BSPP annual scientific meeting

16th of December 2025

Zebrastraat

Zebrastraat 32

9000 Gent

ABSTRACT BOOK

Meeting program

09:30	Registration	
09:55	Welcome word (Carl de Trez, president)	
Session 1 - Chair: Philippe Van den Steen		
10:00	**Keynote 1: dr. John Gilleard** - DIAGNOSTICS, SURVEILLANCE, AND BEYOND: WHERE NEMABIOME SEQUENCING WILL TAKE US NEXT	
10:25	PETRELLIS G	IL-4 RECEPTOR SIGNALING REGULATES LUNG MACROPHAGES DURING HELMINTH COINFECTION RESULTING IN ENHANCED GAMMAHERPESVIRUS PERMISSIVENESS
10:40	PIEDFORT O	ENTERIC HELMINTH INFECTION IMPAIRS THE MEMORY T CELL RESPONSE TO A RECOMBINANT VESICULAR STOMATITIS VIRUS VECTOR VACCINE
10:55	Pitch ITM: Establishment Malaria consortium in Belgium	
11:00	Coffee break	
Session 2 - Chair: Sarah Hendrickx		
11:30	SADLER R	THE ROLE OF COMPLEMENT FACTOR 3 (C3) IN MALARIA-ASSOCIATED ACUTE KIDNEY INJURY
11:45	JACOBS Y	EARLY NEUTROPHIL RESPONSES UPON INFECTION WITH AFRICAN TRYPANOSOMES
12:00	DE CLEENE W	ELUCIDATING THE MOLECULAR BASIS FOR THE RECOGNITION OF HUMAN BASIGIN BY PLASMODIUM VIVAX TRYPTOPHAN-RICH ANTIGENS
12:15	1-minute poster pitches	
12:45	Lunch	
13:40	BSPP statutory meeting	
Session 3 - Chair: Benjamin Dewals		
14:00	**Keynote 2:dr. Susanne Hart** – ASCARIS: HOST INTERACTION AND BACTERIAL COINFECTIO	
14:25	ZENG L	DEVELOPMENT OF A CAMELID SINGLE-DOMAIN ANTIBODY-BASED ANTIGEN DETECTION ASSAY FOR THE PAN-SPECIFIC DIAGNOSIS OF ACTIVE HUMAN AND ANIMAL TRYPANOSOMA BRUCEI INFECTIONS
14:40	ÁLVAREZ-RODRÍGUEZ A	A CRISPR-CAS-BASED RECOMBINASE POLYMERASE AMPLIFICATION ASSAY FOR ULTRA-SENSITIVE DETECTION OF ACTIVE TRYPANOSOMA BRUCEI EVANSI INFECTIONS
15:55	TIRUNEH A	DEVELOPMENT, FIELD-TESTING, AND OPTIMIZING OF A DIAGNOSTIC TOOL TO QUANTIFY SOIL-TRANSMITTED HELMINTH LIFE STAGES IN PIT LATRINE SLUDGE IN JIMMA TOWN, ETHIOPIA
15:10	Coffee break	
Session 4 - Chair: Johannes Charlier		
15:40	**Keynote 3: dr. Cyril Hammoud** – LONG-TERM CHANGES IN WILDLIFE POPULATIONS: INSIGHTS FROM A META-ANALYSIS	
16:05	KATTENBERG E	HIGHLY MULTIPLEXED AMPLISEQ TARGETED NGS ASSAYS FOR MALARIA GENOMIC SURVEILLANCE IN AFRICA
16:20	MABILLE D	EVALUATING THE ANTITRYPANOSOMAL ACTIVITY AND MODE-OF-ACTION OF PYRAZOLOPYRIMIDINES
16:35	SHIGOLEY M	PARASITE DIVERSITY, PATHOLOGY, AND NEGLECT BY LOCAL STAKEHOLDERS IN NILE TILAPIA (OREOCHROMIS NILOTICUS) FROM UPPER TANA RIVER REGION, KENYA
16:50	DE BOCK S	ASSESSMENT OF INTERNAL AND EXTERNAL HELMINTH EGG CARRIAGE BY FLIES FOLLOWING EXPERIMENTAL EXPOSURE
17:00	Break / Award deliberation	
17:15	**Award session and closing remarks** (Avia-Gis, Huvepharma & Zoetis awards)	
17:30	End	

Poster Presentations

GOOSSENS E	EXCESSIVE MORTALITY IN THE CAPE BUFFALO UNDER HIGH ENVIRONMENTAL STRESS - THE ROLE OF TREMATODES AND INVASIVE SNAILS
ANTONOPOULOS A	NEMABIOME AND MIXED AMPLICON METABARCODING FOR SPECIES IDENTIFICATION AND ANTHELMINTIC RESISTANCE SURVEILLANCE: PRELIMINARY RESULTS FROM ON SITE SEQUENCING TRIAL IN GREECE AND POLAND
WIJTEN A	HARNESSING NANOBODIES TOWARDS POINT-OF-CARE TRYPANOSOMIASIS DETECTION
PRESENT C	EXPLORING ANTIGEN PRESENTATION ON BONE MARROW CELLS AFTER LEISHMANIA INFECTION
CRANSHOFF Y	RUXOLITINIB PROTECTS AGAINST MALARIA-ASSOCIATED ACUTE RESPIRATORY DISTRESS SYNDROME
BERGHMANS F	MOLECULAR BASIS FOR INFECTION AND QUIESCENCE OF LEISHMANIA IN THE BONE MARROW
SCHOTTE F	A SYSTEMATIC REVIEW OF AVAILABLE DETECTION METHODS FOR ANISAKID ALLERGENS IN FOOD PRODUCTS
GOES J	TOWARDS SUSTAINABLE WORM CONTROL: FIELD VALIDATION OF A DECISION SUPPORT TOOL FOR TARGETED TREATMENT OF GASTROINTESTINAL NEMATODES IN FIRST-SEASON GRAZING DAIRY CATTLE
DE MEULENAERE K	A PLASMODIUM KNOWLESI A1-H.1 TRANSCRIPTOME TIME COURSE FOCUSING ON THE LATE ASEXUAL BLOOD STAGES
ALI K	PROTEOMIC ANALYSIS AND IDENTIFICATION OF IMMUNOGENIC PROTEINS IN EXCRETORY/SECRETORY MATERIAL OF ASCARIDIA GALLI WORMS: TOWARDS RECOMBINANT-BASED SERODIAGNOSTIC ASSAYS FOR LAYING HENS
VAN DEN BROECK L	IN VIVO SELECTION OF A LEISHMANIA COSMID LIBRARY REVEALS CANDIDATE GENES INVOLVED IN SAND FLY TRANSMISSION
COOLS L	THE BIOINDICATION POTENTIAL OF PARASITIC FAUNA INFECTING MACROINVERTEBRATES IN AFRICAN WETLANDS
DE VOCHT L	IDENTIFICATION OF POTENT SINGLE-DOMAIN ANTIBODIES AGAINST THE MALARIA SPOROZOITE THROUGH SYNTHETIC SINGLE-DOMAIN ANTIBODY LIBRARIES CONTAINING UNCONVENTIONAL DIVERSIFICATION STRATEGIES
BRYS M	HIGH PREVALENCE OF CHORIOPTES BOVIS: AN IMPORTANT FACTOR IN CHRONIC PROGRESSIVE LYMPHEDEMA IN BELGIAN DRAFT HORSES
TOPIC M	DIVERSITY OF PARASITIC COPEPODS FROM ESTUARINE FISHES IN A SOUTH CAROLINA ESTUARY
ABAWARI MJ	EXPLORING THE CONTEXT AND OPPORTUNITIES FOR OPTIMIZING WATER, SANITATION AND HYGIENE FOR THE PREVENTION OF SOIL TRANSMITTED HELMINTHIASIS IN URBAN PRIMARY SCHOOLS: A PHOTOVOICE STUDY, ETHIOPIA
MONSIEURS P	DERMOTROPIC LEISHMANIA DONOVANI IN NEPAL: (RE-) EMERGENCE OF 'PRUDENT' PARASITES AND THREAT FOR ELIMINATION?

GEENS R	TRYPTACKLE – TACKLING THE LIVESTOCK PARASITE TRYPANOSOMA CONGOLENSE BY TARGETING INVARIANT SURFACE GLYCOPROTEINS
GABRIEL S	INTEGRATED ANTHELMINTIC-BASED CONTROL OF TAENIA SOLIUM CYSTICERCOSIS/TAENIASIS, SOIL-TRANSMITTED HELMINTHIASIS AND SCHISTOSOMIASIS: SAFETY, EFFECTIVENESS, AND IMPLEMENTATION STRATEGIES: 3SI-CONTROL
ARAUJO S	IMMUNOLOGICAL MECHANISMS OF RESPIRATORY CO-INFECTIONS: THE IMPACT OF TRYPANOSOMES ON SECONDARY INFECTIONS CAUSED BY MAJOR LOWER RESPIRATORY TRACT PATHOGENS
ARAUJO S	UNVEILING GLIAL CELLS IN THE NASAL MUCOSA AS HOST CELLS FOR LEISHMANIA WITH POTENTIAL IMPLICATIONS IN DISEASE OUTCOMES
WONG S	ADAPTIVE CO-EVOLUTION IN VISCERAL LEISHMANIASIS: THE ROLE OF HOST MIF CYTOKINE AND PARASITE MIMICRY
VRANKEN N	FROM SLIDES TO BYTES: DIGITAL PARASITES INNOVATE EDUCATION AND PROMOTE AWARENESS
VAN ACKER L	DEVELOPMENT OF A COST-UTILITY MODEL FOR EPILEPSY MANAGEMENT: INTEGRATING IMMUNODIAGNOSIS FOR NEUROCYSTICERCOSIS DETECTION
MATOKA T	NGAL DIPSTICK AS A PROGNOSTIC MARKER FOR PEDIATRIC MALARIA-ASSOCIATED ACUTE KIDNEY INJURY IN A RESOURCE-LIMITED SETTING
MWANGI H	ROAD TOWARDS SUSTAINABLE ANTHELMINTIC USE IN GRAZING RUMINANTS: INSIGHTS FROM STAKEHOLDER ENGAGEMENT

Abstracts keynote presentations

Dr John Gilleard - BVsc PhD DipACVM DipEVPC FCAHS FRCVS

Dr. John Gilleard is a veterinarian and Professor of Parasitology at the University of Calgary Faculty of Veterinary Medicine (UCVM). He spent a large part of his early career in the UK and was Professor of Veterinary Parasitology at the University of Glasgow before moving to Canada in 2008. His research program focuses on anti-parasitic drug resistance, molecular diagnostics and effective, targeted and sustainable parasite control. He has a particular interest in parasitic nematodes in ruminant livestock but also has active projects in companion animal and human parasites. He is a Fellow of the Canadian Academy of Health Sciences (CAHS), a Fellow of the Royal College of Veterinary Surgeons (RCVS), former President of the American Association of Veterinary Parasitology (2018) and Associate Dean Research, Faculty of Veterinary Medicine (2012-2018, 2022-2025).

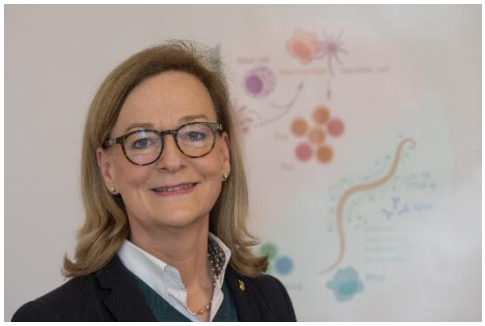
DIAGNOSTICS, SURVEILLANCE, AND BEYOND: WHERE NEMABIOME SEQUENCING WILL TAKE US NEXT

John Gilleard.

Faculty of Veterinary Medicine , University of Calgary, Calgary, Alberta Canada

Advances second and third-generation sequencing technologies over the last decade have revolutionised the analysis of microbial communities and are increasingly used in helminthology research. Although, high quality reference genomes are increasingly available for helminth species, there are still some significant challenges to applying genome-wide approaches to study helminth population genetics and molecular epidemiology in the field. Metabarcoding and targeted deep sequencing have an important role, particularly when dealing with large sample numbers, low amounts and/or low quality template DNA (for example in stool samples), and complex and/or poorly defined helminth communities. This talk will focus on current resources and future directions in both short- and long-read metabarcoding and targeted deep sequencing to study gastrointestinal nematode communities (eg. nemabiome metabarcoding). Specific examples will be used from both animal and human health to illustrate the power of these targeted sequencing approaches to study anthelmintic drug resistance and parasitic nematode molecular epidemiology. Also, the potential role of these methods in the development and application of molecular diagnostics will be discussed.

Prof. Dr. rer. nat. Susanne Hartmann



Susanne Hartmann is Professor for Infection Immunology and Director of the Institute of Immunology at the Department of Veterinary Medicine at Freie Universität Berlin. Her research focuses on infections with parasitic worms (nematodes) in humans and animals. She has demonstrated immunoregulatory processes between parasites and host immune cells and identified the responsible proteins of the parasites. Further, she investigated the potential of parasites and their regulatory products for the treatment of inflammatory processes. Her current research focuses on i) the impact of dietary supplements in the immune response to nematodes in pigs; ii) the interaction of nematodes with co-infections in pigs; and iii) the interaction of gut nematodes with the hosts microbial environment. Susanne Hartmann was president of the German Society for Parasitology from 2017 - 2021 and spokesperson of the DFG-funded Research Training Group “Parasite Infections” from 2015 - 2024. Currently she is chair of the scientific advisory board of the Friedrich-Loeffler-Institute, the federal research institute for animal health and spokesperson of the DFG research training group “One Health approach to soil-transmitted helminths”, funded from 2026 - 2031. Since 2018 Susanne Hartmann is member of the German National Academy of Sciences Leopoldina.

ASCARIS – HOST INTERACTION AND BACTERIAL COINFECTION

Susanne Hartmann

Institute of Immunology, Faculty of Veterinary Medicine, Freie Universität Berlin, Berlin, Deutschland

Soil-transmitted helminths (STH) are among the most common infections worldwide, affecting humans, animals and the environment alike. Within STH infections, Ascaris is the most abundant, with approximately 800 million people being globally infected with Ascaris lumbricoides. Ascaris suum in pigs and Ascaridia galli in chickens are highly prevalent in our important farm animals. We study A. suum infections in their natural host, the pig. These pathogens reside in their host's gut, living in close contact with the host gut microbiota and harbouring bacteria within their own intestines. Questions such as whether and how Ascaris modulates the host gut microbiome and what the nematode microbiome is like and where it is derived from will be discussed. Furthermore, coinfections represent the natural situation such as Ascaris and non-typhoid Salmonella spp. infections are highly prevalent in low- and middle-income countries. Both pathogens are also extremely widespread in commercial pig farming, posing a serious zoonotic threat to human health. Information on the consequences of an acute A. suum infection for the control of a Salmonella Typhimurium challenge in pigs will be discussed. The talk will illustrate the impact on innate cells, such as macrophages and natural killer cells, and their modulated effector functions, as well as antibody responses. Finally, a new graduate school funded by the German Research Foundation on the topic of "One Health Approach to Soil-Transmitted Helminths" will be presented.

Dr. Cyril Hammoud

Cyril Hammoud is an ecologist passionate about parasite biodiversity in general. He studied biology and earth sciences at UCLouvain and the University of Amsterdam, developing strong interests in biogeography and community ecology along the way. Afterward, he pursued a PhD with Ghent University and the Royal Museum for Central Africa (Tervuren), investigating patterns of wildlife trematode diversity in Ugandan crater lakes using molecular approaches. Cyril currently works at the Royal Netherlands Institute for Sea Research as a postdoctoral researcher with the IMPACT Biodiversa+ project focused on European fish parasites. Within this project and together with his collaborators, he is investigating spatial and temporal patterns in fish parasite biodiversity, as well as evaluating the potential for fish parasite conservation in Europe.

LONG-TERM CHANGES IN WILDLIFE PARASITE POPULATIONS: INSIGHTS FROM A QUANTITATIVE SYNTHESIS

C. Hammoud1, J. A. Balbuena2, I. Blasco-Costa3, K. O'Dwyer4, R.A. Paterson5, T. Scholz6, C. Selbach7, 8, B. Sures8, 9, 10, D. Thieltges1, 11

1. Coastal Systems department, Royal Netherlands Institute for Sea Research, The Netherlands; 2. Cavanilles Institute of Biodiversity and Evolutionary Biology, University of Valencia, Spain; 3. Department of Invertebrates, Natural History Museum of Geneva, Switzerland; 4. Marine and Freshwater Research Centre, Atlantic Technological University, Ireland; 5. Norwegian Institute for Nature Research, Norway; 6. Institute of Parasitology, Biology Centre of the Czech Academy of Sciences, Czech Republic; 7. Department of Arctic and Marine Biology, UiT – The Arctic University of Norway, Norway; 8. Water Research Group, Unit for Environmental Sciences and Management, North-West University, South Africa; 9. Aquatic Ecology and Centre for Water and Environmental Research, University of Duisburg-Essen, Germany; 10. Research Center One Health Ruhr, Research Alliance Ruhr, University Duisburg-Essen, Germany; 11. Groningen Institute for Evolutionary Life-Sciences (GELIFES), University of Groningen, The Netherlands

Parasites occur in every ecosystem, affecting nearly all organisms, and despite the dire medical and economic burden caused by a few species, they contribute significantly to biodiversity and support ecosystem stability by performing essential ecological functions. For all their significance, our understanding of how parasitic organisms are affected by anthropogenic environmental changes remains poor, and overarching long-term trends in the abundance or diversity of parasites have yet to be identified. However, for decades, parasite ecologists have recorded changes in infections levels of parasites in a variety of study systems. Here, we attempt to perform a first quantitative synthesis of long-term changes in parasite populations, focusing on wild animal hosts. We compiled a dataset of 1591 time series of changes in the infection levels of parasite populations from 111 studies. Using this dataset, we assessed whether the prevalence of these parasite populations increased, declined, or remained stable through time. Then, we used meta-regressions to identify overarching directional temporal trends across these time series, and to investigate whether ecological or environmental factors could be linked with a propensity to increase or decline in prevalence. Finally, we discuss the significance of these results in the context of the current biodiversity crisis, and provide recommendations for future research in the field.

Abstracts Oral Presentations

IL-4 RECEPTOR SIGNALING REGULATES LUNG MACROPHAGES DURING HELMINTH COINFECTION RESULTING IN ENHANCED GAMMAHERPESVIRUS PERMISSIVENESS

Petrellis G.[1], Rolot M.[1], Preure A.[1] , Lenoir L. [1], Leemans S., [1] Wathieu C.[2], Dougall A.[1] , Chetty A.[3] , Chariot A.[2,4,5], Horsnell W.[3], Dewals B. G.[1,5]

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Viral infections are often studied in pathogen-free settings, yet in regions where helminth infections are endemic, host susceptibility and viral control can be can profoundly altered. To investigate this interaction, we examined murid gammaherpesvirus 4 (MuHV-4) infection in helminth-coinfected C57BL/6 mice, where viral replication in the lungs was markedly enhanced. MuHV-4 has a natural tropism for alveolar macrophages (AlvMs) *in vivo,* and we observed an increased permissiveness restricted to AlvMs and monocyte-derived macrophages after helminth infection. Phenotypic changes in lung macrophages, after helminth exposure, consisted of an *Il4ra* signalling dependent disappearance of SiglecF^{+} macrophages and concomitant enrichment of CD11b^{+} macrophages of monocytic origin, in the lung. Using single-cell RNA sequencing and flow cytometry, we demonstrated that after helminth infection, lung macrophages upregulated proteoglycans known to be decorated with heparan sulfates, potentially improving virion attachment. We further observed a dampened type-I interferon stimulated gene signature in mice coinfected with helminths and MuHV-4. Finally, we found that airway macrophages isolated from helminth-infected C57BL/6 mice were more permissive to low doses of MuHV-4 infection *ex vivo* and that IL-4 and IL-13 could potentiate macrophages susceptibility to MuHV-4 infection. In conclusion, we propose that helminth infection causes phenotypical and functional IL-4Rα-dependent restructuration of the airway and lung macrophages, significantly increasing their permissiveness to gammaherpesvirus coinfection.

ENTERIC HELMINTH INFECTION IMPAIRS THE MEMORY T CELL RESPONSE TO A RECOMBINANT VESICULAR STOMATITIS VIRUS VECTOR VACCINE

Ophelie Piedfort[1,2], Olivier Botman[1], Justine Javaux[1], Remy Sandor[1], Sylvain Leemans[1], Benedicte Machiels[1,*], Benjamin Dewals[1,2,*]

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Vaccination relies on the induction of effective antibody responses, together with the priming and maintenance of long-lived antigen (Ag)-specific memory T lymphocytes. Gastro-intestinal helminths have been associated with reduced vaccine efficacy, through various immunoregulatory mechanisms. However, how helminth infection would impact the dynamics and response of memory T cells remains elusive. Here, we used both a recombinant vesicular stomatitis virus (rVSV) vector and an mRNA-based vaccine to investigate the memory T cell responses to the chicken ovalbumin (Ova) model Ag during enteric infection with the parasite nematode *Heligmosomoides polygyrus*. Naïve or *H. polygyrus* -infected mice were vaccinated at day 14 after infection with rVSV-Ova or mRNA-Ova intramuscularly. We observed a significant reduction of total, Ag-specific $CD8^+$ T cells and adoptively transferred Ova-specific OT-I cells in blood and spleen when vaccination occurred during helminth infection. To further investigate the response to vaccination during helminth infection, splenocytes at day 3 or 7 after rVSV-Ova administration were depleted of B cells, NK cells and neutrophils before single-cell (sc)RNA sequencing, including T cells, monocyte/macrophages and dendritic cells. While a cluster of *Cd4*-expressing T cells upregulated *Gata3, Cxcr6, Il4* and *Il1rl1* (T1/ST2) after *H. polygyrus* infection, a cluster of *Cd8a/Cd8b1*-expressing T cells upregulated effector/memory signature genes (*Gzmb, Cx3cr1, Klrg1, Itga4*) and was strongly enriched from day 3 to day 7 after vaccination. Strikingly, effector/memory $CD8^+$ T cells following vaccination of *H. polygyrus*-infected mice showed significantly lower enrichment, with reduced cytotoxic (*Gzma, Gzmb, Gzmk*) and activation (*Ccl5, Klrk1, Cx3cr1*) gene signatures. Finally, when challenged intratracheally with a murid gammaherpesvirus 4 recombinant strain expressing the OT-I epitope, vaccinated and *H. polygyrus*-infected mice showed a defective response in the lung of Ag-specific $CD8^+$ T cells and adoptively transferred OT-I cells. These results demonstrate that helminth infection impacts memory T cell responses and opens new avenues for mechanistic insights.

THE ROLE OF COMPLEMENT FACTOR 3 (C3) IN MALARIA-ASSOCIATED ACUTE KIDNEY INJURY

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Malaria remains a global life-threatening disease, with 260 million clinical cases and ~600 000 deaths annually. Malaria-associated acute kidney injury (MAKI) is a leading cause of morbidity and mortality. An estimate of 24-59% of African children with severe malaria develop acute kidney injury (AKI). Currently, renal replacement therapy is the only effective treatment for MAKI, yet it is often inaccessible in malaria-endemic regions.

The pathogenesis of MAKI remains poorly understood. Elevated levels of immune complexes (ICs) and activation of the complement system have been reported in severe malaria. Given the established role of complement activation in various kidney diseases, this study investigated the role of complement component 3 (C3) in a mouse model of MAKI.

Wild-type (WT) and C3 knock-out (KO) C57BL/6 mice were infected with *Plasmodium berghei* NK65 (*Pb*NK65) and monitored from six days post infection (dpi) onwards. C3 deficiency did not affect the clinical progression of infection. *Pb*NK65-infected WT and C3 KO mice developed mild MAKI alongside lethal malaria-associated acute respiratory distress syndrome (MA-ARDS), with comparable lung pathology and impaired lung function. Glomerular and tubular injury were equivalent between infected groups, as shown by comparable proteinuria, glomerular collapse, and renal expression of injury markers lipocalin-2, heme oxygenase-1 and interferon-γ. Upon infection, circulating ICs were lower in C3 KO compared to WT mice. Despite the absence of C3, plasma levels of the soluble membrane attack complex (sC5b-9) were similarly increased as in WT-infected mice, suggesting C3-independent complement activation.

Our findings indicate that MAKI and MA-ARDS occur independently of C3. The increased levels of sC5b-9 suggest activation of the complement cascade through a C3-independent mechanism. Ongoing investigations using a C5 inhibitor, acting further downstream of C3, aim to evaluate the potential therapeutic benefit of complement inhibition in MAKI.

EARLY NEUTROPHIL RESPONSES UPON INFECTION WITH AFRICAN TRYPANOSOMES

Mabille D.[1], **Jacobs Y.[1]**, Yu T.[1], De Kock J.[1], Maes L.[1], Tacchini-Cottier F.[2], Baeza Garcia A.[1], Wullaert A.[3], Caljon G.[1]

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Human African Trypanosomiasis (HAT), also known as sleeping sickness, is a tsetse fly transmitted parasitic disease indigenous for the African continent. Trypanosome transmission occurs during the blood feeding of infected flies, resulting in the inoculation of parasites in the skin. Insights into the early immunological events upon an infectious bite could potentially provide a scientific basis for the development of novel transmission-blocking approaches. Neutrophils were shown to exhibit a detrimental role during trypanosome infections. Depletion experiments and infections in neutrophil-deficient mouse models demonstrated an exacerbating role in the early parasite dissemination phase towards systemic infection establishment. In addition, damage to the spleen microarchitecture from neutrophil metalloproteinase was shown to hamper infection control.

To further dissect the trypanosome-neutrophil interactions, human neutrophils were isolated from the peripheral blood of healthy donors and exposed in vitro to naïve tsetse fly saliva and live bloodstream form (BSF) parasites. Co-culture experiments were also conducted in the presence of pharmacological inhibitors of toll-like receptor (TLR) and purinergic receptor activation. In vivo infection experiments were performed in multiple genetically modified mouse models (PADi4 -/-, TNF -/- and MPO -/- mice), combined with bioluminescent imaging, to determine the role of neutrophils in parasite dissemination and systemic infection.

Exposure of human neutrophils to naive tsetse fly saliva and BSF parasites significantly prolonged neutrophil survival in vitro. This prolonged survival was shown to be mediated by TLR and purinergic receptor activation. Furthermore, BSF parasites induced a robust cellular activation with degranulation of azurophilic granules and neutrophil extracellular trap (NET) formation. However, limited parasite cell death and phagocytosis in the presence of neutrophils demonstrated that the parasite is not hampered by activated neutrophils. Notably, infections in mice with a systemic deficiency in NET formation resulted in a tenfold higher peak parasitaemia and accelerated mortality indicating differential contributions of neutrophil cellular processes in early infection establishment and systemic parasite control.

ELUCIDATING THE MOLECULAR BASIS FOR THE RECOGNITION OF HUMAN BASIGIN BY PLASMODIUM VIVAX TRYPTOPHAN-RICH ANTIGENS

Witse De Cleene[1], Lien Boeykens[1], Dalia Diaz[2], Katlijn De Meulenaere[2], Natalia Smiejkowska[1], Anna Rosanas-Urgell[2], Yann G.J. Sterckx[1]

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Plasmodium vivax is the most widespread human-infective malaria parasite. Despite having a severe socio-economic impact, the progress in battling *P. vivax* is slow. Problems are worsened due to low-efficacy vaccines, drug-resistant parasites and disease (re-)emergence. This calls for active research into *P. vivax* biology. Invasion of a host reticulocyte (retic) by the merozoite (MRZ) is an essential event in the parasite's life cycle. Yet, our understanding of interactions at the MRZ-retic interface is limited. The PvTRAgs are MRZ surface antigens mediating retic binding. PvTRAg35.2 and PvTRAg38 are known to interact with basigin (hBSG), but many aspects of this interaction are yet to be investigated: 1) the structural basis for hBSG recognition is unknown, 2) the molecular determinants underlying the versatility displayed by PvTRAg-hBSG interactions remain enigmatic, and 3) how these events relate to retic invasion is unclear. Given the knowledge gap in *P. vivax* biology and the importance of PvTRAgs in MRZ biology, tackling these issues is expected to generate many novel findings that may support *P. vivax* specific vaccine design efforts. The recombinant production and purification of these PvTRAgs and hBSG have been established within our research group and the first steps towards unraveling the molecular basis for the interaction between PvTRAg38 and hBSG have been taken, by grating coupled interferometry, isothermal titration calorimetry and macromolecular X-ray crystallography. Furthermore, llamas have been immunized with PvTRAg38. The generated sdAbs, interaction and non-interaction blocking, could be of significant interest for further applications and can be used as tools for structural studies.

DEVELOPMENT OF A CAMELID SINGLE-DOMAIN ANTIBODY-BASED ANTIGEN DETECTION ASSAY FOR THE PAN-SPECIFIC DIAGNOSIS OF ACTIVE HUMAN AND ANIMAL *TRYPANOSOMA BRUCEI* INFECTIONS

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Trypanosoma brucei (*T. brucei*) infections cause African trypanosomiasis, also known as sleeping sickness in humans and nagana in animals, presenting a significant global health and economic burden, especially in sub-Saharan Africa. Animal trypanosomiasis also affects the economic development in Asia and South America. Accurate diagnosis of diseases caused by infection with the parasite of *T. brucei* group remains a major challenge due to the persistence of infection-induced antibodies long after parasite clearance, complicating serological discrimination between active and past infections. To address this limitation, we developed a sensitive and specific antigen-detection assay targeting *T. brucei* enolase (*Tbr*ENO), using a panel of camelid single-domain antibodies (sdAb aka nanobodies). Among the candidates, the sdAbsR3-77/sdAbR2-103 sandwich ELISA exhibited robust performance in detecting circulating *Tbr*ENO in plasma from experimentally infected mice. Additionally, this assay showed strong potential as a "test-of-cure" tool by monitoring real-time antigenemia throughout a chronic *T. brucei* infection course. We further validated the assay's diagnostic utility in human clinical samples, detecting *T. b. rhodesiense* infections at both early and advanced stages and *T. b. gambiense* infections in advanced stage. The sdAbsR3-77H/sdAbR2-103HA ELISA targeting *Tbr*ENO shows potential for point-of-care (POC) pan-diagnosis of active *T. brucei* infections (including *T. b. brucei*, *T. b. gambiense*, *T. b. rhodesiense*, *T. b. evansi* and *T. b. equiperdum*) in both animals and humans. Therefore, this assay addresses gaps in current diagnostic capabilities by overcoming the key limitations of antibody-based tests, offering a promising tool for improved disease control.

A CRISPR-CAS-BASED RECOMBINASE POLYMERASE AMPLIFICATION ASSAY FOR ULTRA-SENSITIVE DETECTION OF ACTIVE TRYPANOSOMA BRUCEI EVANSI INFECTIONS

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Control of *Trypanosoma brucei evansi* (*T. b. evansi*) infections remains a significant challenge in managing Surra, a widespread veterinary disease affecting both wild and domestic animals. In the absence of an effective vaccine, accurate diagnosis followed by treatment is crucial for successful disease management. However, existing diagnostic methods often fail to detect active infections, particularly in field conditions. Recent advancements in CRISPR-Cas technology, combined with state-of-the-art isothermal amplification assays, offer a promising solution. This approach has led us to the development of a *Tev*RPA-CRISPR assay, a highly sensitive and specific *T. b. evansi* diagnostic tool suitable for both laboratory and field settings.

First, the *Tev*CRISPR-Cas12b cleavage assay was developed and optimized, and its analytical sensitivity was evaluated. Next, this technology was integrated with the *Tev*RPA to create the *Tev*RPA-CRISPR test, with the reaction conditions being optimized and its analytical sensitivity and specificity assessed. Finally, the test's accuracy in detecting both active and cured *T. b. evansi* infections was evaluated.

The optimized *Tev*CRISPR-Cas12b cleavage assay demonstrated the ability to detect *T. b. evansi* target DNA at picomolar concentrations. Integrating *Tev*CRISPR-Cas12b with RPA in Two-Pot and One-Pot *Tev*RPA-CRISPR tests achieved up to a 100-fold increase in analytical sensitivity over RPA alone, detecting attomolar concentrations of *T. b. evansi* target DNA, while maintaining analytical specificity for *T. b. evansi*. Both assays exhibited performance comparable to the gold standard *Tev*PCR in experimental mouse infections, validating their effectiveness for detecting active infections and assessing treatment efficacy.

The *Tev*RPA-CRISPR tests prove highly effective for diagnosing active infections and assessing treatment efficacy, while being adaptable for both laboratory and field use. Thus, the *Tev*RPA-CRISPR assays emerge as a promising addition to current diagnostic tools, offering efficient and reliable detection of active *T. b. evansi* infections.

DEVELOPMENT, FIELD-TESTING, AND OPTIMIZING OF A DIAGNOSTIC TOOL TO QUANTIFY SOIL-TRANSMITTED HELMINTH LIFE STAGES IN PIT LATRINE SLUDGE IN JIMMA TOWN, ETHIOPIA

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Worldwide, schoolchildren are dewormed to control the morbidity attributable to soil-transmitted helminthiasis (STH). To monitor the effectiveness of these control programs, WHO recommends to periodically assess the prevalence and intensity of STH infections across children in sentinel schools. Yet, these surveys are often resource demanding and even disturb the school activities. A potentially cost-saving alternative that does not involve children, is to process sludge samples from school latrines. However, little is known on how to best process samples and how many samples are required to make reliable program decisions.

We developed a diagnostic method and conducted series of spiking experiments to explore the diagnostic performance (analytical sensitivity and egg recovery rate) of our egg-counting method. Second, we estimated the variation in egg counts from 40 sludge samples collected at different depths across 24 pits from eight latrines (used by boys and/or girls) in six primary schools in Jimma Town (Ethiopia). Each sample was processed four times, and four aliquots per sample were screened. Finally, these field data were used to inform an egg count simulation model to quantify variation in egg counts and determine the sampling (number of samples per school) and diagnostic strategy (number of sample preps and aliquots).

Our spiking experiment demonstrated that the diagnostic method had an analytical sensitivity of 10-20 eggs per gram of sludge. In the field samples, six medically important worm species were detected, including *Ascaris* and *Trichuris*. Based on our simulation study, it is recommended to sample three sludge samples per school and process one sample prep and one aliquot per sample.

We have thoroughly validated our tool and optimized both the sampling and diagnostic strategy. In a follow-up study we will sample sludge samples from 25 schools and compare the mean egg counts at school level with the STH prevalence based on the examination of stool of schoolchildren (52 children per school).

Key words: deworming programs, soil-transmitted helminths, monitoring and evaluation, sludge, pit latrines

HIGHLY MULTIPLEXED AMPLISEQ TARGETED NGS ASSAYS FOR MALARIA GENOMIC SURVEILLANCE IN AFRICA

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Emerging drug and diagnostic resistance highlight the urgency of continuous surveillance of malaria. Additionally, malaria vaccine rollout exerts unprecedented selection pressure on parasite populations. Genomic surveillance, which uses genetics to track diseases, aids in decision-making for malaria control and elimination. We developed a highly-multiplexed sequencing assay (AmpliSeq) for cost-effective targeted deep sequencing of *Plasmodium falciparum*. This assay integrates phenotypic and population genetic markers to study parasite dynamics in Sub-Saharan Africa and offers advantages over other methods through its large number of targets, full-length gene coverage, and exceptional sensitivity to low-density infections. The standardized, rapid protocol has been successfully piloted in Burkina Faso, the Democratic Republic of Congo, and Kenya.

We demonstrate the application of the assay to investigate the prevalence and spread of markers associated with chloroquine, sulfadoxine-pyrimethamine and artemisinin resistance, and explore the rate of *hrp2/3* deletions associated with false negative rapid diagnostic tests in West, Central and East Africa. In addition, we present the diversity of *Pfcsp*, the target included in malaria vaccines (both RTS,S and R21) and compared this diversity between continents, including also samples from Latin America and South East Asia.

Our approach can effectively differentiate and characterize parasite isolates over time and space, and is easily adaptable to diverse epidemiological contexts. We show how results from these analyses can guide surveillance and implementation of control interventions such as chemotherapy and vaccine deployment strategies. The priority is to make this tool available to key actors in endemic countries to increase ownership and ensure data usage for decision-making and policy.

Key words: Malaria, genomic surveillance, molecular epidemiology, NGS, *Plasmodium falciparum*, *Plasmodium vivax*.

EVALUATING THE ANTITRYPANOSOMAL ACTIVITY AND MODE-OF-ACTION OF PYRAZOLOPYRIMIDINES

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Human African trypanosomiasis (HAT), caused by *Trypanosoma brucei*, leads to severe neurological dysfunction and is fatal if left untreated. Current chemotherapeutic options are limited and often associated with toxicity or emerging resistance. Pyrazolopyrimidines have recently emerged as a promising compound class with potent anti-trypanosomal activity. This study investigates the mode-of-action of two lead compounds, NPD-2975 and NPD-3519, against *T. brucei*.

An inducible genome-wide RNA interference (RNAi) library was employed to identify genes modulating compound sensitivity. Candidate targets were validated *in vitro*. Comparative metabolomic profiling was used to uncover affected metabolic pathways, and Ligand Gaussian accelerated molecular dynamics simulations were performed to evaluate compound-target interactions.

Compound exposure resulted in multinucleated parasites, indicative of cytokinesis failure. RNAi screening implicated the *leucine carboxy methyltransferase* (*lcmt*) gene as a key mediator of compound activity. Metabolomic analysis revealed perturbations in S-adenosyl-L-methionine metabolism which is consistent with impaired methyltransferase function, while the L-glutamine pathway was also affected. Molecular dynamics simulations demonstrated that NPD-2975 destabilizes the interaction between LCMT and its co-factor PP2A, consistent with the observed cellular phenotype.

Our results indicate that NPD-2975 exerts it anti-trypanosomal activity by interfering with LCMT-PP2A regulation and associated metabolic pathways, leading to cytokinesis arrest and parasite death. These findings identify PP2A regulation as a critical vulnerability in *T. brucei* and establish pyrazolopyrimidines as compelling leads for next-generation HAT therapeutics.

PARASITE DIVERSITY, PATHOLOGY, AND NEGLECT BY LOCAL STAKEHOLDERS IN NILE TILAPIA (*OREOCHROMIS NILOTICUS*) FROM UPPER TANA RIVER REGION, KENYA

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Nile tilapia (*Oreochromis niloticus*), a globally important farmed species, faces under-reported parasitological challenges. This study adopted an integrated, mixed-methods approach in Kenya's Upper Tana River region, combining detailed biological health assessments with a qualitative survey of fish health management. Between mid-January and mid-February 2024, we sampled and examined 157 Nile tilapia hosts to document parasite diversity and assess tissue-level impacts. Gill, intestine, and liver samples were examined histologically, and parasites from external and internal organs were identified using light microscopy, molecular barcoding, and scanning electron microscopy. We documented a diverse assemblage of parasites including members of *Clinostomum*, *Euclinostomum*, *Cichlidogyrus*, *Scutogyrus*, and *Acanthogyrus* with variable prevalence and intensity among hosts. Histopathological examination commonly revealed notable gill tissue alterations, including hypertrophy, epithelial hyperplasia, lamellar fusion, vascular congestion of the secondary lamellae and associated inflammation. Complementing this, we engaged 48 stakeholders (36 fish farmers, 11 fisheries and extension officers, and one veterinarian) to understand local fish health knowledge and practices. Qualitative findings revealed a persistent neglect of parasites in farm decision-making, a "don't know, don't care" dynamic driven by low awareness, weak diagnostic capacity, and limited institutional feedback. Farmers rarely linked observed disease signs to parasitism, while training gaps among extension officers eroded trust and encouraged improvised treatments. Framed by the concept of "undone science," this local neglect is structural rather than individual. We propose a Challenge and Reconstruct Learning (ChaRL) framework that uses parasitological evidence to prompt collective reflection and coordinated action among stakeholders. By making biologically confirmed infections visible both scientifically and socially, this study bridges pathology and practice, providing a practical way to improve tilapia health management in this region.

ASSESSMENT OF INTERNAL AND EXTERNAL HELMINTH EGG CARRIAGE BY FLIES FOLLOWING EXPERIMENTAL EXPOSURE

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Synanthropic flies are suspected mechanical transmitters of fecal-borne helminth diseases. To better understand their role in disease transmission, controlled experimental studies are required. This study investigates the carriage of helminth eggs on the exoskeleton and inside the digestive tract of house flies (*Musca domestica* L.) following experimental exposure to feces containing eggs of *Ascaris suum* and *Taenia saginata*.

Forty-eight house flies, allocated to eight cages, were exposed to non-human primate feces spiked with 10,000 *A. suum* eggs/g of feces (EPG) and 5,000 *T. saginata* EPG under controlled environmental conditions (30 °C, 60% relative humidity). After three hours, eggs were recovered from the flies' exoskeletons and digestive tracts and quantified using previously validated protocols. Additionally, fly contact time with the feces was recorded at the cage level.

A total of 227 helminth eggs, comprising 46 *A. suum* and 181 *T. saginata* eggs, were recovered from 48 flies. Significantly more eggs were recovered from the digestive tracts (range: 0–21 eggs/fly; mean: 4.6 eggs/fly; 33/48 flies tested positive) compared to the exoskeletons (range: 0–2 eggs/fly; mean: 0.1 eggs/fly; 4/48 flies tested positive). Female flies (mean: 6.7 eggs/fly) carried approximately twice as many eggs as male flies (mean: 3.2 eggs/fly). At the cage level, the total time spent by the six flies in contact with the feces ranged from 31 to 207 minutes but was not significantly associated with the total number of eggs recovered per cage (range: 5–46 eggs/cage; Spearman's $\rho = 0.47$, $p = 0.243$).

This study confirms the potential for house flies to carry helminth eggs under favorable conditions, primarily acquired through ingestion. To further clarify their contribution to disease transmission, future studies should evaluate the subsequent deposition of ingested eggs onto relevant surfaces and translate these experimental observations to field settings.

Abstracts poster presentations

EXCESSIVE MORTALITY IN THE CAPE BUFFALO UNDER HIGH ENVIRONMENTAL STRESS - THE ROLE OF TREMATODES AND INVASIVE SNAILS

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Trematodes are flatworm parasites with complex life cycles involving freshwater snails and a wide range of definitive hosts, including humans and other mammals. While they are part of natural ecosystems, trematodes can threaten wildlife conservation when transmission intensifies due to human or livestock activity, or when wildlife populations are already suffering under environmental stress. We investigated trematode infections in African buffalo (*Syncerus caffer*) in a small Zimbabwean game reserve following excessive mortality in the herd.

Adult liver and stomach flukes were collected from a sick buffalo that was euthanized. Simultaneously, freshwater snails from nearby dams and streams were screened for larval trematodes (cercariae) through shedding and DNA analysis. Morphological analysis combined with COI and ITS sequencing of 22 adult parasites, nine cercariae, and 11 infected snails suggests the presence of both known and unknown trematode species. The liver fluke *Fasciola gigantica* was found in the buffalo but not in snails, despite the presence of *Pseudosuccinea columella*, an invasive snail linked to fasciolosis. However, we genetically linked stomach flukes (*Calicophoron* aff. *microbothrium*) from the buffalo to larval stages infecting *P. columella*, for the first time, highlighting this invasive snail's role in potential parasite spillback. Furthermore, *Echinostoma revolutum* and *Plagiorchis maculosus* were identified in *Radix natalensis* and *P. columella* snails, respectively. We also recorded an unidentified planorbid snail, which was infected with an unknown trematode. The closest sequence matches corresponded to an exotic Asian snail species, but we suspect it may be an overlooked endemic planorbid. Further analyses should resolve this issue and shed more light on these mysterious hosts and their parasites. Our findings highlight the importance of monitoring parasite transmission in the face of environmental change, and particularly the invasion of non-native snails and the possibility of parasite spillback, as increased transmission could put extra pressure on populations already under stress.

Key words: trematodes, wildlife, invasive snails, parasite spillback

NEMABIOME AND MIXED AMPLICON METABARCODING FOR SPECIES IDENTIFICATION AND ANTHELMINTIC RESISTANCE SURVEILLANCE: PRELIMINARY RESULTS FROM ON SITE SEQUENCING TRIAL IN GREECE AND POLAND

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INTRODUCTION: Collectively, helminth infections are one of the most significant sources of production losses and animal welfare issues in Europe. Treatment relies on regular administration of broad-spectrum anthelmintics from a limited number of classes. Reports of resistance are now widespread. Sustainable approaches to helminth control are essential, but require accurate diagnostics. Next-generation sequencing technologies have been extensively demonstrated for Nemabiome and resistance surveillance mainly using Illumina. Here we present findings from an initial pilot study examining the use of on-site Nanopore sequencing for Nemabiome and anthelmintic resistance monitoring in Greece and Poland.

METHODOLOGY: In this study we undertook a rapid mixed amplicon nanopore sequencing approach to detect larval isolates to the species level, and to survey anthelmintic resistance associated SNPs for BZ and LEV. We amplified the ITS2 region, *beta-tubulin* isoform-1, and exon 4 of the *acr-8* gene, for species identification and benzimidazole and levamisole resistance marker detection respectively. Amplicons were sequenced using the Oxford Nanopore MK1D sequencer.

RESULTS: We initially examined pooled samples at the regional level in Northern Greece, and found that *H. contortus* was the dominant species (>95%) in ¾ regions. We further found a high relative frequency of BZ resistance alleles, but we did not detect LEV resistance alleles. These results are broadly in line with Illumina-seq data. We then examined 36 goat herds in Poland at the farm level. We found *H. contortus* was the dominant species in the majority of herds, although we also found significant presence of *Trichostrongylus* spp. We further noted widespread presence at high relative frequency of the F200Y BZ resistance mutation in both *H. contortus* and *Trichostrongylus* spp, in addition to LEV resistance allele S168T present at 40% relative frequency in several herds.

DISCUSSION: Nanopore sequencing presents a valuable opportunity for improving epidemiological monitoring of anthelmintic resistance, particularly in Central and Southern Europe. Here we successfully demonstrate its use for simultaneous Nemabiome and anthelmintic resistance marker detection in multiple species. We report widespread BZ resistance, and emerging LEV resistance in Poland. However, poor read depth for some species and amplicons needs to be addressed in future work.

HARNESSING NANOBODIES TOWARDS POINT-OF-CARE TRYPANOSOMIASIS DETECTION

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African trypanosomiasis is a neglected tropical disease endemic to Africa, Asia, and South America and frequently found in Europe. The World Health Organization (WHO) has prioritized the elimination of this disease by 2030, emphasizing the need for improved surveillance and diagnostic tools.[1] Current antibody-based methods cannot differentiate between active and past infections. Biomarkers like *T. congolense* pyruvate kinase (TcoPYK), which indicate active infection, offer a more targeted diagnostic approach. This work explores the use of camelid single-domain antibodies in a photoelectrochemical (PEC) platform for TcoPYK detection. While these nanobodies have shown promise in ELISA assays, these are time-consuming and require specialized personnel. Here, we propose a sensitive, user-friendly, and point-of-care-compatible PEC device integrating nanobodies through two distinct detection strategies.

Both approaches rely on photosensitizer-mediated singlet oxygen ($^1O_2$) generation, which reacts with a redox mediator (e.g., hydroquinone) to produce a quantifiable photocurrent.[2] In the indirect method, nanobodies are fused with alkaline phosphatase, which enzymatically converts fluorescein diphosphate into active fluorescein a type II photosensitizer. In the direct method, fluorescein is site-specifically conjugated to the detection nanobody via sortase labeling in a sandwich-type assay format.

To enable selective target capture and improve performance in complex matrices, the capture nanobody is functionalized with biotin and immobilized on streptavidin-coated magnetic beads, facilitating easy sample handling and reducing matrix effects. This dual-strategy PEC platform underscores the versatility of nanobodies in biosensing and advances the development of rapid, robust, and deployable diagnostics for African trypanosomiasis.

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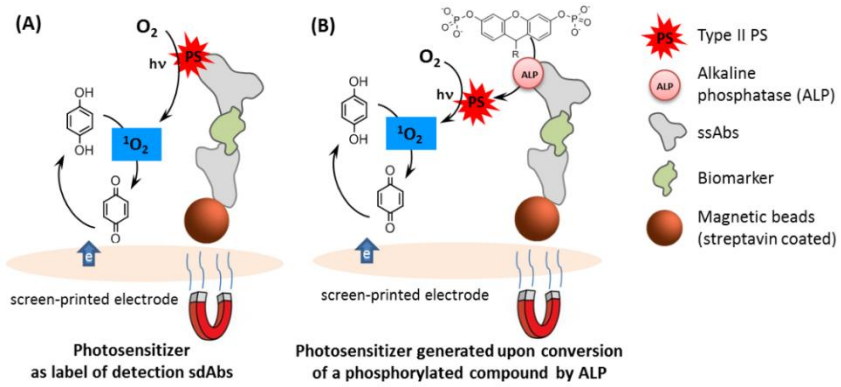
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phosphatase (ALP)
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Biomarker
Magnetic beads
(streptavin coated)

EXPLORING ANTIGEN PRESENTATION ON BONE MARROW CELLS AFTER *LEISHMANIA* INFECTION

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INTRODUCTION: Visceral leishmaniasis (VL) is a neglected tropical disease caused by the protozoan *Leishmania* parasites. It is transmitted through the bites of infected female sand flies, primarily in tropical and subtropical regions. Many current therapeutic drugs face important challenges such as limited availability, toxicity and resistance development. Moreover, we recently discovered relapse occurring as a result of quiescent *Leishmania* parasites surviving in stem cells (LT-HSC) of the bone marrow. Given the infection-induced upregulation of *transporter associated with antigen processing 1* (TAP1) in LT-HSC, we hypothesize that infection triggers antigen presentation. Therefore, our aim is to study the expression of Major Histocompatibility Complex proteins I and II (MHCI and MCHII) on different cell types in the bone marrow upon infection *in vivo* and *ex vivo*.

METHODOLOGY: Antibody panels were designed for the identification and characterization of various immune and progenitor subsets in the bone marrow using a MACSQuant Analyzer 10 flow cytometer. For the *ex vivo* experiments, bone marrow cells were isolated from BALB/c mice, infected and measured at three different time points: 24 hpi, 48 hpi and 72 hpi. For the *in vivo* experiments, BALB/c mice were infected for three weeks, before isolating the bone marrow and analysing the cellular composition and activation. To avoid autofluorescence as a potential confounding factor and to extend the panel of markers, spectral flow cytometry using the Cytek Aurora Northern Lights was also initiated. The choice of compatible fluorochromes, titrations of fluorescent antibodies, and inclusion of appropriate single stains and fluorescence minus one controls was part of the panel optimization.

RESULTS: *In vivo* infection resulted in an upregulation of MHCI expression on LT-HSCs. It is important to note that non-infected and infected cells cannot be discriminated based on fluorescence in the bone marrow of infected mice. In the *ex vivo* immunophenotyping studies, this distinction is possible based on the DsRed signal from the used reporter parasite line. While overall MHCI expression levels increase, separation based on the cellular infection status demonstrates reduced MHCI and MHCII expression levels on infected compared to non-infected LT-HSCs. The functional implications for the induction or avoidance of CD4+ proliferative responses and CD8+ cytolytic activity will be explored as well.

RUXOLITINIB PROTECTS AGAINST MALARIA-ASSOCIATED ACUTE RESPIRATORY DISTRESS SYNDROME

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Malaria remains a major global health burden, causing hundreds of millions of cases and nearly 600,000 deaths annually, primarily in young children in sub-Saharan Africa. While current antimalarial therapies effectively eliminate blood-stage parasites, they often fail to reverse associated organ or tissue damage. Consequently, severe malaria continues to result in high mortality, largely due to complications such as cerebral malaria, severe malarial anemia, malaria-associated acute respiratory distress syndrome (MA-ARDS), and metabolic imbalances.

Our previous work demonstrated that Ruxolitinib, an inhibitor of the JAK1/2-pathway, is able to rescue glucocorticoid receptor knockout mice from malaria-induced lethal hypoglycemia and hyperinflammation[1].

Here, we further evaluate the potential of Ruxolitinib to inhibit MA-ARDS pathogenesis in a mouse model of severe malaria with *Plasmodium berghei* NK65. While treatment with Ruxolitinib did not influence parasitemia, it alleviated severe malaria pathology. In particular, it significantly improved clinical scores, restored normoglycemia, and reduced lung pathology, as indicated by reduced lung weight, decreased alveolar edema, and enhanced lung ventilation function. Collectively, these results highlight the potential of Ruxolitinib as an adjunctive therapy to improve outcomes in severe malaria.

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MOLECULAR BASIS FOR INFECTION AND QUIESCENCE OF LEISHMANIA IN THE BONE MARROW

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Leishmaniasis is a major neglected parasitic disease caused by different *Leishmania* species and transmitted by blood-feeding sand flies. Visceral leishmaniasis, one of the major diseases caused by these parasites still leads to 20,000-30,000 deaths annually. The treatments that are currently available are scarce and suffer limitations in terms of logistics, route of administration, and toxicity. The development of resistance as well as treatment failure have been on the rise for decades. It was recently found that one of the causes of treatment failure/post-treatment relapse relates to infection of long-term hematopoietic stem cells (LT-HSCs) in the bone marrow, as a protective niche for *Leishmania* parasites. This niche protects them from elimination by the immune system and anti-leishmanial drugs. The parasite has also been observed to acquire a quiescent phenotype inside this niche which makes them even harder to eliminate. A transcriptional profile of the infected LT-HSCs was recorded, the "*StemLeish*" signature, defined by an upregulated TNF/NF-kB and CXCR4/RGS1/TGF-ß/SMAD/SKIL signaling as well as a downregulated oxidative burst.

In this collaborative FWO-funded project the first goal is to characterize the stem cell niche and the underlying pathogen-host interactions, with techniques such as single cell RNA-sequencing, spatial transcriptomics and high dimensional flow cytometry. Secondly, to get more insights into the mechanisms of parasitic quiescence, the effects of targeted gene editing on infection and quiescence will be studied. Thirdly, the "*StemLeish*" signature as well as new discoveries coming from the characterization of the niche and quiescence will be used to target specific genes *in vivo* using pharmacological inhibitors and nanobodies. It is expected that in-depth molecular understanding of the stem cell niche and parasite quiescence will advance treatment.

A SYSTEMATIC REVIEW OF AVAILABLE DETECTION METHODS FOR ANISAKID ALLERGENS IN FOOD PRODUCTS

Faust Schotte

The Anisakidae family consists of zoonotic nematode species such as Anisakis and Phocanema. Detection of Anisakid nematodes is considered an important component in ensuring seafood safety and quality. Currently methods used in the industry are mainly based on visual inspection. However, detection of Anisakid material cannot be solely based on visual methods, as allergens can be present in food products through transmission in the food chain. The need for more sensitive, rapid, and scalable techniques has led to the development and application of a range of analytical methods.

A systematic review was conducted to provide a comprehensive overview of current methodologies, and identify the best performing methods for routine testing or scientific research. The literature search identified 25 papers describing at least one detection method. There are 3 main categories that can be distinguished, namely DNA-based (n = 10), immunochemistry-based (n = 14), and mass spectrometry-based (MS-based) detection methods (n = 5). DNA-based methods consisted of conventional PCR, real-time PCR (RT-PCR), high throughput sequencing (HTS) and loop-mediated isothermal amplification (LAMP). The targeted region was mainly the mitochondrial cytochrome c oxidase I or II gene (COI & COII), or the internal transcribed spacer (ITS) region. The LOD was as low as 4*10-6 ng larval DNA per ng total DNA. Immunochemistry-based methods used ELISA tests, immunostaining, and immunohistochemistry. These mainly employed rabbit polyclonal antibodies against whole larvae extract. The lowest LOD reported was 0.5 ng/mL. MS-based detection varied in chromatographic separation methods, and mass spectrometers, making direct comparison on performance challenging. The lowest reported LOD for these methods was 2.5 ng of Ani s 4 protein per gram of fish, making it the most sensitive method. A key observation in this systematic review is the unharmonized manner of reporting performance parameters such as detection limit, specificity, sensitivity and inter- or intra-assay variation.

TOWARDS SUSTAINABLE WORM CONTROL: FIELD VALIDATION OF A DECISION SUPPORT TOOL FOR TARGETED TREATMENT OF GASTROINTESTINAL NEMATODES IN FIRST-SEASON GRAZING DAIRY CATTLE

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Introduction: Gastrointestinal nematode infections in cattle are often controlled by routine anthelmintic treatments without diagnostic support, contributing to anthelmintic resistance and threatening the long-term efficacy of available drugs. Targeted treatment (TT), where only at-risk herds receive treatment, offers a sustainable alternative.

Objectives: This study aims to validate a non-invasive decision support tool to implement TT in first-season grazing dairy heifers, in order to reduce anthelmintic use while maintaining growth and controlling infection levels.

Methods: A field study was conducted on 78 dairy farms in Belgium, monitoring 1233 heifers in 153 groups. Risk scores for gastrointestinal nematode exposure were calculated per group at the beginning of the pasture season, based on pasture management and previous anthelmintic use. Treatment advice for TT groups (N = 77) was tailored to these risk scores, whereas no treatment advice was provided to the control groups (N = 76). Outcomes (anthelmintic use, serum pepsinogen levels, average daily weight gain) were compared between TT and conventionally treated heifers.

Results: TT reduced anthelmintic use by 21%, with no significant difference in weight gain or serum pepsinogen between groups. 'Cumulative egg-free days' was used as a quantitative measure of anthelmintic use, i.e. the total number of days animals remained free of egg-shedding following treatment. Despite the shorter grazing period in the control group, cumulative egg-free days were significantly lower in the TT group. Moreover, TT heifers experienced 16.7% more grazing days without anthelmintic pressure, supporting improved sustainability. Both groups' mean serum pepsinogen values (1.4 U Tyr) fell within the desired reference range (1.2 - 3.5 U Tyr).

Conclusion: This study demonstrates the potential of a non-invasive decision support tool to reduce anthelmintic use without compromising performance.

A PLASMODIUM KNOWLESI A1-H.1 TRANSCRIPTOME TIME COURSE FOCUSING ON THE LATE ASEXUAL BLOOD STAGES

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The zoonotic parasite *Plasmodium knowlesi* is closely related to *Plasmodium vivax*, the second leading cause of human malaria. *P. knowlesi* line A1-H.1 can be maintained in human erythrocytes and is an experimental model for *P. vivax* biology. We present the first transcriptome time course of the intraerythrocytic developmental cycle (IDC) of *P. knowlesi* A1-H.1 grown in human normocytes.

Bulk RNA-sequencing was performed at five highly synchronous IDC stages (5 hpi rings, 14 hpi mid-trophozoites, 20 hpi late trophozoites, 24 hpi mid-schizonts, and 27 hpi late schizonts), enabling identification of housekeeping genes and stage-specific biomarkers, and investigation of temporal expression patterns of multigene families. Comparative analysis with *P. vivax* orthologues revealed strong genome-wide transcriptional conservation, and orthology of invasion-associated genes and ApiAP2 transcription factors was analysed in depth. To support comparative analyses, we developed an interactive web tool to explore *P. knowlesi – P. vivax* orthologue expression across the IDC. This platform facilitates rapid identification of candidate genes with conserved transcription, streamlining functional studies in which *P. knowlesi* A1-H.1 serves as a model for *P. vivax*.

This time course dataset and visualisation tool provide a reference transcriptome framework for *P. knowlesi* A1-H.1 and a resource for comparative *Plasmodium* biology.

PROTEOMIC ANALYSIS AND IDENTIFICATION OF IMMUNOGENIC PROTEINS IN EXCRETORY/SECRETORY MATERIAL OF ASCARIDIA GALLI WORMS: TOWARDS RECOMBINANT-BASED SERODIAGNOSTIC ASSAYS FOR LAYING HENS

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Due to the ban imposed by the EU on conventional battery cage systems, there is now an increased trend towards non-caged housing systems for laying hens. However, due to this ban, infections with worms such as *Ascaridia galli*, *Heterakis gallinarum* and *Capillaria* spp. have reappeared in the poultry industry at a massive rate. These worm infections are mostly diagnosed by the worm counts (WC) after necropsy and the fecal egg counts (FEC), both of which have their own shortcomings. The best possible alternative diagnostic method is serology. A recently developed new ELISA based on excretory/secretory (ES) material of *A. galli* showed the capability to detect both *A. galli* and *H. gallinarum* antibodies in serum following infection. Little information about the proteins present in the ES material of *A. galli* and their functionality is present to date. Hence, the current study aimed to look into the protein composition of ES material of *A. galli* for its characterization, and the identification of immunogenic proteins. Eighty unique individual proteins from the whole ES material have been identified. The proteins have functions related to lipid transport, carbohydrate and amino acid metabolism, nematode larval development, protein translation, electron transport chain, evasion of host immune responses, nervous system development of worms, signal transduction, regulation of gene expression, cytoskeleton organization, DNA damage response and RNA splicing, as found through Gene Ontology database. Protein bands that showed strong immunoreactivity on Western blot were excised from corresponding Coomassie-stained gel and subjected to liquid chromatography–tandem mass spectrometry (LC–MS/MS) for protein identification. Ongoing work focuses on the recombinant expression of proteins identified as immunogenic, including vitellogenin-6, Ancylostoma-secreted protein, a C-type lectin protein, polyprotein ABA-1, an SXP/RAL-2 family member, and a serpin.

IN VIVO SELECTION OF A LEISHMANIA COSMID LIBRARY REVEALS CANDIDATE GENES INVOLVED IN SAND FLY TRANSMISSION

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INTRODUCTION: Due to the absence of a protective human vaccine and the various challenges related to vector control, the management of leishmaniasis currently relies heavily on early and effective diagnosis and treatment. Despite compelling evidence of the importance of the vector, research has predominantly focused on parasite interactions with the vertebrate host. Studies in the invertebrate host have been limited and mostly based on loss-of-function approaches. The development of *Leishmania* promastigotes inside the sand fly vector is a complex, yet critical step in the parasite life cycle, shaped by intricate parasite-vector interactions that may offer interesting novel targets for disease management strategies.

METHODOLOGY: A genome-wide cosmid library of *Leishmania donovani* was subjected to *in vivo* selection through *Lutzomyia longipalpis* sand flies and subsequent mouse macrophage infection. This approach was used to identify candidate genes that provide a gain-of-function and enhance the parasite transmission potential.

RESULTS: Preliminary screening identified several genomic regions potentially conferring an advantage to parasite survival, metacyclogenesis and transmission.

CONCLUSIONS: This unique approach provides a genome-wide strategy for identifying parasite genes essential for vector transmission, deepening our understanding of parasite-vector interactions and offering potential targets for transmission-blocking interventions.

Current efforts focus on narrowing down candidate genes and validating their contribution to sand fly infection establishment, differentiation and macrophage infection through targeted mutagenesis.

THE BIOINDICATION POTENTIAL OF PARASITIC FAUNA INFECTING MACROINVERTEBRATES IN AFRICAN WETLANDS

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The growing communities of Sub-Saharan Africa draw vital support from the region's lakes and rivers. These precious freshwater areas, including wetlands, are threatened by climate change and the increase of anthropogenic exploitation. To ensure sustainable management and protection of these vital water resources, continuous water quality monitoring protocols must be established. As part of water quality monitoring practices worldwide, the community composition of freshwater macroinvertebrates is used to inform on the biological health of wetland ecosystems. Different groups of macroinvertebrates are more or less sensitive to changes in the physicochemical properties, making them suitable for use in bioindication. Parasites infecting the macroinvertebrate hosts can act as sensitive bioindicators, serving as a magnifying lens for ecosystem change. Parasites can also provide valuable insights into the overall state of the ecosystem, as their reliance on specific transmission pathways offers information about their host organisms and the impacts of anthropogenic stressors. Due to the often overdispersed nature of their populations, parasites appear to be more sensitive to subtle environmental changes than their hosts. As such, they can integrate the effects of stressors (e.g. through heavy metal accumulation in soft tissues) throughout time. Within the AfroWetMaP programme, this PhD project aims to add parasitological perspectives into water quality monitoring. We promote an ecologically holistic and One Health–aligned approach to conservation by considering the role parasites and their macroinvertebrate hosts play in ecosystem function. We aim to develop and refine ecosystem health monitoring techniques such as barcoding protocols that will make eDNA-based monitoring possible. As a starting point, we are identifying and barcoding macroinvertebrates and their associated parasite fauna collected from diverse wetland localities in Burundi including Rusizi National Park.

IDENTIFICATION OF POTENT SINGLE-DOMAIN ANTIBODIES AGAINST THE MALARIA SPOROZOITE THROUGH SYNTHETIC SINGLE-DOMAIN ANTIBODY LIBRARIES CONTAINING UNCONVENTIONAL DIVERSIFICATION STRATEGIES

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Despite several control and elimination strategies, malaria is still a global health problem. Today's global community is in desperate need of novel tools to tackle the malaria parasite. Antibodies (Abs) are potent tools for parasite neutralisation. Besides conventional Abs, the natural immune repertoire of mammals contains so-called unconventional diversification strategies. Interestingly, unconventional Abs appear to excel in neutralising highly sophisticated pathogens. Camelid single-domain Abs (sdAbs) are prime examples of such unconventional Ab fragments. Extensive knowledge on the camelid sdAb structure-function relationship enables the construction of highly diverse synthetic libraries that offer several advantages over immune libraries. This project aims to harness the potential of synthetic sdAb libraries with unconventional diversification strategies to tackle the malaria sporozoite. The synthetic sdAb libraries will be constructed by grafting bovine VH CDR3 "stalk-and-knob" and human LAIR1 domains onto the CDR3 of a camelid sdAb scaffold. Key positions in these grafted domains will be randomised to generate two synthetic sdAb libraries ("SyCAbs-KNOB" and "SyCAbs-LAIR1"). The design of SyCAbs-LAIR1 has been completed, resulting in a theoretical library diversity of $1.1x10^{13}$. Non-randomised SyCAbs-LAIR1 linker variants were designed by grafting a LAIR1 variant binding to RIFIN#6 onto the sdAb scaffold. These variants were tested for their recombinant production in and subsequent purification from *E. coli,* thermostability by thermal shift assay (TSA), flexibility and structural integrity by small-angle X-ray scattering (SAXS), and antigen interaction by ELISA. Similar experiments will be performed for non-randomised SyCAbs-KNOB variants containing 2-4-6-8 Cys in their knob once the design of SyCAbs-KNOB has been completed.

HIGH PREVALENCE OF *CHORIOPTES BOVIS*: AN IMPORTANT FACTOR IN CHRONIC PROGRESSIVE LYMPHEDEMA IN BELGIAN DRAFT HORSES

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A cross-sectional study was conducted to estimate the prevalence of chorioptic mange affecting the distal legs of horses in Belgium, focusing on the association between mange and chronic progressive lymphedema (CPL) in Belgian draft horses. Clinical examinations and skin scrapings were performed on the distal legs of 156 Belgian draft horses and 142 Belgian warmblood horses. In the Belgian draft horse breed, 144 (92.31%) horses were infested with *Chorioptes bovis* mites, and 126 (80.77%) displayed clinical signs of CPL. CPL prevalence in draft horses aged <1 year was 17.86%, while mites were detected in 85.71% of this age group, with infestations observed as early as 6 days of age. In a subset of horses aged ≥1 year, CPL and mange prevalence amounted to 94.53% and 93.75%, respectively. In contrast, no mites or CPL were detected in the Belgian warmblood horses examined.

Statistical analysis revealed a strong association between *C. bovis* and CPL (prevalence odds ratio: 7.37; p = 0.002). The prevalence of CPL was approximately twice as high in horses with mites compared to non-infested horses (prevalence ratio: 2.02). Furthermore, the prevalence risk difference of 42.36%, indicates a substantial absolute increase in CPL prevalence among infested horses.

This study demonstrates the high prevalence of *C. bovis* and its breed-specific predilection in Belgian draft horses. The strong association between mange and CPL highlights the potential role of *C. bovis* as a contributing factor in CPL pathogenesis.

DIVERSITY OF PARASITIC COPEPODS FROM ESTUARINE FISHES IN A SOUTH CAROLINA ESTUARY

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Copepods are one of the most diverse groups of aquatic organisms, with more than 15 000 accepted species. They can be found in a number of different freshwater and marine habitats. Parasitism has evolved independently in 14 copepod clades; currently, more than 6 000 species of parasitic copepods are known. These crustaceans can be found parasitizing on a range of fish taxa, and in a range of aquatic habitats, including transitional habitats like estuaries. In 2023, a parasite 'BioBlitz' project was conducted in the Stono Preserve, part of the Stono River estuary in South Carolina, USA. The aim of the project was to assess the diversity of different parasitic taxa in that area, and part of the project included sampling of fishes, in order to look into the diversity of fish parasites. For this project, 125 fish specimens of 12 genera (17 species) were caught and inspected for parasites; when it comes to parasitic copepods, members of only 3 genera (four species) were infected: *Fundulus* (*F. heteroclitus*, *F. majalis*), *Menidia* (*Menidia* sp.), and *Mugil* (*M. cephalus*). From these hosts, 18 parasitic copepods were collected and identified based on their morphology and genetic data (sequences of 28S and 18S rDNA, and mitochondrial COI marker). In our samples, we found four species of *Ergasilus* (Ergasilidae), and one per genus for *Bomolochus* (Bomolochidae), *Caligus* (Caligidae), and *Naobranchia* (Lernaeopodidae). From all screened hosts, *M. cephalus* hosted the highest number of species, with one species of *Bomolochus*, two *Ergasilus* species and one species of *Naobranchia* infecting this fish. The diversity of copepod parasites in the Stono Preserve corresponds to previously reported trends in estuarine habitats, with ergasilids as the most species-rich taxon. Furthermore, *Bomolochus* and *Ergasilus* species are new to science, but assessing the relationship of these species to other members of their respective families through molecular methods proves challenging, due to a pronounced gap in available molecular data.

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EXPLORING THE CONTEXT AND OPPORTUNITIES FOR OPTIMIZING WATER, SANITATION AND HYGIENE FOR THE PREVENTION OF SOIL TRANSMITTED HELMINTHIASIS IN URBAN PRIMARY SCHOOLS: A PHOTOVOICE STUDY, ETHIOPIA

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Background: Poor water, sanitation and hygiene (WASH) contributes to the transmission of various infectious diseases such as soil-transmitted helminthiasis. Today, ensuring WASH services remains an important challenge in Ethiopian schools. Addressing these challenges requires a comprehensive understanding of WASH issues from the perspectives of all stakeholders. In this study used photovoice, a participatory photography-based method, to explore both WASH challenges and opportunities, and to engage stakeholders in collective reflections to identify ways to optimize WASH in urban primary schools of Ethiopia.

Methods: First, we conducted a photovoice in two purposively selected public primary schools in Jimma town. Across these two schools, 10 teachers and 10 students were invited to participate in the study. Photos and narratives emerging from the data were analysed using a thematic analysis. Second, we organized an advocacy event, where the findings were presented and discussed with 75 stakeholders, including representatives of local authorities (e.g., education office, health office and water and energy office).

Results: Despite the presence of WASH facilities, improper use of the toilets, open defecation, and a weak ownership persisted. Additionally, participants linked poor WASH condition to broader system factors such as infrequent monitoring of school WASH facilities, inadequate maintenance , and unclear institutional roles regarding school WASH management responsibilities. Importantly, the photovoice method acted as an advocacy tool beyond data collection by providing a platform for direct discussion between the school community and local authorities. The study process thereby inspired the community towards immediate behavioral changes, such as cleaning the school environment.

Conclusions: This study revealed behavioral, infrastructural, and systemic factors contribute to poor WASH conditions in schools. The advocacy event bridged power hierarchies by enabling direct interaction between school communities and local officials. Photovoice can be used as a useful research tool and an effective intervention method for optimizing WASH practices and disease prevention in urban primary schools of Ethiopia.

Keywords: Photovoice, WASH, behaviour change, advocacy, infection prevention, Ethiopia

DERMOTROPIC LEISHMANIA DONOVANI IN NEPAL: (RE-) EMERGENCE OF 'PRUDENT' PARASITES AND THREAT FOR ELIMINATION?

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Visceral leishmaniasis (VL) in the Indian sub-continent is due to *L. donovani* and is lethal in the absence of treatment. During the last epidemic of VL in Nepal, prior to the commencement of the current elimination initiative in 2005, the disease was confined mostly to the Terai region in the southeast of the country. Although reported VL case numbers are now much lower than before, a shift in foci to hilly regions in western Nepal has been observed. There has been a concurrent emergence of *L. donovani*-associated cutaneous leishmaniasis (CL), a form of the disease that was not reported in the past, mostly also distributed across western Nepal.

We aimed to (i) track the evolutionary origin of these dermotropic *L. donovani* and (ii) understand to what extent parasites causing CL may be contributing to persistence of VL in the Indian subcontinent. To do this, we sequenced the genome of *L. donovani* parasites directly from DNA extracted from clinical samples from VL and CL patients in western Nepal and assessed relatedness with published 'pre-elimination' genomes from the region. Our main findings were: (1) In Nepal, elimination seems to have been associated with replacement of the historically predominant viscerotropic parasite populations by rare variants that expanded in the last decade; (2) Their progeny constituted at least two new genetic groups, (pseudo-)clonally propagating, but hybrids were observed; and (3) both epidemic 'clones' and hybrids were associated with CL and VL. Genomes of contemporary and closely related CL and VL variants are being scrutinized to find potential functional differences between the two forms and results will be presented at the conference.

We explain those results by two alternative hypotheses: (i) low virulent (CL) parasites would have emerged from a main pre-elimination population of aggressive *L. donovani* (causing lethal VL); this kind of 'prudent' parasites would spare their host and contribute to transmission to other hosts, or (ii) CL parasites would have been present in Nepal for decades -but not detected- and recently, they would have re-emerged, like in Sri-Lanka. Whatever the correct evolutionary scenario, there seems to be transmission chains involving both CL and VL parasites in Nepal. Therefore, we strongly recommend implementing genomic surveillance in the region and including CL management in the elimination program.

TRYPTACKLE – TACKLING THE LIVESTOCK PARASITE *TRYPANOSOMA CONGOLENSE* BY TARGETING INVARIANT SURFACE GLYCOPROTEINS

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African trypanosomes are extracellular protozoans transmitted by tsetse flies (*Glossina* spp.), causing deadly diseases in sub-Saharan Africa: sleeping sickness in humans (human African trypanosomiasis, HAT) and nagana in livestock (animal African trypanosomiasis, AAT). AAT, primarily caused by *Trypanosoma congolense* and *T. vivax,* poses a major barrier to socio-economic development in the region, with annual losses estimated at ~$5 billion. This makes it an important cause of food insecurity and poverty. Besides controlling the insect vector, farmers heavily rely on trypanocides to fight AAT. However, recent reports highlight increasing animal treatment failures due to ineffective drug use and the rise and spread of drug-resistant parasite strains. Until recently, the development of a vaccine against African trypanosomes was considered to be unreachable due to the parasite's sophisticated immune evasion strategies, including antigenic variation. However, the recent discovery of an invariant surface antigen that induces significant immune protection to experimental *T. vivax* infection challenged this paradigm and revived the hope for an effective vaccine against AAT. Therefore, our research consortium has focused on *T. congolense*, the predominant tsetse-transmitted parasite affecting livestock. We sequenced the transcriptomes of metacyclic and early bloodstream forms, the parasite stages present at the onset of infection. Using genomic mining and structural bioinformatics, we identified the gene repertoire encoding secreted or surface-attached antigens expressed during these early parasite forms. Potential vaccine candidates were recombinantly produced and purified for immunisation trials in mice. We are currently assessing the impact of vaccination on infection dynamics by challenging the immunised mice via the bite of infected tsetse flies.

INTEGRATED ANTHELMINTIC-BASED CONTROL OF TAENIA SOLIUM CYSTICERCOSIS/TAENIASIS, SOIL-TRANSMITTED HELMINTHIASIS AND SCHISTOSOMIASIS: SAFETY, EFFECTIVENESS, AND IMPLEMENTATION STRATEGIES: 3SI-CONTROL

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Taenia solium cysticercosis/taeniasis, soil-transmitted helminthiasis (STH) and schistosomiasis (SCH) are neglected tropical diseases (NTDs). The efficacy and effectiveness of STH and SCH control programmes is troubled challenged by low and failing drug efficacy and growing concerns of anthelmintic resistance. The novel fixed-dose co-formulation (FDC) including albendazole and ivermectin has proven to be safe and to tackle most of the challenges in drug efficacy for STH. For *T. solium* currently there are no countries routinely implementing control.

The main objective of the EDCTP3 JU (EU) funded 3SI-CONTROL project is to evaluate the safety and effectiveness of mass drug co-administration (MDA) of FDC and praziquantel (PZQ) in sub-Saharan African co-endemic areas, and to develop implementation strategies to ensure uptake.

A multicentric cluster randomised controlled trial will be conducted to evaluate the safety at the community level in Tanzania and Zambia. Communities in the study areas will be randomly assigned to one of the two treatment arms (5,500 participants per arm): arm (i) simultaneous administration of FDC and PZQ, or arm (ii) sequential administration of FDC followed by PZQ after a 14-day interval. To monitor the effectiveness, a second MDA will be conducted after 12 months, with impact evaluated in people, pigs and the environment, applying a One Health approach, joined by a model-based approach comprehensively assessing the potential (long term) epidemiological and economic impact. Additionally, the project will develop country specific tailored implementation strategies in Mozambique, Tanzania and Zambia.

Results from 3SI-CONTROL will provide a scientific evidence base on the safety, effectiveness and implementation of integration of *T. solium* control with STH and SCH. Tackling these three top ranking NTDs will reduce the individual, social and economic burdens of resource poor rural populations.

IMMUNOLOGICAL MECHANISMS OF RESPIRATORY CO-INFECTIONS: THE IMPACT OF TRYPANOSOMES ON SECONDARY INFECTIONS CAUSED BY MAJOR LOWER RESPIRATORY TRACT PATHOGENS

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Cumulative evidence highlights the complex effects of insect-borne protozoan parasites on respiratory immunity, yet their role in co-infection dynamics remains poorly understood. *Trypanosoma brucei*, the causative agent of African trypanosomiasis, profoundly alters systemic immune responses, but its impact on pulmonary defences has not been fully explored. We hypothesise that trypanosome exposure modulates epithelial immune signalling, potentially increasing susceptibility to secondary infections by major lower respiratory tract pathogens, such as respiratory syncytial virus (RSV), *Streptococcus pneumoniae*, and *Mycobacterium abscessus*. To address this, we established an *in vitro* co-infection model using A549 alveolar epithelial cells and compared single infections to co-infections with *T. brucei* at physiologically relevant multiplicities of infection. Transcriptional profiling was performed by RT-qPCR under MIQE 2.0 standards, including efficiency-corrected quantification with prediction intervals, validated normalisation using multiple reference transcripts, and full QC reporting. Our analyses aim to characterise the immunopathology of these polymicrobial interactions, focusing on pathways involved in antiviral defence, inflammation, and metabolic regulation. This multidisciplinary approach bridges parasitology, virology, and bacteriology, providing insights into virulence, host responses, and potential biomarkers. By integrating hypothesis-driven experimentation with rigorous molecular analysis, this study seeks to advance understanding of protozoan-bacterial-viral interplay and inform strategies to mitigate the impact of co-infections in endemic regions.

Acknowledgements

This work was supported by Horizon Europe funding [HORIZON-MSCA-2023-PF-01 Project 101152054 -RespiriCO, S.A.], the Fonds Wetenschappelijk Onderzoek [www.fwo.be; grant numbers G033618N and G013118N (G.C.)], and the University of Antwerp [www.uantwerpen.be; Bijzonder Onderzoeksfonds (BOF) support of S.A., P.C., P.D., and G.C.]. LMPH is a partner of the Excellence Centre 'Infla-Med' (www.uantwerpen.be/infla-med) and participates in COST (European Cooperation in Science and Technology) Action CA21111.

UNVEILING GLIAL CELLS IN THE NASAL MUCOSA AS HOST CELLS FOR LEISHMANIA WITH POTENTIAL IMPLICATIONS IN DISEASE OUTCOMES

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Neurological involvement in leishmaniasis remains underexplored, despite reports of meningoencephalitis, peripheral neuropathy, and neuroinflammation in visceral and mucocutaneous forms. While macrophages are recognised as the primary host cells, emerging evidence suggests that *Leishmania* can exploit immune-privileged niches beyond classical phagocytes. Here, we investigate whether olfactory ensheathing cells (OECs), specialised glia in the nasal mucosa, constitute a previously unrecognised host cell population. These cells form a neuroimmune interface that bridges the peripheral and central nervous systems, potentially offering a sanctuary for parasite persistence. Using an *in vitro* infection model, we compared OECs to PMA-differentiated U937 macrophage-like cells upon exposure to *L. braziliensis*, *L. major*, and *L. infantum*. Transcriptional profiling was performed by RT-qPCR following MIQE 2.0 guidelines, including efficiency-corrected quantification with prediction intervals, validated normalisation using multiple reference transcripts, and full QC reporting. Parasite viability was confirmed by SL-RNA quantification, supporting the hypothesis that OECs can harbour *Leishmania* under conditions mimicking mucosal infection. Distinct immune signatures were observed in OECs, with modulation of NFκB-driven inflammatory pathways, interferon axis transcripts, and regulatory/metabolic mediators, suggesting a unique immunological phenotype compared to macrophages. These findings introduce OECs as a candidate reservoir that may contribute to subclinical persistence, relapse, and treatment failure, particularly in mucocutaneous disease. Beyond expanding the cellular tropism of *Leishmania*, this work underscores the need to consider neuroimmune interactions in disease pathogenesis and therapeutic strategies. Recognising non-traditional host cells could reshape our understanding of chronic parasitic infections and open new avenues for targeted interventions.

Acknowledgements

This work was supported by Horizon Europe funding [HORIZON-MSCA-2023-PF-01 Project 101152054 -RespiriCO, S.A.], the Fonds Wetenschappelijk Onderzoek [www.fwo.be; grant numbers G033618N and G013118N (G.C.)], and the University of Antwerp [www.uantwerpen.be; Bijzonder Onderzoeksfonds (BOF) support of S.A. and G.C.]. LMPH is a partner of the Excellence Centre 'Infla-Med' (www.uantwerpen.be/infla-med) and participates in COST (European Cooperation in Science and Technology) Action CA21111.

ADAPTIVE CO-EVOLUTION IN VISCERAL LEISHMANIASIS: THE ROLE OF HOST MIF CYTOKINE AND PARASITE MIMICRY

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INTRODUCTION: Visceral leishmaniasis (VL) is transmitted by sand flies and varies from an asymptomatic to a life-threatening condition with limited treatment options. The *Leishmania*-host interaction is shaped by an evolutionary arms race involving inflammatory host responses and parasite immunomodulatory molecules. The macrophage migration inhibitory factor (MIF) is a key immune regulator produced by both the mammalian host (hMIF) and the parasite (pMIF). Upon infection, hMIF induces an anti-parasitic, proinflammatory state, whereas in cutaneous leishmaniasis, *L. major mif* (*Lm*MIF) supports parasite survival and persistence in macrophages. Although *Lm*MIF has been extensively studied, research regarding the role of hMIF and its parasite orthologs in VL is lacking. To address this knowledge gap, a search for orthologs in *L. infantum* identified two genes, *Linf*259 (LINF_330025900) and *Linf*260 (LINF_330026000). Overexpression and deletion of these genes were used to elucidate their individual roles.

METHODOLOGY: Single overexpressors (OE) were generated through gene cloning and introduction into the chromosomal *ssu* locus, whereas single and double knockout (KO) lines were obtained through CRISPR-Cas9 gene editing. All transgenic lines were characterized *in vitro* by assessing (i) their growth and metacyclogenesis rates, and (ii) their ability to infect, differentiate into amastigotes and multiply in primary peritoneal macrophages.

RESULTS: No distinct differences could be observed in promastigote proliferation, metacyclogenesis, and parasite survival in culture. Deletion of *Linf*MIF caused profound suppression of macrophage infection and *in situ* proliferation.

FROM SLIDES TO BYTES: DIGITAL PARASITES INNOVATE EDUCATION AND PROMOTE AWARENESS

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Conservation of wildlife parasites is important and urgent –about 30% of parasite species are estimated to become extinct by the end of the century– but it still receives little recognition and support. Digital technologies offer a powerful, yet underutilized, opportunity for parasitology education and public engagement which can support parasite conservation.

Interactive web-based resources provide several benefits. They are accessible to a wide audience independently of their geographical location and sociocultural context, contributing to capacity building in the Global South. Digital tools can also help in attracting aspiring researchers into parasitology, which is an underappreciated career path, addressing the generational gap of parasitologists. Moreover, digital tools can engage the general public, helping to shift their often narrow conception of parasites from solely disease-inducing to organisms that are known to provide a broad variety of ecosystem services. Furthermore, digital resources are adaptable as open-source content can be translated, localised and tailored to specific needs. They provide an ethical and sustainable alternative or addition to traditional dissections, complying with the 3-Rs: Replacement, Reduction and Refinement.

Hasselt University, in collaboration with the Royal Belgian Institute of Natural Sciences, introduces a set of openly available, interactive, web-based resources designed to unlock parasitology for students and non-parasitologists. These include interactive e-learning modules illustrating parasite life cycles and anatomy, and annotated 3D μCT scans providing immersive insights into parasite morphology. We present future plans for the bridging of parasitological data and complementary modules on host histology and anatomy. Besides these elements, which are primarily aimed at students, we also plan to build educational game and story-telling modules for children and young adults. These can be shown during outreach events to engage and inform the public.

By moving from slides to bytes, parasitology can become more engaging and inclusive, bridging education, research, and conservation through digital innovation.

DEVELOPMENT OF A COST-UTILITY MODEL FOR EPILEPSY MANAGEMENT: INTEGRATING IMMUNODIAGNOSIS FOR NEUROCYSTICERCOSIS DETECTION

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INTRODUCTION: Neurocysticercosis, caused by the metacestode stage of the pork tapeworm (*Taenia solium*), is a major cause of acquired epilepsy in endemic areas. Currently, specific management protocols for epileptic patients are lacking in resource-limited healthcare systems. Patients diagnosed with epilepsy typically receive only symptomatic treatment -if any at all- without further diagnostic work-up, as awareness of potential etiologies is limited and diagnostic tools often unavailable. Introducing immunodiagnosis for detection of neurocysticercosis into the diagnostic pathway may offer an effective strategy to identify a fraction of people with epilepsy who could benefit from targeted neurocysticercosis management. Costs associated with immunodiagnosis, confirmatory neuroimaging, and anthelmintic treatment of neurocysticercosis patients, are expected to be outweighed by potential health gains and improvements in quality of life.

METHODOLOGY: A combined decision tree and Markov model was constructed to assess cost-utility of a strategy incorporating immunodiagnosis and consecutive treatment of neurocysticercosis patients, versus current practice without immunodiagnosis. The modelled population is representative of patients aged 18 and above, with epilepsy and attending a healthcare facility in sub-Saharan Africa. The decision tree represents the 2-month period following the first-time epilepsy-related healthcare visit, whereas the Markov model simulates lifetime effects of living with epilepsy, reflecting disease management, seizure control, and mortality. Costing data to be input includes diagnostic procedures, treatment, and direct and indirect patient expenses related to epilepsy. Health outcomes can be expressed in quality-adjusted life years, derived from EQ-5D-5L data. The model was constructed in Excel and R, allowing for visual and analytical exploration of model assumptions and outcomes.

CONCLUSION: This modelling framework provides a transparent and adaptable platform for evaluating the potential economic and health benefits of integrating immunodiagnosis into epilepsy management in endemic settings. Once populated with empirical data, it will enable estimation of incremental cost-effectiveness ratios and identification of key drivers influencing cost-utility.

NGAL DIPSTICK AS A PROGNOSTIC MARKER FOR PEDIATRIC MALARIA-ASSOCIATED ACUTE KIDNEY INJURY IN A RESOURCE-LIMITED SETTING

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Introduction: Malaria-associated acute kidney injury (MAKI) is one of the leading causes of mortality in severe malaria. Early identification of children at risk of deterioration is essential in low-resource settings. The Urinary Neutrophil Gelatinase-Associated Lipocalin (uNGAL) is an early biomarker of tubular injury and may help identify children at the highest risk. We assessed whether uNGAL dipstick testing predicts disease severity and outcomes in pediatric MAKI.

Methods: We conducted a prospective study from May 2024 to July 2025 among children aged 6 months to 16 years admitted with MAKI at the University Hospital of Kinshasa in the Democratic Republic of Congo. Malaria was defined according to WHO guidelines for malaria and AKI per KDIGO 2012. uNGAL was measured at admission using the BioPorto dipstick (Denmark); the NGAL dipstick provides three semi-quantitative thresholds: 50, 150, and 300 ng/ml reflecting increasing levels of tubular injury severity, and elevated NGAL was defined as ≥150 ng/mL. Outcomes included KDIGO stage, renal replacement therapy (RRT) requirement, and survival.

Results: Among 119 enrolled children, the median age was 3 years (IQR 2–6) and 56.5% were male. NGAL results were available for 110/119 patients; 85.5% (94/110) had elevated NGAL. Elevated NGAL was strongly associated with KDIGO stage 3 ($p < 0.001$) and RRT requirement ($p < 0.001$). Overall mortality was 33.6% (37/110) and a non-significant trend toward higher mortality was observed ($p = 0.078$) among children with elevated uNGAL.

Conclusion: A simple bedside uNGAL dipstick test reliably identifies children at high risk of severe AKI and poor outcomes. It could serve as an affordable triage and prognostic tool to guide clinical decision making and resource allocation in low resource settings.

Note: BioPorto Diagnostics A/S (Denmark) supported this research by providing the NGALds materials under a research agreement.

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ROAD TOWARDS SUSTAINABLE ANTHELMINTIC USE IN GRAZING RUMINANTS: INSIGHTS FROM STAKEHOLDER ENGAGEMENT

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Sustainable worm control (SWC) strategies advocate for evidence-based treatment of livestock, guided by diagnostics and epidemiological indicators, to mitigate anthelmintic resistance and optimize herd health. While parasitological outcomes of SWC are well-documented, its economic implications remain insufficiently explored. This study investigated stakeholder perceptions on the economic feasibility and value of SWC interventions across diverse livestock systems.

An online survey, translated into 11 languages and disseminated across 13 countries (Feb–Mar 2025), yielded 932 responses from 12 countries. Findings indicate that over 25% of farmers' animal health budgets are allocated to worm control, with Southern Europe reporting the highest expenditure (52%). Although 32% of respondents perceived diagnostic testing as too costly, 93.8% agreed that SWC practices can enhance farm profitability.

The most economically endorsed SWC strategies were: (i) targeted anthelmintic use (87% vets, 83% farmers), (ii) grazing management (84% vets, 80% farmers), and (iii) quarantine and strategic screening (80% vets, 75% farmers). Support for alternative approaches—such as bioactive feed additives, multispecies pastures, and vaccination—was more variable. Notably, vaccination was considered economically viable by only 64% of veterinarians and 58% of farmers, reflecting limited availability and awareness.

A substantial proportion of "I don't know" responses, particularly from farmers in Southern and Western Europe. This uncertainty suggests that implementation barriers stem more from unfamiliarity than resistance. These findings underscore the need for targeted extension services, practical training, and clearer communication of cost-benefit evidence. Targeting younger and more engaged farmers, who formed a substantial share of respondents, may accelerate adoption.

In conclusion, while the biological benefits of SWC is well-established, its perceived economic value is high but unevenly understood. Bridging this gap through stakeholder-actionable guidelines and field validation is essential to facilitate broader uptake and sustainable implementation.

We acknowledge our project partners for their support in survey dissemination.

A special thanks to