of dietary Tryptophan supplementation on behaviour and gut microbial composition in pullets" <u>Gesa Krueger - IfI</u> "Deciphering responses to Mycobacterium tuberculosis and Mycobacterium bovis in a 3D model for bovine tuberculous granulomas"
15:30 - 15:45	Coffee break
15:45 - 16:30	Keynote Lecture 2 Dr. Christoph Staubach - <i>Friedrich Loeffler Institute, Institute of Epidemiology</i> tba
16:30 - 16:45	Coffee break
16:45 - 17:45	Talks II <u>Assem Tara - ING</u> „Development of Highly Active Ligninase Genes for Future Ruminants with Reduced CO2 Footprint" <u>Nele Lechleiter - IfE</u> "Metagenomic analyses reveal seasonal dynamics and AMR presence in the gut microbiome of red deer (Cervus elaphus)"
18:00 - 19:45	Dinner in the canteen
20:30 - 22:00	Pub Quiz at the „Studiclub Kiste e.V.

Thursday, 16/10/25

09:00 - 09:30	Welcome, Information, Poster hanging
09:30 - 10:15	Keynote Lecture 3 Dr. Fee Zimmermann - <i>Helmholtz Institute for OneHealth</i> “One Health Exploratories - Developing ecology informed solutions for public health”
10:15 - 11:00	"Fresh Brews & Fresh Views"
11:00 - 11:45	Talks III <u>Nils Tadewaldt - IMED</u> “Viral Interference During Flavivirus Coinfections” <u>Constantin Lorenz - IVD</u> “Too Weak to Protect, Too Virulent to Use: FMDV Vaccines with No Therapeutic Window”
11:45 - 12:45	Picture + guided Island Tour
12:45 - 13:30	Lunch
13:30 - 14:15	Keynote Lecture 4 Prof. Dr. Lars Kaderali - <i>University of Greifswald</i> “Applications of AI in Virology“
14:15 - 14:30	Short Coffee Break
14:30 - 15:30	Talks IV <u>Laura Schmid - IMVZ</u> “Prefusion Stabilization of Rabies Virus Glycoprotein Impairs Viral Fusion and Enhances Safety” <u>Ender Menges - IfE</u> “PigVirScan: A Peptide Library for Sero-Epidemiological Surveillance of Swine Viruses” <u>Lina Spieß - INNT</u> “Identifying Risk Factors for Classical Scrapie in Icelandic Sheep: Results from a countrywide farm survey”
15:30 - 16:30	"Fresh Brews & Fresh Views"
16:30 - 17:15	Keynote Lecture 5 Dr. Christian Nawroth - <i>Research Institute for Farm Animal Biology (FBN)</i> “Farm animal cognition: How Smart Are Goats, and Why Should We Care?”
17:00 - 17:30	Poster feedback and ranking
17:45 - 22:00	BBQ in the canteen

Friday, 17/10/25

09:00 - 09:15	Welcome
09:15 - 10:15	Talks V <u>Marie-Luise Eweleit - IMED</u> “West Nile virus in focus: Does host background influence the clinical outcome on cellular level?” <u>Lisa Hildebrand - ITT</u> “Keel bone fractures in low- and high-performing chicken genotypes” <u>Marine-Noel Klamke - IMVZ</u> “Characterization of Tacaribe virus matrix protein phosphorylation and its impact on protein functions”
10:15 - 10:30	Coffee break, Talk ranking
10:30 - 11:15	Keynote Lecture 6 Dr. Alexandra Bahr - <i>Helmholtz Institute for OneHealth</i> “From Local Observations to Global Insights: Why AMR Surveillance Matters in the One Health Era”
11:15 - 11:45	Selection of the new PhD representation Poster/Speaker Price
From 12:00	Lunch packages and Farewell

“Fresh Brews and Fresh Views” Schedule (Poster Session)

For detailed information, please see “Fresh Brews and Fresh Views” (Poster Session) in the Chapter “General Information”

Odd numbers (e.g. 1, 3, 5...) will be displayed in the early poster session (Thursday, 16 October 10:15 -11:00).

Even numbers (e.g. 2, 4, 6...) will be displayed in the late poster session (Thursday, 16 October, 15:30 - 16:30).

Name	Poster Number	Name	Poster Number
Patricia Scholz	1	Andrei Lazar	20
Jessica Raabe	2	Joaquin Neumann-Heise	21
Anna Fingerhut	3	Kira Wisnewski	22
Sophia Brunnbauer	4	Annemarie Knief	23
Chimei Sun	5	Kristina Kissner	24
Felix Fischer	6	Filipe Tomaz	25
Katharina Zavyalova	7	Laura Rzepa	26
Sarah Jahn	8	Franziska Neffgen	27
Deliah Tamsyn Winterfeld	9	Marie Krebs	28
Nick Laurenz Kaiser	10	Maxi Hertel	29
Cindy Nyanzi	11	Alina Anton	30
Nadine Herberl	12	Nihal Telli	31
Janine Benthin	13	Alix Drobinia	32
Christin Unruh	14	Elisabeth Striese	33
Janet Held	15	Judith Wedemeyer	34
Stina Auerbach	16	Jessica Junker	35
Johanna Pauline Wiethoff	17	Josefine Wassermann	36
Alaleh Mohammadi Aghbash	18	Bidiepta Saha	37
Abhishek Sharma	19	Louisa Schroeter	38

Explanation

Animal Nutrition
Animal welfare and
behaviour
Viruses & Screening

Genetics
Animal husbandry and
biosecurity
Bacteria & Parasites

AI/ Machine Learning
Immunology

Abstracts

Oral Presentations

1. Cross-species analysis of Ebola virus and Sudan virus replication components identifies a novel polymerase-nucleoprotein interaction

Nina Böttcher, Jana Schmitz, Falk Butter, and Thomas Hoenen

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Many orthoebolaviruses cause severe hemorrhagic fever with high case fatality rates. The best-known of these is Ebola virus (EBOV), whose detailed characterization has led to the successful development of approved vaccines and therapeutics. However, for other closely related species, such as Sudan virus (SUDV), such countermeasures do not yet exist, and there are considerable knowledge gaps with respect to the details of their biology. Therefore, we sought to characterize the understudied SUDV. As a first step towards this, we developed and optimized reverse genetics-based molecular tools, including both life cycle modeling (LCM) systems and systems for the rescue of recombinant viruses. The application of these LCM systems, in particular, have considerable potential to contribute to the elucidation of details of the molecular biology of SUDV, as they allow us to dissect the viral life cycle into its individual steps under BSL1 conditions. Specifically, we have used these systems to compare the requirements and interspecies compatibilities of the ribonucleoprotein complex (RNP) components required for viral RNA synthesis of EBOV and SUDV. Using T7-based monocistronic minigenome systems, we showed that EBOV and SUDV RNP proteins are largely compatible across species and can, in most cases, be interchanged without consequence. However, we identified an incompatibility between the EBOV polymerase L and the SUDV nucleoprotein NP. Chimeric NP proteins carrying the EBOV-specific N-terminal region of NP could rescue viral RNA synthesis. To narrow down the involved regions within NP, further deletional studies were performed, which demonstrated that the NP region between amino acids 240-517, encompassing the second lobe within the NP-NTD, appears to be responsible for the observed interspecies incompatibility. This finding suggests a novel interaction between L and NP involving the N-terminal region of NP.

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2. Influence of dietary Tryptophan supplementation on behaviour and gut microbial composition in pullets

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Tryptophan (Trp) is an essential amino acid which is metabolized to e.g. Serotonin and Kynurenine, either peripherally or after transport to the brain. These metabolites act as neuromodulators, thereby affecting behaviour in e.g. pigs and broilers. Moreover, the gut microbiota is known to affect peripheral Tryptophan metabolism, indirectly affecting the neuromodulatory functions of Tryptophan metabolites. The present study explored the effect of dietary Tryptophan supplementation on the interaction between the gut microbiota and behaviours including fearfulness, locomotor activity and sociality during pullet development. Two-hundred one-day-old chicks (brown/white hybrids) were fed either a standard 0.2% Tryptophan or a surplus 0.5% Tryptophan diet until week-of-life (wol) 23. Each treatment (hybrid × Trp-concentration) consisted of 5 pens with 10 hens each. Weekly feed consumption measurements confirmed that the surplus groups ingested 2-3 times more Tryptophan than the standard groups. To monitor development, body weight was recorded repeatedly (wol: 6, 9, 12, 15). Tonic immobility tests were conducted on 140 hens (= 7 per pen) at three timepoints (wol: 6, 9, 12) to measure fearfulness. Over three periods (wol: 7-8, 10-11, 13-14), locomotor activity of hens in their home pens was assessed using an electronic antenna-transponder system and sociality through video observations. Corresponding to behavioural measures, blood samples were collected from the same hens (wol: 6, 9, 12) to determine peripheral concentrations of Tryptophan, Serotonin and Kynurenine through ELISAs. At the same timepoints, faecal samples were collected to sequence the gut microbiome. Data were analyzed using linear mixed effects models. Data on peripheral concentrations, fearfulness and locomotor activity have been analyzed and will be presented. In a nutshell, Tryptophan supplementation in the present study increased peripheral concentrations of its metabolites but had only limited effects on behaviour, possibly owing to limited effects on metabolite concentration in the brain.

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3. Deciphering responses to *Mycobacterium tuberculosis* and *Mycobacterium bovis* in a 3D model for bovine tuberculous granulomas

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Tuberculosis (TB) remains a threat for human and livestock health. Mycobacteria causing TB are host-adapted pathogens which occasionally spillover to other species. *Mycobacterium bovis* (Mbv) causes bovine TB, a well-known zoonosis. *Mycobacterium tuberculosis* (Mtb) is adapted to humans, may trigger symptomatic infection in cattle yet these are resistant to experimental challenge. A hallmark of TB in all hosts are multicellular tissue lesions termed granulomas. We designed a new three-dimensional model termed in vitro granuloma-like structures (IVGLS) which resemble different stages of granulomas. We aimed to investigate early dynamics of the TB granuloma, defining trajectories and microenvironments of IVGLS elicited by Mbv and Mtb. The matrix-free spheroids contain bovine monocyte-derived macrophages alone, resembling an early stage of TB granulomas, or also lymphocytes, reflecting a mature stage. Using host-adapted Mbv and host-unadapted Mtb, we observed different replication patterns of Mtb and Mbv. Mtb replicated within the IVGLS whereas the Mbv stopped replicating early post-infection. Abundances of reactive oxygen species and cell viability were similar, yet Mbv triggered higher and earlier interferon-gamma responses as well as prompt production of nitric oxide. Transmission electron microscopy revealed a higher cell density in Mbv-IVGLS indicating remodeling of cell-cell interactions in case of host-adapted bacteria. Taken together, IVGLS facilitate a deepened understanding of intra-granulomatous responses against host-adapted mycobacteria. These insights are essential for the development of vaccines for cattle and interventions such as host-targeted therapies for humans.

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4. Development of Highly Active Ligninase Genes for Future Ruminants with Reduced CO2 Footprint

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The open-field burning of agricultural residues such as rice straw is a widespread practice, particularly in East Asia. While it offers a quick and inexpensive way to clear fields, it releases greenhouse gases and toxic by-products, contributing significantly to environmental pollution. An alternative strategy is to repurpose straw as animal feed, which would reduce the dependence on quality cereals for animal

fodder. However, the high lignin content in straw restricts its digestibility due to its association with and complexation of cellulose and hemicellulose. Since the rumen microbiome does not produce lignolytic enzymes, our project aims to engineer transgenic goats that secrete optimized ligninase through their saliva, enabling efficient breakdown of lignocellulose. This approach would not only improve straw digestibility for ruminants but also offers a sustainable solution to reduce CO₂ emissions from straw burning. We are leveraging AlphaFold, an AI-based protein structure prediction tool, to optimize the evolutionary procedure for ligninase gene. This involves analysis of several hundred fungal and bacterial ligninase genes and their comparison with the previously validated Dyp1 gene (Hyder and Kues, 2023). The goal is to identify conserved enzymatic pockets critical for activity and select the most efficient sequence for ligninase activity, which will then be tested for functionality in vitro. Simultaneously, we are working towards the characterization and immortalization of fetal goat tongue epithelial cells (ZZ-R 127), aiming to induce pluripotency and differentiate them into salivary gland-like cells. These will serve as a physiologically relevant in vitro model for testing ligninase expression and activity. The two collaborating partners in India\$, involved in this BMGF-funded project will further develop transgenic goats expressing ligninase and conduct physiological studies and straw feeding trials. By combining machine learning with genetic engineering, this work is expected to provide a data-driven approach to improving ruminant nutrition and sustainability in livestock production.

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Reference: Hyder I, Kues WA 2023. Transgenic mammalian salivary cells expressing ligninase as a proof-of-concept model for enhanced lignocellulose degradation to generate future resilient livestock. J Cleaner Production 293, 136226.

\$Funding: Bill and Melinda Gates Foundation - Grant to Dharmendra Kumar, Iqbal Hyder, Wilfried A. Kues

5. Metagenomic analyses reveal seasonal dynamics and AMR presence in the gut microbiome of red deer (*Cervus elaphus*)

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Background As the largest native mammal in Germany, Red deer (*Cervus elaphus*) is a culturally and economically important game species. With the growing concerns about antimicrobial resistances (AMR) worldwide, wild animals are increasingly considered as an element in the maintenance and dispersal of AMR-carrying microorganisms. The gut microbiome of red deer, for instance, has been shown to harbour resistance genes. Since the composition of the microbiome is dependent on environmental factors, we sampled hunt-harvested red deer in one location over the course of a year to investigate the dynamics of the microbiome and resistome by shotgun metagenomic sequencing. Results and Discussion 76 animals were sampled from August 2023 to August 2024. We found significant differences of the bacterial

composition between summer and autumn but no differences in the resistome. The bacterial composition was similar to that found in other investigations on red deer but showed a high amount of rare species in the microbiome. Bacillota dominated in most samples with 41% (± 8), on family level, Oscillospiraceae (18.85% (± 6)), Lachnospiraceae (6.83% (± 2.06)) and Clostridiaceae (5.41% (± 6.4)) mostly represented this Phylum. Normalized abundance of resistance gene reads was 0.17 ± 0.81 , which is in line with similar investigations. Over 50% of the resistance gene reads was assigned to the class of Macrolides, lincosamides and streptogramins. The observed difference between the seasons might be caused by a shift in taxa of low abundance, while a stable core microbiome, relating to the unchanged abundance of AMR genes, persists year-round.

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6. Viral Interference During Flavivirus Coinfections

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Superinfection exclusion describes the phenomenon of closely related viruses being unable to simultaneously coinfect the same cells. Despite being reported across a wide range of virus families, no general mechanism responsible has been identified. To study how closely related viruses interact, it's often necessary to use tagged or otherwise genetically modified viruses, since classic detection methods like antibodies or RNA/DNA probes are usually not specific enough to differentiate between them. This, however, comes with potential caveats: Depending on the virus used, tagging it can lead to reduced replication rates, which could influence viral interactions. Additionally, any genetic modifications to a virus's genome, even if not influencing its replication rate, could influence its interaction with the host or other viruses. In this study, we describe an antibody-based staining method capable of differentiating the two closely related mosquito-borne Orthoflaviviruses WNV and USUV without any genetic modifications. We use it to characterize their interference with one another during simultaneous and sequential coinfections, finding evidence for superinfection exclusion occurring between them. We supplement our findings with qRT-PCR showing that reduced viral infection rates translate to reduced genome replication.

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7. Too Weak to Protect, Too Virulent to Use: FMDV Vaccines with No Therapeutic Window

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Foot-and-mouth disease (FMD) is one of the most economically significant viral diseases in livestock farming worldwide. Inactivated whole-virus vaccines have

historically enabled eradication of the disease in many industrialized countries. Recent outbreaks in Europe prove that re-entry of FMDV into formerly free countries can happen at any time with rapid spread and significant economic damage. While inactivated vaccines are effective, they can be expensive and dangerous to produce en masse. Live attenuated vaccines (LAV) may overcome these disadvantages and are a promising strategy for controlling FMDV. However, attenuation must be balanced between safety and sufficient immunogenicity. We evaluated the efficacy and safety of two rationally designed, genetically engineered LAV candidates for FMDV in pigs: a codon pair deoptimized (CPD) virus and a high-fidelity polymerase (HiFi) mutant with a reduced replication error rate. The CPD virus, administered at 10^5 TCID₅₀ per animal (n=6) intradermally into the heel bulb, failed to establish detectable infection or induce seroconversion. A higher dose of 4.7×10^6 TCID₅₀, however, successfully established infection and caused clinical FMD and virus shedding indistinguishable from wild type viruses, indicating that the attenuation was insufficient at immunogenic doses. The HiFi mutant, tested at 10^5 TCID₅₀ per animal (n=6) via the same route, resulted in clinical disease in 50% of the animals. Clinical signs and virus shedding again mirrored wild-type infection. However, two of the infected animals exhibited a delayed disease onset. The other half of the group remained uninfected and seronegative throughout the study. Overall, both vaccine candidates demonstrated a lack of a viable immunization window: ineffective at lower doses, but pathogenic at higher ones. These findings highlight the limitations of single-mechanism live attenuation strategies for FMDV and suggest that future approaches will require more sophisticated, multi-layered attenuation to achieve both safety and protective efficacy in natural hosts.

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8. Prefusion Stabilization of Rabies Virus Glycoprotein Impairs Viral Fusion and Enhances Safety

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Rabies virus (RABV) is a neurotropic virus causing lethal encephalitis in humans and other mammals. Viral entry depends on the RABV glycoprotein (G), which undergoes pH-dependent conformational changes to mediate membrane fusion after endocytosis. The prefusion conformation of RABV-G is the principal target of neutralizing antibodies and therefore a focus for vaccine design. While stabilizing G in the prefusion state inhibits membrane fusion in cell-cell assays, its effect in the context of infectious virus remained unclear. To address this, we engineered a recombinant RABV, “SAD lock”, with a double mutation in G (H261L;H270P) to stabilize the prefusion state and assessed its impact on viral entry, infectivity in cell culture but also immunogenicity and safety in an animal model. SAD lock could bind and enter cells but failed to mediate fusion or spread. Complementation with a heterologous (VSV) G enabled virus amplification. However, upon passaging, escape mutants emerged that had restored fusion activity without reverting to histidine residues, indicating alternative fusion pathways. In BALB/c mice, SAD lock exhibited

a markedly improved safety profile compared to the live attenuated vaccine strain SAD L16. All SAD lock-inoculated mice survived both intramuscular (i.m.) and intracerebral (i.c.) inoculation, whereas all i.c. infected SAD L16 mice succumbed to infection. Notably, despite being replication-deficient, SAD lock induced a robust antibody response similar to that of SAD L16. Our findings provide insight into RABV fusion mechanisms and support the potential of incorporating prefusion-stabilized G into live virus vaccine platforms for safer rabies vaccines with robust immunogenicity.

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9. PigVirScan: A Peptide Library for Sero-Epidemiological Surveillance of Swine Viruses

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Immune responses play a crucial role in defining a pig's susceptibility to viral infections. Following exposure to viruses, specific subsets of B cells develop as part of the immune response to produce antibodies that bind unique viral structures, known as epitopes. These specific antibodies are secreted into the blood and facilitate immunity against encountered viruses. Conventional serological tests examine single virus-antibody-interactions, limiting their ability to reveal the full landscape of viral infections within livestock populations. As a result, it remains unknown which other viruses are circulating and which viral structures are actually targeted by antibodies found in convalescent animals. Here we design a library of peptide sequences from porcine viruses for the comprehensive characterisation of antibody repertoires in pig populations. We first conduct a systematic literature review to identify currently known pig-infecting viruses and extracted over 50,000 literature records using a large language model. Of the identified pig viruses, we then retrieve viral proteomes from public repositories for phylogenetic analysis. Representative proteomes are chosen for each virus and bioinformatically broken down into small peptides, which are further selected to minimise sequence homology among all generated peptides. As a result, we created the PigVirScan library, a pool of short peptide sequences, that resemble all linear B cell epitopes and cover the entire known pig virome. PigVirScan can now be used to develop an advanced sero-epidemiological diagnostic tool by utilising a high-throughput sequencing technology (phage immuno-precipitation sequencing, PhIP-seq) that can detect humoral immune response profiles to a wide range of viruses. This information enables comprehensive surveillance, supports effective vaccination strategies and disease management practices that sustain resilient pig populations. This project, part of the European Partnership on Animal Health and Welfare, is co-funded by the European Union's Horizon Europe Project 101136346 EUPAHW.

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10. Identifying Risk Factors for Classical Scrapie in Icelandic Sheep: Results from a countrywide farm survey

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Classical Scrapie is a progressive and fatal prion disease affecting sheep. It poses an ongoing challenge to animal health and threatens the long-term sustainability of sheep farming in Iceland. Despite control strategies being in place for decades, new outbreaks continue to emerge in certain regions. This study aims to identify potential transmission routes and contributing risk factors by analysing data from a nationwide questionnaire survey that includes of both Scrapie-affected and unaffected farms. Between April 2024 and April 2025, sheep farmers from four distinct regions in Iceland participated in the survey. The survey gathered information on land use, flock size, seasonal management practices, livestock movements, the use of communal grazing areas, human interactions and biosecurity measures. Regional comparisons were included to identify area-specific risk factors. The descriptive analysis highlights several practices that are associated with an increased disease risk. These include frequent contact with external personnel, the use of shared machinery, animal movements between farms and the use of common mountain pastures with communal sheep gatherings. Larger flock sizes and on-farm carcass disposal are also more common among affected farms. In contrast, protective factors include limited contact with external people and animals, the use of artificial insemination, genetic selection for resistance traits, and secure carcass disposal. Regional differences in farm structure and the level of biosecurity also appear to influence the spread of disease. Overall, the findings highlight the importance of enhancing on-farm biosecurity, supporting resistance breeding initiatives, ensuring safe carcass management, and raising regional awareness of particular risk factors, with the aim of strengthening future eradication strategies for Classical Scrapie in Iceland.

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11. West Nile virus in focus: Does host background influence the clinical outcome on cellular level?

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The mosquito-borne West Nile virus (WNV) belongs to the Flaviviridae family. It circulates between mosquitoes and birds, the virus reservoir. Horses and humans are dead-end hosts. Disease progression varies between hosts. Some birds show no

symptoms, while others die from multiple organ failure. Most infected mammals are asymptomatic, but some develop neurological diseases like encephalitis or meningitis, which can be fatal¹. This project aims to investigate possible reasons at cellular level for different host reactions to a WNV infection. Kidney cell lines were analyzed from three different hosts: human, horse, and goose. Geese were chosen for their high viremia among German poultry². Vero, HEK-293, and GN-R cells show high WNV titers after 24 hpi, whereas virus titer in PFN cells is tenfold lower, indicating less efficient virus replication in horse cells. This observation is supported by immunofluorescence and transmission electron microscopy. Virus genome detection by qPCR shows the same tendency having a guess that maturation of infectious particle is comparable in all cell lines. As expected, virus particles could be detected in endoplasmic reticulum and Golgi apparatus. Mass spectrometry based proteome analysis of WNV-infected Vero cells showed the upregulation of components of signal transduction pathways targeting antiviral immune response, cytokine production and metabolism of small molecules. Infection of kidney cells resulted in weaker response, however, a small but detectable increase of immunological factors was noticed. With this study we investigated kidney cell lines originating from the three main host organisms of WNV for the first time and shed light on possibly different regulatory mechanisms against WNV. 1Byas, A. D. et al. (2020): Comparative pathology of West Nile virus in humans and non-human animals. *Pathogens*. 9: 48. 2Holicki, C. M. et al. (2020): Pathogenicity of West Nile Virus Lineage 1 to German Poultry. *Vaccines*. 8: 1-22.

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12. Keel bone fractures in low- and high-performing chicken genotypes

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Skeletal integrity is an important animal welfare measure in laying hens. It has been suggested that the selection for laying performance was accompanied by physiological changes that increase the susceptibility to keel bone fractures in modern laying hens.

Therefore, the aim of our study was to compare two low-performing chicken genotypes, Sumatra (Su) and Red Junglefowl (RJF), with the high-performing hybrid layer Lohmann Selected Leghorn (LSL) in terms of keel bone damage and potentially related blood and bone parameters.

The animals were housed in small groups of 11 to 20 hens accompanied by at least one rooster. X-ray and blood sampling took place in the 16th, 25th, 33rd, 50th/52nd, and 70th/72nd weeks of age (WOA). The evaluation of the X-ray images involved assessing keel bone fractures, radiographic density, and ossification. The plasma concentrations of reproductive hormones, calcium, and phosphate were measured using commercial ELISA kits and a clinical chemistry analyzer.

Keel bone fractures were found in LSL hens, but not in the other two genotypes. Ossification was completed significantly earlier in RJF and LSL than in Su ($p < 0.05$). Radiographic density increased over the first three sampling dates in all three genotypes. Taking into account the time when the first egg was laid (LSL: 18th WOA, RJF: 24th WOA, Su: 31st WOA), this suggests that keel bone ossification and mineralization were more advanced in RJF and Su at the onset of the laying than in LSL, which could affect susceptibility to fractures later on. The statistical analysis of the blood parameters is in progress. Further analysis will show whether differences in laying performance and onset of lay are reflected in the plasma levels of reproductive hormones. Overall, the study contributes to the understanding of internal influencing factors which may help to reduce keel bone fractures in the long term.

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13. Characterization of Tacaribe virus matrix protein phosphorylation and its impact on protein functions

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Arenaviruses are important zoonotic pathogens capable of causing severe disease in humans. Despite their clinical significance, these viruses are structurally simple, encoding only 4 viral proteins. As a result, each protein has to fulfill multiple roles during the viral life cycle. One such protein, the matrix protein Z, is essential for viral assembly, nucleocapsid trafficking, and viral budding. Additionally, Z inhibits viral polymerase activity and antagonizes the host interferon response. However, the regulatory mechanisms that control these diverse functions remain poorly understood. Post-translational modifications (PTMs) are commonly employed to regulate protein activity. To explore potential PTMs of the Z protein, we generated arenavirus virus-like particles and also infected HEK-293T cells. Using mass spectrometry combined with phospho-enrichment techniques, we identified extensive phosphorylation of Z, particularly within its flexible N- and C-terminal arms, regions known to mediate protein-protein interactions. To investigate the functional impact of these phosphorylated sites, we performed site-directed mutagenesis and assessed the effects of these mutations on Z's known activities. This included conventional budding assays and life-cycle modeling systems. Through these approaches, we identified three phosphorylation sites, Y38, Y57, and T82

which are important for virus-like particle production and the release of infectious particles in a trVLP system. These mutations were also associated with reduced localization to the plasma membrane, as observed via immunofluorescence assays, suggesting disrupted intracellular trafficking. Moreover, mutations at Y57 and T82 altered the incorporation of nucleoprotein into VLPs, further highlighting the functional importance of these phosphorylation events. Collectively, our results demonstrate that phosphorylation at multiple sites across the Z protein significantly influences its activity, contributing to a broader understanding of how arenaviruses regulate their multifunctional proteins.

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Poster Presentation

1. Evaluation of pea protein as an alternative to soya in the diet of fattening Holstein bulls: effects on growth performance, feed intake, health and metabolism

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Adequate supply of protein for agricultural livestock can currently only be ensured with imported soya resulting in deforestation and high carbon footprints. At the same time, there is a growing demand for sustainable agriculture, climate and environmental protection, and animal welfare, including GMO-free diets. There are national efforts for a protein crop strategy in response to this, which calls for increased use of alternative and regional protein feed crops, among other things. This two-part study aimed to evaluate the long-term effects of using peas as an alternative to soya in concentrate feed (CF) on the growth performance, feed intake, health and metabolism of Holstein bulls from rearing to fattening, with a particular focus on the fattening period. Forty-nine male calves were randomly assigned to one of four feeding groups, receiving either skimmed or whole milk-based milk replacers, and either a soy- or pea-based CF. At 230 days (± 28 d) of age, all bulls started an 11-week fattening trial. They were fed a total mixed ration (TMR) of 70 % maize silage and 30 % CF ad libitum. Their individual feed and water intake was recorded daily with weighing troughs. Live weight, growth parameters, faeces, urine and saliva were collected before the start, after five weeks and at the end of the fattening trial. Blood samples were collected before the start and six times during the trial, at 2-weekly intervals. Feed components and TMR were regularly sampled for determination of dry matter and Weender analyses. Current evaluations aim to gain insight into the long-term dietary effects of feeding either soya or peas to bulls, considering feed intake and efficiency, growth performance and health parameters such as haematology, liver metabolism, and the endocrine system, based on the assumption that peas are an equivalent alternative to soya.

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2. Enrichment: Texture of pecking blocks seems to influence the behavior of male fattening turkeys (*Meleagris gallopavo* f. dom.)

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A major problem in fattening turkey husbandry is injurious pecking by conspecifics, frequently leading to high amounts of injured birds and deaths. This behavior can be

attributed to various causes. On the one hand, the low-stimulus environment can lead to turkeys pecking conspecifics out of boredom. On the other hand, the onset of sexual maturity causes hierarchical disputes that can result in severe agonistic interactions between turkeys. Common practice is to provide pecking blocks (PB) as enrichment material to divert the birds' attention from conspecifics. But, previous observations at the FLI-ITT showed a low consumption of PB with high hardness during the fattening phase. Therefore, this study intends to examine the acceptance of male fattening turkeys (B.U.T 6, not beak-trimmed) towards PB with low hardness in comparison to usually hard textured used (both PB by Vilovoss, Deutsche Vilomix Tierernährung GmbH, Neuenkirchen-Vörden, Germany). Turkeys were kept at the FLI-ITT for 20 weeks according to the "Federal Guidelines for a Voluntary Agreement on the Husbandry of Turkeys for Fattening". In five trials, a total of 2,356 day-old chicks were randomly divided into groups of either of 10, 33 or 44 birds per pen (floor area: 18 m²). 2,001 animals were offered PB of a higher hardness grade (PECKStone "fresh"). PB of a lower hardness grade (PECKStone "MeidArom") were used for 355 turkeys. Low hardness PB seem to be a much more attractive enrichment device for the turkeys. Preliminary results show a significantly higher consumption ($P < 0.001$) of low hardness PB ($M = 150$ g/bird) compared to hard textured PB ($M = 10.2$ g/bird). However, the prevalence of injured turkeys caused by pecking was reduced in the first half of the fattening period, when PB of lower hardness were used. Further trials will examine the reproducibility of these preliminary results.

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3. Physiological and metabolic adaption of lactating dairy cows to an increase or a decrease in the proportion of concentrate in the ration

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Change in dietary energy content can have immediate effects on health, performance and energy metabolism of dairy cows. The project aimed to investigate how these physiological parameters adapt following a sudden change in energy supply via the concentrate feed, focusing on measurable physiological and metabolic indicators.

A six-week feeding trial was conducted at the Experimental Station of the Institute of Animal Nutrition, Friedrich-Loeffler-Institut, Braunschweig with 46 lactating Holstein cows that were fed according to two different feeding regimes aimed at changing their energy intake via their ration. One group (HE/LE) received a ration of 11.45 MJ ME/kg dry matter (DM), which was reduced to 10.80 MJ ME/kg DM. The other group (LE/HE) received a ration of 10.85 MJ ME/kg DM, which was changed to 11.5 MJ ME/kg DM.

Daily feed intake and milk yield were documented individually. Blood samples for clinical-chemical analysis were collected before the ration change, after a three-week adaption period and again two weeks later.

Feed change resulted in a significant, contrasting DM intake in both groups, with stable uptake in the LE/HE group after three weeks, a week ahead of the other group. In the HE/LE group, a significant decrease in milk yield was observed immediately after the ration change, which remained constant over time. Conversely, the LE/HE group responded with a significant increase in milk yield only in the second week after the feed change, with variations over time. In the HE/LE group, non-esterified fatty acids remained unchanged, while levels decreased in the LE/HE group at week 5, causing significant group differences. At the same time, BHB increased and glucose decreased in HE/LE, highlighting marked metabolic divergence between groups.

These findings suggest that metabolic adaptations in response to a change in ration were more pronounced in the energy-restricted group and varied according to group and time.

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4. Impact of selected algae species on ruminal methanogenesis in vitro using the ANKOM gas production system

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The rapid increase in global greenhouse gas emissions, dominated by methane (CH₄) and carbon dioxide (CO₂), is making it crucial for environmental protection to find sustainable mitigation strategies. An emerging approach might be the use of potential feed materials that could influence the ruminal microbiome and therefore might have an impact on methane mitigation. This study aimed to investigate the effect of six different regional and local cultivable algae species on methane and total gas production in ruminants, using the in vitro ANKOM gas production system. All algae were tested incorporated into a dairy cow diet (TMR), consisting of 60% roughage (70% maize silage and 30% grass silage) and 40% concentrate, in a concentration of 4% of the dry matter (DM). Test substrates consisted of 1 g of the base TMR with an addition of either 4% DM of the different algae (A1-6) or 4% DM of the TMR itself in the control (CON) and were incubated for 48 hours per run in buffered rumen fluid, which was previously collected from three fistulated German-Holstein cows that were adapted to a ration similar to the TMR for at least two weeks. The ruminal fluid was pooled and added to the buffer solution at a concentration ratio of 1:4. Each substrate (A1-6; CON) was tested in triplicate per run, doing four runs in total. During incubation, cumulative gas pressure was measured continuously, while gas composition, pH, volatile fatty acid concentration, and apparent dry matter degradability (ADMD) were measured after 48 hours.

Preliminary results indicate that there were no significant changes ($p < 0.05$) in gas- and methane production after 48 hours of incubation.

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5. Genetics Underlying Growth Curve Characteristics in German Local Chicken Breeds

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Biodiversity has long been global concern, especially in light of increasing environmental dynamics. Although the high efficiency of commercial chicken breeds plays a crucial role in poultry industry, there has been a growing shift towards understanding and integrating local breeds, who are rich in genetic diversity, which offers critical resilience for future breeding strategies. The RegioHuhn project, which is dedicated to preserving and revitalizing local chicken breeds by developing economically viable dual-purpose animals, rather than conserving these breeds in isolation, aims to support this concept. RegioHuhn promotes a functional approach, to strategically cross local breeds with high-performance commercial parental lines and explore their economic values through niche markets focused on organic and sustainable production in general. In this study, we analysed growth performance in three local chicken breeds, the Altsteirer, Bielefelder, and Ramelsloher, using longitudinal body weight measurements. Individual growth curves were fitted with the Gompertz model to derive key parameters: asymptotic weight, growth rate, and age at inflection. Principal component analysis of these parameters reveals partial separation between the three breeds. To explore the genetic architecture underlying these parameters, we conducted both single-trait GWAS and multi-trait metaGWAS, identifying significant variants on several chromosomes. Genomic best linear unbiased prediction models were used to estimate parameters heritabilities via different settings. Despite relatively high standard errors due to the limited sample size, estimated heritabilities ranged from 0.04 to 0.69 (0.2 on average), highlighting the potential for within-breed selection while maintaining genetic diversity.

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6. Towards a Stillbirth Monitoring System for Dairy Cattle in the MGPT Project

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Animal health and the welfare of farm animals are increasingly coming into the focus of public attention. In particular, the stillbirth rate in dairy cows is being critically examined, as its causes have so far not been sufficiently clarified on a broad scale. In this context, breeding strategies are being increasingly discussed. To tackle the problem, a reliable data basis needs to be created in order to better understand phenotypic and genotypic causes of stillbirths and to identify risks, e.g. deleterious mutations which need to be identified at an early stage before they can spread unnoticed within the population. At the same time, specific challenges arise in collecting such data, as stillborn calves have no economic value for the farmer and therefore in-depth investigations or genotyping are often not carried out. Additionally, the causes of stillbirths are of varying aetiology and therefore require a comprehensive and differentiated recording of relevant influencing factors. In this context, the project MGPT (Monitoring of Genetic and Phenotypic Trends) aims in establishing a detailed monitoring system of stillborn calves in the German Holstein Friesian dairy cattle population. Central phenotypic characteristics, such as time of death (intra partum or post partum), the course of delivery, birth weight, and the body condition of the dam are systematically recorded on farms participating in the project via a mobile app. The data collection is significantly more detailed than the five-level ordinal scale previously used in practice for assessing calving difficulty. This allows for classification based on potentially common causes. In addition, farms systematically collect data on further environmental factors and operational characteristics such as housing, calving supervision and management, as well as feeding during the dry period. This is accompanied by SNP-genotyping stillborn calves (their parents are genotyped routinely). Based on this information, phenotypic and genotypic causes of stillbirth shall be identified, and a prospective risk monitoring system for defective alleles shall be developed, enabling early breeding intervention.

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7. Automated Detection of Insufficient Stunning in Pigs: Evaluating (Un)consciousness Indicators for AI Applications

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Failures during the stunning of animals for slaughter cannot be completely avoided, even under the best technical conditions. Therefore, it is crucial to swiftly identify animals that have retained consciousness or are at risk of regaining it, in order to minimize pain, fear, distress and suffering by applying immediate re-stunning. Video-based detection methods, especially when using deep learning-based computer vision, show promising results for animal monitoring in various agricultural applications, including the monitoring of stunning effectiveness during slaughter. However, to our knowledge none of the commercially available automated solutions have been scientifically validated yet. Any such validation and / or developing new prototypes must be preceded by a comprehensive literature study, the results of which can be found in the current presentation.

There are a number of indicators for assessing (un)consciousness in pigs during / after stunning and at slaughter. These indicators - such as vocalizations, the presence of a corneal reflex, and gasping - vary in their timing and probability of occurrence as well as discriminatory power between consciousness and unconsciousness. They also vary in how feasible they are for detecting insufficient stunning. This is because video-based detection does not involve handling the animals, and thus indicators based on human intervention (such as inducing the pain withdrawal reflex by an ear pinch) cannot be used in an AI-based tool. Moreover, the indicators to be observed differ between pigs stunned electrically and those stunned with CO₂.

The current presentation provides a comprehensive overview of the indicators of (un)consciousness in pigs, focusing on their suitability for integration into an AI-based tool for automated detection of insufficient stunning, based on relevant scientific literature.

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8. Moo-vie Star - Calves and their Behavior in the Spotlight

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The behavior of calves is a central indicator of their health status. Early detection of behavioral changes in calves is therefore important to ensure healthy growth. In this context, automated and objective methods for behavioral monitoring are gaining scientific relevance, offering the potential to enhance early detection strategies. Keypoint estimation is an innovative approach to automatically. The deep learning algorithm YOLOv8-pose is utilized for the detection and analysis of anatomical keypoints, including head features, shoulders and extremities, within the video. For

this purpose, a total of 2223 individual images were extracted from video recordings of pairs of calves housed separately. Five classes were defined, three with focus on body posture (standing, lying in prone or lateral position) and two on consumption behavior (drinking and eating). A total of 30 keypoints were identified and each calf was manually annotated for each image. The data set was then divided into training and validation data sets, with the ratio of training data set to validation data set at 80:20. The YOLOv8-pose model was adapted accordingly and trained with the training data. The validation images were utilized to assess the model's precision. A mean accuracy for the classes was 98.3%, while the keypoint estimation achieved an average accuracy of 90.3%. This method provides a non-invasive approach to the continuous monitoring of animal welfare during calf rearing. Beyond the ability to localize calves within the barn, the YOLOv8-pose model enables the extraction of activity patterns, resting phases, and feeding behavior. Deviations from these behavioral patterns may serve as indicators of discomfort or compromised well-being.

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9. Reinforcement Learning in human and veterinary medicine - a systematic review

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Artificial intelligence has experienced significant growth in recent years. Reinforcement Learning (RL) is also gaining importance in human and veterinary epidemiology. RL, a machine learning approach where an agent learns optimal strategies through environment interaction, holds promise for enhancing health outcomes through improved surveillance and intervention strategies. This systematic review aims to evaluate the current state of research on the application of RL in human and veterinary epidemiology. It seeks to investigate which specific RL methods have been applied and which epidemiological questions have been addressed. For the literature search, the databases Web of Science, PubMed, and Scopus will be utilized to generate a comprehensive data basis. Only peer-reviewed articles that address RL methods in an epidemiological context will be included. The identified studies will be analysed and categorized based on their methodological approaches, areas of application, and results. The free web tool CADIMA will be used for conducting and documenting the review. The collected information offers potential for the use of RL methods in new application areas and for developing solutions to challenges related to RL applications. In the long term, this can contribute to developing more precise intervention strategies and optimizing the surveillance of disease outbreaks.

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10. Refining Machine Learning Approaches to Improve Nanopore Basecalling Accuracy

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Nanopore sequencing is a powerful technology that enables real-time, long-read DNA and RNA sequencing. A protein nanopore is embedded in a membrane that separates two electrolyte-filled compartments. By applying a constant voltage across the membrane, a steady ionic current is driven through the pore. This current is continuously measured by a sensor. When a single-stranded nucleic acid passes through the pore, the bases inside the pore partially obstruct the flow of ions. This causes characteristic changes in the current signal, which reflect the identity of the nucleotides occupying the pore at a given time. Decoding this signal into the corresponding nucleotide sequence – known as basecalling – is a central computational task, but remains highly challenging. The signal is inherently noisy, context-dependent, and influenced by variable translocation speeds, making accurate and robust basecalling particularly difficult in homopolymer regions and low-quality signal segments. Basecalling is typically performed using machine learning models. These models are trained to map raw current signals to nucleotide sequences of varying lengths, without requiring explicit alignments. In our work, we explore potential improvements to the training methods and model architectures. A central focus is the loss function: current state-of-the-art approaches often rely on a task-specific linear-chain Conditional Random Field during training. We investigate alternatives based on customised Connectionist Temporal Classification losses. In addition, we examine the role of decoding strategies in translating the model output into the final nucleotide sequence, as improvements at this stage can directly impact basecalling accuracy and robustness. Finally, we explore modifications to the network architecture itself, including the use of auxiliary loss layers to introduce intermediate supervision, as well as the optimisation of transformer layers to better model nanopore signal data.

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11. Effects of elevated platforms on the behaviour, health and growth of fattening pigs in different group constellations

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In pig farming, elevated platforms accessible via a ramp could provide an opportunity to better structure pens and ideally increase the space available per pig. However,

how well elevated platforms are used by pigs has not been monitored in detail and is an important question in verifying their value for animal welfare. This will be investigated under different stocking densities to address potentially conflicting interests between animal welfare and farmers' economical demands. Four experimental variants shall be tested in one pen each, in which A) 0 %, B) 50 % or C) 100 % of the elevated platform is counted as usable floor space or D) the pigs do not have access to the platform (control). This shall be achieved by adjusting the number of pigs (17, 20 or 23 pigs) in the four equally sized pens. In six approximately ten-week fattening trials (12th-22nd week of life), the behaviour, health and growth of pigs with and without previous experience with elevated platforms during rearing (5th-11th week of life) will be examined. This shall include employing Ultra-High Frequency Radio Frequency Identification (UHF RFID) technology and video cameras to continuously record the pigs' use of the platform and other pen areas as well as fitted air quality sensors to continuously monitor temperature, humidity, ammonia and carbon dioxide levels. Skin lesions, lameness, salivary cortisol levels, soiling (of pigs and pens) and weight shall be recorded weekly. Furthermore, the effects of an elevated platform on the pigs' motor skills when walking across a wobble board and their behaviour during loading for slaughter will be analysed once for each pig. By determining if and how fattening pigs use elevated platforms and the effects thereof, the results of this study may inform discussions on whether such platforms could contribute to animal welfare under commercial settings.

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12. Identifying and evaluating the validity of indicators to assess positive welfare in poultry

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Animal welfare is increasingly focusing on promoting positive experiences and a good life (= positive welfare), rather than merely minimizing suffering and factors negatively affecting welfare. To improve the lives of farm animals such as poultry in terms of positive welfare, it is necessary to understand how positive affective states (emotions and moods) are expressed. In order to validate indicators of positive affect, first situations in which poultry are likely to experience such states must be identified. Therefore, a first systematic literature review (empirical studies only) was conducted to identify situations and resources that are preferred or valued by poultry (i.e. laying hens, broiler, turkeys and quails), based on preference and motivation tests. The review follows the PRISMA-P (Preferred Reporting Items for Systematic Reviews and Meta-Analyses-Protocols) guidelines. The literature search was conducted in Web of Science using a predefined search string. The retrieved set of publications was imported into the systematic review software Covidence, where title and abstract screening as well as full-text screening were performed. An extraction template was developed for data extraction, interrater reliability tests were conducted prior to each phase. After identifying preferred and valued

situations, a second systematic literature review was conducted to identify animal-based indicators of positive affective states in poultry and assess their validation status. Indicators were classified as behavioral, cognitive or physiological. All reported validation steps were critically evaluated with respect to the sensitivity and specificity of each indicator. The resulting databases from both systematic reviews provide a foundation to identify knowledge gaps in the validation of positive welfare indicators. In addition to creating these databases, both reviews will serve as the basis for systematic review publications summarizing indicators that can be used to assess positive welfare in poultry.

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13. Influence of feeding place design on agonistic behavior and body condition in dairy goats on organic farms

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Agonistic interactions are common in goat husbandry, especially at the feeding place where competition for resources is high. Thus, appropriate feeding place design is critical to ensure undisturbed access to feed for all animals. While experimental studies have provided recommendations for improved feeding place design, many farms use self-constructed systems whose effects on goat welfare remain insufficiently understood. This study investigates how feeding place design influences agonistic interactions, feed intake and body condition in dairy goats on organic farms. Two common feeding barrier types, neck rail and palisade, are compared, as well as different milking management systems (seasonal lactation vs. year-round milking). Data will be collected on 19 organic farms in both summer (barn and pasture) and winter (barn only) seasons. Animal-specific metrics include body weight, body condition score, health variables (e.g. skin lesions) and morphological measurements (height at the withers, shoulder width, height of the point of the shoulder). Group-level data involve video analysis of agonistic interactions at the feed barrier during feeding and feeding place occupancy. Furthermore, feeding place width, feeding table height, feeding partition and entrance height are recorded to evaluate the adequacy of feeding space. The animal-to-feeding place ratio is also calculated. In light of the increased parasite exposure from pasture access, fecal samples are analyzed to assess parasite load as a potential factor influencing body condition. This study aims to identify feeding place characteristics that promote adequate feeding conditions and reduce social stress in dairy goat herds.

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14. Development of a statistical model for estimating methane emissions from dairy cows on farms

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The project "NEMUR" was funded by the Federal Ministry of Food and Agriculture as part of the German Climate Protection Programme 2022.

The reduction of greenhouse gas emissions from agriculture is becoming increasingly relevant in the context of climate change. Methane emissions from dairy farming, in particular, represent a major challenge. To enable accurate monitoring and effective mitigation, the development of robust estimation models is essential. Over a period of three years, a feeding trial was conducted with nine different periods, each lasting 6 weeks with the aim of creating a data set covering a wide range of feed rations. Methane emissions were measured using the GreenFeed system (C-Lock Inc., Rapid City, USA). Several parameters that influence methane production and are accessible under practical farm conditions were identified, including physically effective neutral detergent fiber (peNDF) and metabolizable energy (ME) of the ration, temperature-humidity index (THI), lactation number, week of lactation, body condition score (BCS), locomotion score and energy-corrected milk yield (ECM) of the cows. A linear mixed-effects model was developed in R following the top-down modeling strategy described by Zuur et al. (2009). The final model included week of lactation and THI as smooth terms; ECM, ME, peNDF, and interactions between THI and lactation number as fixed effects; and individual animal as a random effect. An autoregressive moving average (ARMA) covariance structure was applied. This model provided the best fit for estimating methane emissions based on the available parameters on farms.

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15. Final destination: HPAI-freedom through biosecurity? Please mind the KAP.

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In Germany, highly pathogenic avian influenza (HPAI) has now become endemic. Despite similar risks for virus introduction, not all farms are equally affected. Many studies describe risk factors regarding increased probabilities of occurrence and aspects of biosecurity, but only few describe the underlying decision-making of farmers. In a case-control study in the German federal state of Lower Saxony 15 poultry farms affected by HPAI several times since 2016 were compared with 30 unaffected farms. A "KAP"-questionnaire was used to semi-quantitatively record the knowledge, attitude and practices regarding biosecurity of the farmers. In addition, existing biosecurity measures were evaluated using a checklist. Data collection is complete. Subsequently, correlations between the assessed biosecurity and the questionnaire results, as well as differences between the case and control farms will

now be statistically investigated. The results will be available by October. In a second part of the project the “health belief model” will be used in a cross-sectional study to understand the behavior of farmers regarding the implementation of biosecurity measures more comprehensively and nationwide. This includes the assessment of the perceived risk of HPAI, the own susceptibility, effectiveness of measures, obstacles and motivators. Based on the results of both study parts, impediments regarding the effective implementation of biosecurity measures are to be identified, elimination points created and poultry protected from HPAI in long term.

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16. Bio●Logically●Safe - An intervention study on ASF prevention in organic pig farms

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African swine fever (ASF) poses a constant threat to pig farming in Europe. Organic farms are particularly vulnerable due to open housing systems and mandatory outdoor access, which promote high animal welfare but also increase the risk of disease transmission through potential contact with wild boar or contaminated material. This creates a critical challenge for disease prevention in organic pig farming. Therefore, practical, effective and specifically tailored prevention strategies are urgently needed for organic pig production. As part of the FLI project “Living in Harmony - Transformative strategies for the control of African swine fever on the interface between wild boar and organic pig farming,” a holistic, systemic approach to ASF control is being developed, focusing especially on the interface between wild boar and organically raised pigs in Germany. A central component of the project is an intervention study investigating how targeted information and training measures can influence farmers’ knowledge, risk perception, and behavior regarding ASF. To this end, an awareness campaign has been developed to raise awareness of ASF and the importance of biosecurity measures in organic pig farming. The campaign includes three intervention levels, with at least 20 farms participating in each group:

- An information group receives materials such as brochures and digital content.
- A training group additionally participates in webinars and lectures.
- A control group without specific intervention.

Farm recruitment takes place across Germany and includes only organic pig farms. The effectiveness of the measures is evaluated using a before-and-after comparison with a standardized KAP (Knowledge, Attitude, Practice) questionnaire. The intervention study explores how different educational approaches affect farmers’ knowledge, attitudes, and practices regarding ASF. The results aim to support the development of effective prevention strategies and biosecurity measures to ensure the long-term viability of organic pig farming, even under disease pressure.

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17. Knowledge, Attitudes and Practices (KAP) of Ruminant Livestock farmers related to Rift Valley fever and Q fever in Zanzibar, Tanzania

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The Zanzibar Archipelago, mainly consisting of the two main islands Unguja and Pemba, is part of the United Republic of Tanzania and located 40 km off of Tanzania's mainland in the Indian Ocean. The level of knowledge and awareness of ruminant livestock farmers in Zanzibar with regard to zoonotic pathogens is largely unknown. The primary objective of this study was to investigate ruminant livestock keepers' knowledge, attitudes and practices (KAP) linked to infection risk. The questionnaire focuses on Rift Valley fever and Q-fever. A previously conducted study found 24.50% seropositivity for Rift Valley Fever and 12.33% for Q fever in Zanzibar's local cattle (Wiethoff et al., submitted). The gain of knowledge on farmers' KAP can enhance data on human exposure and risk factors for zoonotic diseases. A further, targeted effect of the study will be to increase awareness among animal keepers regarding the presence of zoonotic diseases. A total of 451 interviews were conducted on Unguja and Pemba from January to April 2025. Preliminary results of the data analysis suggest that the level of knowledge is generally low. It appears that many livestock keepers are not familiar with the diseases mentioned, and some do not believe in the existence of these diseases. The present study can assist in the formulation of recommendations for the prevention and control of future zoonotic outbreaks, and thereby improve preparedness for such events. Outreach activities will be addressed once the final results are available, allowing the identification of key areas for intervention and the development of targeted awareness efforts.

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18. Functional Analysis of Coxiella burnetii-specific Antibodies Using Ovine Target Cells

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Coxiella (C.) burnetii is a highly infectious, obligate intracellular bacterium causing Q-fever in humans and reproductive disorders in ruminant livestock, such as sheep, cattle, and goats. This pathogen presents serious threats to both public health and agricultural sustainability. Effective disease control strategies require the development of next-generation vaccines that are protective but also capable of distinguishing infected from vaccinated animals (DIVA). This project, conducted within the framework of the EU-funded REPRODIVAC project, focuses on the functional characterization of antibodies generated in sheep immunized against

selected *C. burnetii* antigens. Specifically, the study investigates whether these antibodies can neutralize infection by blocking bacterial entry into host cells, disrupting intracellular replication, and triggering immune responses. To this end, two ovine cell models were established: the epithelial PT cell line (derived from sheep kidney) and primary monocyte-derived macrophages (MDM), cultured in two-dimensional (2D) (PT; MDM) monolayers and three-dimensional (3D) spheroids (PT only). *C. burnetii* strain NMII stocks were prepared using L929 murine fibroblasts and axenic culture, then quantified via *icd*-qPCR and confirmed contamination-free (mycoplasma; blood agar). Initial infection studies across multiple time points identified day 7 post-infection as a critical window for evaluating infection dynamics. Moreover, mRNA analysis of LPS-stimulated PT cells indicated significant induction of pro-inflammatory cytokines, such as TNF- α , validating the cell model's general responsiveness in inflammatory signaling. However, efforts focus on optimizing neutralization assays in 96-well plate format using sheep sera at varying dilutions to determine dose-dependent inhibition. Additional analyses will include complement activation studies and transcriptomic profiling to assess broader immune effects. Future phases will validate promising sera in 3D cultures and potentially in cattle and goat cell lines to explore cross-species applicability. Collectively, this research supports the development of innovative DIVA-compatible vaccines and deepens our understanding of host-pathogen interactions, with implications for both animal and human health.

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19. PLB-985 cells partially recapitulate human neutrophil features

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Mycobacterial infections, including tuberculosis, remain a global health concern. Current studies underline the importance of neutrophils in clearance of mycobacteria and shaping the disease outcome. A major limitation in advancing the understanding of neutrophil-pathogen interactions relies in the lack of suitable cell line systems amenable to genetic manipulation. Several myeloid cell lines undergo neutrophil-like differentiation *in vitro*. Here, we assessed the feasibility of using PLB-985 cells, a confirmed derivative of human promyelocytic leukaemia-derived HL60, as a model to study mycobacterial infection. Differentiated PLB-985 (dPLB) cells recapitulated many aspects of human neutrophil biology. Both dimethylsulfoxide (DMSO) and N, N'-dimethylformamide (DMF), two commonly used inducers of differentiation, led to the segmentation of nuclei and expression of the neutrophil-activation marker CD11b during the course of differentiation. dPLB cells generated abundant reactive oxygen species, formed neutrophil extracellular traps upon activation, and phagocytosed opsonized mycobacteria. Surprisingly, using fluorescence and electron microscopy, we observed that dPLB cells failed to develop neutrophil-specific granules. Finally, we evaluated the viability of dPLB cells under hypoxia, a hallmark of tuberculous

granuloma. Unlike primary human neutrophils, dPLB cells exhibited reduced viability under oxygen restriction. Taken together, dPLB cells represent a tractable model to study certain neutrophil responses to pathogens. However, they are not suitable for studying granule-linked biology or hypoxia-driven responses of neutrophils.

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20. Age-dependent immunome and functional roles of $\gamma\delta$ T cells in pigs

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Lymphocytes can generate a huge repertoire of antigen receptors while being organized into 3 compartments, two T-cell like ($\alpha\beta$ and $\gamma\delta$ T cells) and one B-cell like. Species that have high $\gamma\delta$ T cell frequencies (30% in pigs) show expansion of γ and δ chain genes but also increased diversity in co-receptors indicating important functional roles of $\gamma\delta$ T cells. To interrogate these roles, a $\gamma\delta$ KO pig animal model was produced via CRISPR Cas9 technology. We are investigating the immune system of this animal model in order to understand better the roles of the $\gamma\delta$ T cell compartment. We developed a lymphocyte panel and a myeloid panel for flow cytometry that will be used to quantify the frequencies of the main immune subpopulations but also to see if the empty niche is compensated. We will later use scRNA sequencing to analyse in detail white blood cell composition and functional states. We will also perform functional assays by evaluating phagocytic capacity and measuring cytokine release.

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21. Comparison of Enrichment Methods for the Detection of Avian Influenza Virus in Water Samples

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Low Pathogenicity Avian Influenza viruses (LPAIV) typically induce mild, if any, disease in birds; yet subtypes H5/H7 may mutate into High Pathogenicity Avian Influenza virus (HPAIV). HPAIV subtype H5 is circulating globally, causing outbreaks in wild birds, poultry, dairy cattle (United States only), and mass mortality in seals. Concerns have been raised about its pandemic potential. Passive surveillance is more effective than active surveillance in detecting HPAIV cases among wild birds. In contrast, active surveillance in wild bird populations tends to detect LPAIV. LPAIV frequently reassort with HPAIV. Understanding the evolutionary traits of HPAIV H5 also requires active wild bird surveillance. Due to its time- and resource-intensive nature, a simplified, non-invasive approach is needed for more efficient monitoring.

We introduced a new concept of non-invasive water sampling using feed-baited water bins for active avian influenza surveillance. Mallards (*Anas platyrhynchos*), present year-round at the Baltic shallows of the Greifswalder Bodden, Germany, are attracted to wheat submerged in 5 L of water in small bins. Ducks feeding on the wheat have close oropharyngeal contact with the water and may release viral particles if infected. Water samples are collected two to three times per week and analyzed for AIV by pan-Influenza A RT-qPCR. To maximize sensitivity, different virus and RNA enrichment methods from water samples are compared using H4N6-spiked samples (A/rhea/Ger-MV/AI06884/2023) as a model. a) Swab (no enrichment; used as reference) b) PEG precipitation c) Activated carbon capture d) Activated carbon capture + PEG precipitation e) Electronegative membranes For each method, variables that could influence their effectiveness are considered, such as the use of freshwater versus Bodden water, and the presence or absence of wheat in the water samples. Semi-quantitative data collected by RT-qPCR Ct values referenced against previously defined standard curves of H4N6 dilutions are compared and will be presented.

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22. Publish the Negative: Knowledge Gaps in FMDV Research Revealed by the Failure of Oral Inoculation in Pigs

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Foot-and-mouth disease virus (FMDV) is one of the first animal viruses to be discovered and was the focus of intensive research for many decades. Yet despite this, key questions remain unanswered, particularly regarding its pathogenesis in pigs. FMDV is often referred to as one of the most infectious viruses, and while this may hold true for cattle, the situation in pigs is far less clear. During our search for a suitable live-attenuated vaccine candidate, we encountered unexpected challenges: repeated failures to infect pigs via intra-oropharyngeal (IOP) exposure with high virus doses (up to 10^7 TCID₅₀), even when using a pig-adapted strain that had previously caused devastating outbreaks in pig farms. This raised fundamental questions: How do pigs become infected in nature? What role do they play in outbreaks? And what physiologically relevant infection routes can we realistically mimic under laboratory conditions? Surprisingly, we found that similar failed attempts had been made by others, but their negative data had never been published. Our work underscores the importance of reporting negative findings, not only to prevent redundant efforts, but also to address long-standing knowledge gaps in FMDV infection dynamics in pigs. If negative findings were systematically published, not only time and resources could be saved, but more importantly, it could contribute to the reduction of animal experiments in accordance with the 3Rs principle.

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23. Development and preclinical testing of a vaccine candidate against the Nipah virus - project presentation

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Nipah virus (NiV) is a highly pathogenic zoonotic RNA virus of negative-polarity belonging to the genus *Henipavirus* within the *Paramyxoviridae* family. In humans, NiV infection can cause severe, often fatal, respiratory and neurological disease. NiV was first identified in 1998/1999 during an outbreak in Malaysia. Numerous individuals with close contact with pigs developed an acute febrile illness affecting the central nervous system (CNS) and the respiratory tract. Domestic pigs, infected via spillover from *Pteropus* fruit bats, were identified as the intermediate host. Since 2001, recurrent outbreaks have occurred in Bangladesh and India, this time without the involvement of pigs. These infections are instead linked to the consumption of raw date palm syrup contaminated with infectious secretions from *Pteropus* bats. Due to its high pathogenicity, NiV is included in the World Health Organization (WHO) R&D Blueprint Prioritization list. The EU-funded project “VICI DISEASE” aims to develop a safe and effective vaccine candidate for human use and to evaluate its efficacy in small animal models under BSL4 conditions at the FLI. These studies will contribute to a better understanding of NiV pathogenesis, host immune response and protective effect of the vaccine candidates. As part of my doctoral thesis, I will investigate organ samples from in vivo experiments using histopathology, immunohistochemistry, in situ hybridization, RT-qPCR, and ELISA to assess NiV pathogenesis of NiV and the protective effect of the vaccine candidates.

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24. Development of an efficient vaccine against porcine reproductive and respiratory syndrome virus

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Infection with the porcine reproductive and respiratory syndrome virus (PRRSV), which belongs to the *Arteriviridae*, results in great losses in pig farms worldwide. The significant variety of circulating strains and the immunosuppressive characteristics of the virus limit the impact of currently available vaccines on disease prevention and control. Our aim is to develop a highly protective vaccine against PRRSV by identifying conserved antigenic regions in viral surface proteins and expressing them in the alphaherpesvirus pseudorabies virus live-attenuated vaccine strain Bartha (PrV-Ba) to generate a bivalent vaccine. PrV-Ba is suitable as a DIVA

(differentiating infected from vaccinated animals) vaccine and has contributed to eradication of Aujeszky's disease (AD) in domestic pigs in many European countries, North America and Australia. However, AD is still a problem in most other parts of the world. PrV-Ba-PRRSV recombinants were generated by two different approaches: (1) antigenic PRRSV peptide sequences were inserted into the non-essential N-terminal region of the essential PrV fusion protein gB, and (2) full-length genes of PRRSV envelope proteins were expressed either singly or in different combinations from the non-essential glycoprotein gG gene locus. Most of the tested recombinants replicated comparably to PrV-Ba. PRRSV protein expression was confirmed by immunoblotting and indirect immunofluorescence assays. Intracellular protein localisation is currently being analysed. Additionally, a pilot in vivo study in pigs was conducted to evaluate the immunogenicity of a PrV-PRRSV recombinant expressing either the full-length PRRSV gp4 or only a predicted immunogenic peptide in PrV-gB. Key results will be discussed.

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25. Uncovering Nairobi Sheep Disease Virus Glycoprotein Precursor (GPC) Processing

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Crimean-Congo haemorrhagic fever virus (CCHFV) and Nairobi sheep disease virus (NSDV), are tick-borne pathogens with significant impact in public and veterinary health. These viruses encode a viral glycoprotein precursor (GPC) that undergoes a complex proteolytic processing. For CCHFV, the cleavage product GP38 has recently been described in several studies with regards to its immunogenicity in infected hosts or its potential role in viral pathogenesis. Nonetheless, the processing of GPC, and the function of GP38 in other orthonairoviruses such as NSDV remains largely unexplored. Proteolytic cleavage of CCHFV GP38 has been described to involve the proprotein convertases furin and subtilisin/kexin-isozyme-1 (SKI-1). However, little is known about the processing of NSDV GPC and the putative proteases involved in GP38 maturation. To elucidate on the proteolytic processing of NSDV GPC, with a particular focus on identifying the proteases involved in GP38 cleavage, different cell lines will be transfected using a full-length NSDV GPC plasmid. The subsequent analysis of the transient expression of proteins will be carried out with NSDV-specific monoclonal antibodies targeting the viral glycoproteins enabling to observe the different cleavage products via Western Blot. In parallel, using different protease inhibitors, we plan to gain further knowledge of which host-proteases are involved in GPC proteolytic processing. Additionally, site-directed mutagenesis of predicted cleavage sites will be performed to confirm the host proteases involved. Finally, in vitro infection experiments with NSDV will be carried out to validate the results from the transfection-based studies. Overall, advancing our understanding of GPC

processing and specifically, GP38 maturation and function across orthonairoviruses, will provide new insights into their replication strategies and pathogenic mechanisms.

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26. Visualization of the Spatial Organization of Henipavirus Entry Protein Complexes by Correlative Light and Electron Microscopy

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Henipaviruses, a genus within the Paramyxoviridae family, are zoonotic pathogens capable of infecting a broad range of mammalian hosts. Infections with these viruses can lead to severe respiratory or neurological disease with high fatality rates, highlighting the urge for deeper understanding of their entry mechanisms. Viral entry is mediated by the coordinated action of two surface glycoproteins: the attachment protein G, which binds to host cell ephrins, and the fusion protein F, which facilitates membrane fusion. Successful entry requires clustering of F and G on the viral surface as well as multimerization of ephrins on the host cell membrane. However, the precise molecular interactions among F, G, and ephrins triggering fusion remain poorly defined. To investigate this, we employ high-resolution confocal fluorescence microscopy, transmission electron microscopy (TEM), and correlative light and electron microscopy (CLEM), which integrates the strengths of both approaches. Fluorescence microscopy will enable visualization of viral and host proteins. For this purpose, we generated fluorescently tagged viral proteins and perform immunostaining. In contrast, TEM will offer ultrastructural detail at nanometer scale. TEM samples are prepared applying epoxy resin embedding followed by ultramicrotome sectioning and contrasting. Combining both methods through CLEM, we intend to localize viral F and G proteins in relation to host ephrins with high spatial resolution. We aim to identify a specific spatial arrangement or ratio of F and G proteins in relation to ephrins that is necessary to trigger membrane fusion. This may allow us to define a threshold for fusion activity and identify key organizational patterns critical for viral entry. These findings could advance our understanding of molecular prerequisites for henipavirus entry and potentially lead to novel antiviral strategies.

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27. Investigating Orthonairovirus GP38 as an Antigen Candidate in CCHFV and AIGV Serology

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Aigai Virus (AIGV) is a zoonotic, tick-borne orthonairovirus of the family Nairoviridae with livestock serving as silent but amplifying hosts. Unlike its better-known relative, Crimean-Congo haemorrhagic fever virus (CCHFV), which is highly pathogenetic with case fatality rates up to 30% or higher in humans, AIGV appears to be less pathogenic with only two confirmed clinical cases to date. Both AIGV and CCHFV are classified as BSL4 pathogens, making serological diagnostics preferable, as they do not necessarily require handling live virus. Up to now, most commercial serological assays are based on the highly conserved nucleoprotein (N) of CCHFV. However, due to their close genetic relatedness, N-based assays cannot distinguish between AIGV and CCHFV, leading to cross-reactivities. Such possible cross-reactivity has been observed in N-based assays detecting CCHFV-seropositive individuals, despite the absence of clinical symptoms, in regions like Greece and Bulgaria, where both viruses have been described to circulate. This raises the possibility that AIGV as a less pathogenic orthonairovirus, may be responsible for the observed seropositivity. A similar concern arises for CCHFV-seropositive ruminants in areas without any human CCHF cases. To enable serological differentiation between AIGV and CCHFV, novel viral antigens are needed. The viral glycoprotein GP38 of orthonairoviruses, which is immunogenic in patients but less conserved may serve as a suitable candidate. In this study, we aim to evaluate the potential of CCHFV- and AIGV-derived GP38 antigens to reliably distinguish between AIGV and CCHFV infections in serological assays. Therefore, we tested N-seropositive human sera from asymptomatic individuals from Bulgaria, sera from hospitalized CCHF patients from Turkey. Furthermore, we plan to test N-seropositive ruminant sera from AIGV-endemic areas in Greece.

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28. Characterization of CD46-dependent infectivity of bovine viral diarrhea virus field isolates from Germany

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Bovine viral diarrhea virus (BVDV), a member of the genus Pestivirus and the family Flaviviridae, is of major economic importance for the cattle industry worldwide. Host animals also include goat, sheep, pig, and other cloven-hoofed animals. Acute infections in non-pregnant animals are typically subclinical whereas infection of seronegative pregnant cows may lead to embryonic loss, abortion, congenital

malformations, or the birth of persistently infected (PI) calves. PI animals continuously shed virus and thereby play a central role in sustaining infection chains. The interaction between the viral glycoprotein E2 and the cellular transmembrane receptor CD46 was identified as a major mediator for BVDV entry into host cells. Under selective pressure through lack of CD46, some BVDV isolates can escape the CD46-dependency. At the genetic level, several amino acid substitutions within the viral ERNS protein were identified as potential causes for this shift in infectivity. These processes have primarily been studied with isolates extensively passaged in cell culture. This work aims to characterize the CD46-dependency of different field isolates from BVD cases in Germany. These isolates originate from diagnostic samples submitted to the German National Reference Laboratory for BVD between 2019 and 2024 and have not undergone additional passage in cell culture after the initial isolation. A stable Madin-Darby Bovine Kidney (MDBK) wild-type and a CD46-knockout cell line are being used for growth studies. Sequencing data is generated on the PromethION 2 platform (Oxford Nanopore Technologies) for subsequent genetic analysis of BVDV isolates with different growth characteristics.

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29. Pathogenesis and persistence of rustrela virus infection: cross-species analysis of the central nervous system and verification of in vitro models

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Since 2020, rustrela virus (RusV) is identified as the cause of sporadic cases of fatal encephalitis in mammals of various taxa in Germany, Austria, Sweden and the USA. RusV was also found in the brains of apparently healthy yellow-necked field mice (*Apodemus flavicollis*) and wood mice (*Apodemus sylvaticus*) that are considered as putative reservoir hosts. The broad spectrum of affected mammals raises concerns about a zoonotic potential. Among other investigations, in situ hybridization (ISH) identified in healthy reservoir hosts and also in spill-over hosts neurons as main target cells. The major histopathologic feature in spill-over hosts is a non-suppurative meningoencephalomyelitis. Initial in vivo experiments at the FLI in wood mice and rats (*Rattus norvegicus*), inoculated with RusV-positive brain homogenates, have indicated persistent infection of the brain (unpublished data). To date, questions regarding the pathogenesis, the mechanisms of RusV persistence, and the virus's entry into the central nervous system (CNS) remain unanswered. Organ samples from experimentally inoculated ferrets (*Mustela putorius furo*), rabbits (*Oryctolagus cuniculus*), guinea pigs (*Cavia porcellus*), house mice (*Mus musculus*) and wood mice will be systematically processed and evaluated by histopathological (hematoxylin-eosin staining), immunohistochemical, in situ hybridization and immunofluorescence methods. The aim is to identify initial indications of replication

sites, target cells, invasion routes into the brain and a potential lesion profile. In addition, the project includes the establishment and verification of in vitro models using murine brain organoids and brain precision cut slices. The direct comparison of in vivo versus in vitro data enables the assessment of the suitability and limitations of the models (for reduction/replacement of animal experiments). Our results will support the development of infection models and contribute to a better understanding of RusV-pathogenesis across spill-over and putative reservoir hosts.

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30. Influence of Seasonality and Host Demography on Zoonotic Pathogens in Bank Vole (*Myodes glareolus*) Populations in Northern Sweden

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In order to mitigate disease emergence risks in changing environments, it is crucial to understand how landscape composition, seasonal changes and rodent community demography influence the prevalence of zoonotic pathogens. In this study, we investigated the presence of multiple zoonotic pathogens in bank voles (*Myodes glareolus*) from two boreal forest sites in northern Sweden: Vindeln and the coastal region of Västerbotten. These populations are subject to well-characterised multi-annual density cycles (3-4 years), which offers a unique opportunity to examine pathogen dynamics across temporal and demographic gradients. We focused on pathogens known or suspected to have rodent reservoir dynamics, including Orthohantavirus puumalaense (PUUV), *Leptospira* spp., *Toxoplasma gondii*, *Yersinia pseudotuberculosis* and Hepatitis E virus. We applied a disease freedom analysis to a subset of samples, using published prevalence as baseline parameters for each pathogen. After detection, a true prevalence analysis was conducted. Negative results were considered to indicate a pathogen free population. Among the 519 bank voles screened, 83 (15.99%) tested positive for PUUV with no significant difference between sites. PUUV prevalence increased over time with 6% (6/100) in 2020, 13.44% (25/186) in 2021 and 23.31% (52/223) in 2022 correlating with host population growth, suggesting a density-dependent transmission pattern. Sex-based were also observed: in Vindeln, male prevalence was 21.35% (206/339) compared to 9.77%

(133/339) in females; at the coast, 20.79% (102/180) males were infected versus 5.12% (78/180) females. Furthermore, a notable finding was the difference in prevalence in spring 28.49% (55/193) compared to 8.58% (28/326) in autumn. Additionally, only one individual tested positive for *Y. pseudotuberculosis*; all other pathogens were undetected. These results suggest that host demographics, particularly sex and population density, as well as seasonal dynamics, play a significant role in maintaining and transmitting pathogens.

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31. Monitoring of mosquitoes (Diptera: Culicidae) collected in northeastern Germany for mosquito-borne pathogens

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Globalization as well as climatic and environmental changes support the global spread of vectors and vector-borne diseases. Europe has recently been facing a rise in mosquito-borne diseases, following improving conditions for both mosquitoes and mosquito-borne pathogens. To monitor the circulation of mosquito-borne disease agents, zoos and wildlife parks have increasingly been recognized as appropriate early warning sites. Their structurally diverse habitats, the presence of numerous water containers, and a high density and variety of vertebrate blood host species, provide favourable conditions for both mosquito development and pathogen circulation. To study mosquito populations and associated pathogens in northeastern Germany, BG sentinel traps were set up at 8 zoos and wildlife parks in the vegetative seasons (spring to autumn) of 2024 and at 13 zoos and wildlife parks in the vegetative seasons of 2025. Furthermore, overwintering mosquito females were collected in 130 dungeons, bunkers and cellars in winter 2024/2025. Among a total of around 5,500 mosquitoes collected in the vegetative seasons of 2024, 14 species were identified, mainly from the *Culex pipiens* complex (ca. 3,400 individuals). Preliminary RT-PCR analyses of single mosquitoes or pools with up to ten specimens resulted in the detection of various viruses in that specific complex: Sindbis virus in a single mosquito from one location, West-Nile virus in two pools from one location and Usutu virus in three pools from three locations. Mosquitoes collected in the vegetative seasons of 2025 are not available yet for analyses. More than 34,000 mosquito females of six taxa, including 29,800 specimens of the *Cx. pipiens* complex, were collected in winter 2024/2025. Some 4,600 were identified to species so far. Virus screening of overwintering mosquitoes is still pending.

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32. Surveillance of Invasive Arthropods and Vector Borne Pathogens in Tropical Imports at Frankfurt International Airport

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The continuous increase in imports from tropical and exotic regions, coupled with global climate change, has significant ecological consequences. Numerous studies have documented the introduction and spread of invasive arthropod species, particularly plant pests, resulting in substantial agricultural losses. Additionally, imported vector species have been increasingly reported across Europe in recent years. Given this context, the surveillance of potential points of entry such as airports, seaports and ground borders, may help identify introduction pathways and design control measures. At the Perishable Center of Frankfurt International Airport, targeted inspections of containers importing vegetables, fruits, and ornamental plants originating from tropical countries are therefore conducted five to six times per year, each over a multi-week period. The protocol involves inspecting and sampling the incoming containers and their cargo using handheld vacuum devices. In parallel, BG-Pro insect traps are deployed in various strategic locations throughout the Perishable Center. These traps operate continuously and are checked on a daily basis. The objective of the project is to collect and identify, morphologically or genetically, arthropod specimens found through active sampling and trapping. Particular emphasis is placed on potential vectors, of which selected specimens will undergo Next-Generation-Sequencing to characterize their microbiome and to screen for associated pathogens. Sampling activities have started in spring 2025. Taxonomic classification at the order level is completed for the first two two-week sampling periods, while molecular species identification is in progress. A total of about 400 arthropod specimens were collected so far, representing nine zoosystematic orders and including four specimens of families containing vectors of vertebrate disease agents. Upon completion of the collection, identification and sequencing phases, a risk assessment will be conducted. This assessment will serve as a scientific basis for developing strategies to prevent future introductions and mitigate potential public health, animal health and agricultural risks.

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33. Influence of climate-induced forest conversion on rodent-borne zoonoses

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As result of climate change and pest outbreaks, forest conversion is currently being carried out in Germany. Wildlife, especially rodents, are the reservoir for various zoonotic pathogens. The implementation of climate change-appropriate forest conversion will result in changes to habitats that impacts the existing reservoir species. In addition, there is potentially increased wildlife-human contact during the

necessary forestry work. This project is therefore investigating the effects of forest conversion on wildlife reservoirs and the human pathogens they transmit. The project partners (Julius Kühn Institute, Northwest German Forest Research Institute, University of Münster) select investigation areas with different tree species compositions and catch rodents. Subsequently, the species are determined morphologically and genetically. After dissection, food and microbiome analyses are carried out using metabarcoding. In the sub-project at FLI, specific pathogens will be examined from already existing samples in order to make a final selection for the screening. Next-generation sequencing (NGS) is carried out to identify additional pathogens. The investigations focus on pathogens such as Puumala virus, Tula virus, tick-borne encephalitis virus, cowpox virus, *Leptospira*, *Borrelia*, *Bartonella* and *Rickettsia* spp. The samples are screened to determine the prevalence and pathogen patterns in the individuals and populations. The viral pathogens are molecularly typed using sequencing and phylogenetic analysis to draw conclusions about their spread through the various animal populations. Bacteria are sequenced or characterized by multi-locus sequence typing. To identify further, previously unknown viruses, pool samples of the respective species (red-backed vole, common vole, wood mouse and yellow-necked vole) will be examined. In addition, pathogen-specific screening or NGS of large mammal faecal samples from selected trapping areas is also carried out. These data provide an important contribution to understanding the influence that a changed tree population can have on biodiversity and zoonoses and enables a direct analysis of future climate-related health risks.

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34. Analyzing the impact of husbandry on the microbiome and resistome in pigs

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Antimicrobial resistance is a growing threat to human and animal health. The microbiome of farm animals such as pigs can act as a reservoir for antimicrobial resistance genes. In addition to the use of antibiotics, other factors such as husbandry can have an influence on the microbiome and thus possibly on the totality of all resistance genes (resistome). In this study, we investigated the influence of husbandry on the microbiome and resistome of pigs. Fecal samples were collected regularly in one pig farm at pen level over a period of two years. The samples were collected around the sixtieth day of the pigs' lives, at the end of rearing. One group of animals has access to an outdoor run (organic husbandry), while the other group is kept according to conventional standards. Shotgun metagenomic sequencing was performed on the samples. Kraken2 was used for taxonomic profiling of reads and the AMR++ pipeline for detection of resistance genes. ARG abundance was normalized by the 16S DNA content of each sample. We observed significant differences in alpha and β -diversity in the microbiome. Several families were

enriched in the organic group including Prevotellaceae, Lachnospiraceae and Cellulosilyticaceae, while Methanobacteriaceae showed a higher abundance in the conventional group. In the resistome, the differences were smaller, dominant genes were the same in both groups and only a few ARGs showed different frequencies. However, there was a significant difference in the overall composition based on beta diversity. In addition, the overall frequency of ARG, normalized by the number of 16S rRNA genes, was on average higher in the organic group. Our data indicate that the type of husbandry can have an influence on the microbiota and the composition of ARG in the microbiome of pigs.

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35. ESBL-producing *E. coli*: Hidden Guest in the Chicken House

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Background One of the biggest global problems is the increase in antimicrobial resistance (AMR), which makes the treatment of bacterial infections more difficult. A long-term study was launched in December 2023 to investigate how ESBL-*Escherichia coli* are introduced into stables and whether they can become established there. In order to investigate the dynamics of the occurrence of ESBL-*E. coli* across different groups, a long-term study was started in December 2023, in which faecal samples are regularly taken from two different poultry farms over a period of two years. **Results** The sequencing of a total of 113 isolates revealed that 21 isolates are *E. coli*, most of which carry the blaTEM-52B gene. The blaTEM-1B and blaCTX-M-15 genes were also identified in isolated cases. Of a total of 81 isolates from the fattening farm from 55 samplingpoints, 17 isolates were ESBL-positive. In 32 isolates from 26 samplingpoints from the laying hen farm, 4 ESBL-*E. coli* isolates were identified. Sequencing of the 21 ESBL-positive isolates revealed that in most cases the blaTEM-52B gene was involved. The blaTEM-1B and blaCTX-M-15 genes were also identified in isolated cases. The biofilm analysis with crystal violet showed that only a few isolates form biofilms. **Conclusion** Full genome sequencing of the isolates from fattening and laying hen farms showed that a proportion of the samples contained ESBL-positive *E. coli* strains, with the blaTEM-52B gene being identified most frequently. Individual isolates also carried the blaTEM-1B and blaCTX-M-15 genes. Biofilm analysis revealed that only a few of the isolates were able to form biofilms. Overall, the data show a low prevalence of ESBL-producing *E. coli* in the investigated poultry farms and a limited biofilm-forming capacity of the identified strains.

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36. Prevalence and risk factors of *Echinococcus multilocularis* infections in wild carnivores on the island of Rügen

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Background The fox tapeworm *Echinococcus multilocularis* is a cestode that lives in the small intestine of its definitive hosts and is known as the causative agent of alveolar echinococcosis in humans. The red fox (*Vulpes vulpes*) contributes significantly to the parasite's transmission in Europe, but other wild carnivore species, such as raccoon dogs (*Nyctereutes procyonoides*), could also play a role. *Echinococcus multilocularis* is endemic in Germany, but the epidemiological situation in Mecklenburg-Western Pomerania, especially on the touristic island of Rügen, is unknown. **Methods** In an ongoing study, foxes and raccoon dogs from Rügen are sampled with support from the local hunting community for a period of three years. The exact coordinates of each animal's location are recorded to enable environmental risk factor analyses using biotope maps. Following necropsy, intestinal mucosa, swabs, tissue, serum and fecal samples are collected from all animals and examined for various zoonotic pathogens. Diagnostic testing for the presence of *E. multilocularis* in the carnivore's intestine is performed through microscopic and biomolecular methods. **Results** So far, 443 foxes and 249 raccoon dogs have been examined since the start of the study in November 2023 and our results confirm the presence of *E. multilocularis* in both host species on the island of Rügen. Spatial analysis of environmental risk factors by multivariable logistic regression indicated that increasing proportions of farmland in proximity of fox capture sites correlated with an increased risk of *E. multilocularis* infection ($P < 0.01$), while increasing proportions of scrubland ($P < 0.05$) were associated with a lower risk of infection. These initial findings suggest that landscape structures could play an important role in shaping transmission risk for *E. multilocularis* on the island of Rügen.

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37. Tunneling Nanotubes (TNTs): An Export/Import Strategy for *Chlamydia* via Direct Cell-to-Cell Communication

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The mechanism by which *Chlamydia* exits infected host cells remains poorly understood. While a lytic mechanism has been proposed and observed in vitro, it is not seen in vivo. Our research indicates that *Chlamydia* utilizes Tunneling Nanotubes (TNTs)—actin- and microtubule-based cellular protrusions—for intercellular transfer,

providing a non-lytic, immune-evasive exit strategy. Normally, TNTs mediate organelle and vesicle exchange between cells, but recent evidence suggests that pathogens exploit these structures for their propagation. Combining immunofluorescence and electron microscopy and various proteomic and transcriptomic methods we aim to elucidate the bacterial and host proteins regulating Chlamydia transfer via TNTs. Our current data identify the host protein LST-1, a key TNT regulator, as being modulated in expression by chlamydial cell infection. We also observed the localization of the bacterial protein IncA within TNTs and its potential role in influencing LST-1 function. These findings provide novel insights into Chlamydia pathogenesis and may inform the development of targeted interventions to block TNT-mediated bacterial dissemination.

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38. Studying Strain Specific Host Cell-Pathogen Interactions in ex vivo Lung Models after Mycobacterial Infection

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Mammalian Tuberculosis (mTB) is an infectious zoonotic disease caused by different species of mycobacteria belonging to the *Mycobacterium tuberculosis* complex (MTBC), most importantly *M. bovis*, *M. caprae* and *M. tuberculosis*. It has economic importance in cattle due to high animal losses in infected populations alongside with significant trade restrictions for animals and animal-derived food from the affected country and represents an ongoing zoonotic threat. Despite intensive eradication and surveillance programs in various countries, mTB remains a serious on-going threat worldwide and a constant burden in low- and middle-income countries. mTB can have different outcomes, as the susceptibility varies significantly across the different host species and even between different breeds. Some hosts develop a latent or active infection while others are naturally resistant. Those outcomes depend on various factors e.g., host immune response, genetic factors and strain variability. Therefore, we investigate early host-pathogen interactions during MTBC infection using precision-cut lung slices (PCLS) obtained from three different host species (cattle, goat, sheep). PCLS are an ex vivo tissue model that maintains the 3D architecture of the lung tissue including blood vessels, airways, parenchyma and resident lung cells such as alveolar macrophages. Therefore, it is a powerful tool to investigate respiratory infections on a cellular and subcellular level in a representative yet controlled environment. To gain in-depth knowledge on the early interaction between different MTBC and the host species, our studies include analyses of the cellular responses (inter alia cell viability, secreted cytokines, inflammatory transcriptomic signatures) as well as bacterial loads up to 14 days after inoculation. By fluorescence microscopy and histology, we visualize immune cell locations and structural changes in alveolar morphology. Our study will integrate ten different strains from three different MTBC-species to achieve a detailed insight to

strain- and host cell specific processes influencing susceptibility and disease development.

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