BSP

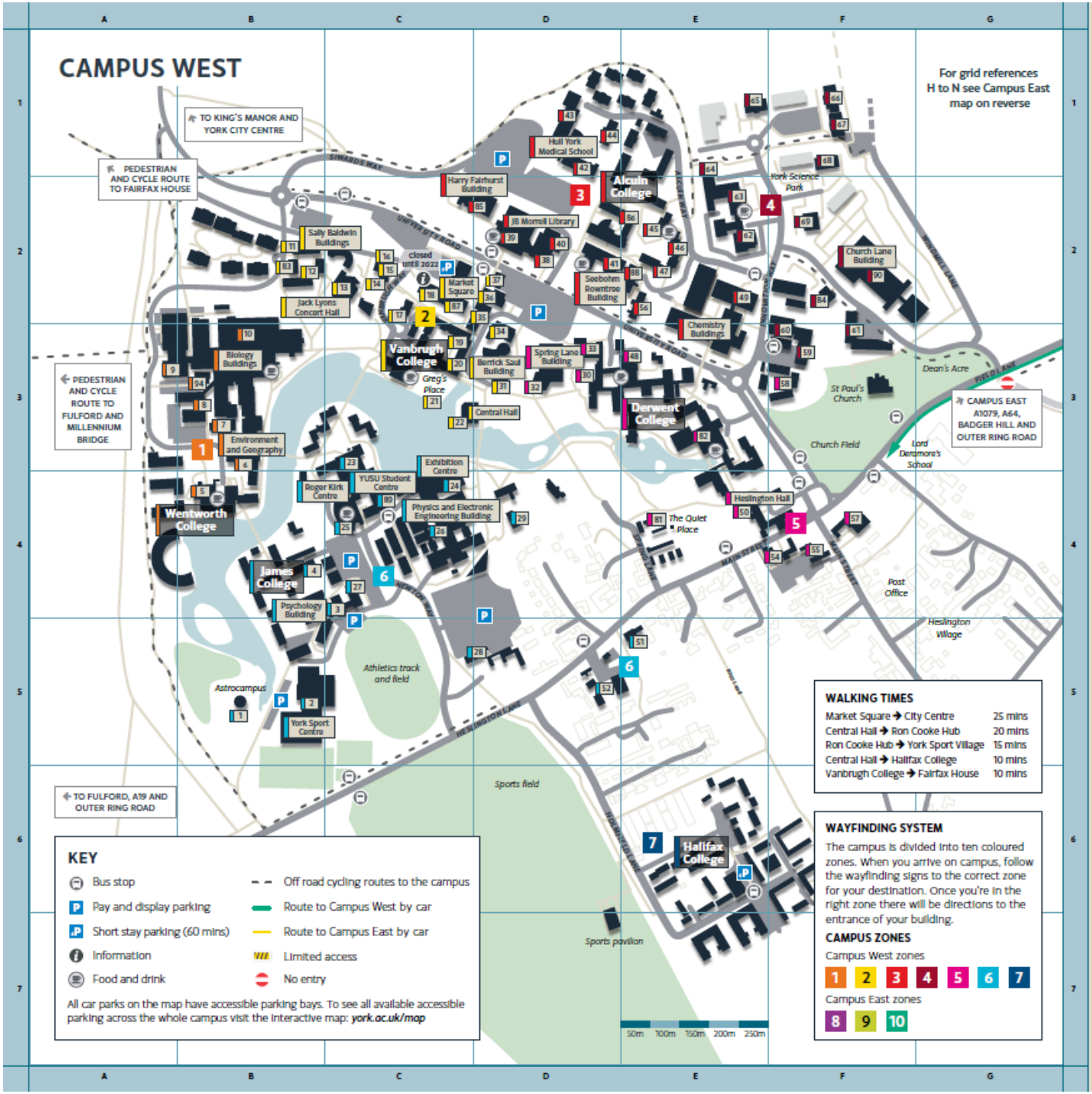
UNIVERSITY of York

"Together Again"
Reuniting our Community

2022

BRITISH SOCIETY FOR PARASITOLOGY

BSP



Key

Dear BSP60York Delegates,

We are delighted to welcome you to the 60th Anniversary Spring Meeting of the British Society for Parasitology, hosted by the University of York.

This year's theme, "*Together Again: Reuniting our Community*" hopes to reunite our international scientists and experts from across the globe, each of us with unique contributions to the field of Parasitology. It will be an excellent opportunity to hear first hand about the latest new discoveries, form new partnerships and collaborations, and share knowledge for one common goal: to better comprehend and combat parasitic diseases.

We are particularly pleased that the experimental hybrid format for this meeting promotes opportunities for those who could not join us in person for part or all of the event. This schedule flexibility has enabled better representation from colleagues who have travel obstacles such as parents and carers, and is in direct response to the findings from our inaugural Diversity sessions in the BSP Online meeting last June. Ours is a beautifully diverse community, and as such, the BSP believes that race, gender, nationality, sexual orientation, age and career status should never hinder professional opportunities. We therefore encourage and welcome your suggestions and participation in the sessions and workshops designed to promote these ethos and brainstorm productive ways to better realise this. We are great scientists and global citizens united by our dedication to combat global infectious diseases.

We hope that, in addition to the excellent scientific presentations and social events, you will enjoy seeing some of the sights in the spectacular city of York. The core of this meeting is held within the Department of Biology at the University of York's scenic campus, which hosts the 1st and 3rd largest man-made lakes in Europe and serves as a bespoke bird sanctuary. Our Young Parasitologists will enjoy a special event organized by and for our ECRs at Las Iguanas within our beautiful York City Centre. York city walls are accessible and enable some beautiful glimpses of our city's best gardens. The city's name herald from its history as the Viking Capital Yorvik, which was formerly the Roman city Eboracum. The first Christian Roman Emperor Constantine was crowned Emperor of the Roman Empire where the York Minster now sits. The York Minster and Railway Museum are both impressive destination tourist sites not to be missed. Our Conference Reception and Dinner Gala will be held after the BSP Award ceremony Thursday at the magnificent Castle Howard, a jewel of Yorkshire with ornate interiors, landscaped gardens and tremendous historical significance for the region.

The organising committee hope you find this an enjoyable, memorable and inclusive meeting. Please feel free to contact us with any questions or concerns.

We look forward to seeing you at this inspiring event celebrating 60 years of our cherished Society!

Pegine Walrad and James LaCourse

On behalf of the BSP60York Organising committee

Day (Date)	Mon (21/3)	Tues (22/3)				Wed (23/3)				Thurs (24/3)				Fri (25/3)			
		K/018	T/005	P/X001	T019	K018	T005	PX001	T019 Workshops	K018	T005	PX001	T019 Workshops	K018	T005	PX001	T019 Workshops
9am	JCPiL	Welcome and History of York Parasitology: (P/X001)				Morning Announcements				Morning Announcements				AGM BSP Council Meeting - All welcome!			
10am		Combative strategies: vaccines	Parasite Molecular Genetics	BES-Sponsored Parasite Ecology: Coinfections		Mathematical Modeling	Tissue Tropism	Vectors and Parasitology	ECR Career Workshop	Combative strategies: Drug Discovery	Diversity in Science II	Parasite Cell Biology II	Controlled Human Infection Models CHIM	Evolutionary Genomics	Parasite Gene Expression	Parasite Trafficking / Signalling	Social Sciences and Parasitology Workshop
11am																	
12pm																	
1pm		Lunch: Virtual Posters T/005				Lunch: Virtual Posters T/005				Lunch				Lunch			
2pm	Registration	Diversity in Science I	Parasite Cell Biology I	BES-Sponsored: Wild Parasitology		Parasite Biochemistry	Parasite Immuno-pathology	Parasite Genetic Architecture	Scientific Publishing	Presidents medal and CA wright medal talks				Prizes/Farewell/Close			
3pm																	
4pm										4-4:30pm Buses to Castle Howard for Reception/Grounds							
5pm	ial/Opening(Atriu	In Person Posters (Atrium)				In Person Posters (Atrium)				Reception at Castle Howard							
6pm	Drinks/Nibbles									30pm Buses back from Castle Howard for non-attendees of dinn							
7pm																	
8pm										Conference Dinner at Castle Howard							
9pm										Ebor Choir entertainment							
10:00 PM																	
11:00 PM										11pm Buses back from Castle Howard							

Contents

The Programme with Abstract Titles by Session

Day 1 – Mon 21st Mar 2022 –

Registration Opens *(Atrium)* – Mon 21st Mar 15:00 - 17.00)

Social/Opening – Drinks and nibbles *(Atrium)* – Mon 21st Mar 17:00 - 19.00)

Day 2 – Tue 22nd Mar 2022 -

Registration *(Atrium)* – Tue 22nd Mar 08:00 - 09.00)

Morning Announcements *(Lecture Theatre P/X001)* – Tue 22nd Mar 09:00 - 09.20)

Welcome and Opening Plenary Session: "Welcome and History of York Parasitology". Prof Paul Kaye *(Lecture Theatre P/X001)* – Tue 22nd Mar 09:20 - 10.00)

Session 1 – Tue 22nd Mar 10:00 - 11.20

Day 2 – Combative Strategies: Vaccines *(Lecture Theatre K/018)*

22-March-2022, at 10:00 to 11:00
Chairs - Prof Gavin Wright & Dr Mohamed Osman
10:00 (20 mins) - A26225 - Towards Broadly-Neutralising Blood-Stage Vaccines against Human Malaria Parasites (Simon Draper)
10:20 (10 mins) - A26080 - Pre-erythrocytic and transmission-blocking multi-stage malaria vaccine indicates synergic sterile protection effect in a murine model with *Plasmodium berghei* transgenic parasite (Tetsushi Mizuno)
10:30 (10 mins) - A26009 - An exploratory study to verify the safe and reproduceable use of aseptic purified cryopreserved *Plasmodium falciparum* sporozoites for the induction of controlled human malaria infection in healthy malaria-naïve adults at hVIVO medical research unit. (Anand Odedra)

11:00 (20 mins) - A26450 - Question Time - Combative Strategies: Vaccines

Day 2 – Parasite Molecular Genetics *(Lecture Theatre T/005)*

22-March-2022, at 10:00 to 11:00
Chairs - Dr Natalia Teles & Dr Pegine Walrad
10:00 (20 mins) - A26224 - RNA communication in helminth-host interactions (Amy Buck)
10:20 (10 mins) - A25940 - Developmental incompetence in selected and naturally occurring trypanosome isolates (Guy Oldrieve)
10:30 (10 mins) - A25960 - Accessing the variability of multicopy genes in complex genomes using unassembled short reads: the case of *Trypanosoma cruzi* multigene families (Joao Cunha)

11:00 (20 mins) - A26451 - Question Time - Parasite Molecular Genetics

Day 2 – Parasite Coinfections *(Lecture Theatre P/X001)*

22-March-2022, at 10:00 to 11:00
Chairs - Prof Colin Sutherland & Mr John Archer
10:00 (20 mins) - A26216 - Multiple-fronts costs of defence: the case of the swarming T-helper cells (Andrea Graham)
10:20 (10 mins) - A25511 - Evolution of leishmaniaviruses with a focus on LRV2 (Vyacheslav Yurchenko)
10:30 (10 mins) - A25968 - Effect of delayed calf-dam separation on Cryptosporidium infection in dairy cattle. (Olivia Ingle)

11:00 (20 mins) - A26453 - Question Time - Parasite Coinfections

Session 2 – Tue 22nd Mar 11:50 - 13.00

Day 2 – Combative Strategies: Vaccines *(Lecture Theatre K/018)*

22-March-2022, at 11:50 to 12:40
Chairs - Prof Gavin Wright & Dr Mohamed Osman
11:50 (10 mins) - A26154 - An invariant *Trypanosoma vivax* vaccine antigen eliciting protective immunity (Delphine Autheman)
12:00 (10 mins) - A26145 - The Leishmania *donovani* ortholog of the GPI-anchor biosynthesis co-factor PBN1 is essential for host infection (Adam Roberts)
12:10 (10 mins) - A25916 - Laboratory evaluation of the miniature direct-on-blood PCR nucleic acid lateral flow immunoassay (mini-dbPCR-NALFIA), a simplified molecular diagnostic test for malaria (Norbert van Dijk)
12:20 (10 mins) - A26152 - Vaccination is probably considered the most efficient tool for preventing current and future threats from parasitic diseases. (John Ellis)

12:30 (10 mins) - **Turbo Talk – 5 min each talk -**
A26144 - Systematic identification of genes encoding cell surface and secreted proteins that are essential for in vitro growth and infection in Leishmania *donovani*. (Adam Roberts) (5 mins) -

12:40 (20 mins) - A26455 - Question Time - Combative Strategies: Vaccines

Day 2 - Parasite Molecular Genetics *(Lecture Theatre T/005)*

22-March-2022, at 11:50 to 12:35
Chairs - Dr Natalia Teles & Dr Pegine Walrad
11:50 (10 mins) - A26008 - The Helminth Antimicrobial Peptidome: a novel opportunity for parasite control? (Allister Irvine)
12:00 (10 mins) - A26017 - No more mutants: lack of canonical resistance mutations in *Ascaris* β-tubulin isotypes. (Ben Jones)
12:10 (10 mins) - A26049 - Structure and selection: Insights into the evolution of host-parasite interactions of Tetraspanin 23 in *Schistosoma* turkestanicum. (Camilla Eldridge)

12:35 (15 mins) - A26454 - Question Time - Parasite Molecular Genetics

12:50 (10 mins) - **Turbo Talks – 5 min each -**
22-March-2022, at 12:50 to 13:00
12:50 (5 mins) - A26001 - Molecular epidemiology and evolution of the Antigen Coding Gene (ACG) TSP-23, from the multi-host parasite *Schistosoma japonicum* (Daniel Parsons)
12:55 (5 mins) - A26005 - The emergence of hybrid Fasciolids in central Vietnam. (Philip Best)

Day 2 – Parasite Coinfections *(Lecture Theatre P/X001)*

22-March-2022, at 11:50 to 12:35
Chairs - Prof Colin Sutherland & Mr John Archer
11:50 (10 mins) - A25512 - *Ovale* malaria - unknown knowns, known unknowns and other mysteries (Colin Sutherland)
12:00 (10 mins) - A25979 - How much does innate immunity impact rodent malaria infection dynamics? A meta-analytic approach (Alejandra Herbert Mainero)
12:10 (10 mins) - A26019 - Integrating ecological perspectives into anthelmintic resistance management (Samuel Brown)
12:20 (10 mins) - A26089 - Co-Infections with Malaria, Urinary Schistomiasis, Typhoid Fever and Hepatitis B Virus Among School Children in Ogbese, Ise-Ekiti, South-Western Nigeria (Charles Ologunde)

12:35 (15 mins) - A26452 - Question Time - Parasite Coinfections

12:50 (10 mins) - **Turbo Talks – 5 min each talk -**
22-March-2022, at 12:50 to 13:00
12:50 (5 mins) - A26165 - First observation of Parasitic viruses in Trichomonas gallinae (Dalal Ardan)
12:55 (5 mins) - A26088 - Malaria Co – Infection with Urinary Schistosomiasis, Typhoid Fever, Hepatitis B Virus, and Human Immunodeficiency (HIV) Virus in three Local government areas of Ekiti-State, South Western Nigeria (Charles Ologunde)

Lunch + Online Posters *(T/005 + Online)* (Tue 22nd Mar 13:00 - 14.00)

Poster themes -

- Combative Strategies: Vaccines
- Parasite Molecular Genetics
- Parasite Coinfections
- Parasite Cell Biology I
- Wild Parasitology: into the field

Session 3 – Tue 22nd Mar 14:00 - 15.20 for '*Parasite Cell Biology I*' and '*Wild Parasitology: into the field'*; **14:30 – 15:20** for *'Diversity in Science I: Conversations toward inclusion and equity'.*

Day 2 – **14:00** - **15.20** Parasite Cell Biology I *(Lecture Theatre T/005)*

22-March-2022, at 14:00 to 15:00
Chairs - Dr Eden Ramalho Ferreira & Dr Rachel Neish
14:00 (20 mins) - A26229 - Breaking (down) the chain – structure/function studies of apicomplexan respiratory complexes (Lilach Sheiner)
14:20 (10 mins) - A25963 - A map of PFR proteins and dissection of their functions in motility and assembly of the *Trypanosoma brucei* flagellum. (Heloisa Berti Gabriel)
14:30 (10 mins) - A26118 - TbHD82 is important for the maintenance of nucleotide homeostasis in *Trypanosoma brucei* (Pablo Antequera)

15:00 (20 mins) - A26458 - Question Time - Parasite Cell Biology I

Day 2 - **14:00** - **15.20** Wild Parasitology: into the field *(Lecture Theatre P/X001)*

22-March-2022, at 14:00 to 15:00
Chair - Prof Matthew Thomas
14:00 (20 mins) - A26227 - The ecology of helminth infection and immunity in a wild rodent model (Amy Pedersen)
14:20 (10 mins) - A26148 - What you eat it is what you get: parasite-reptile interactions in a human-dominated landscape (Liesbeth Frias)
14:30 (10 mins) - A25671 - Toxocara sp. egg contamination of allotment-grown vegetables in the UK: A pilot study (Sara Healy)
14:40 (10 mins) - A25590 - Fish Faecal Xenomonitoring as a potential tool for schistosomiasis transmission monitoring (Zikmund Bartonicek)
14:50 (10 mins) - A26135 - The role of sylvatic rodents in transmission of Toxocara canis in NE Poland (Martyna Krupińska)

15:00 (20 mins) - A26459 - Question Time - Wild Parasitology: into the field

Day 2 – **14:30 – 15:20** Diversity in Science I: Conversations toward inclusion and equity *(Lecture Theatre K/018)*

22-March-2022, at 14:30 to 15:20
Chairs - Dr Giulia Bandini & Dr Sabrina Absalon
14:30 (20 mins) - A26222 - BIPOC in Parasitology: an anti-racist, equitable, and inclusive community of parasitologists. (Sabrina Absalon)
14:50 (20 mins) - A26335 - Building Bridges using the Universal Language of Science (Omar Harb)

Session 4 – Tue 22nd Mar 15:50 - 17.00

Day 2 – Diversity in Science I: Conversations toward inclusion and equity *(Lecture Theatre K/018)*

22-March-2022, at 16:20 to 16:25
Chairs - Dr Giulia Bandini & Dr Sabrina Absalon
15:50 (20 mins) - A26350 - Building a globally inclusive and equitable parasitology conference and community (Deepali Ravel)
16:20 (40 mins) - A26470 - RoundTable **Discussion**s - Diversity in Science I: Conversations toward inclusion and equity

Day 2 - Parasite Cell Biology I *(Lecture Theatre T/005)*

22-March-2022, at 15:50 to 16:25
Chairs - Prof Derrick Robinson & Dr Eden Ramalho Ferreira
15:50 (10 mins) - A26177 - Divergent metabolism between *Trypanosoma congolense* and T. *brucei* underlies differential sensitivity to metabolic inhibitors (Pieter Steketee)
16:00 (10 mins) - A26153 - Bioinformatic and functional characterisation of Trichomonas tenax GH30 homologues that potentially target fungal glycans (Lenshina Mpeyako)
16:10 (10 mins) - A25984 - Deletion of the P21 gene triggers changes in the invasion and replication of *Trypanosoma cruzi* (Thaise Teixeira)

16:25 (15 mins) - A26456 - Question Time - Parasite Cell Biology I

Turbo Talks – 5 min each talk –
16:40 (5 mins) - A26184 - Role of RDK2 and its interacting protein kinases in Leishmania *mexicana* differentiation. (Rachel Neish)
16:45 (5 mins) - A26115 - Characterising Heat Shock in *Trypanosoma congolense* (Marianne Aelmans)
16:50 (5 mins) - A26134 - *Galba truncatula* and Helminths, the Importance of Microbes (Peter McCann)
16:55 (5 mins) - A26105 - Expression and characterization of a mitochondrial fucosyltransferase from *Trypanosoma cruzi* and use of monoxenous parasite Crithidia fasciculata as an enzymatic source for synthesis of radioactive GDP-Fucose. (Jose Carlos Paredes-Franco)

Day 2 – Wild Parasitology: into the field *(Lecture Theatre P/X001)*

22-March-2022, at 15:50 to 16:40
Chair - Prof Matthew Thomas
15:50 (10 mins) - A25678 - Parasitic Platyhelminthes of Sparus aurata (Sparidae, Teleosteans): first report of the Digeneans Macvicaria obovata Molin, 1859 and Allopodocotyle pedicellata Stossich, 1887 off Algerian coast. (Fatima Zohra Zedam)
16:00 (10 mins) - A26129 - Development of a Recombinase Polymerase Amplification (RPA) for the detection of *Schistosoma mansoni* infection (Silvia Mesquita) - 16:10 (10 mins)
A25513 - Development of a computer visualization program for the taxonomic identification of Free Living Amoebas (FLAs) (Otavio Thiemann)
16:20 (10 mins) - A26011 - The role of sylvatic rodents in transmission of *Toxoplasma gondii* in NE Poland (Joanna Nowicka)
16:30 (10 mins) - A26174 - Genetic diversity and population structure analysis of various Taenia multiceps isolates from definitive and intermediate hosts worldwide (Ibrahim Abbas)

16:40 (20 mins) - A26457 - Question Time - Wild Parasitology: into the field

On-site Poster Session - 22nd Mar 17:00 - 18.00 - *(Atrium)* for '*Poster themes - Combative Strategies: Vaccines, Parasite Molecular Genetics, Parasite Coinfections Parasite Cell Biology I, Wild Parasitology: into the field.*

Day 3 – Wed 23rd Mar 2022 -

Session 5 – Wed 23rd Mar 10:00 - 11.20

Day 3 – Mathematical Modelling of Parasites *(Lecture Theatre K/018)*

23-March-2022, at 10:00 to 11:00
Chair - Dr Laurence Wilson
10:00 (20 mins) - A26161 - Can Mass Drug Administration of Moxidectin Accelerate Onchocerciasis Elimination in Africa? (Maria-Gloria Basanez)
10:20 (10 mins) - A26168 - Spatial determinants of water contact in Mayuge, Uganda: a cross-sectional study exploring exposure risk for Schistosomiasis (Max Eyre)
10:30 (10 mins) - A25921 - Public health policy pillars for the sustainable elimination of zoonotic schistosomiasis (Eva Janoušková)

11:00 (20 mins) - A26441 - Question Time - Mathematical Modelling of Parasites

Day 3 - Host:Parasite interactions : Tissue Tropism *(Lecture Theatre T/005)*

23-March-2022, at 10:00 to 11:00
Chairs - Dr Cecile Crosnier & Dr James Hewitson
10:00 (20 mins) - A26230 - Manipulation of inflammasome activation and host cell death by Leishmania parasites (Dario Zamboni)
10:20 (10 mins) - A26133 - Incomplete Recruitment of Protective T Cells Is Associated with *Trypanosoma cruzi* Persistence in the Mouse Colon (Martin Taylor)
10:30 (10 mins) - A26013 - *Plasmodium* sporozoites homing to the liver: exploring the interplay between parasite and host factors (Mónica Sá)

11:00 (20 mins) - A26442 - Question Time - Host:Parasite interactions : Tissue Tropism

Day 3 – Parasite:Vector Biology *(Lecture Theatre P/X001)*

23-March-2022, at 10:00 to 11:00
Chairs - Dr Álvaro Acosta-Serrano & Dr Poppy Lambert
10:00 (20 mins) - A26228 - A sense of direction: how African trypanosomes orient themselves in their insect host (Isabel Roditi)
10:20 (10 mins) - A26068 - Variation in water contact behaviour and risk of *Schistosoma mansoni* (re)infection among Ugandan school-aged children in an area with persistent high endemicity (Poppy Lamberton)
10:30 (10 mins) - A26003 - Xeno-monitoring of molecular drivers of artemisinin and partner drug resistance in P. *falciparum* populations in malaria vectors across Cameroon (Nkemngo Francis Nongley)

11:00 (20 mins) - A26443 - Question Time - Parasite:Vector Biology

Day 3 - ECR Career Workshop *(Lecture Theatre T/019)*

22-March-2022, at 10:00 to 11:20
Chair - TBD
Session Content TBD

Session 6 – Wed 23rd Mar 11:50 - 13.00

Day 3 - Mathematical Modelling of Parasites *(Lecture Theatre K/018)*

23-March-2022, at 11:50 to 12:40
Chair - Dr Laurence Wilson
11:50 (10 mins) - A26064 - The changing face of schistosome infection may help explain conflicting outcomes among malaria-schistosome coinfection studies (Sarah Rollason)
12:00 (10 mins) - A26132 - ScTralign: a computational method to align and compare biological development trajectories across conditions from single cell RNA sequencing data (Ross Laidlaw)
12:10 (10 mins) - A26234 - High speed, three-dimensional imaging to inform biophysical modelling in parasitology (Laurence Wilson)

12:40 (20 mins) - A26438 - Question Time - Mathematical Modelling of Parasites

Day 3 - Host:Parasite interactions : Tissue Tropism *(Lecture Theatre T/005)*

23-March-2022, at 11:50 to 12:40
Chairs - Dr Cecile Crosnier & Dr James Hewitson
11:50 (10 mins) - A25931 - Daily rhythms in malaria hosts and parasites influence artemisinin drug efficacy (Aliz Owolabi)
12:00 (10 mins) - A26162 - Histopathological and molecular diagnosis of eight clinical human hydatidosis from Gaza Strip, Palestine (Adnan Al-Hindi)
12:10 (10 mins) - A26121 - Novel *Trypanosoma brucei* heterogeneity is associated to tissue invasion and adaptation (Mariana De Niz)

12:40 (20 mins) - A26439 - Question Time - Host:Parasite interactions : Tissue Tropism

Day 3 – Parasite:Vector Biology *(Lecture Theatre P/X001)*

23-March-2022, at 11:50 to 12:40
Chair - Dr Álvaro Acosta-Serrano & Dr Poppy Lambert
11:50 (10 mins) - A25959 - Investigation of a protein kinase signalling pathway required for haptomonad differentiation in Leishmania *mexicana* (Nicola Baker)
12:00 (10 mins) - A25549 - Optimisation of in vitro feeding and long-term storage of the hematophagous mite Dermanyssus gallinae. (Francesca Nunn)
12:10 (10 mins) - A26203 - The MISP family of surface glycoproteins from *Trypanosoma brucei* is co-expressed with VSG and BARP in the metacyclic trypomastigote stage, adopts a triple helical bundle structure, and is not essential for the colonization of the tsetse salivary glands (Aitor Casas-Sanchez)
12:20 (10 mins) - A25683 - CTL4 controls TEP1-independent melanization of human malaria parasites (Maria Luisa Simoes)

12:40 (20 mins) - A26440 - Question Time - Parasite:Vector Biology

Day 3 - ECR Career Workshop *(Lecture Theatre T/019)*

23-March-2022, at 11:50 to 13:00
Chair - TBD
Session Content TBD

Lunch + Online Posters *T/005 + Online)* (Wed 23rd Mar 13:00 - 14.00)

Poster themes -

- Mathematical Modeling of Parasites
- Host:Parasite interactions : Tissue Tropism
- Parasite:Vector Biology
- Parasite Biochemistry
- Parasite ImmunoPathology
- Parasite Genetic Architecture

Session 7 – Wed 23rd Mar 14:00 - 15.20

Day 3 - Parasite Biochemistry *(Lecture Theatre K/018)*

23-March-2022, at 14:00 to 15:00
Chairs - Prof Anthony Wilkinson & Dr Michael Plevin
14:00 (20 mins) - A26226 - tba (Chi-Min Ho)
14:20 (10 mins) - A26204 - Invariant surface glycoprotein 65 of *Trypanosoma brucei* is a complement C3 receptor important for virulence (Alexander Cook)
14:30 (10 mins) - A25538 - A *Toxoplasma gondii* oxopurine transporter binds nucleobases and nucleosides using different binding modes. (Harry De Koning)

15:00 (20 mins) - A26444 - Question Time - Parasite Biochemistry

Day 3 - Parasite ImmunoPathology *(Lecture Theatre T/005)*

23-March-2022, at 14:00 to 15:00
Chairs - Dr Damian Perez Mazliah & Dr Jillian Barlow
14:00 (20 mins) - A25409 - Multi-omic approaches reveal a dynamic crosstalk between plasma cells and Cx3cr1+ microglia in the murine brain during chronic *Trypanosoma brucei* infection (Juan Fernando Quintana Alcala)
14:20 (10 mins) - A26131 - Dissecting side-by-side *Trypanosoma cruzi*-specific and cardiac-specific B cell responses in Chagas disease (Damian Perez Mazliah)
14:30 (10 mins) - A26107 - Experimental digestive Chagas disease: spatio-temporal infection dynamics, immunopathological mechanisms and the prospect of functional cure with trypanocidal benznidazole chemotherapy. (Michael Lewis)
14:40 (10 mins) - A25934 - A library of cell-surface and secreted *Schistosoma mansoni* proteins to investigate host:parasite interactions (Cecile Crosnier)

15:00 (20 mins) - A26445 - Question Time - Parasite ImmunoPathology

Day 3 – Parasite Genetic Architecture *(Lecture Theatre P/X001)*

23-March-2022, at 14:00 to 15:00
Chairs - Dr Nathaniel Jones & Dr Joana Faria
14:00 (20 mins) - A26192 - Thousands of messenger RNA untranslated regions reprogram post-transcriptional gene expression profiles in trypanosomes (David Horn)
14:20 (10 mins) - A25669 - Heterogeneous elongation of RNA polymerase I transcription at the active VSG expression site in *Trypanosoma brucei* (James Budzak)
14:30 (10 mins) - A25922 - The *Trypanosoma brucei* RNA/DNA hybrid interactome reveals a role for RAD51 in R-loop homeostasis and repair of VSG-localised DNA breaks during antigenic variation (Mark John Girasol)

15:00 (20 mins) - A26446 - Question Time - Parasite Genetic Architecture

Day 3 – Scientific Publishing *(Lecture Theatre T/019)*

23-March-2022, at 14:00 to 15:20
Chair - Dr Kevin Tyler
Speakers – Dr Kevin Tyler (Virulence, CRPVD), Prof Russ Stothard (Parasitology), Dr Lucy Jones (Elsevier Life Sciences), Prof Ariel Silber (Experimental Parasitology), Prof Richard McCulloch (MBP), Prof John Ellis (Parasitology), Prof Maria Gloria Basanez (PLoS NTD and others), Prof Filipe Dantas Torres (P&V), Prof Martin Llewllyn (Proc R. Soc), Dr Joanne Power (BSP Social Media Secretary).
Questions and Answers from 3.20

Session 8 – Wed 23rd Mar 15:50 - 17.00

Day 3 - Parasite Biochemistry *(Lecture Theatre K/018)*

23-March-2022, at 15:50 to 16:40
Chairs - Prof Anthony Wilkinson & Dr Michael Plevin
15:50 (10 mins) - A26146 - Biochemical investigations revealed the inhibitory mechanisms of novel inhibitors of Trypanosome Alternative Oxidase active against human and animal African trypanosomiasis (Godwin Ebiloma)
16:00 (10 mins) - A25983 - Investigating the role of glycosylation in *Toxoplasma gondii* protein homeostasis (Giulia Bandini)
16:10 (10 mins) - A26309 - The disorderly behaviour of Leishmania hydrophilic acylated surface proteins (HASPs) (Michael Plevin)

16:40 (15 mins) - A26447 - Question Time - Parasite Biochemistry

Turbo Talks – 5 min each talk –
23-March-2022, at 16:55 to 17:05
16:55 (5 mins) - A26116 - Biophysical and biochemical characterisation of the interaction between Leishmania braziliensis PRMT1 and PRMT3 (Edward Nay)
17:00 (5 mins) - A26002 - Investigating a galactokinase orthologue from Leishmania *donovani* (Hasana Baber)

Day 3 - Parasite ImmunoPathology *(Lecture Theatre T/005)*

23-March-2022, at 15:50 to 16:40
Chairs - Dr Damian Perez Mazliah & Jillian Barlow
15:50 (10 mins) - A26163 - Investigating the roles of the cell regulator TRIM24 during visceral leishmaniasis (Edward Muscutt)
16:00 (10 mins) - A26155 - Mapping the immune response in schistosomiasis – insights from controlled human infection models. (Emma Houlder)
16:10 (10 mins) - A26183 - Chronic schistosome infection remodels bone marrow haematopoiesis (James Hewitson)

16:40 (20 mins) - A26448 - Question Time - Parasite ImmunoPathology

Day 3 – Parasite Genetic Architecture *(Lecture Theatre P/X001)*

23-March-2022, at 15:50 to 16:40
Chairs - Dr Nathaniel Jones & Dr Joana Faria
15:50 (10 mins) - A26167 - RNase H1, a R-loop resolving enzyme, acts to suppress R-loop mediated DNA replication and limit genome instability in Leishmania (Jeziel Dener Damasceno)
16:00 (10 mins) - A26143 - The open chromatin profile changes at genome compartments and at tDNA loci in *Trypanosoma cruzi* life formstract (Julia Pinheiro Chagas Cunha)
16:10 (10 mins) - A25926 - Leishmania Bromodomain Factor 5 is an Essential Transcriptional Regulator (Nathaniel Jones)
16:20 (10 mins) - A25994 - A kinetoplastid-specific subunit of the Origin Recognition Complex? (Catarina Marques)

16:40 (20 mins) - A26449 - Question Time - Parasite Genetic Architecture

Day 3 – Scientific Publishing *(Lecture Theatre T/019)*

23-March-2022, at 15:50 to 17:00
Chair - Dr Mario de Bono
15:45 (30 mins) - *Tapeworm tumours: how not to make a helminth* (Peter Olson)
16:15 (15 mins) - *Detecting Molecular Similarities Between Allergenic And Metazoan Parasitic Proteins: Allergy In The Light of Immunity* (Nicholas Furnham)
16:30 (15 mins) - *Akt signalling in the human parasite Schistosoma mansoni* (Maxine Mckenzie)

On-Site Poster Session - Wed 23rd Mar 17:00 - 18.00 - *(Atrium)* for '*Poster themes - Mathematical Modeling of Parasites, Host:Parasite interactions : Tissue Tropism, Parasite:Vector Biology, Parasite Biochemistry, Parasite ImmunoPathology, Parasite Genetic Architecture*

Day 4 – Thu 24th Mar 2022 -

Session 9 – Thu 24th Mar

10:00 - 11.20 for '*Combative Strategies: Drug Discovery*', '*Parasite Cell Biology II', and CHIM workshop: Controlled Human Infection Models;*

10:30 – 11:20 for *'Diversity in Science II: Conversations toward inclusion and equityI'.*

Day 4 - Combative Strategies: Drug Discovery *(Lecture Theatre K/018)*

24-March-2022, at 10:00 to 11:00
Chair - Dr Elmarie Myburgh
10:00 (20 mins) - A26231 - A platform for drug target deconvolution and exploitation (Susan Wyllie)
10:20 (10 mins) - A26109 - SMGBs as novel in vitro and in vivo anti-infective agents for Acanthamoeba spp. infections. (Alemao Gustavo Carpinteyro Sanchez)
10:30 (10 mins) - A26085 - First Insights into the Autophagy Machinery and its Induction by Imatinib in *Schistosoma mansoni* (Mudassar Niaz Mughal)
10:40 (10 mins) - A26156 - How do Schistosomes Breathe? (Adam Burgess)

11:00 (20 mins) - A26460 - Question time - Combative Strategies: Drug Discovery

Day 4 - Parasite Cell Biology II *(Lecture Theatre P/X001)*

24-March-2022, at 10:00 to 11:00
Chairs - Dr James LaCourse & Dr Eden Ramalho Ferreira
10:00 (10 mins) - A26093 - The unique mRNA decapping enzyme ALPH1 of trypanosomes (Paula Andrea Castañeda Londoño)
10:00 (20 mins) - A26264 - Intrabody induced cell killing by targeting an essential cytoskeletal protein in T. *brucei* (Derrick Robinson)
10:20 (10 mins) - A26265 - Characterisation of the essential trypanosome protein TbSmee1 reveals that endocytosis is required for flagellar pocket access of surface-bound cargo (Brooke Morriswood)

11:00 (20 mins) - A26461 - Question Time - Parasite Cell Biology II

Day 4 - CHIM workshop: Controlled Human Infection Models *(Lecture Theatre T/019)*

24-March-2022, at 10:00 to 11:20
Chair - TBD
Session Content TBD

Day 4 - Diversity in Science II: Conversations toward inclusion and equity *(Lecture Theatre T/005)*

24-March-2022, at 10:30 to 11:20
Chair - Dr Giulia Bandini
10:30 (20 mins) - A26330 - Women in science: a picture of the Brazilian scenery (Angela Kaysel Cruz)
10:50 (20 mins) - A26331 - Is Open Access Inclusive? (Ariel Silber)

Session 10 – Thu 24th Mar 11:50 - 13.00.

Day 4 - Combative Strategies: Drug Discovery *(Lecture Theatre K/018)*

24-March-2022, at 11:50 to 12:40
Chair - Dr Elmarie Myburgh
11:50 (10 mins) - A25951 - Investigating Bromodomain Proteins as Targets for Anti-Leishmanial Drug Discovery (Catherine Russell)
12:00 (10 mins) - A26178 - The development of an oral oleylphosphocholine treatment for cutaneous leishmaniasis (Katrien Van Bocxlaer)
12:10 (10 mins) - A26102 - CPSF3 and beyond…a Systematic View of the Mode-of-Action of Benzoxaboroles in African Trypanosomes (Ning Zhang)
12:20 (10 mins) - A26078 - Oligo targeting for profiling drug resistance mutations in the parasitic *Trypanosoma*tids (Melanie Ridgway)

12:40 (20 mins) - A26463 - Question Time - Combative Strategies: Drug Discovery

Day 4 - Parasite Cell Biology II *(Lecture Theatre P/X001)*

24-March-2022, at 11:50 to 12:40
Chairs - Dr James LaCourse & Dr Eden Ramalho Ferreira
11:50 (10 mins) - A25970 - Single-cell RNA-Sequencing Analysis of Life and Cell Cycle Progression in L. *mexicana* (Felix Warren)
12:00 (10 mins) - A25679 - *Schistosoma mansoni* α-N-acetylgalactosaminidase (SmNAGAL) regulates coordinated parasite movement and egg production (Ben Hulme)
12:10 (10 mins) - A25958 - Mistargeting of aggregation-prone mitochondrial proteins activates a nucleus-mediated posttranscriptional quality control pathway in trypanosomes (Caroline Dewar)
12:20 (10 mins) - A25547 - Characterisation of a host receptor for *Plasmodium falciparum*-infected erythrocyte rosette formation (Molly Carlier)

12:40 (20 mins) - A26462 - Question Time - Parasite Cell Biology II

Day 4 - CHIM workshop: Controlled Human Infection Models *(Lecture Theatre T/019)*

24-March-2022, at 11:50 to 13:00
Chair - TBD
Session Content TBD

Day 4 - Diversity in Science II: Conversations toward inclusion and equity *(Lecture Theatre T/005)*

24-March-2022, at 11:50 to 12:15
Chair - Dr Giulia Bandini
11:50 (20 mins) - A26332 - Autism: myths and realities: a disability, a neurotype, and challenges for inclusion and equity. (Mariana De Niz)

12:20 (40 mins) - A26471 - RoundTable **Discussion**s - Diversity in Science II: Conversations toward inclusion and equity

Lunch *T/005* (Thu 24th Mar 13:00 - 14.00)

Session 11 – Thu 24th Mar 14:00 - 16.00 - *Presidents Medal & CA Wright Medal awards*

Day 4 - Presidents Medal and CA Wright Medal awards and acceptance talks *(Lecture Theatre P/X001)*

Chair - Prof Colin Sutherland & Prof Jo Hamilton

President's Medal Award - Dr Joana R. Correia Faria. (25 min)
Wright Medal Award - Dr Álvaro Acosta-Serrano (45 min)

Session 12 – Thu 24th Mar 16:00 - 18.30 - *Castle Howard Afternoon Reception*

Day 4 – Castle Howard Afternoon Reception

Buses to Castle Howard (4.00 -4.30PM
Reception at Castle Howard (5.00-6.30PM)
Buses back from Castle Howard (6:30-7.00PM from reception).

Session 13 – Thu 24th Mar 19:00 - 23.30 - *Castle Howard Dinner*

Day 4 – Castle Howard Dinner

Conference dinner 7.00-11.00PM.
Buses back from Castle Howard 10:30-11.00PM.

Day 5 – Fri 25th Mar 2022 -

Session 14 – Fri 25th Mar 09:00 - 10.00 - *BSP Council Annual General Meeting*

Day 4 - BSP Council Annual General Meeting *(Lecture Theatre P/X001)*

25-March-2022, at 09:00 to 10:00
Chairs Prof Colin Sutherland, Prof Jo Hamilton & Dr Kevin Tyler

According to Agenda TBD

Session 15 – Fri 25th Mar 10:00 - 11.20

Day 4 - Parasite Evolutionary Genomics *(Lecture Theatre K/018)*

Chairs - Dr Daniel Jeffares & Dr Joao Cunha
10:00 (20 mins) - A26223 - Comparative Genomics in *Trypanosoma*tids: genetic diversity of isolates in endemic regions in Brazil and identification of virulence factors (Daniella Bartholomeu)
10:20 (10 mins) - A26060 - Population genomics and geographic dispersal in Chagas disease vectors: Landscape drivers and evidence of possible adaptation to the domestic setting. (Martin Llewellyn)
10:30 (10 mins) - A26159 - Aneuploidies are an ancestral feature in *Trypanosoma*tids and could be related to parasite adaptation (Samuel Carvalho)
10:40 (10 mins) - A26110 - Assessing LRV1 role as risk factor for mucosal leishmaniasis occurrence and its relationship with TRL3 polymorphism (Maria Echeverry)

11:00 (20 mins) - A26467 - Question Time - Parasite Evolutionary Genomics

Day 4 - Parasite Gene Expression *(Lecture Theatre T/005)*

25-March-2022, at 10:00 to 11:00
Chairs - Dr Nathaniel Jones & Dr Joana Faria
10:00 (20 mins) - A25990 - Roles and interactions of the specialized initiation factors EIF4E2, EIF4E5 and EIF4E6 in *Trypanosoma brucei*: EIF4E2 maintains the abundances of S-phase mRNAs (Christine Clayton)
10:20 (10 mins) - A26176 - RNA binding proteins as trans-regulators impacting surveillance and infectivity in Leishmania (Natalia Teles)
10:30 (10 mins) - A25988 - Characterisation of a new Apicomplexa-specific zinc-finger protein family in *Plasmodium* with a key role across different stages of the life cycle. (Lauren Carruthers)
10:40 (10 mins) - A26160 - Post-transcriptional iron regulatory mechanisms in *Trypanosoma brucei* (Calvin Tiengwe)

11:00 (20 mins) - A26468 - Question Time - Parasite Gene Expression

Day 4 - Trafficking, Signaling *(Lecture Theatre P/X001)*

25-March-2022, at 10:00 to 11:00
Chairs - Dr Romina Nievas & Dr Nicola Baker
10:00 (20 mins) - A26206 - Heavy Metal: The role of iron storage in *Toxoplasma gondii* (Clare Harding)
10:20 (10 mins) - A25985 - *Ascaris suum* Pseudocoelomic Fluid: A Peptide-rich Biofluid that Modulates Nematode Motility (Darrin McKenzie)
10:30 (10 mins) - A25761 - Decoding heat shock signalling in the African trypanosome (Michael Urbaniak)
10:40 (10 mins) - A25839 - Exploiting Omics Approaches to Unravel Endocannabinoid Biology in *Strongyloides* Parasites (Luke Cadd)

11:00 (20 mins) - A26464 - Question Time - Trafficking, Signalling

Day 4 - Social Sciences and Parasitology Workshop *(Lecture Theatre T/019)*

Chair - Dr James LaCourse
10:30 Prof Helen Price, Keele University, and members of the ECLIPSE team. Insights from the ECLIPSE programme on cutaneous leishmaniasis
11:20-11.50 Coffee break

Session 16 – Fri 25th Mar 11:50 - 13.00

Day 4 - Parasite Evolutionary Genomics *(Lecture Theatre K/018)*

25-March-2022, at 11:50 to 12:40
Chairs - Dr Daniel Jeffares & Dr Joao Cunha
11:50 (10 mins) - A25502 - Morphological and PCR Screening of *Schistosoma* Hybrid infecting humans in communities around the Oyan River Dam Area, Ogun State, Nigeria (Uwemedimo Ekpo)
12:00 (10 mins) - A26059 - The clonal dynamics and molecular epidemiology of Amoebic Gill Disease in Salmo salar via multiplex amplicon sequencing (Bachar Cheaib)
12:10 (10 mins) - A26045 - Parasite genotype strongly influences mortality risk in visceral leishmaniasis. (Daniel Jeffares)

12:40 (20 mins) - A26466 - Question Time - Parasite Evolutionary Genomics

Day 4 - Parasite Gene Expression *(Lecture Theatre T/005)*

25-March-2022, at 11:50 to 12:40
Chairs - Dr Nathaniel Jones & Dr Joana Faria
11:50 (10 mins) - A25919 - Control of variant surface glycoprotein expression by CFB2 in African trypanosomes and quantitative proteomic connections to translation and cytokinesis (Gustavo Bravo Ruiz)
12:00 (10 mins) - A26179 - The mobile genome – transposable elements in *Schistosoma mansoni* and *Fasciola hepatica* (Anna Protasio)
12:10 (10 mins) - A25379 - Using nanopore sequencing to identify transcript variation between different life cycle stages of the parasitic nematode *Strongyloides* ratti (Dominika Lastik)
12:20 (10 mins) - A26095 - A single-cell atlas of the free-living miracidium larva of *Schistosoma mansoni* (Teresa Attenborough)

12:40 (20 mins) - A26465 - Question Time - Parasite Gene Expression

Day 4 - Trafficking, Signalling *(Lecture Theatre P/X001)*

25-March-2022, at 11:50 to 12:40
Chairs - Dr Romina Nievas & Dr Nicola Baker
11:50 (10 mins) - A26027 - K13-associated endocytic structures in *Toxoplasma* are required for plasma membrane homeostasis rather than parasite growth (Brandon Mercado)
12:00 (10 mins) - A25929 - Protein Kinase Involvement in Leishmania Cell Cycle Regulation Revealed Using Chemical Genetics (Juliana Brambilla Carnielli Trindade)
12:10 (10 mins) - A26114 - Investigating the role of unique kinesin-2 motors in intraflagellar transport in trypanosomes (Aline Araujo Alves)

12:40 (20 mins) - A26469 - Question Time - Trafficking, Signaling

Day 4 - Social Sciences and Parasitology Workshop *(Lecture Theatre T/019)*

25-March-2022, at 11:50 to 13:00
Chair - Prof Helen Price
11:50-12:10 Gregory Milne (A25500) Secular changes in exposure to *Toxoplasma gondii*: Implications for congenital disease in human populations
12:10-12:30 Jo Widdecombe The economic evaluation of Cystic echinococcosis control strategies focused on zoonotic hosts: A scoping review
12:30-13:00 **Discussion**: Gaps and barriers in the control of parasite diseases

Lunch - *T/005* (Fri 25th Mar 13:00 - 14.00)

Prizes/Farewell/Close (*Lecture Theatre P/X001*- Fri 25th Mar 14:00 - 15.00)

Full Abstracts by Sessions

Session 1 – Tue 22nd Mar 10:00 - 11.20

Day 2 – Combative Strategies: Vaccines *(Lecture Theatre K/018)*

22-March-2022, at 10:00 to 11:00
Chairs - Prof Gavin Wright & Dr Mohamed Osman

10:00 (20 mins) - A26225 - Towards Broadly-Neutralising Blood-Stage Vaccines against Human Malaria Parasites

Author - Prof Simon Draper
University of Oxford

Abstract - *Plasmodium falciparum* malaria affects 200-300 million people annually, resulting in the death of about 0.5 million individuals. Another species of malaria parasite, P. *vivax*, also causes severe and relapsing malaria illness, but less mortality. P. *vivax* is more geographically widespread than P. *falciparum*, and is found largely in South America and South-East Asia. Thus, despite increasing implementation of control measures, the burden of malarial death and disease remains far too high. The most advanced subunit vaccine against P. *falciparum*, called RTS,S/AS01, has shown modest short-term efficacy against clinical disease in young children, whilst no effective vaccine exists for P. *vivax*. More recently, calls have been made for a second generation vaccine to exert 75% efficacy over two years against both species of parasite. If this ambitious rhetoric is to be realised, new approaches to malaria vaccine design are required. Vaccines against the parasite's asexual blood-stage have the potential to complement RTS,S/AS01, and reduce mortality, morbidity and transmission of malaria, however such a vaccine has proved elusive.Recently, we have developed next-generation vaccines targeting the reticulocyte-binding protein homologue 5 from P. *falciparum* (PfRH5) and the Duffy-binding protein from P. *vivax* (PvDBP), both of which mediate essential invasion pathways into the human red blood cell. Critical to this work has been the elucidation of how human antibody responses are able to neutralise parasite invasion to guide rational vaccine design, coupled with human experimental studies. This talk will describe our on-going work and present data from our most recent Phase I/II clinical trials both in the UK and Africa.

10:20 (10 mins) - A26080 - Pre-erythrocytic and transmission-blocking multi-stage malaria vaccine indicates synergic sterile protection effect in a murine model with *Plasmodium berghei* transgenic parasite

Authors - T Mizuno[2]; AM Blagborough[1]; M Niikura[3]; AA Hasyim[4]; M Iyori[4]; Y Yamamoto[4]; A Sakamoto[4]; H Mizukami[5]; H Shida[6]; S Yoshida[4];
[1] Department of Pathology, University of Cambridge, UK; [2] Department of Global Infectious Diseases, Kanazawa University, Japan; [3] Department of Infectious Diseases, Kyorin University, Japan; [4] Laboratory of Vaccinology and Applied Immunology, Kanazawa University, Japan; [5] Division of Gene Therapy, Jichi Medical University, Japan; [6] Institute for Genetic Medicine, Hokkaido University, Japan.

Abstract - The Malaria Vaccine Technology Roadmap 2013 (World Health Organization) aims to develop safe and effective vaccines by 2030. It targets at least 75% protective efficacy. Recently, we've established a highly effective multistage vaccine against *Plasmodium falciparum*. This heterologous prime-boost viral-vectored vaccine induces both humoral and cellular immune responses effectively and durable. This notable vaccine is targeting multi-stage in *P. falciparum* life cycles. The vaccine encodes both a pre-erythrocytic stage antigen circumsporozoite protein (PfCSP) and one sexual stage antigens s25 (Pfs25). The combination between the pre-erythrocytic vaccine (PEV) effect and transmission-blocking vaccine (TBV) effect brings the synergy to enhance the vaccine more potent in malaria prevention than single antigen target strategy. In this study, we evaluated both PE and TB efficacies of our newly-developed vaccine in a rodent model. As a result, our vaccine showed 100% protection as PEV against PfCSP-transgenic *P. berghei* sporozoites and more than 90% oocyst reduction in mosquito midgut as TBV against Pfs25-transgenic *P. berghei*. Next, we evaluated its synergy of TBV+PEV on a double transgenic *P. berghei* expressing both PfCSP and Pfs25. We performed the assay in conditions that weaken our vaccine effect to measure the benefit of synergy. As a result, while the individual efficacy decreased to about 50% in TBV and about 60% in PEV respectively, the synergy of TBV+PEV remained over 90%. These findings propose the potential of our vaccine as a "next-generation malaria vaccine" to achieve the landmark goals of the malaria vaccine technology roadmap.

10:30 (10 mins) - A26009 - An exploratory study to verify the safe and reproduceable use of aseptic purified cryopreserved *Plasmodium falciparum* sporozoites for the induction of controlled human malaria infection in healthy malaria-naïve adults at hVIVO medical research unit.

Authors - **A Odedra**[3]; A Wildfire[3]; A Anandakumar[3]; J Myott[3]; M Leers[3]; R Danaf[3]; L Stewart[2]; D Nolder[2]; H Liddy[2]; CJ Sutherland[1];
[1] London School of Hygiene and Tropical Medicine, UK; [2] London Schhool of Hygiene & Tropical Medicine, UK; [3] hVIVO Limited, UK

Abstract - Controlled human malaria infection (CHMI) studies involving the introduction of malaria parasites into healthy volunteers have been successfully employed in the investigation of new antimalarial drugs, vaccine candidates and diagnostic assays. Here we describe the initiation of CHMI studies at a new site in London, to facilitate research on parasite biology and host responses in a UK cohort. In a proof-of-principle, pilot study, we challenged two malaria naive, healthy human subjects with ~3,200 aseptic purified cryopreserved *Plasmodium falciparum* Sporozoites (PfSPZ Sanaria®) by direct venous inoculation (DVI) on Day 1. Subjects remained as in-patients within the hVIVO Ltd clinical research unit for 48 hours post-inoculation (Day 3) after which they were followed up as outpatients and underwent daily monitoring from Day 7 until proven negative by quantitative polymerase chain reaction, targeting the 18S rRNA gene (18S-qPCR). Testing was carried out at United Kingdom Health safety agency (UKHSA) malaria reference laboratory at the London School of Hygiene and Tropical Medicine (LSHTM). Volunteers were rescued with a 3-day course of artemether-lumefantrine within 24 hours of reaching 18S-qPCR threshold ≥250 parasites/mL. Volunteers were exempted from daily clinic visits and were deemed to have achieved 'test of cure' following two consecutive negative 18S-qPCR signals. Safety assessments comprised clinical and laboratory monitoring of vital signs, adverse events and biochemical markers. One female and one male volunteer were successfully inoculated with PfSPZ Sanaria®. Parasitological and clinical histories of both participants will be presented. There were no severe or serious

adverse events and no evidence of *P. falciparum* recrudescence. hVIVO Ltd and LSHTM have safely and successfully conducted a CHMI study in London. The model can now be used to better understand and develop new interventions against the *P. falciparum* parasite.

Day 2 – Parasite Molecular Genetics *(Lecture Theatre T/005)*
22-March-2022, at 10:00 to 11:00
Chairs - Dr Natalia Teles & Dr Pegine Walrad

10:00 (20 mins) - A26224 - RNA communication in helminth-host interactions

Amy Buck
University of Edinburgh

RNA is a dynamic molecule that underpins biological complexity through diverse mechanisms of gene regulation. In plants RNA can transmit information between diverse parasites and plant cells. Research in the last five years demonstrates that parasitic nematodes also release RNAs that get internalized by host (mammalian) cells. We previously showed that the gastrointestinal nematode Heligmosomoides polygyrus bakeri (Hpb) exports small RNAs in extracellular vesicles that are internalized by mouse epithelial and macrophage cells, and these suppress host immune responses in vitro and in vivo. We further showed that raising antibodies against the EVs confers protection to infection, suggesting that EVs are an important mechanism of host modulation. More recently we found that the dominant class of RNAs in EVs is siRNAs generated by RNA-dependent RNA polymerases inside the nematode and there is a specific enrichment in siRNAs derived from novel repetitive and transposable elements. Small RNA sequencing of mouse epithelial cells following EV treatment suggests a subset of the nematode siRNAs and miRNAs are present at functionally relevant concentrations and may interact with host genes with high degrees of complementarity. We further characterize one specific vesicular Argonaute protein (exWAGO) that associates with the siRNAs and mediates their export. Using immunoprecipitation techniques we have developed a method to capture exWAGO in vivo and identify the guide RNAs with which it associates under different environmental conditions. We have also developed methods for directly detected the exWAGO protein and parasite EVs in vivo, providing evidence for RNA transmission from nematode to mouse under physiological conditions.

10:20 (10 mins) - A25940 - Developmental incompetence in selected and naturally occurring trypanosome isolates

Authors
G Oldrieve[1]; M Cayla[1]; F Van den Broeck[2]; K Matthews[1];
[1] Institute of Immunology & Infection Research, University of Edinburgh, UK; [2] Department of Microbiology, Immunology and Transplantation, Rega Institute, Belgium

Abstract
Trypanosoma brucei, a parasitic species which causes human and animal African trypanosomiasis, presents a complex digenic life cycle that requires transmission between mammalian hosts by its insect vector, the tsetse fly. In its mammalian host, the proliferative 'slender' bloodstream-form morphs into a transmissible 'stumpy' form, which is adapted to survive in the tsetse fly. The slender to stumpy transition occurs in a density dependent quorum sensing (QS) like process for which key molecular regulators have been identified (e.g. Mony *et al*; doi:10.1038/nature12864). Some *T. brucei* subspecies (*T. b. evansi* and *T. b. equiperdum*) that circulate in the field are less able to transition from the slender to stumpy morphotype and so are described as 'monomorphic'. These subspecies have forgone their tsetse vector and have expanded their geographic range outside of Sub-Saharan Africa. Monomorphic *T. brucei* are transmitted mechanically, between livestock and wildlife, via biting flies (*T. b. evansi*) or during equine coitus (*T. b. equiperdum*). They cause the diseases Surra or Dourine, respectively. Lacking population growth control, they can also display increased virulence compared to pleomorphic *T. brucei*. Using tools developed during the elucidation of the QS pathway, we aim to understand how development has been disrupted in naturally occurring monomorphs. In parallel, we aim to identify additional regulatory mechanisms, through the selection of monomorphic *T. brucei* in the laboratory, which will assist in determining how monomorphism is selected in the field.

Utilising whole genome sequences of 41 naturally occurring monomorphic isolates, we corroborate previous studies in identifying at least four independent monomorphic *T. brucei* clades. We found clear lineage-specific variation in the selection efficacy and heterozygosity of these lineages, supporting their distinct evolutionary histories. Using genomic variants, we highlighted genes which are under positive selection in monomorphic lineages, but not pleomorphic ones. Variants unique to monomorphic lineages were explored for their contribution to monomorphism. Prioritisation was based on previous identification of the gene as a QS regulator, selection pressure acting on the gene and the position of the variant in relation to predicted domains. Thereafter, monomorphic gene sequences were synthesised for each of the monomorphic lineages and used to replace wild type alleles, via CRISPR-Cas9, in developmentally competent *T. brucei*. We found that two of the mutant genes analysed to date cause reduced responsiveness to the QS signal, as determined using an *in vitro* differentiation assay. Further identified variants are being tested using this analytical pipeline.

In a complementary approach, we selected 40 clonal monomorphic cell lines *in vitro* from a pleomorphic parental cell line via serial passage. Samples were stored periodically across the selection series allowing the tracking of molecular changes associated with the loss of pleomorphism over time. Initial experiments, using a single cell line, highlighted significant differential expression of transcripts from known QS genes during the selection of monomorphism. Once the loss of differentiation has been validated for the 40 clonal monomorphs, transcriptomes and genomes will be analysed to identify common molecular changes which accrue during the progression from pleomorphism to monomorphism.

By combining results from naturally occurring and selected monomorphic *T. brucei*, we have begun to highlight how monomorphism can arise, providing insight into the molecular control of the QS process and diagnostic tools to anticipate increased virulence in the field.

10:30 (10 mins) - A25960 - Accessing the variability of multicopy genes in complex genomes using unassembled short reads: the case of *Trypanosoma cruzi* multigene families

Authors
JL Reis-Cunha[1]; A Coqueiro-dos-Santos[4]; SA Carvalho[4]; GF Rodrigues-Luiz[3]; RP Baptista[2]; LV Almeida[4]; NR Honorato[4]; FP Lobo[4]; VG Fraga[4]; LL Bueno[4]; RT Fujiwara[4]; MS Cardoso[4]; D Bartholomeu[4]; GC Cerqueira[5];

[1] University of York, UK; [2] University of Georgia, United States; [3] Campinas State University (UNICAMP), Brazil; [4] Federal University of Minas Gerais - UFMG, Brazil; [5] Personal Genome Diagnostic, United States

Abstract

Multicopy genes and other repetitive elements cause assembly fragmentation in complex eukaryotic genomes, limiting the study of their variability. The genome of *Trypanosoma cruzi*, the protozoan parasite that causes Chagas disease, has a high repetitive content, which consist of multigene families, transposable elements, tandem repeats, and satellite sequences. Although many *T. cruzi* multigene families encode surface proteins that play pivotal roles in host-parasite interactions, their variability is currently underestimated, as their high repetitive content results in collapsed gene variants, even in current long-read assemblies. Also, there are few studies comparing multigene family's variability among Discrete Typing Units (DTUs), which are usually performed at the level of assembled genomes, using a limited number of strains. To estimate sequence variability and copy number variation of multigene-repetitive families, we have developed a whole-genome-sequencing read-based approach that is independent of gene-specific mapping and *de novo* assembly. Reads from each parasite isolate are mapped in a reference containing genomic sequences from representative strains, and reads that map to any given gene of a family of interest are recovered and fragmented in 30 nucleotide long k-mers. These k-mers are clustered based on sequence similarity to reduce redundancy. Finally, sums of counts of all k-mers in each cluster are assumed as the cluster copy number. This methodology was used to estimate the copy number and variability of MASP, TcMUC and Trans-Sialidase (TS), the three largest *T. cruzi* multigene families, in 36 *T. cruzi* strains, including members of all six parasite DTUs. This analysis has shown that *T. cruzi* multigene families present a specific pattern of variability and copy number among the distinct parasite DTUs. TcI isolates had the lowest, while hybrid strains present the highest sequence variability, suggesting that maintaining a larger content of their members after hybridization could be advantageous. There were differences observed between the hybrid strains CL Brener and Tulahuen, which suggests that they could have resolved the hybridization differently. The three evaluated multigene families vary in antigenicity in murine model, where the antibody response to MASP and TS had respectively the highest and lowest diversification with chronification. The reactivity of sera from chronic Chagasic human patients was focused on TS antigens, suggesting that targeting TS conserved sequences could be a potential avenue to improve diagnosis and vaccine design against Chagas disease. Finally, the proposed approach can be applied to study multicopy genes in any organism, providing new possibilities to access sequence variability in complex genomes.

Day 2 – Parasite Coinfections *(Lecture Theatre P/X001)*

22-March-2022, at 10:00 to 11:00

Chairs - Prof Colin Sutherland & Mr John Archer

10:00 (20 mins) - A26216 - Multiple-fronts costs of defence: the case of the swarming T-helper cells

Author - Prof Andrea Graham

Princeton University

Abstract - Individual-level heterogeneity is abundantly clear in host-parasite interactions, and all the more so during a global pandemic: hosts exhibit remarkable heterogeneity in the strength, speed and specificity of their immune responses, which in turn generates varied susceptibility to infectious and autoimmune diseases and varied propensity to transmit disease. Why do hosts vary so much? One important category of explanation for host variation arises from cost-benefit analysis of defence. I will illustrate how multiple-fronts costs of defence, for example, can help to explain host heterogeneity in natural populations subject to co-infections. I will focus on a particular mechanism generating multiple-fronts costs in mammals: when T-helper cells polarize into worm-clearing and germ-clearing factions. Subtle differences in initial conditions can be amplified by the collective behavior of T-helper cells to generate radically different system-level responses in different hosts. I will outline how adaptive systems analysis is beginning to shed light on the adaptive value of "agile swarms" of these cells during infections, even if acute worm-germ co-infections may prove costly to hosts.

10:20 (10 mins) - A25511 - Evolution of leishmania viruses with a focus on LRV2

Authors - V Yurchenko[1]; AY Kostygov[1]; D Grybchuk[2]; A Zakharova[1]; A Saura[1]; ES Gerasimov[3];

[1] University of Ostrava, Czech Republic; [2] Central European Institute of Technology, Masaryk University,, Czech Republic; [3] Moscow State University, Moscow, Russian Federation

Abstract - *Leishmania* spp. are important pathogens causing a vector-borne disease with a broad range of clinical manifestations from self-healing ulcers to the life-threatening visceral forms. Presence of *Leishmania RNA virus* (LRV) confers survival advantage to these parasites by suppressing antileishmanial immunity in the vertebrate host. The two viral species, LRV1 and LRV2 infect species of the subgenera *Viannia* and *Leishmania*, respectively. Here, we investigated co-phylogenetic patterns of leishmaniae and their viruses on a small scale (LRV2 in *L. major*) and demonstrated their predominant coevolution, occasionally broken by intraspecific host switches. This contrasts with a co-phylogenetic pattern of LRV1 and *Leishmania* (*Viannia*). To better understand how LRVs are intermingled with host metabolism, we established LRV1- and LRV2-negative strains of *L. guyanensis* M4147 and *L. major* TG44, respectively by 2'-C-methyladenosine treatment. The direct comparison of virus-positive and -negative strains by RNA-seq has revealed that LRV1 and LRV2 affect their respective hosts differently. While only 2 transcripts were found to be differentially expressed between *L. guyanensis* LRV1$^+$ and LRV1$^-$, this number in the case of *L. major* was substantially higher (67 up- and 20 down-regulated genes in LRV2$^-$ cells). The GO enrichment analysis has shown that they may be involved in numerous cell processes associated with nucleosome and DNA-protein complexes, while scrutiny of the KEGG pathways indicated that LRV2$^-$ *L. major* cells undergo autophagy. We concluded that in comparison to LRV1, LRV2 is more integrated into the *Leishmania* metabolism and its ablation is more detrimental for the host cells. In addition, our analysis of the two viral genes, encoding the capsid and RNA-dependent RNA polymerase (RDRP), revealed them to be under the pressure of purifying selection, which was considerably stronger for the former gene across the whole tree. The selective pressure also differed between the LRV clades and correlated with the frequency of interspecific host switches.

10:30 (10 mins) - A25968 - Effect of delayed calf-dam separation on Cryptosporidium infection in dairy cattle.

Authors

OK Ingle[1]; S McPherson[2]; A Sinnott[2]; E Kennedy[2]; E Morgan[1];
[1] Queen's University Belfast, UK; [2] Teagasc, Ireland

Abstract

Cryptosporidiosis is a globally important diarrhoeal disease in both animals and people. Livestock, particularly young calves, are vulnerable to cryptosporidiosis and have been identified as a major source of environmental *Cryptosporidium* transmission. Calf vulnerability can also have a major impact on the cattle industry through the costs of diagnosis and treatment, production losses, poor future performance, and mortality. Cattle are commonly infected with four species of *Cryptosporidium*, with the zoonotic *Cryptosporidium parvum* often associated with clinical disease in calves. The effect of altered calf management, notably delayed calf removal from the dam, on *Cryptosporidium* infections has, however, not been investigated.

We collected faecal samples from individual cattle over several weeks during spring 2021 to determine whether delayed calf removal affects infection timing in three experimental groups: 1) Control group - dam and calf separated on the day of birth with no further contact, 2) Full-time group - dam and calf together 100% of the time until weaning at eight-weeks, 3) Part-time group - dam and calf together 50% of the time until weaning at eight-weeks. We used nested PCR amplification and further sequencing on extracted faecal DNA collected from both calf and dam samples. Infection with *Cryptosporidium* spp. was identified in all three groups at different levels, with a higher incidence in the control group. Improving our knowledge on *Cryptosporidium* transmission is essential to mitigate disease risk and to support a One Health approach to human and animal cryptosporidiosis.

Session 2 – Tue 22nd Mar 11:50 - 13.00

Day 2 – Combative Strategies: Vaccines *(Lecture Theatre K/018)*

22-March-2022, at 11:50 to 12:40

Chairs - Prof Gavin Wright & Dr Mohamed Osman

11:50 (10 mins) - A26154 - An invariant *Trypanosoma vivax* vaccine antigen eliciting protective immunity

Authors

D Autheman[3]; C Crosnier[3]; S Clare[2]; DA Goulding[2]; A jackson[1]; GJ Wright[3];
[1] University of Liverpool, UK; [2] Wellcome Sanger Institute, UK; [3] Department of Biology, University of York, UK, UK

Discussion

Animal African Trypanosomiasis (AAT) has a significant impact on animal agriculture in Sub-Saharan Africa by threatening the livelihood of farmers and food security in endemic countries, and is mainly caused by two species of African trypanosomes, *Trypanosoma congolense* and *T. vivax*. While vaccination would be an ideal solution to manage AAT, effective vaccines against African trypanosomes were considered unachievable due to the sophisticated system of antigenic variation employed by these pathogens to elude the host immune response. By using a systematic genome-led vaccinology approach and a murine model of *Trypanosoma vivax* infection, we have shown that protective invariant subunit vaccine antigens can be identified. Vaccination with a single recombinant protein comprising the extracellular region of a conserved cell surface protein localised to the flagellum membrane termed "invariant flagellum antigen from *T. vivax*" (IFX) induced long-lasting protection. Immunity was passively transferred with immune serum, and recombinant monoclonal antibodies to IFX could induce sterile protection and revealed multiple mechanisms of antibody-mediated immunity, including a major role for complement. Our discovery identifies a vaccine candidate for an important parasitic disease that has constrained the socioeconomic development of sub-Saharan African countries and provides evidence that highly protective vaccines against trypanosome infections can be achieved.

12:00 (10 mins) - A26145 - The *Leishmania donovani* ortholog of the GPI-anchor biosynthesis co-factor PBN1 is essential for host infection

Authors

AJ Roberts[2]; R Nagar[4]; C Brandt[3]; K Harcourt[3]; S Clare[3]; MA Ferguson[1]; GJ Wright[2];
[1] Wellcome Trust Centre for Anti-Infectives Research, School of Life Sciences, University of Dundee, UK; [2] Wellcome Sanger Institute, UK; [3] Sanger Institute, UK; [4] Wellcome Centre for Anti-Infectives Research, Division of Biological Chemistry and Drug Discovery, University of Dundee, Dundee, UK, UK

Discussion

Leishmania *donovani*, a kinetoplastid parasite for which no licenced vaccine is available. To identify potential vaccine candidates, we systematically identified genes encoding putative cell surface and secreted proteins essential for parasite viability and host infection. We identified a protein encoded by *LdBPK_061160* which, when ablated, resulted in a remarkable increase in parasite adhesion to tissue culture flasks. Here, we show that this phenotype is caused by the loss of glycosylphosphatidylinositol (GPI)-anchored surface molecules, and that *LdBPK_061160* encodes a non-catalytic component of the *L. donovani* GPI-mannosyltransferase I (GPI-MT I) complex. GPI-anchored surface molecules were rescued in the *LdBPK_061160* mutant by the ectopic expression of both human genes *PIG-X* and *PIG-M*, but neither gene could complement the phenotype alone. From further sequence comparisons, we conclude that *LdBPK_061160* is the functional orthologue of yeast *PBN1* and mammalian *PIG-X*, which encode the non-catalytic subunits of their respective GPI-MT I complexes and we assign *LdBPK_061160* as *LdPBN1*. The *LdPBN1* mutants could not establish a visceral infection in mice, a phenotype that was rescued by constitutive expression of *LdPBN1*. Although mice infected with the null mutant did not develop an infection, exposure to these parasites provided significant protection against subsequent infection with a virulent strain. In summary, we have identified the orthologue of the PBN1/PIG-X non-catalytic subunit of GPI-MT I in *Trypanosoma*tids, shown that it is essential for infection in a murine model of visceral leishmaniasis, and demonstrated that the *LdPBN1* mutant shows promise for the development of an attenuated live vaccine.

12:10 (10 mins) - A25916 - Laboratory evaluation of the miniature direct-on-blood PCR nucleic acid lateral flow immunoassay (mini-dbPCR-NALFIA), a simplified molecular diagnostic test for malaria

Laboratory evaluation of the miniature direct-on-blood PCR nucleic acid lateral flow immunoassay (mini-dbPCR-NALFIA), a simplified molecular diagnostic test for malaria*A25916*

Authors
NJ van Dijk[1]; Z Piets[1]; S Menting[1]; HD Schallig[1]; PF Mens[1];
[1] Amsterdam UMC, Netherlands
AbstractPoint-of-care diagnosis of malaria is currently based on microscopy and rapid diagnostic tests. However, both techniques have their constraints, including poor sensitivity for detection of low parasite densities. Hence, more accurate diagnostic tests for field use and routine clinical settings are warranted. The miniature direct-on-blood PCR nucleic acid lateral flow immunoassay (mini-dbPCR-NALFIA) is an innovative, easy-to-use molecular assay for the diagnosis of malaria in resource-limited settings. Unlike other simplified molecular methods, such as LAMP, the mini-dbPCR-NALFIA does not require DNA extraction and makes use of a handheld, portable thermal cycler that can be powered with a solar-charged power pack. Reading of results is done using a rapid lateral flow strip enabling differentiation of *Plasmodium falciparum* and non-*falciparum* malaria infections. Laboratory validation was performed to assess the performance of the mini-dbPCR-NALFIA for the diagnosis of pan-*Plasmodium* and *P. falciparum* infections in whole blood. Diagnostic accuracy was determined by testing a set of confirmed *Plasmodium*-positive blood samples from returned travellers (n=29), and confirmed *Plasmodium*-negative blood samples from returned travellers with suspected malaria (n=28), the Dutch Blood Bank (n=19) and intensive care patients at the Amsterdam University Medical Centers (n=16). The overall sensitivity and specificity of the assay were determined at 96.6% (95% CI, 82.2% - 99.9%) and 96.8% (95% CI, 89.0% - 99.6%). The limit of detection for *P. falciparum* was two parasites per microlitre of blood, as measured in dilution series of three *P. falciparum*-positive clinical blood samples. The repeatability of the assay was 92.0%. In conclusion, the mini-dbPCR-NALFIA is a sensitive, specific and relatively easy method for accurate detection of *Plasmodium* infections in whole blood and differentiation of *P. falciparum*. A phase-3 field trial is being performed to evaluate the potential implementation of this assay in malaria control programmes in both high- and low-transmission settings.

12:20 (10 mins) - A26152 - Vaccination is probably considered the most efficient tool for preventing current and future threats from parasitic diseases.

Authors
SJ Goodswen[1]; PJ Kennedy[2]; **JT Ellis**[1];
[1] School of Life Sciences, University of Technology Sydney, Australia; [2] School of Computer Science, Faculty of Engineering and Information Technology and the Australian Artificial Intelligence Institute, University of Technology Sydney, Australia
Discussion
Vaccination is probably considered the most efficient tool for preventing current and future threats from parasitic diseases. Immunogenic proteins sourced from parasites are often considered worthwhile vaccine components (subunits) and studied further for their vaccine potential. Very few parasite proteins have achieved proof of concept trials in animal models as vaccines and so be considered as being true vaccine candidates for future development. Publications on parasites with 'subunit vaccine' in their title have accumulated to thousands over the last three decades. However, there are possibly thousands more reporting immunogenicity results without mentioning 'subunit' and/or 'vaccine'. The exact number is unclear given the non-standardised keywords in publications. The aim of this study was to identify parasite proteins that induce a protective response in an animal model as reported in the scientific literature within the last 30 years using machine learning and natural language processing.Ultimately, we believe this data set has future value in the development of reverse vaccinology approaches for parasites.

12:30 (10 mins) - **Turbo Talk – 5 min each talk -**

A26144 - Systematic identification of genes encoding cell surface and secreted proteins that are essential for in vitro growth and infection in *Leishmania donovani*.

Authors
AJ Roberts[2]; H Ong[2]; S Clare[2]; C Brandt[2]; K Harcourt[2]; S Franssen[1]; J Cotton[1]; GJ Wright[2];
[1] Wellcome Trust Sanger Institute, UK; [2] Sanger Institute, UK
Discussion
Leishmaniasis is an infectious disease caused by protozoan parasites belonging to the genus *Leishmania* for which there are no approved human vaccines. Infections localise to different tissues in a species-specific manner with the visceral form of the disease caused by *Leishmania donovani* and *L. infantum* being the most deadly in humans. Although *Leishmania* spp. parasites are predominantly intracellular, the visceral disease can be prevented in dogs by vaccinating with a complex mixture of secreted products from cultures of *L. infantum* promastigotes. With the logic that extracellular parasite proteins make good subunit vaccine candidates because they are directly accessible to vaccine-elicited host antibodies, here we attempt to discover proteins that are essential for *in vitro* growth and host infection with the goal of identifying subunit vaccine candidates. Using an *in silico* analysis of the *Leishmania donovani* genome, we identified 92 genes encoding proteins that are predicted to be secreted or externally anchored to the parasite membrane by a single transmembrane region or a GPI anchor. By selecting a transgenic *L. donovani* parasite that expresses both luciferase and the Cas9 nuclease, we systematically attempted to target all 92 genes by CRISPR genome editing and identified four that were required for *in vitro* growth. For fifty-five genes, we infected cohorts of mice with each mutant parasite and by longitudinally quantifying parasitaemia with bioluminescent imaging, showed that nine genes had evidence of an attenuated infection although all ultimately established an infection. Finally, we expressed two genes as full-length soluble recombinant proteins and tested them as subunit vaccine candidates in a murine preclinical infection model. Both proteins elicited significant levels of protection against the uncontrolled development of a splenic infection warranting further investigation as subunit vaccine candidates against this deadly infectious tropical disease.

Day 2 - Parasite Molecular Genetics *(Lecture Theatre T/005)*
22-March-2022, at 11:50 to 12:35
Chairs - Dr Natalia Teles & Dr Pegine Walrad

11:50 (10 mins) - A26008 - The Helminth Antimicrobial Peptidome: a novel opportunity for parasite control?

Authors
A Irvine[1]; L Atkinson[1]; SA Huws[1]; A Mousley[1];
[1] Queen's University Belfast, UK

Discussion

Antimicrobial Peptides (AMPs) are ancient innate immune components that play key roles in defending against diverse microbial pathogens including bacteria, fungi and viruses. AMPs are ubiquitous in nature and act as the first line of defense against pathogens. Whilst AMPs are known to be critical for the survival of many invertebrates, the role of AMPs in helminth biology remains uncharacterised. Parasitic helminths, living in specific host niches, likely produce a diverse AMP arsenal with bioactivity against host microbiota. Indeed, many gastrointestinal helminths have been shown to modify the host gut microbiome, causing some aspects of disease pathology. Characterisation of helminth derived AMPs is key to deciphering their role in host-worm-microbiome interactions and may reveal novel targets for parasite control. In this study we characterised the pan-phylum AMP profiles of nematodes and flatworms through homology directed approaches. Here we reveal that phylum Nematoda is AMP-rich and -diverse, where >5000 genes encode AMPs. The data demonstrate that some nematode species e,g. *Trichinella/Trichuris* genera and the filarial parasites, possess a reduced AMP profile, whereas others encode more expanded AMPs cohorts. Using public transcriptomic datasets, we also show that nematode AMPs are transcriptionally active and are upregulated in key parasitic life stages. In contrast, flatworms do not appear to be as AMP-rich or -diverse. In this study we also employed machine learning tools to mine helminth genomic datasets for novel AMPs that are not detectable via homology-based approaches. These data reveal eight novel helminth AMPs which possess variable antibacterial properties against key gram-negative and gram-positive bacterial pathogens. This study provides valuable insights into the complexities of the Helminth Antimicrobial Peptidome and expose the system as a novel target for helminth control.

12:00 (10 mins) - A26017 - No more mutants: lack of canonical resistance mutations in *Ascaris* β-tubulin isotypes.

Authors

BP Jones[3]; AH van Vliet[3]; EJ Lacourse[1]; S Roose[2]; P Geldhof[2]; M Betson[3];

[1] Liverpool School of Tropical Medicine / UoL, UK; [2] Ghent University, Belgium; [3] University of Surrey, UK

Discussion

Ascaris lumbricoides and *Ascaris suum* are intestinal roundworms that infect humans and pigs respectively, and cause the disease known as ascariasis. Ascariasis affects half a billion people, with chronic infections leading to reduced growth and cognitive ability. Ascariasis affects pigs worldwide and can reduce production yields via decreased growth and condemnation of livers. The predominant drugs used to treat ascariasis are the benzimidazoles (BZ). Despite the farming industry using these drugs for decades, and BZ resistance occurring in numerous livestock helminths, there has been little work on the development of BZ resistance in pig ascariasis. Benzimidazoles work by interacting with β-tubulin, and the mutations causing resistance are known in ruminant nematodes. In most nematodes there are multiple β-tubulin isotypes. Only a few of these are expressed at high levels, with others being restricted to specialised cells or specific developmental stages. Recent work in other ascarids has shown that the canonical benzimidazole-resistance associated mutations, originally identified in ruminant nematodes, are not found in the β-tubulins of *Ascaridia* or *ParAscaris*, even in phenotypically resistant populations. We have conducted widespread screening of two *Ascaris* β-tubulin isotypes highly expressed in adult worms from *Ascaris* samples from pigs and humans across the world using deep amplicon sequencing techniques. We then used these data to study the population dynamics of *Ascaris* and highlighted the differences in diversity between the two isotypes, and the differences between genotypes found in each species. Screening of both human- and pig-derived *Ascaris* isolates found no evidence of any canonical BZ resistance mutations. Overall, there was a clear difference seen in the genetic diversity of each isotype with differences seen in the distribution of β-tubulin genotypes between human- and pig-derived isolates. This work suggests that resistance via the canonical β-tubulins mutations is not a problem in *Ascaris* and BZ are likely to be an effective means of control for the near future. However, alternative modes of resistance may emerge therefore continued monitoring of drug efficacy will be required.

12:10 (10 mins) - A26049 - Structure and selection: Insights into the evolution of host-parasite interactions of Tetraspanin 23 in *Schistosoma turkestanicum*.

Authors

C Eldridge[1]; A Juhasz[2]; G Majoros[6]; AM Emery[3]; RT Cook[5]; D Kidd[5]; SP Lawton[4];

[1] Kingston University, UK; [2] Liverpool School of Tropical Medicine, UK; [3] Natural History Museum, UK; [4] Scottish Rural University College, UK; [5] Kingston University London, UK; [6] University of Veterinary Medicine, Budapest, István u. 2, 1078 Hungary, Hungary

Discussion

Schistosoma turkestanicum is a widespread Asian veterinary parasite of domestic livestock and a causative agent of cercarial dermatitis in humans. Since its discovery in Hungary in 2010 the population of *S. turkestanicum* from the Gemenc region has been of particular interest as an undisturbed schistosome population that has escaped anthelmintic treatment with praziquantel. Currently there are no vaccines to protect against schistosomiasis and it has been hypothesised that high levels of parasite diversity in host populations is responsible for the current lack of protective efficacy in vaccine candidate trials. One promising vaccine candidate Tetraspanin-23, an immune evasion protein that has hypothesised non-immune IgG binding function in the large extracellular loop region (LEL), has so far had limited success in clinical trials. In this study protein function and site selection analyses were combined with inter-host population sequencing of the Tsp-23 gene to study host-parasite interactions and selective pressures that may influence population diversity at this locus. Tetraspanin-23 is highly conserved across the schistosome phylogeny and analysis of Tsp-23 orthologs predicted a single site under diversifying positive selection in the LEL region. Further sequencing of individual *S.turkestanicum* worms taken from five red deer hosts in the Gemenc, Hungary identified evidence of balancing selection at the same site. In addition structural and antigenicity score prediction identified this site to be associated with structural antigenic variation both between species and between individuals in host populations. Interestingly this site was predicted on the outer region of the LEL and not in the FC binding domain which was found to be entirely conserved in the population sample and predicted to be under purifying selection from between species selective site analysis. This suggests that host-parasite interactions with the Tsp-23 LEL region may be occurring at two levels, a conserved FC binding motif which functions to bind to the less variable host non-immune IgG FC region as well as diversifying positive selection acting on the exposed region of the LEL that is physically interacting with the more variable components of the host immune system. With the numerous strategies of immune modulation and evasion employed by schistosomes at the tegument surface this study on Tsp-23 suggests that protein function is likely an important factor in the assessment of vaccine candidate suitability for schistosomes.

12:50 (10 mins) - **Turbo Talks – 5 min each -**
22-March-2022, at 12:50 to 13:00

12:50 (5 mins) - A26001 - Molecular epidemiology and evolution of the Antigen Coding Gene (ACG) TSP-23, from the multi-host parasite *Schistosoma japonicum*

Authors
D Parsons[3]; AM Emery[1]; AJ Walker[3]; J Buxton[3]; F Allan[1]; JP Webster[2]; SP Lawton[4];
[1] Natural History Museum, UK; [2] Royal Veterinary College, University of London, UK; [3] Kingston University, UK; [4] Scottish Rural University College, UK

Abstract
The zoonotic nature of schistosomiasis in Southeast Asia, caused by *Schistosoma japonicum*, is arguably the most challenging factor hindering schistosomiasis control and elimination in the region. S. *japonicum* infects > 40 different mammalian hosts, however, the precise impact of wild and domesticated animals acting as reservoirs of infection, in maintaining transmission cycles, and promoting parasite diversification, remains unclear. Zoonotic pathogen interactions within hosts likely impact and alter pathogen evolution and may have implications for control and elimination goals. It is generally thought that parasite populations that infect multiple hosts can evolve novel, host-specific, Antigen Coding Genes (ACGs), and can become increasingly virulent when infecting different host species. In other pathogens, the diversification of ACGs has been implicated in alterations in the ability of the parasite to evade detection, and thus the ability of the host's immune system to recognise infection. This work focuses on the variation and genetic diversity of the tegumental-surface antigen tetraspanin-23 (SjTSP-23) within and between definitive host species. SjTSP-23 is integral to parasite survival in the definitive host and has been shown to interact directly with the definitive host's immune system via their Large Extracellular Loop (LEL) domains, and is thus regarded as a potential vaccine candidate.
Sequencing SjTSP-23-LEL domains from 81 FTA-archived S. *japonicum* miracidia from four definitive host species (human, dog, cat, and pig), has enabled the variation and frequency of parasite ACG genotypes from human and animal host populations, and the identification of shared antigen variants, to be investigated. It is expected that SjTSP-23-LEL sampled from human hosts would be more genetically diverse than those sampled from other host species, and that this increased variation will have structural, functional, and antibody-binding consequences, possibly leading to challenges with downstream vaccine design and development.
Through investigating the phylogenetic relationships and distribution of SjTSP-23-LEL haplotypes among definitive hosts, it was found that although humans contain the greatest frequency of unique, divergent haplotypes, there is significant sharing of antigen variants between hosts, with antigens predicted to be more genetically variable within host populations than between them. Furthermore, the selection pressures acting on sites within the LEL domain of SjTSP-23 from all hosts induced amino acid changes and antigenic variation in and around predicted antibody binding-sites, suggesting that host-derived selection pressures driving amino acid changes may serve as 'escape mutations', acting to reduce SjTSP-23-LEL antigenicity as an immune evasion mechanism.

12:55 (5 mins) - A26005 - The emergence of hybrid Fasciolids in central Vietnam.

Authors
P Best[3]; L Phuong[5]; J Campbell[5]; T Hien[5]; R Hanna[4]; J LaCourse[1]; K Cwiklinski[6]; JP Dalton[2]; MW Robinson[3];
[1] Liverpool School of Tropical Medicine, UK; [2] National University of Ireland, Galway (NUI Galway), Ireland; [3] Queens University Belfast, UK; [4] Agri-Food Biosciences Institute, Belfast, UK; [5] Oxford University Clinical Research Unit, Centre for Tropical Medicine, Ho Chi Minh City, Vietnam; [6] Institute of Infection, Veterinary and Ecological Sciences, University of Liverpool, UK

AbstractAlthough traditionally regarded as a disease of livestock, fasciolosis is now recognised as an emerging zoonotic disease with at least 2.4 million people infected worldwide. The major causative agents of fasciolosis are *Fasciola hepatica* and *F. gigantica*, in regions with temperate and tropical climates respectively, although hybrid forms have also emerged, notably in Southeast Asia. Whilst animal fasciolosis is endemic throughout this region, central Vietnam has emerged as a particular "hotspot" for human infection. In a bid to identify the predominant *Fasciola* species in this area, liver flukes were collected from cattle at abattoirs in the Phu Yen and Binh Dinh provinces and subjected to molecular speciation via multiplex-PCR and restriction fragment length polymorphism of phosphoenolpyruvate carboxykinase (*pepck*) and DNA polymerase delta (*pold*) genes respectively. Out of 326 adult flukes, 36 (11%) were identified as *F. gigantica* whilst 290 (89%) were identified as hybrid/intermediate forms. No flukes were identified as *F. hepatica* using these genetic markers. The reproductive strategy of the flukes was also assessed by aceto-orcein squash and histological observation. The majority of hybrid flukes were found to be aspermic, and thus likely to reproduce via parthenogenesis. However, a small number of hybrids did contain limited numbers of sperm but they displayed abnormal morphology and are unlikely to be viable. Our data show that hybrid *Fasciola* are the dominant form in the Phu Yen and Binh Dinh provinces. Given the absence of *F. hepatica* in this region, it is likely that hybrids were introduced to central Vietnam via import of infected livestock. However, further molecular analysis is required to confirm this.

Day 2 – Parasite Coinfections *(Lecture Theatre P/X001)*

22-March-2022, at 11:50 to 12:35
Chairs - Prof Colin Sutherland & Mr John Archer

11:50 (10 mins) - A25512 - *Ovale* malaria - unknown knowns, known unknowns and other mysteries

Authors
CJ Sutherland[1]; HP Fuehrer[2]; OJ Watson[1]; S Campino[1];
[1] London School of Hygiene & Tropical Medicine, UK; [2] Med Vet University, Wien, Austria

AbstractThe advent of molecular genetic and genomic analyses over the past 25 years has greatly increased our understanding of the parasites that cause *ovale* malaria in humans. It is now widely accepted that these comprise two distinct, non-recombining but fully sympatric species, a hypothesis first presented at the BSP Spring Meeting in Edinburgh in 2009. It has become clear also that, across the African continent, *ovale* malaria is ubiquitous yet under-reported, most frequently occurring as asymptomatic and/or mixed species infections with *Plasmodium*

falciparum. In these mixed infections, *ovale* parasites are invariably the minor species in terms of peripheral blood densities, and so easily over-looked. There is also a growing body of compelling data supporting the view that the two species differ in the latency period exhibited by putative quiescent hypnozoite forms residing in the liver following primary infection. Finally, new genomic studies utilising bespoke selective amplification protocols are beginning to shed comprehensive light on the degree of gene diversity within and between the dimorphic forms of *ovale* parasites. We present a summary of recent clinical, epidemiological and genomic studies of *Plasmodium ovale* curtisi and P. *ovale* wallikeri and, in response to the incontrovertable weight of evidence that these parasites represent two distinct species, propose a new binomial nomenclature.

12:00 (10 mins) - A25979 - How much does innate immunity impact rodent malaria infection dynamics? A meta-analytic approach

Authors

A Herbert Mainero[1]; S Reece[1]; T Kamiya[2];

[1] University of Edinburgh, Institute of Evolutionary Biology, UK; [2] Center for Interdisciplinary Research in Biology (CIRB), Collège de France, CNRS, INSERM, Université PSL, Paris, France, France

Abstract

Plasmodium parasites are intraerythrocytic parasites that elicit various immune responses in their vertebrate host. Molecular and cellular studies point to various mechanisms of innate immune regulation. However, how much and to what extent, innate immunity impacts the rate of replication of these parasites within their host is not fully understood. Here, we applied a meta-analytic approach to quantify the effects of innate immunity against rodent malaria and identified covariates that affect the extent of immune control. We used four malaria species of rodent hosts as models because over 4 decades of primary research provides an opportunity for a rigorous quantitative synthesis. We found a small significant effect across different *Plasmodium* spp. (Cohen's h 0.15 – 0.42) meaning experimental manipulation of innate immunity has a small, yet consistently impact on malaria infection dynamics. Additionally, we explored the role of specific components in the innate immune system and various methodological approaches. We found that the type of manipulation (methodological approach) impacts the mean magnitude of innate immune interventions, and this could be helpful to design experiments because you could maximise the chances of observing even small differences. We also show that the pace at which innate immunity affects parasite growth is parasite-specific and covaries with functional innate immune components (receptors, regulators, and effectors). Finally, the direction in specific mechanisms in the literature reported effects to be beneficial or detrimental, similarly, we show no directionality in the effects. We conclude that in rodent *Plasmodium* spp. host innate immune components impact parasite early replication, but not all components have the same impact; for example, we could not detect a difference in magnitude between pro-inflammatory and anti-inflammatory interventions in P. chabaudi. Our findings could be combined with knowledge about disease severity to generate strategies that have the biggest impact on parasite growth with little immunopathological effects.

12:10 (10 mins) - A26019 - Integrating ecological perspectives into anthelmintic resistance management

Authors

S Brown[2]; A Morrison[1]; J McIntyre[2]; E Devaney[2]; B Mable[2]; D Bartley[1]; R Laing[2];

[1] Moredun Research institute, UK; [2] Institute of Biodiversity, Animal Health and Comparative Medicine, University of Glasgow, UK, UK

Abstract

Despite the large body of research on anthelmintic resistance (AR) in parasitic nematodes of livestock, the interactions between AR and parasite ecology are poorly understood. The relative costs and benefits of resistance could be influenced both by environment and parasite life history traits. This project aims to investigate how interactions between environment, life history traits and ivermectin treatment could influence AR in *Haemonchus contortus,* an economically important gastrointestinal nematode of sheep.

Donor sheep were orally infected with either a multidrug resistant (MHco18) or susceptible (MHco3) isolate of *H. contortus.* Based on faecal egg counts, MHco3 infections reached patency earlier than MHco18. Sheep were treated with a standard dose of ivermectin (0.2mg/kg; MHco18), a low dose of ivermectin (0.012mg/kg; MHco3) or left as untreated controls. Adult worms were collected at post-mortem. Adult worm body size showed no significant difference associated with isolate or treatment, but the effect of the donor sheep was significant, implying that host-parasite interactions impact size. Faecal eggs were collected pre- and post-ivermectin treatment, with egg hatch and larval development assays conducted across a range of temperatures. There was no significant effect of isolate or treatment on egg hatch and larval development success. However, over the course of the experiment egg hatch and larval development rates varied at the temperature limits, suggesting time to patency can influence larval survival.

The presentation will highlight the complex interaction of factors influencing adult size and larval survival, highlighting possible mechanisms by which ecological factors could be utilised in AR management.

12:20 (10 mins) - A26089 - Co-Infections with Malaria, Urinary Schistomiasis, Typhoid Fever and Hepatitis B Virus Among School Children in Ogbese, Ise-Ekiti, South-Western Nigeria

Charles Ologunde

TBD

12:50 (10 mins) - **Turbo Talks – 5 min each talk -**
22-March-2022, at 12:50 to 13:00

12:50 (5 mins) - A26165 - First observation of Parasitic viruses in *Trichomonas gallinae*

Authors

[1]Dalal Ardan, [2]Marlene Benchimol, [3]Sally Warring, [3]Neil Hall, [1] Diana Bell, [1]Kevin M. Tyler

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Discussion

Trichomonas gallinae is a single cell protozoan parasite that causes avian trichomonosis in a diverse array of birds especially pigeons and doves. Numerous studies show that viruses can reside within protozoan pathogens and contribute towards pathogen virulence and the closely related *Trichomonas vaginalis,* can be infected with a double-stranded RNA (dsRNA) virus which enhances its pathogenicity. However, the presence of *T. gallinae* has remained hitherto undiscovered. We screened a cryobank containing hundreds of UK passerine columbid and raptor isolates of *T. gallinae* with a wide range of genotypes for the presence or RNA virus. An initial Agarose gel-based screen of extracted RNA from different isolates of *T. gallinae* revealed an extra band of RNA in two isolates (C3 and C10). This band of RNA is consistent with the size of viral dsRNA and indicative of viral infection in *T. gallinae*. The presence of dsRNA was further verified using immune fluorescence monoclonal antibodies J2 specific to dsRNA viruses. Both these isolates were from infections which lacked demonstrable pathology and which were considered to be avirulent strains. To characterize the effect of the virus on *Trichomonas gallinae* we compared these strains with two virulent strains strains which lacked virus namely (A1 and C4). We observed that virus infected cells of *T. gallinae* were smaller and grew less well than non-infecting cells. Moreover, using (scanning and transmission) electron microscopic methods, we found evidence of plasma membrane disruption and granular structures which may be virus budding from the cell surface. Using negative staining of supernatants, we found icosahedral structures which may be virions. Using RNA transcriptomics, we were able to show expression of viral RNAs with 70% RNA identity to Trichomonas Virus 1. Overall our study offers new insight into parasitic pathogenesis of*T. gallinae* which in contrast to *Trichomonas vaginalis* correlates with low virulence of strains. It is to be hoped that knowledge of the virus may provide a route to novel intervention strategies for avian trichomonosis in birds.

12:55 (5 mins) - A26088 - Malaria Co – Infection with Urinary Schistosomiasis, Typhoid Fever, Hepatitis B Virus, and Human Immunodeficiency (HIV) Virus in three Local government areas of Ekiti-State, South Western Nigeria

Charles Ologunde

TBD

Session 3 – Tue 22nd Mar 14:00 - 15.20 for '*Parasite Cell Biology I*' and '*Wild Parasitology: into the field';* **14:30 – 15:20** for *'Diversity in Science I: Conversations toward inclusion and equity'.*

Day 2 – 14:00 - 15.20 Parasite Cell Biology I *(Lecture Theatre T/005)*
22-March-2022, at 14:00 to 15:00
Chairs - Dr Eden Ramalho Ferreira & Dr Rachel Neish

14:00 (20 mins) - A26229 - Breaking (down) the chain – structure/function studies of apicomplexan respiratory complexes

Author
Dr Lilach Sheiner
University of Glasgow

Discussion

The mitochondrial respiratory chain is of central importance for energy and metabolism in most eukaryotes. This is true also for the apicomplexan parasites that cause diseases like toxoplasmosis and malaria, where in fact the respiratory chain is a known target for clinically used and newly developing drugs. Yes, while the respiratory complexes are well studied in mammalians, the hosts of apicomplexans, their composition or structure/function relationship was largely unknown. Our team has been using biochemical, proteomics, genetics, and structural biology methods to discover, validate and understand the function of novel components and features of the respiratory complexes in apicomplexan. We primarily use *Toxoplasma gondii* as a model system, while novel insights are being validated in *Plasmodium falciparum*. My talk will describe the discovery of 20 new components of the *Toxoplasma* respiratory chain complexes, the validation of new components of complex II, III and V, and the structural insights of how some of the new components contribute to corresponding new functions, with focus on multimeric complex formations.

14:20 (10 mins) - A25963 - A map of PFR proteins and dissection of their functions in motility and assembly of the *Trypanosoma brucei* flagellum.

Authors
H Berti Gabriel[1]; **JD Sunter**[1];
[1] Department of Biological and Medical Sciences, Oxford Brookes University, Oxford, UK

AbstractAlongside the microtubule axoneme in many flagellated cells there are additional protein structures such as the paraflagellar rod (PFR) of *T. brucei*. The PFR has an intricate structure composed of three distinct domains (inner, middle and outer) that runs next to the axoneme within the flagellum and is important for flagellar beat regulation and cell motility. However, the specific contribution of each of the PFR domains to motility is unknown. The TrypTag project highlighted the complexity of the PFR with 146 proteins found in this structure. We determined by bioinformatics analysis, conservation patterns of these proteins across Euglenozoa and identified two major conserved sets of PFR proteins. The first set was conserved across Euglenozoa; whereas, the second set was conserved but not present in organisms with a reduced PFR, such as *Angomonas deanei*. A semi-automated analysis of TrypTag images, measuring the distance between the kinetoplast and the start of the PFR signal revealed a discontinuous start to the PFR and predicted the domain to which a PFR protein localised. These predictions were confirmed for a subset of 15 proteins by analysing the distance to known flagellum proteins. We analysed the function of these 15 proteins in cell motility and depletion of four of them disrupted motility, reduced cell growth and led to a 'blob' forming at the flagellum tip. The motility phenotypes were only observed in proteins present in the inner and very outer PFR domains. However, for three of these proteins despite changes in motility, there were no obvious changes to the PFR ultrastructure, suggesting these proteins are not required for PFR assembly. These results suggest that the

motility function of the PFR can be separated from its assembly and that there are domains and sub-structures of the PFR with a specific role in motility. This is an important step in assigning specific functions to the individual PFR domains.

14:30 (10 mins) - A26118 - TbHD82 is important for the maintenance of nucleotide homeostasis in *Trypanosoma brucei*

Authors

P AntequeraVM Castillo-Acosta[1]; M Yague-Capilla[1]; C Bosch-Navarrete[1]; LM Ruiz-Perez[1]; D gonzalez-pacanowska[1];

[1] Instituto de Parasitologia y Biomedicina 'Lopez-Neyra', CSIC. Granada, Spain; [2] Instituto de Parasitología y Biomedicina , Spain

Discussion

DNA integrity, replication and repair depend on an appropriate homeostasis of dNTPs pool. Degradation of dNTPs takes place through catabolic pathways, where enzymes such as 5'-nucleotidases, nucleoside phosphorylases, and deaminases mediate the break-down of dNTPs to deoxynucleosides or nucleobases. dNTP break-down products can be reutilized in the salvage pathway or are transported to other organelles in order to maintain intracellular homeostasis. In humans, one of the most relevant proteins in the control of dNTP levels is sterile alpha motif and histidine-aspartic acid domain-containing protein 1 (SAMHD1), a triphosphohydrolase that catalyzes the degradation of the four canonical dNTPs to their corresponding nucleosides. SAM domains are known to function as protein interaction or RNA-binding modules, whereas several characterized HD domain proteins have been shown to possess phosphodiesterase, phosphatase, dGTP triphosphatase, or nuclease activities. The purified HD of human SAMHD1 has been shown to possess dGTP-stimulated dNTP triphosphohydrolase activity. Two unique SAMHD1 putative orthologs have been identified in *Trypanosoma brucei:* TbHD82 and TbHD52, according to their molecular mass. While TbHD52 is mitochondrial, TbHD82 is a nuclear protein and both lack a canonical SAM domain. The quantification of dNTPs levels in bloodstream TbHD82-knockout cells revealed a significant accumulation of dATP and dCTP, which is reverted with the expression of an ectopic copy of the gene. Additionally, TbHD82-knockout cells present increased phosphorylation of *Trypanosoma*l histone H2A indicating the occurrence of DNA damage as a consequence of dNTP imbalance. Finally, *in vivo* analysis in mice evidenced that TbHD82 deficient cells are less infective that parental cells, which could be linked to a need for adequate dNTP levels during the early stages of infection. In conclusion, we suggest that TbHD82 plays a central role in dNTP homeostasis in *Trypanosoma brucei* and that while not essential for *in vitro* growth, its function is important for the progression of infection in animal models of the disease.

Day 2 - 14:00 - 15.20 Wild Parasitology: into the field *(Lecture Theatre P/X001)*

22-March-2022, at 14:00 to 15:00

Chair - Prof Matthew Thomas

14:00 (20 mins) - A26227 - The ecology of helminth infection and immunity in a wild rodent model

Dr Amy Pedersen

University of Edinburgh

Despite great concern about the global health threat of infectious diseases in humans and domestic animals, we still don't have a clear understanding about how ecological heterogeneity determines infection burdens, disease, transmission, or how to successfully control infections in variable populations. Our reliance on highly controlled, laboratory models may underlie some of our failures to adequately manage disease burdens in real-world settings, where individuals compete for food, mates and space; endure seasonal and spatial environmental variability; and are exposed to a vast array of parasites and pathogens. We have established a hybrid wild/laboratory rodent model system in order to investigate the causes and consequences of this ecological heterogeneity for host-parasite interactions. Specifically, this work focuses on Heligomosoides polygyrus and its natural host, the wood mice (*Apodemus sylvaticus*), and addresses the following questions: (i) what determines susceptibility and resistance to H. polygyrus in the wild? and (ii) how does nutritional availability and coinfection with other gastrointestinal parasites impact H. polygyrus infection and immunity. Our results highlight how pairing both the lab and natural setting provides a unique and powerful opportunity to understand the causes and consequences of ecological heterogeneity on infection, immunity and disease control.

14:20 (10 mins) - A26148 - What you eat it is what you get: parasite-reptile interactions in a human-dominated landscape

Authors

S Guerrero Sanchez[1]; L Frias[2];

[1] Institute of Borneo Studies, University College Sabah Foundation, Malaysia; [2] Asian School of the Environment, Nanyang Technological University, Singapore

Discussion

Agricultural expansion in Southeast Asia has converted most natural landscapes into mosaics of forest interspersed with plantations, dominated by the presence of generalist species that benefit from resource predictability. Dietary shifts resulting from these adaptations can alter the structure of host parasite communities and ultimately impact the fitness and survival of their populations. Our study focuses on the Asian water monitor lizard (*Varanus salvator*), one of the largest predators in Asian wetlands, as a model species to understand the health consequences of dietary shifts in an oil palm dominated landscape in Sabah, Malaysian Borneo. We evaluated the influence of diet diversity on the parasite species richness and prevalence of lizards living in forest patches and oil palm plantation estates. We observed that lizards feeding on less diverse diets, mostly dominated by rodents in oil palm plantations, hosted less diverse parasite communities with overall higher parasite prevalence. However, parasites with complex transmission modes, such as cestodes and trematodes, were more prevalent in forested areas. By working with a widely distributed generalist carnivore as model species, we outline how human-dominated landscapes can pose a negative effect on wildlife species whose diet may be altered by the influence of human activities, such as farming and extensive agriculture.

14:30 (10 mins) - A25671 - Toxocara sp. egg contamination of allotment-grown vegetables in the UK: A pilot study

Authors
S Healy[2]; M Betson[2]; E Morgan[1]; JM Prada[2];
[1] Queen's University Belfast, UK; [2] University of Surrey, UK
Abstract
Toxocara canis and *Toxocara cati* are zoonotic roundworm parasites of dogs, cats and foxes. These definitive hosts pass eggs in their faeces, which contaminate the environment and can subsequently be ingested via soil or contaminated vegetables. In humans, infection with *Toxocara* can have serious health implications, including brain disorders and blindness. This proof-of-concept study investigated the presence of *Toxocara* eggs on vegetables sampled from allotments (community gardens) in southern England between May 2021-July 2021. This is the first time that vegetable contamination with *Toxocara* eggs has been investigated in the UK, or in allotments anywhere.
Sixteen allotment sites participated, providing 82 vegetable samples for testing. For eight of the vegetable samples, sufficient soil from where the sample originated was additionally available in the sample bag for analysis. Study participants also completed an anonymous questionnaire on observed visits to the sites by definitive hosts of *Toxocara*. Comparison of egg recovery methods was performed using lettuce samples spiked with a series of *Toxocara* egg concentrations, with sedimentation and centrifugal concentration retrieving the highest number of eggs. This method was subsequently used to analyse the vegetable samples collected from the allotment sites.
Two lettuce samples tested positive for *Toxocara* eggs, and one sample among the eight soil samples tested was also positive. Questionnaire data revealed that 88% of respondents had seen a definitive host species or the faeces of a definitive host on their site. This is the first study demonstrating the presence of *Toxocara* eggs on vegetables grown in the UK, as well as within the soil where these vegetables originated, highlighting the biosecurity risks in allotment sites.
This pilot study provides a method for assessment of *Toxocara* eggs on vegetable produce and paves the way for larger-scale investigations of *Toxocara* egg contamination of field-grown vegetables.

14:40 (10 mins) - A25590 - Fish Faecal Xenomonitoring as a potential tool for schistosomiasis transmission monitoring

Authors
Z Bartonicek[1]; J Dvorak[4]; F Allan[2]; AM Emery[1]; JJ Day[3]; BL Webster[1];
[1] Natural History Museum, UK; [2] University of St Andrews, UK; [3] University College London, UK; [4] Center of Infectious Animal Diseases, Faculty of Agrobiology, Food and Natural Resources, Czech University of Life Sciences, Czech Republic
Abstract
Background:
Appropriate surveillance methods are needed to detect and monitor schistosomiasis transmission, particularly as ongoing interventions decrease the disease prevalence. *Schistosoma mansoni* is a parasitic trematode causing intestinal schistosomiasis in humans. It has a complex lifecycle depending on two zooplanktonic larvae (cercariae and miracidia) and an intermediate freshwater snail host. Traditionally, environmental transmission monitoring is achieved via malacological surveys where snails are collected and screened for emerging *Schistosoma* cercariae or the parasite DNA. Although informative, these methods can be laborious and insensitive. Other molecular methods, such as detecting environmental DNA (eDNA), could be a more efficient and sensitive tool. Therefore, this study develops a new, potentially more informative, DNA-based approach relying on detection of the *S. mansoni* DNA in the faeces of natural predators – Fish Faecal Xenomonitoring (FFX). To develop this method, we used juvenile cichlid Nile tilapia (*Oreochromis niloticus*) that has previously been demonstrated to consume cercariae and miracidia of *S. mansoni*.
Methods:
We conducted multiple laboratory and microcosm fish feeding experiments under variable conditions, offering 1-900 *S. mansoni* cercariae to juvenile *O. niloticus* fish (SL 2-5 cm). The fish faecal samples were analysed using a multiplex FFX-qPCR assay, targeting *S. mansoni* and *O. niloticus* DNA (internal control), developed and tested as part of this study. In lab feeding experiments, we analysed the effects of fish size, the number of offered larvae, or the availability of alternative fish food sources on the presence of *S. mansoni* DNA in the fish faeces after consumption. Additionally, we tested the gut passage times of the *S. mansoni* DNA and the method's sensitivity.
Results:
S. mansoni DNA was detected in the faeces of 67.5% of the fish offered ≥300 cercariae. When fish were offered cercariae in microcosms and pooled in groups of five during faeces collection, the overall sample positivity increased to 83.3%. Positive detection was achieved when fish consumed ≥1 cercaria, although sensitivity decreased with fewer cercariae consumed. The gut passage experiments showed that the *S. mansoni* DNA is expelled within 24 hours of consumption, with a peak excretion between 12-21 hours after feeding. The analytical sensitivity of the FFX-qPCR, which targets the *S. mansoni* 16S mitochondrial DNA region, was 100% for ≥10 DNA copies/reaction and 71% for 1 DNA copy per reaction.
Conclusions:
Our findings show that *S. mansoni* DNA, from consumed cercariae, is detectable in fish faeces within 24 hours after consumption and that fish readily consume cercariae even in the presence of other food sources. This FFX approach could provide a new, complementary method for schistosomiasis transmission monitoring in endemic settings. *S. mansoni* and *O. niloticus* are co-occurring across Sub-Saharan Africa. Our study may thus not only have implications for xenomonitoring approaches of *Schistosoma* spp., but also indicates that *O. niloticus* may play a role in the biological control of *S. mansoni* and other aquatic parasites. Further studies in field settings will enable the evaluation of the FFX methodology for the detection of *S. mansoni* transmission.

14:50 (10 mins) - A26135 - The role of sylvatic rodents in transmission of *Toxocara canis* in NE Poland

Authors
M Krupińska[3]; D Antolová[5]; K Tołkacz[4]; A Goll[3]; J Nowicka[3]; A Bajer[2]; JM Behnke[1]; M Grzybek[3];
[1] University of Nottingham, UK; [2] University of Warsaw, Poland; [3] Medical University of Gdansk, Poland; [4] Institute of Biochemistry and Biophysics, PAS, Warsaw, Poland, Poland; [5] Slovak Academy of Sciences, Košice, Slovakia
Discussion
Toxocara canis is a cosmopolitan nematode parasite of carnivores, notably canids, both wild and domestic, which act as definitive hosts. Eggs consumed by paratenic hosts, such as rodents, cannot develop further into the adult stage, but infective larvae can persist in their tissue for an

extended time, constituting a reservoir of *T. canis* for canids. Small mammals are suspected as contributing to the dissemination of *T. canis* and helping with the survival of the parasite during periods when there is a temporary absence of suitable definitive hosts. They can also play a role as an indicator of environmental contamination with Toxocara. While the primary aim of the current study was the assessment of seroprevalence of Toxocara spp. infections in wild rodents in Poland, we also explored the role of intrinsic (sex, age) and extrinsic factors (study site, year of study) influencing the dynamics of this infection. We trapped 577 rodents belonging to four species (*Myodes glareolus, Microtus arvalis, Microtus agrestis, Alexandromys oeconomus*) in northern eastern Poland. Blood was collected during parasitological examination, and serum was frozen at -72°C until further analyses. A bespoke enzyme-linked immunosorbent assay was used to detect antibodies against *T. canis*. We found T. canis antibodies in the sera of all four rodent species with an overall seroprevalence of 2.8% [1.9-4.1]. There was a significant difference in seroprevalence between vole species ($\chi 2/3> = 29.4$; p= 0.001) with the grassland species (M. arvalis, M. agrestis, and A. oeconomus) showing 16-fold higher seroprevalence (15.7% [8.7-25.9]) than the forest dwelling, M. glareolus (0.98% [0.5-1.8]). We hypothesise that seroprevalence of T. canis differs between forest and grassland rodents because of higher contamination of grasslands by domestic dogs and wild canids. Our results underline the need for wide biomonitoring of both types of ecosystems to assess the role of rodents in spreading zoonotic nematodes. This research was funded through the 2018–2019 BiodivERsA joint call for research proposals, under the BiodivERsA3 ERA-Net COFUND program; the funding organizations ANR (France), DFG (Germany), EPA (Ireland), FWO (Belgium), and NCN (Poland). JN, MG and AG were supported by the National Science Centre, Poland, under the BiodivERsA3 programme (2019/31/Z/NZ8/04028). MK was supported by the National Science Centre, Poland, under the Preludium BIS programme (2020/39/O/NZ6/01777).

Day 2 – 14:30 – 15:20 Diversity in Science I: Conversations toward inclusion and equity *(Lecture Theatre K/018)*
22-March-2022, at 14:30 to 15:20
Chairs - Dr Giulia Bandini & Dr Sabrina Absalon

14:30 (20 mins) - A26222 - BIPOC in Parasitology: an anti-racist, equitable, and inclusive community of parasitologists.

Dr Sabrina Absalon
Indiana University

Summer 2020 in the United States of America was marked by nationwide protests voicing the fight for racial equity and racial justice. The long-overdue awakening to systemic racism in American society triggered the self-assessment of funding agencies, scientific institutions, research groups, and scientists. As a result, we witnessed a significant rise in Diversity, Equity, Inclusion (D.E.I.) statements, plans, and articles in the following months. In addition, 2020 introduced a wide range of grassroots-style, often trainee-organized groups, that arose without any formal structure or support to fill the much-needed gap in support of scientists from historically marginalized communities. For instance, virtual symposia and trending hashtags, including #BlackinMicrobiology, #BlackinImmunology, and our group #BIPOCinParasitology, were prominent. These organizations have stepped up to fill a very critical void within our field. Notably, groups historically excluded from academic research and administrative positions have often been initiated to support their communities (rather than 'top-down, these initiatives are 'ground up' in nature). BIPOC in Parasitology (BiP) was conceived after the 2020 "Black in Microbiology Week," and our community has grown to 122 members. We are a community of Black, Indigenous, and People of Color studying parasites. In partnership with our non-BIPOC colleagues in the field of parasitology, our goal is to provide a safe, professional, collaborative, and collegial space to engage with each other. Furthermore, BiP strives to promote the contributions of BIPOC parasitologists and cultivate an anti-racist, equitable, and inclusive environment within the broader parasitology community. Our signature event, "Parasite Hour with BiP," is a monthly research seminar series that provides a space for BIPOC parasitologists to share their journeys in science, showcase their contribution to parasitology, and provide invaluable advice to future parasitologists.
We are looking forward to providing more opportunities for professional development and community building in years to come.

14:50 (20 mins) - A26335 - Building Bridges using the Universal Language of Science

Dr Omar Harb
University of Pennsylvania

Sadly, human history is marred with conflict. Indeed, most manuscripts punctuate their retelling of a history with battles, wars and human strife. The Middle East and Africa have not been spared from this and in recent history exemplify the long-term consequences of colonialism. While people engage in conflict and the ensuing cultural, political, and national separatism, pathogens do not. In addition, social and physical boarders may inhibit cross-cultural interactions but do not prevent the spread of disease. Many of the diseases endemic to this area of the world are parasitic in nature resulting in devastating illnesses such as Malaria, Leishmaniasis and Schistosomiasis. To effectively combat these diseases, and to maximize the existing knowledge of parasite biology, there is a need to establish effective lines of scientific communication that is divorced from the political infrastructure. Established in 2016, The Middle East Biology of Parasitism (MeBoP) course was conceived of as a venue where young scientists from across the region can interact with each other while also being exposed to cutting edge knowledge and skills. The course runs over a two-week period during which participants attend morning lecture by eminent parasitologists, engage in laboratory practical experience in the afternoons and get to spend social time together in the evenings. MeBoP has to main **Objective**s:

Session 4 – Tue 22nd Mar 15:50 - 17.00
Day 2 – Diversity in Science I: Conversations toward inclusion and equity *(Lecture Theatre K/018)*
22-March-2022, at 16:20 to 16:25
Chairs - Dr Giulia Bandini & Dr Sabrina Absalon

15:50 (20 mins) - A26350 - Building a globally inclusive and equitable parasitology conference and community

Prof Deepali Ravel
Harvard T. H. Chan School of Public Health

Discussion

Over the last two years, increased attention to inequities in research funding and access as well as increased comfort with virtual engagement have prompted many conferences and professional societies to make changes to become more inclusive and equitable. This session will focus on such efforts in the context of the annual Molecular Parasitology Meeting (MPM), which is traditionally hosted in Massachusetts in the US. Through support from the Burroughs Wellcome Fund, we are redesigning MPM as a hybrid meeting with the goal of ensuring that both in-person and virtual attendees have opportunities to share research and build mentoring and collaboration networks through the meeting. I'll share lessons learned from our 2020 virtual meeting and 2021 hybrid meeting as well as our process and plans for the future. I'll also talk about our additional work focused on creating new virtual between-meeting science, professional development, and mentoring activities for all trainees in the community.

Day 2 - Parasite Cell Biology I *(Lecture Theatre T/005)*
22-March-2022, at 15:50 to 16:25
Chairs - Prof Derrick Robinson & Dr Eden Ramalho Ferreira

15:50 (10 mins) - A26177 - Divergent metabolism between *Trypanosoma congolense* and *T. brucei* underlies differential sensitivity to metabolic inhibitors

Authors
PC Steketee[6]; E Dickie[4]; K CrouchG Awuah-Mensah[2]; E Paxton[6]; HP De KoningC Gadelha[3]; B Wickstead[5]; MP Barrett[7]; LJ Morrison[6];
[1] University of Glasgow , UK; [2] University of Nottingham, UK; [3] Queen's Medical Centre, University of Nottingham, Nottingham, UK; [4] Wellcome Centre for Molecular Parasitology, Institute of Infection, Immunity and Inflammation, University of Glasgow, UK; [5] Queen's Medical Centre, University of Nottingham, UK; [6] The Roslin Institute, University of Edinburgh, UK; [7] Wellcome Centre for Integrative Parasitology, Institute of Infection, Immunity and Inflammation, University of Glasgow, UK

Discussion

Animal African Trypanosomiasis (AAT) is a debilitating livestock disease prevalent across sub-Saharan Africa. One of the main causes is the protozoan parasite *Trypanosoma congolense*. Whilst the closely related *T. brucei* has been studied for decades, there is a major paucity of knowledge regarding the biology of *T. congolense*. In this study, we have used a combination of omics technologies and novel genetic tools to characterise core metabolism in *T. congolense* mammalian-infective bloodstream-form parasites. In addition, we have tested whether metabolic differences compared to *T. brucei* impact upon sensitivity to metabolic inhibition. Like the bloodstream stage of *T. brucei*, glycolysis plays a major part in *T. congolense* energy metabolism. However, the rate of glucose uptake is significantly lower in bloodstream stage *T. congolense*. Furthermore, the primary glycolytic endpoints are succinate, malate and acetate. Transcriptomics analysis showed higher levels of transcripts associated with the mitochondrial pyruvate dehydrogenase complex, acetate generation, and the glycosomal succinate shunt in *T. congolense*, compared to *T. brucei*. To validate the metabolic similarities and differences, both species were treated with metabolic inhibitors, Strikingly, *T. congolense* exhibited significant resistance to inhibitors of fatty acid synthesis, including a 780-fold higher EC_{50} for the lipase and fatty acid synthase inhibitor Orlistat, compared to *T. brucei*. These data highlight that bloodstream form *T. congolense* diverges from *T. brucei* in key areas of metabolism, with several features that are intermediate between bloodstream- and insect-stage *T. brucei*. These results have implications for drug development, mechanisms of drug resistance and host-pathogen interactions.

16:00 (10 mins) - A26153 - Bioinformatic and functional characterisation of Trichomonas tenax GH30 homologues that potentially target fungal glycans

Authors
L Mpeyako[1]; M Schindler[1]; E Lowe[1]; NS Jakubovics[2]; R Hirt[1];
[1] Newcastle University Biosciences Institute, Newcastle University, UK; [2] School of Dental Sciences, Newcastle University, UK, UK

Discussion

Trichomonas tenax is an oral anaerobic/microaerophilic flagellated protist that is reported to be associated with periodontitis and pulmonary trichomoniasis. Microbial dysbiosis, a taxonomic and functional shift in the oral microbial population towards a more pathogenic state, drives an excessive host inflammatory response and leads to periodontitis, an irreversible damage of the tissues around the teeth that is associated with aging but that can affect people of any age, including children. Periodontitis affects over 47% of adults aged ≥30 years and 5-15% of adults worldwide. It remains the leading cause of edentulism and has a major impact on the quality of life through the loss of tooth function and associated issues with eating and speaking. Difficulties with eating often lead to nutritional deficits in older people. Periodontitis is also increasingly recognised to be associated with systemic conditions including arthritis and dementia/Alzheimer.
T. tenax is positively associated with periodontitis and could contribute to the dysbiotic state associated with periodontitis through its interactions with the oral microbiota, including bacteria, *Mycoplasma* species, fungi and viruses. However, the precise mechanism underlying tissue damage in this pathology is still poorly understood.
We have identified a list of conserved genes of bacterial origins encoding candidate enzymes targeting bacterial peptidoglycans (PG) or fungal cell walls in the species *T. vaginalis* and *T. gallinae*, which are closely related to *T. tenax*. This suggests that targeting of bacteria and fungi is essential for these parasites to thrive in various mucosal surfaces from different animal hosts ranging from birds to mammals including pet animals (dogs and cats) and humans. Using co-culture experiments we were able to show that *T. tenax* dynamically interacts with *Candida albicans*, both yeast forms, which are phagocytosed by the parasite, and hyphae forms. This is similar to published data for *T. vaginalis*. Notably, targeting of bacterial and fungal cell walls can liberate pro-inflammatory molecules, hence *Trichomonas* species could contribute to the damaging inflammation indirectly through the targeting of the cell walls of members the microbiota. BT_3312; an endo-1,6-β-glucanase belonging to the Glycosyl Hydrolase family 30 subfamily 3 (GH30_3) in *Bacteroides thetaiotaomicron* was shown to degrade the 1,6-β-glucans of yeast cell walls. Homologues of BT_3312 were identified among *T. vaginalis* and *T. gallinae* annotated genes and sequence comparisons indicated that these share

all the key residues from BT_3312 that mediates its specificity to fungal 1,6-β-glucans. Here we identified a GH30_3 homologue in *T. tenax* (TtGH30) and investigated its phylogenetic relationship with trichomonads and bacterial homologues. As the TtGH30 sequence also possess all key residues of the BT_3312 1,6-β-glucanase we expressed it and purified a recombinant version of the enzyme and tested its activity against 1,6-β-glucans. These comparative investigations highlight a lateral gene transfer into the Trichomonas lineage from a bacterial origin that encode functional enzymes and that might contribute to target fungal cell wall *in vivo* and by doing so help the parasites to thrive at mucosal surfaces, including *T. tenax* in the periodontal pocket. By doing so it could also contribute at liberating the strongly pro-inflammatory fungal 1,3-β-glucans. Dual transcriptomics of *T. tenax-C. albicans* co-culture are currently being processed to gain a more global perspective on the molecular basis of *T. tenax* targeting of the fungi, including the range of enzymes potentially targeting fungal cell wall and the parasite metabolism more generally.

16:10 (10 mins) - A25984 - Deletion of the P21 gene triggers changes in the invasion and replication of *Trypanosoma cruzi*

Authors

T Teixeira[2]; MA Chiurillo[3]; N Lander[3]; CC Rodrigues[4]; TS Onofre[2]; ER Ferreira[1]; CM Yonamine[2]; JG Santos[4]; RA Mortara[2]; CV Silva[4]; JF Silveira[2];

[1] University of York, UK; [2] Federal University of Sao Paulo, Brazil; [3] University of Cincinnati, United States; [4] Federal University of Uberlandia, Brazil

Discussion

P21 is a protein encoded by a single-copy gene and no orthologs are found in other *Trypanosoma*tids. Previous studies demonstrated the role of P21 during the infection of *Trypanosoma cruzi.* P21 operates as a signal transducer molecule, triggering a signaling cascade, which results in the alteration of the actin cytoskeleton of host cell, increasing the internalization of parasites. In addition, *T. cruzi* infected mice treated with recombinant P21 have shown increasing in leukocytes chemotaxis and fibrosis, but reduced angiogenesis and replication of intracellular amastigotes, indicating the role of P21 in pathogenesis of Chagas disease. However, the mechanisms underlying the role of P21 remain poorly understood. In this study, we generated P21 knockout parasites using CRISPR/Cas9 and analysed the phenotypic effects of the deletion of this gene. Our results showed that ablation of P21 reduced the growth rate of epimastigotes evaluated for 14 days. Furthermore, P21 knockout epimastigotes showed an increase in the length of the G1 phase and reduction in the S phase, resulting in a delay on cell cycle progression. Invasion assays performed with metacyclic trypomastigotes revealed that P21 knockout impairs parasites ability to invade HeLa cells when compared to the wild-type control. In contrast, intracellular replication rate of amastigotes is increased in P21 knockout parasites, observed after 72 hours of infection. Taken together, our data reveals the involvement of P21 during the different life stages of *T. cruzi*, demonstrating its importance throughout the parasite life cycle. Support: FAPESP 2016/15000-4, FAPESP 2019/05049-4, FAPEMIG APQ-00971-17, CNPq and CAPES.

Turbo Talks – 5 min each talk –

16:40 (5 mins) - A26184 - Role of RDK2 and its interacting protein kinases in Leishmania *mexicana* differentiation.

Authors

RP Neish[1]; V Geoghegan[1]; K Newling[1]; K Hogg[1]; J Smith[1]; J Mottram[1];

[1] University of York, UK

Discussion

The protein kinase RDK2 (Repressor of differentiation 2) has been proposed to be involved in differentiation of *L. mexicana*. To investigate this, we generated an *RDK2* null mutant (Δ*rdk2*)using CRISPR-Cas9 genome editing. In *T. brucei,* RNA inference of RDK2 promotes differentiation from bloodstream stumpy form to procyclic form. In *L. mexicana in vitro*, the Δ*rdk2* cells were able to undergo normal differentiation from promastigote to amastigote. To attempt to quantitatively measure this differentiation using flow cytometry, we generated a mNeonGreen-dynein line where the newly formed flagellum can be a measured fluorescently. We found Δ*rdk2* was able to transform from amastigote to promastigote in vitro, albeit at a slower rate compared to the control, during the initial phase of differentiation.

The Δ*rdk2* cell line was screened within a protein kinase knockout library using BarSeq to identify genes with a loss of fitness in differentiation from lesion-derived amastigotes to promastigotes. Like the *in vitro* assays, Δ*rdk2* had a delayed differentiation from amastigote to promastigote. We generated an N-terminal MYC tagged RDK2 and carried out immunoprecipitation and mass spectrometry to identify potential interacting proteins. From this we identified other protein kinases that may be involved in the same pathway as RDK2. From these screens, we are attempting to unravel the role of RDK2 and its interactome in *L. mexicana* differentiation.

16:45 (5 mins) - A26115 - Characterising Heat Shock in *Trypanosoma congolense*

Authors

M Aelmans[1]; C Dewar[1]; M Urbaniak[1];

[1] Division of Biomedical and Life Sciences, Faculty of Health and Medicine, Lancaster University, UK

Discussion

Trypanosoma congolense causes significant economic burden across Sub-Saharan Africa, as it is the etiological agent of Animal African Trypanosomiasis (AAT), a wasting disease affecting cattle which currently has no pharmaceutical treatment. *T. congolense* is a close relative of *Trypanosoma brucei*, they co-infect the same hosts so have been exposed to similar evolutionary selective pressures, and it is expected they will show similarities in host interactions. While *T. brucei* is a well-studied model organism, very little experimental work has been performed using *T. congolense* as tools have only recently been developed for genetic manipulation, but now is the time to use them to investigate the survival and infection mechanisms of the parasite. One of the major symptoms of AAT is a high fever which *T. congolense* responds to by eliciting the heat shock (HS) response, an important virulence factor which allows the parasite to survive in the host. The aim of this project is to characterise the *T. congolense* HS response, as understanding the mechanisms involved could pave the way for discovering novel drug targets in this parasite.
A bioinformatic analysis looking into the conservation of proteins, phosphorylation sites and active sites involved in the HS response between *T. brucei* and *T. congolense* has been conducted. Key proteins involved in the HS response are being tagged and analysed with immunofluorescence microscopy to see if they localise in the same way in *T. brucei* and *T. congolense* during HS. Flow cytometry is being used to characterise cell cycle

progression through HS. RNAi knockdown of ZC3H11 will be performed to see its effect on the cells ability to survive HS. Preliminary results of this work will be shown.

16:50 (5 mins) - A26134 - *Galba truncatula* and Helminths, the Importance of Microbes

Authors

P McCann[4]; C McFarland[3]; J Megaw[3]; C Cantacessi[2]; G Rinaldi[1]; G Gobert[3];

[1] Wellcome Trust Sanger Institute, UK; [2] Department of Veterinary Medicine, University of Cambridge, UK; [3] Queens University Belfast, UK; [4] Queen's Univeristy Belfast, UK

Discussion

Liver fluke (*Fasciola hepatica*) and rumen fluke (*Calicophoron daubneyi*) are endemic in the UK. Liver fluke is estimated to cost the UK agriculture industry approximately £300 million per year, particularly due to lamb deaths and liver condemnations. Rumen fluke is fatal in severe infections and only one flukicide, oxyclozanide, has been shown to effectively reduce rumen fluke burdens. The desirable potency of triclabendazole has stimulated its overuse for liver fluke control resulting in widespread anthelmintic resistance. Therefore, there is an urgent need to develop new control strategies for fasciolosis.

The microbiome is defined as the combined genetic material of the microorganisms inhabiting a particular environment. A host's microbiome is known to play a key role in many aspects of health and disease, including susceptibility to parasitic infection. While most microbiome studies have focused on the mammalian hosts of helminths, their intermediate hosts should also be considered. Recent research of mosquitoes infected with *Wolbachia* shows they cannot transmit dengue fever. As a result, efforts to control dengue fever are being focused on bacterial symbionts that can aid disease elimination. The interaction between the snail microbiome and life stages of parasitic trematodes residing in their intermediate hosts has not been investigated to any large extent.

This project is in its earliest phase. We aim to profile the microbiome of snail species harbouring active helminth infections. We will compare host stress markers, and investigate the role played by bacterial symbionts. Finally, the functional roles played by snail microbiota will be considered using classical microbiological methods.

16:55 (5 mins) - A26105 - Expression and characterization of a mitochondrial fucosyltransferase from *Trypanosoma cruzi* and use of monoxenous parasite Crithidia fasciculata as an enzymatic source for synthesis of radioactive GDP-Fucose.

Authors

JC Paredes-Franco[1]; MA J Ferguson[1];

[1] Wellcome Centre for Anti-Infectives Research, Division of Biological Chemistry and Drug Discovery, University of Dundee, Dundee, UK, UK

Discussion

For all eukaryotes, surface glycoproteins and glycolipids are made in the secretory pathway (i.e., the endoplasmic reticulum and the Golgi apparatus) and, therefore, this is where the vast majority of glycosyltransferases reside.

However, previous research has identified a specific glycosyltransferase enzyme, a fucosyltransferase, in the single mitochondrion of *Trypanosoma brucei* and of *Leishmania major* (Bandini G, et al., 2021, eLife; Guo H, et al., 2021, PNAS). This enzyme, called FUT1, not only has an unusual mitochondrial localization, but also it is essential for both parasites. The presence of FUT1 suggests that some novel type of glycoprotein or glycolipid glycosylation occurs in the mitochondria of *Trypanosoma*tid parasites. Nevertheless, nothing is known about the orthologous enzyme from *Trypanosoma cruzi* (TcFUT1) and so we decided to perform expression and characterization studies with it.

Based on the work done with TbFUT1 (Bandini G, et al., 2021, eLife), initial expression and purification attempts were done using different *E. coli* strains as expression systems. When eventually recombinant TcFUT1 was purified, although in low yields, we performed fucosyltransferase activity assays involving a panel of synthetic glycosidic acceptor substrates and radioactive guanosine diphosphate fucose (GDP-[^{3}H]Fuc) as the donor substrate. However, no activity was detected, and new trials were done using eukaryotic Expi293F cells as the expression system, since HEK293-derived cells have been reported as useful for expressing many human glycosyltransferases. At the same time, we have established a procedure to synthesize our own GDP-[^{3}H]Fuc based on previous knowledge from our group on using the monoxenous parasite *Crithidia fasciculata* as an enzyme source for the generation of this and other GDP nucleotide sugars (Schneider P, et al., 1995, Biochem. J.; Mengeling B J, et al., 1999, Analytical Biochem.). Combining our home-made radioactive donor, and recombinant TcFUT1 obtained from the culture medium of our eukaryotic expression system, we aim to define the substrate specificity of this fucosyltransferase and potentially characterize the fine chemical structure of its product(s).

Day 2 – Wild Parasitology: into the field *(Lecture Theatre P/X001)*

22-March-2022, at 15:50 to 16:40

Chair - Prof Matthew Thomas

15:50 (10 mins) - A25678 - Parasitic Platyhelminthes of Sparus aurata (Sparidae, Teleosteans): first report of the Digeneans Macvicaria obovata Molin, 1859 and Allopodocotyle pedicellata Stossich, 1887 off Algerian coast.

Authors

FZZedam[1]; A Boukadoum[1]; F Tazerouti[1];

[1] University of Sciences and Technology Houari Boumdienne, Faculty of Biological Sciences, Laborratory Biodiversity and Environnement : Interactions and Genomes, BP 32, El Alia Bab Ezzouer, Algiers, Algeria

Abstract

Sparus aurata (sea bream) is a Sparidae marine teleost of great economic interest. Frequently harbor various groups of parasites, particularly Platyhelminthes. Three species of Digenea were identified from the intestine, the collected Digeneas belongs to the same family Opecoelidae

Ozaki, 1925 represented by *Allopodocotyle pedicellata* Stossich, 1887; *Macvicaria obovata* Molin, 1859 and *Macvicaria maillardi* Bartoli, Bray & Gibson, 1989.
Allopodocotyle pedicellata Stossich, 1887 and *Macvicaria obovata* Molin, 1859 were reported for the first time in the Algerian coast.
This study has allowed us to investigate the biodiversity of the parasitic Digenea in Teleostean fish.
Keywords: Biodiversity– Parasites- Digenea– Platyhelminthes- Metazoans- Teleost fish - Algerian coast.

16:00 (10 mins) - A26129 - Development of a Recombinase Polymerase Amplification (RPA) for the detection of *Schistosoma mansoni* infection

Authors
S Mesquita[2]; CT Fonseca[2]; E Lugli[1]; RL Caldeira[2]; B Webster[1];
[1] Natural History Museum, UK; [2] Oswaldo Cruz Foundation, Brazil

Abstract
Schistosomiasis is a parasitic disease associated to poverty and low sanitation condition. It is estimated that nearly 240 million people are infected in the world and over 25 million people live in high-risk areas in the Americas. Almost 1.6 million people are infected in Brazil, allopatric for *Schistosoma mansoni*, that causes the intestinal schistosomiasis and is transmitted by freshwater snails from the genus *Biomphalaria*. Among the strategies currently available, sensitive, and specific diagnostic tests followed by the timely treatment of the population, is a strategy for the prevention of potential complications together with reducing transmission. Also, accurate mapping and monitoring of snail breeding sites to detect active transmission areas are needed. The Recombinase Polymerase Amplification (RPA) is an isothermal method that has been piloted for urogenital schistosomiasis, and its simplicity, low resource needs, and speed have highlighted its utility for use in the field at the point-of-care. This research aims to evaluate the performance of RPA to support the diagnosis of infection with *S. mansoni* in humans and snails. For that, primers and probe targeting a repeated region of *S. mansoni* mitochondrial DNA have been designed and are being tested for the standardisation of the assay. Each RPA reaction is being performed using half volume of the rehydrated pellet from the TwistAmp exo kits, with fluorescent signal detection via a portable reader. The specificity of the assay was accessed using gDNA of *Schistosoma* species, and other helminths that are co-endemic with *S. mansoni*. The analytical sensitivity was determined using serial dilutions of *S. mansoni* gDNA. The use of fresh and frozen crude *S. mansoni* eggs was also tested as well as urine and stool samples spiked with gDNA and eggs, respectively, and infected and noninfected *Biomphalaria* snails. The developed assay, named SmMIT-RPA, presented promising results being specific to *S. mansoni*, sensitive enough to detect a single egg and up to 1fg of DNA. Positive results were also obtained from urine samples spiked with 0.01pg of *S. mansoni* DNA, stool samples spiked with *S. mansoni* eggs, and prepatent experimentally infected *Biomphalaria* snails. Further analysis will be conducted in order to optimise and validate the use of the SmMIT-RPA for *S. mansoni* diagnosis using clinical and field samples collected in low-endemic areas of the state of Minas Gerais (MG), Brazil. Results from this work will be later compared to data from reference tests previously applied to the same samples. It is expected that SmMIT-RPA will allow a more accurate and rapid diagnostic in order to improve the decision-making process for a more appropriate destination of public funding aiming the elimination of schistosomiasis as a public health problem.

A25513 - Development of a computer visualization program for the taxonomic identification of Free Living Amoebas (FLAs)

Authors
MI Issa[1]; DC Oliveira[3]; NK Bellini[2]; **OH Thiemann**[1];
[1] University of São Paulo, Brazil; [2] INSTITUTO BUTANTAN, CELL CYCLE SPECIAL LAB, SÃO PAULO, Brazil; [3] Instituto Federal Goiano, Brazil

Abstract
Free Living Amoebas (FLA) are unicellular eukaryotic microorganisms belonging to the Kingdom Protista widely found in nature, but which, despite most of them living freely in the environment, they have some facultative opportunistic parasite genera, such as *Naegleria* spp.
and *Acanthamoeba* spp. causing infections associated with the Central Nervous System (CNS) whose rapid progression leads patients to death in a short period of time. Morphological analyzes carried out today are not optimized, based on microscopic identification with the taxonomic guide Page (1988), which hinders the efficient classification of the samples. Therefore, we are developing a computer visualization program based on taxonomic keys described on the Page´s guide to be available to the community for future taxonomic identification of FLA. The morphological characteristics of the different amoebas are ordered for the creation of a software for multidimensional data visualization based on parallel coordinates, called Page's Visualization Tool (PVT).
In parallel to the PVT development, we have investigated the presence of FLA species along 5 sites of the Rio Monjolinho in the city of São Carlos, SP employing molecular techniques and phylogenetic classification of the sequenced rDNA target gene fragments.
The limnological analysis identified the impact of urbanization on the water course. The coupled molecular and morphological based methodology enabled us to obtain an appropriate FLA description in which pathogenic (*Naegleria*, *Acanthamoeba* and *Vermamoeba*) and non pathogenic (*Filamoeba*, *Vanella*, and *Stenamoeba*) genera were isolated and characterized. Thus, it has been possible to obtain an environmental overview of the dispersion of these microorganisms in the region, contributing, together with the PVT, to the optimization and complementation of future research on the subject.

16:20 (10 mins) - A26011 - The role of sylvatic rodents in transmission of *Toxoplasma gondii* in NE Poland

Authors
J Nowicka[2]; B Biernat[2]; D Antolová[3]; K Tołkacz[4]; A Goll[2]; M Krupińska[2]; A Bajer[5]; JM Behnke[1]; M Grzybek[2];
[1] University of Nottingham, United Kingdom, UK; [2] Medical University of Gdansk, Poland; [3] Slovak Academy of Sciences, Košice, Slovakia, Poland; [4] Institute of Biochemistry and Biophysics, PAS, Warsaw, Poland, Poland; [5] University of Warsaw, Warsaw, Poland, Poland

Abstract
There is currently considerable interest in understanding the transmission of pathogens and the range of different variables that influence infection dynamics. Wild rodents pose a particular threat to human communities because they constitute the most abundant and diversified group of all living mammals. *Toxoplasma gondii* is an intracellular Apicomplexan parasite with a broad range of intermediate hosts, including humans and rodents. Rodents are considered to be reservoirs of infection for their predators that include cats, pigs and dogs. We conducted a multi-site, long-

term study of T. *gondii* in northeastern Poland. Our **Objective**s were to monitor the seroprevalence of T. *gondii* in the four abundant vole species found in the forests and meadows of the region (Myodes glareolus, Microtus arvalis, Microtus agrestis, Alexandromys oeconomus) and to assess variation in seroprevalence attributable to both intrinsic and extrinsic factors that were quantified. A bespoke enzyme-linked immunosorbent assay was used to detect antibodies against T. *gondii*. We detected T. *gondii* antibodies in the sera of all four rodent species with an overall seroprevalence of 5.5% (3.6% for M. glareolus and 20% for other vole species). Seroprevalence in bank voles varied significantly between host age classes, increasing with host age, and between the sexes, with higher levels recorded in female compared with male voles.
Since T. *gondii* seroprevalence was significantly higher in rodents trapped in meadows, we aimed to assess the prevalence of T. *gondii* in these animals. We trapped 24 rodents comprising Microtus arvalis, Apodemus agrarius and Apodemus sylvaticus in September 2021. We extracted DNA from their brains and femoris muscles. Using PCR and nested-PCR reactions we detected T. *gondii* in 2 samples with an overall prevalence of 8.3% (1.5-26.7). Our results confirm that sylvatic rodents play a role as intermediate hosts and reservoirs of T. *gondii*. Taken together, these results contribute to our understanding of the distribution and abundance of T. *gondii* in rodents in Poland and establish that all the four species sampled in the current study are potential reservoir hosts of T. *gondii*.
This research was funded through the 2018–2019 BiodivERsA joint call for research proposals, under the BiodivERsA3 ERA-Net COFUND program; the funding organizations ANR (France), DFG (Germany), EPA (Ireland), FWO (Belgium), and NCN (Poland). JN, MG and AG were supported by the National Science Centre, Poland, under the BiodivERsA3 programme (2019/31/Z/NZ8/04028). MN was supported by the National Science Centre, Poland, under the Preludium BIS programme (2020/39/O/NZ6/01777).

16:30 (10 mins) - A26174 - Genetic diversity and population structure analysis of various Taenia multiceps isolates from definitive and intermediate hosts worldwide

Authors
I Abbas[1]; E El-Alfy[1];
[1] Faculty of Veterinary Medicine, Mansoura University, Egypt

Discussion

Taenia multiceps circulates in a two-host life cycle including various canids, but primarily dogs, as definitive hosts and a wide range of intermediate hosts, particularly sheep and goats, which carry the larval stage (coenurus cerebralis) in the CNS. However, the coenuri have been occasionally observed outside the CNS (e.g., subcutaneous tissues), particularly in goats. Based on a limited number of analyzed isolates, genetic variants and/or strains of T. multiceps have been proposed for extra-CNS coenuri, and in a specific animal species from a certain geographical region (e.g., cattle from Turkey). The present study provides the first comprehensive genetic analysis for all published T. multiceps nucleotide sequences from various definitive and intermediate hosts. A total of 233 partial cox1 nucleotide sequences that represented 442 T. multiceps isolates (cerebral and non-cerebral) deposited in GenBank were collected after consulting the NCBI website. The retrieved sequences were aligned using the software MEGA6, which was also used to construct the phylogenetic trees. Various genetic indices as well as intra- and inter-population variations were calculated using the software DnaSp6. Haplotype networks were constructed using PopArt1.7. The analyzed sequences displayed 51 haplotypes with 59-point mutations, of which 28 were parsimony informative. High haplotype (0.859 ± 0.00009 SD) and low nucleotide (0.00915 ± 0.00032 SD) diversities were detected. Significant negative values for the neutrality indices; Tajima`s D and Fu`s Fs tests were also determined, which are suggestive for the recent population expansion. Six geographic populations (China, Iran, Turkey, Greece, Italy and Egypt) were defined suitable for population analyses. Five major haplotypes were noticed; 2 of them as well as their associated minor haplotypes were common in China, Iran and Turkey. The 3 other haplotypes and their adjoining minor haplotypes were prevalent in Italy and Egypt. This pattern of haplotype distribution could be related to some factors that probably include the geographical neighborship; for example, Iran and Turkey. However, haplotypes from Greece do not follow this assumption and circulate worldwide, which was then confirmed after population structure analyses; the calculated pairwise distance and gene flow values for the Greek population in comparison to the other populations were comparable. Moreover, the haplotype networks revealed no specific patterns for distribution of the detected haplotypes among various infected hosts, and the wild life shared the same haplotypes with sheep and goats. In addition, our analyses underline the genetic analogy between the cerebral and non-cerebral coenuri isolates. Overall, the existence of genetic variants in T. multiceps is highlighted, but due to the limited number of sequenced isolates of these variants, the relationship of these variants to a specific phenotypic character (e.g., non-cerebral locations of the coenuri) cannot be confirmed.

Day 3 – Wed 23rd Mar 2022 -

Session 5 – Wed 23rd Mar 10:00 - 11.20
Day 3 – Mathematical Modelling of Parasites *(Lecture Theatre K/018)*
23-March-2022, at 10:00 to 11:00
Chair - Dr Laurence Wilson

10:00 (20 mins) - A26161 - Can Mass Drug Administration of Moxidectin Accelerate Onchocerciasis Elimination in Africa?

Authors
MG Basáñez[1]; P Milton[1]; K Kura[1]; J Hamley[1]; M Walker[2];
[1] Imperial College London, UK; [2] Royal Veterinary College, University of London, UK

Discussion

The World Health Organization's 2021–2030 Roadmap on Neglected Tropical Diseases has proposed that elimination (interruption) of transmission (EoT) be achieved and verified in 12 onchocerciasis-endemic countries by 2030. In Africa, epidemiological and modelling studies have suggested that EoT may not be achieved with annual mass drug administration (MDA) of ivermectin alone, particularly in areas of high initial endemicity (characterised by high vector biting rates). Phase II and III clinical trials demonstrated moxidectin's superiority to ivermectin regarding clearance of *Onchocerca volvulus* skin microfilariae. Not only was the amicrofilaridermic period following moxidectin treatment longer, but also there was less inter-individual variation in treatment responses compared to ivermectin. In 2018, moxidectin was approved by the US FDA for treatment

of *O. volvulus* infection in those aged ≥12 years. We used the stochastic EPIONCHO-IBM model to compare the probabilities of reaching EoT between ivermectin and moxidectin MDA for a range of (from hypo- to hyper-) endemicity levels, treatment frequency and adherence, with and without inter-individual response variation. We examined assumptions regarding: a) the modelling of vector biting rates to simulate pre-intervention endemicity levels; b) drugs' efficacy and their effects on *O. volvulus* reproductive biology; c) starting with moxidectin from the outset vs. switching to moxidectin during current ivermectin MDA programmes; d) treating the population aged ≥5 years with moxidectin vs. treating the 5–11-year olds with ivermectin and those aged ≥12 years with moxidectin; and e) random vs. systematic variation in inter-individual treatment responses. EPIONCHO-IBM's projections indicated that annual moxidectin MDA would be roughly equivalent to 6-monthly ivermectin MDA, but 6-monthly moxidectin was the best strategy, leading to substantial reductions in the number of years necessary to achieve EoT, and the only strategy that would help achieve EoT in highly endemic areas. To improve modelling projections, it is crucial to collect further data on: i) patterns of treatment adherence in endemic communities; i) the effect of moxidectin on adult *O. volvulus* worms; iii) the potential prophylactic effect of moxidectin; and iv) patterns of inter-individual variation in treatment responses over subsequent treatment rounds.

10:20 (10 mins) - A26168 - Spatial determinants of water contact in Mayuge, Uganda: a cross-sectional study exploring exposure risk for Schistosomiasis

Authors
M Eyre[1]; G Chami[1];
[1] NDPH, University of Oxford, UK

Discussion

Transmission of *Schistosoma* spp., blood flukes that cause schistosomiasis, is often geographically focal due to fine-scale spatial variation in exposure and contamination. Consequently, understanding the spatial determinants of water contact and individual and population-level factors is important for identifying high risk individuals and areas.

We conducted a cross-sectional household survey of 3,491 households across 17 villages in Mayuge district, Uganda in 2013. We collected sociodemographic and water contact information comprising the number of weekly household visits to Lake Victoria, duration, and activity type. For each of three leisure, occupational and domestic water contact outcomes we fitted a zero-inflated negative binomial generalised linear model to week-hours of water contact. To explore the spatial structure of water contact we then fitted a binomial geostatistical model to a binary lake water contact outcome.

We identified risk factors for high levels of water contact and found that there was residual spatial correlation in binary lake water use after controlling for these variables. These findings provide insights into the determinants of water contact in Mayuge and potential drivers of spatial heterogeneity in *Schistosoma* transmission.

10:30 (10 mins) - A25921 - Public health policy pillars for the sustainable elimination of zoonotic schistosomiasis

Authors
E Janoušková[2]; J ClarkO Kajero[2]; S AlonsoPH LambertonM Betson[2]; JM Prada[2];
[1] University of Glasgow , UK; [2] University of Surrey, UK

AbstractSchistosomiasis is a parasitic disease acquired through contact with contaminated freshwater. The definitive hosts are terrestrial mammals, including humans, with some *Schistosoma* species crossing the animal-human boundary through zoonotic transmission. An estimated 12 million people live at risk of zoonotic schistosomiasis caused by *Schistosoma japonicum* and *Schistosoma mekongi*, largely in the World Health Organization's Western Pacific Region and in Indonesia. Mathematical models have played a vital role in our understanding of the biology, transmission, and impact of intervention strategies, however, these have mostly focused on non-zoonotic *Schistosoma* species. Whilst these non-zoonotic-based models capture some aspects of zoonotic schistosomiasis transmission dynamics, the commonly-used frameworks are yet to adequately capture the complex epi-ecology of multi-host zoonotic transmission. However, overcoming these knowledge gaps goes beyond transmission dynamics modelling. To improve model utility and enhance zoonotic schistosomiasis control programmes, we highlight three pillars that we believe are vital to sustainable interventions at the implementation (community) and policy-level, and discuss the pillars in the context of a One-Health approach, recognising the interconnection between humans, animals and their shared environment. These pillars are: (1) human and animal epi-ecological understanding; (2) economic considerations (such as treatment costs and animal losses); and (3) sociological understanding, including inter- and intra-human and animal interactions. These pillars must be built on a strong foundation of trust, support and commitment of stakeholders and involved institutions.

Day 3 - Host:Parasite interactions : Tissue Tropism *(Lecture Theatre T/005)*
23-March-2022, at 10:00 to 11:00
Chairs - Dr Cecile Crosnier & Dr James Hewitson

10:00 (20 mins) - A26230 - Manipulation of inflammasome activation and host cell death by Leishmania parasites

Prof Dario Zamboni
University São Paulo, Ribeirao Preto

Discussion

Inflammasomes are multimeric protein complexes that assemble in the cytosol of many types of cells, including innate immune cells. The inflammasomes can be activated in response to infection or in response to stress signals that induce damage in the host cell membranes. These platforms trigger inflammatory processes, cell death, and the control of microbial replication. Many inflammasomes have been described so far, including NLRP3, NAIP/NLRC4, caspase-11, and AIM2. The ligand for NLRP3 is still unidentified, but the efflux of K+ is essential for NLRP3 activation. By contrast, inflammasomes, such as those composed of NAIP/NLRC4, caspase-11, and AIM2, can be activated by bacterial flagellin, LPS, and dsDNA. The knowledge of inflammasome biology in response to bacteria has advanced tremendously in the last decade, while inflammasome

activation in response to intracellular parasites, remains less explored. Leishmania infection triggers activation of the NLRP3 inflammasome in macrophages for restriction of intracellular parasite replication. Accordingly, Leishmania can dampen NLRP3 activation as an evasion strategy. In vivo, the NLRP3 inflammasome can promote parasite clearance, but the failure to eliminate parasites in the tissues together with sustained inflammasome activation can promote IL-1β-mediated disease pathology. Data to be presented will highlight recent data regarding activation and consequences of inflammasome activation in response to Leishmania, a process that effectively impacts the development of the disease in humans.

10:20 (10 mins) - A26133 - Incomplete Recruitment of Protective T Cells Is Associated with *Trypanosoma cruzi* Persistence in the Mouse Colon

Authors
MC Taylor[2]; MD Lewis[1]; A Ward[1]; JM Kelly[1];
[1] London School of Hygiene and Tropical Medicine, UK; [2] London School of Hygiene and tropical Medicine (LSHTM), UK

Discussion

Trypanosoma cruzi is the etiological agent of Chagas disease. Following T cell-mediated suppression of acute-phase infection, this intracellular eukaryotic pathogen persists long-term in a limited subset of tissues at extremely low levels. The reasons for this tissue-specific chronicity are not understood. Using a dual bioluminescent-fluorescent reporter strain and highly sensitive tissue imaging that allows experimental infections to be monitored at single-cell resolution, we undertook a systematic analysis of the immunological microenvironments of rare parasitized cells in the mouse colon, a key site of persistence. We demonstrate that incomplete recruitment of T cells to a subset of colonic infection foci permits the occurrence of repeated cycles of intracellular parasite replication and differentiation to motile trypomastigotes at a frequency sufficient to perpetuate chronic infections. The lifelong persistence of parasites in this tissue site continues despite the presence, at a systemic level, of a highly effective T cell response. Overcoming this low-level dynamic host-parasite equilibrium represents a major challenge for vaccine development.

10:30 (10 mins) - A26013 - *Plasmodium* sporozoites homing to the liver: exploring the interplay between parasite and host factors

Authors
M Sá[3]; DM Costa[3]; AR Teixeira[3]; I Loureiro[3]; P Formaglio[1]; S Golba[1]; D Klug[2]; F Frischknecht[2]; R Amino[1]; J Tavares[3];
[1] Institut Pasteur, Paris, France; [2] Universität Heidelberg, Germany; [3] IBMC/i3S, Portugal

Discussion

Hematogenous dissemination followed by tissue tropism is a characteristic of the infectious process of many pathogens. Once in the blood of the mammalian host, *Plasmodium* sporozoites specifically arrest in the hepatic sinusoids before infecting the liver. Two adhesive proteins present on the surface of sporozoites – circumsporozoite protein (CSP) and thrombospondin-related anonymous protein (TRAP) – have been proposed to specifically interact with highly sulphated heparan-sulphate proteoglycans expressed by hepatic cells, but evidence of their importance for the selectivity of the interactions between sporozoites and the hepatic sinusoids is lacking. Noteworthy, the model explaining the homing of sporozoites to the liver is based on indirect experiments interpreting the binding of recombinant proteins to liver cells or the outcome of a liver infection by sporozoites. To investigate the role of TRAP and its critical role for the sporozoite gliding motility in the homing to the liver, we combined live imaging techniques in mice and reverse genetics. We found that *trap* knockout (*trap*-) sporozoites are defective in homing to the liver and the deletion of the cytosolic domain that links host ligands that bind to the ectodomain of TRAP to the actomyosin motor (TRAP_CTD-) also failed to home to the liver. The deletion of this domain renders sporozoites incapable of gliding but does not alter the surface localization of the protein. As these mutant parasite lines do not invade the salivary glands of the mosquito and accumulate in the hemolymph, two additional mutants where changes in the adhesive I domain of TRAP previously reported to do not prevent sporozoites from entering the salivary glands, but impact gliding motility were also investigated. These include a mutant in which TRAP I domain was: i) replaced by the I-domain of MIC2 from *Toxoplasma gondii* (TRAP_MIC2), and ii) specifically mutated to shift the charge around the metal ion-dependent adhesion site (MIDAS) motif (TRAP_RevCh). Homing experiments demonstrate that TRAP_RevCh (and to a less extent TRAP_MIC2) sporozoites have a defect in homing to the liver, contrarily to control sporozoites, indicating that the gliding motility might contribute to the retention of sporozoites in the liver sinusoids. Further experiments are now being conducted to consolidate the contribution of sporozoite motility in this process.

Day 3 – Parasite:Vector Biology *(Lecture Theatre P/X001)*
23-March-2022, at 10:00 to 11:00
Chairs - Dr Álvaro Acosta-Serrano & Dr Poppy Lambert

10:00 (20 mins) - A26228 - A sense of direction: how African trypanosomes orient themselves in their insect host

Prof Isabel Roditi
University of Bern

Discussion

Parasite life cycles are traditionally depicted as static pictures of different life-cycle stages in various tissues. These depictions overlook the question of how parasites home in on these tissues and then move from one to the next, particularly when they need to cross barriers. *Trypanosoma brucei* is an extracellular parasite that sequentially occupies different tissues in the alimentary tract of its tsetse fly host. Our recent work on social motility, the collective migration of procyclic (insect midgut) forms on agarose surfaces, has provided insights into how the parasites both condition and sense their environment. In addition to explaining the self-organising properties of trypanosome communities on plates, we have shown that the same sensors, which are components of the cyclic AMP signalling pathway, are required for efficient colonisation of the tsetse midgut and penetration of the peritrophic matrix.

10:20 (10 mins) - A26068 - Variation in water contact behaviour and risk of *Schistosoma mansoni* (re)infection among Ugandan school-aged children in an area with persistent high endemicity

Authors

S Trienekens[4]; CL FaustF Besigye[1]; L Pickering[2]; EM Tukahebwe[1]; J Seeley[3]; **PH L Lamberton**

[1] Vector Control Division, Ministry of Health, Uganda; [2] Institute of Health & Wellbeing, College of Social Sciences, University of Glasgow, UK; [3] Medical Research Council, Uganda Virus Research Institute, Entebbe, Uganda; [4] Institute of Biodiversity, Animal Health and comparative Medicine, and Wellcome Centre for Integrative Parasitology, University of Glasgow, UK

Discussion

Annual mass drug administration with praziquantel has reduced schistosomiasis transmission in some highly endemic areas, but areas with persistent high endemicity have been identified across sub-Saharan Africa, including Uganda. In these areas many children are rapidly reinfected post treatment, while some children remain uninfected or have low-intensity infections. The aim of this mixed-methods study was to better understand variation in water contact locations, behaviours and infection risk in school-aged children within an area with persistent high endemicity to inform additional control efforts. Data were collected in Bugoto, Mayuge District, Uganda. Two risk groups were identified from a longitudinal cohort, and eight children with no/low-intensity infections and eight children with reinfections were recruited. Individual structured day-long observations with a focus on water contact were conducted over two periods in 2018. In all identified water contact sites, four snail surveys were conducted quarterly over 1 year. All observed *Biomphalaria* snails were collected, counted and monitored in the laboratory for *Schistosoma mansoni* cercarial shedding for 3 weeks. Children came into contact with water for a range of purposes, either directly at the water sources or by coming into contact with water collected previously. Although some water contact practices were similar between the risk groups, only children with reinfection were observed fetching water for commercial purposes and swimming in water sources; this latter group of children also came into contact with water at a larger variety and number of sites compared to children with no/low-intensity infection. Households with children with no/low-intensity infections collected rainwater more often. Water contact was observed at 10 sites throughout the study, and a total of 9457 *Biomphalaria* snails were collected from these sites over four sampling periods. Four lake sites had a significantly higher *Biomphalaria choanomphala* abundance, and reinfected children came into contact with water at these sites more often than children with no/low-intensity infections. While only six snails shed cercariae, four were from sites only contacted by reinfected children.

10:30 (10 mins) - A26003 - Xeno-monitoring of molecular drivers of artemisinin and partner drug resistance in *P. falciparum* populations in malaria vectors across Cameroon

Nkemngo Francis Nongley

TBD

Session 6 – Wed 23rd Mar 11:50 - 13.00

Day 3 - Mathematical Modelling of Parasites *(Lecture Theatre K/018)*

23-March-2022, at 11:50 to 12:40

Chair - Dr Laurence Wilson

11:50 (10 mins) - A26064 - The changing face of schistosome infection may help explain conflicting outcomes among malaria-schistosome coinfection studies

Authors

S Rollason[1]; J Lello[1];

[1] Cardiff School of Biosciences, Cardiff University, UK

Discussion

Malaria and schistosomiasis are two of the most important parasitic diseases and coinfection with their causative parasites is common, particularly in sub-Saharan Africa. These parasites interact with each other via their effects on the host immune system, but studies to date report conflicting results, some suggesting that schistosomes are associated with reduced malaria intensity while others report increased intensity. Schistosomes provoke different immune responses during early vs late infection, which may be a factor in these conflicting results. Using agent-based modelling we explored the effects of schistosomes on blood stage malaria, by simulating the effects of different stages of schistosome infection. We find the intensity and dynamics of malaria infections are greatly influenced by the stage of schistosome infection. Our findings may help to explain the apparent contradictions between studies and will have implications for host health and for the design of parasite control strategies

12:00 (10 mins) - A26132 - ScTralign: a computational method to align and compare biological development trajectories across conditions from single cell RNA sequencing data

Authors

R Laidlaw[1]; R McCulloch[1]; E Briggs[3]; K Matthews[3]; T Otto[1];

[1] University of Glasgow, UK; [2] University of Glasgow , UK; [3] University of Edinburgh, UK

Discussion

Understanding the biological processes underpinning lifecycle transition stages in parasites is crucial not only to further knowledge on the parasite itself, but also in identifying possible targets for therapeutics. Single cell RNA sequencing (scRNA-seq) is becoming a more common approach for studying such life cycling transitions, as it is possible to capture gene expression profiles of individual cells at varying stages of the lifecycle and dissect them from the mixed populations into clusters of similar cells. A crucial element to gain full insight from such scRNA-seq experiments is to compare datasets from different conditions of development, such as comparisons of mutant and wild type parasites, and of different parasite species. A common way to try and integrate such datasets and identify differences between cell clusters across conditions. However, this may force similarity between cells, possibly obscuring processes unique to one of the datasets, and some expression differences might not be properly captured at the cluster level. We present scTralign, a method that allows the alignment of two independently generated linear scRNA-seq trajectories irrespective of whether or not they have differing cell populations and process kinetics. The method creates a common pseudotime

axis for the two datasets, separating cells which have no equivalent on the opposing trajectory from cells that are similar across both, thereby allowing variation to be preserved and identified between both datasets. The user can then identify genes that are differentially expressed between cells undergoing processes shared between the two conditions to see if expression patterns differ between the datasets, which might be causing later splits in development between the conditions. We verify the process of scTralign with simulated scRNA-seq datasets, as well as applying it to two conditions of *Trypanosoma brucei* development to identify genes that allow slender parasites to transition into stumpy forms and to compare the lifecycle of *Plasmodium berghei* in different organs. Compared with other trajectory alignment methods, scTralign correctly identifies the overall alignment of the real and simulated data, identifying where the processes begin to differ from one another. We also compare our method with the current methods used to extract differentially expressed genes across different conditions, showing scTralign captures more information from the dataset than contemporary methods.

12:10 (10 mins) - A26234 - High speed, three-dimensional imaging to inform biophysical modelling in parasitology

Laurence Wilson
University of York

Discussion

Title: High speed, three-dimensional imaging to inform biophysical modelling in parasitology L.G. Wilson 2, R.C. Findlay 1,2, P.B. Walrad 1 1. York Biomedical Research Institute, Department of Biology, University of York, York, UK 2. Department of Physics, University of York, York, UK. The interactions between physics and biology have been a rich seam of scientific advances. Cellular motility is a paradigm for these interactions. Motility is an ancient eukaryotic trait, ubiquitous across phyla with roles in predator avoidance, resource access, and competition. Flagellar motility is seen in various parasitic protozoans, and morphological changes in flagella during the parasite life cycle have been observed. We have developed a unique implementation of holographic microscopy to image swimming cells of various parasites, including Leishmania *mexicana* and *Plasmodium berghei*, in three dimensions and at rates up to 1,000 volumes per second. These measurements give insight into the relationship between flagellar structure and motility, and phenotypic variation, through quantitative data on the reaction of individual cells to an external stimulus. Among our findings, we have seen that the human-infective (metacyclic promastigote) forms of L. *mexicana* display 'run and tumble' behaviour in the absence of stimulus, reminiscent of bacterial motion, and that they specifically modify swimming direction and speed to target host immune cells in response to a macrophage-derived stimulus. Non-infective (procyclic promastigote) L. *mexicana* cells swim more slowly, along meandering helical paths.

Day 3 - Host:Parasite interactions : Tissue Tropism *(Lecture Theatre T/005)*
23-March-2022, at 11:50 to 12:40
Chairs - Dr Cecile Crosnier & Dr James Hewitson

11:50 (10 mins) - A25931 - Daily rhythms in malaria hosts and parasites influence artemisinin drug efficacy

Authors
AT Owolabi[2]; SE Reece[1]; P Schneider[1];
[1] University of Edinburgh, Institute of Evolutionary Biology, UK; [2] The University of Edinburgh, UK

Abstract
Background and Objectives:
Circadian rhythms contribute to treatment efficacy in several non-communicable diseases. However, chronotherapy (administering drugs at a particular time of day) against infectious diseases has been overlooked. Yet, the daily rhythms of both hosts and disease-causing agents can impact the efficacy of drug treatment. We use the rodent malaria parasite *Plasmodium chabaudi,* to test if the daily rhythms of hosts, parasites, and their interactions, affect sensitivity to the key antimalarial, artemisinin.
Methodology:
Asexual malaria parasites develop rhythmically in the host's blood, in a manner timed to coordinate with host daily rhythms. Our experiments coupled or decoupled the timing of parasite and host rhythms, and we administered artemisinin at different times of day to coincide with when parasites were either at an early (ring) or later (trophozoite) developmental stage. We quantified the impacts of parasite developmental stage, and alignment of parasite and host rhythms, on drug sensitivity.
Results:
We find that rings were less sensitive to artemisinin than trophozoites, and this difference was exacerbated when parasite and host rhythms were misaligned, with little direct contribution of host time of day on its own. Furthermore, the blood concentration of haem at the point of treatment correlated positively with artemisinin efficacy but only when parasite and host rhythms were aligned.
Conclusions and implications:
Parasite rhythms influence drug sensitivity *in vivo*. The hitherto unknown modulation by alignment between parasite and host daily rhythms suggests that disrupting the timing of parasite development could be a novel chronotherapeutic approach.

12:00 (10 mins) - A26162 - Histopathological and molecular diagnosis of eight clinical human hydatidosis from Gaza Strip, Palestine

Authors
A Al-Hindi[1]; F Rouk[2]; H Hamada[3]; A Al-Fara[4]; A Lubbad[5]; S Al-Hindi[5];
[1] Islamic University of Gaza, P.O. Box 108, Gaza, Palestine, Palestinian Territory; [2] European Gaza Hospital, Ministry of Health, Gaza, Palestinian Territory; [3] Director of Pathology Unit, Al-Shifa Hospital, Ministry of Health, Gaza, Palestinian Territory; [4] Head of Thoracic Surgery Department at the European Gaza Hospital, Gaza Strip, Palestinian Territory; [5] Faculty of Medicine, Islamic University of Gaza, P.O. Box 108, Gaza, Palestine, Palestinian Territory

Abstract

Objectives: Hydatidosis is a parasitic disease caused by the cestode *Echinococcus granulosus.* The present study focused on the multi-diagnosis of a clinical cases including, histopathology, clinical presentation of the patient, and the molecular diagnosis of the tissue.
Methods: We collected the hydatid cyst diseases tissue samples from patients attending hospitals in Gaza Strip. Investigations are included the clinical presentation of each patient, histopathological studies, and molecular diagnosis.
Clinical presentations: These are 15 clinical cases of hydatidosis of hydatid cysts collected from the five governorates of the Gaza Strip. A total of 46.7% of the examined cases were liver affected. The sequencing and analyses revealed one genotype of *E. granulosus* (G1) responsible for these huma hydatid cysts.
Conclusion: Hydatid cyst disease occurrence is confirmed in the examined human tissue samples and belong to genotype G1.

12:10 (10 mins) - A26121 - Novel *Trypanosoma brucei* heterogeneity is associated to tissue invasion and adaptation

Authors
M De Niz[1]; F Carvalho[1]; F Guegan[1]; L Lopez Escobar[1]; H Machado[1]; A Temudo[1]; N Santos[1]; C Franco[1]; L Figueiredo[1];
[1] Instituto de Medicina Molecular, Portugal

Discussion

Recent research across eukaryotes has demonstrated the importance of heterogeneity for a plethora of biological phenomena, ultimately related to population survival. *Trypanosomes* have very complex life cycles, which rely on the successful adaptation the parasites must display to both the insect vector and the mammalian hosts. In order to investigate parasite heterogeneity in a holistic manner, we used surgical approaches and intravital microscopy on rodent models to characterize in vivo parasite morphology, cell cycle, stumpy formation, PAD1 expression, motility type, and velocity of parasites inside the vasculature and extravascular spaces. We studied 12 organs throughout 20 days of infection. Inside the vasculature, we found that the morphology of parasites can be quite variable (14-28 µm range in length; 1.2-3.5 µm range in width) and that crossing into the extravascular space of several organs, including large reservoirs, is associated to the presence of particularly long and large forms. Characterization of the extravascular parasites first revealed a group of organs in which parasites do not undergo significant changes relative to the blood counterparts, either because they colonize the organ poorly or because they are only transiently colonized. Second, we found that in white adipose tissues, pancreas, spinal cord, yellow marrow, and brain, parasites undergo significant phenotypic changes relative to the blood counterparts but similar within this group, indicating that lipid-rich environments trigger a common phenotypic adaptation. Third, in lymphoid organs including the spleen and RBM, parasites undergo another unique adaptation, different to that observed in lipid-rich tissues. Moreover, by atomic force microscopy, we show that the viscoelastic properties of organs are heavily modified throughout infection. This study reveals a remarkable heterogeneity in the parasite population among and within organs, which probably play key roles in organ invasion and establishment of chronic infection.

Day 3 – Parasite:Vector Biology *(Lecture Theatre P/X001)*
23-March-2022, at 11:50 to 12:40
Chair - Dr Álvaro Acosta-Serrano & Dr Poppy Lambert

11:50 (10 mins) - A25959 - Investigation of a protein kinase signalling pathway required for haptomonad differentiation in *Leishmania mexicana*

Authors
N Baker[2]; C Hughes[2]; J Sadlova[1]; AA Dowle[2]; C Taylor[2]; P Volf[1]; JC Mottram[2];
[1] Charles University, Prague, Czech Republic; [2] Department of Biology, University of York, UK

Abstract
Survival of *Leishmania* throughout its life cycle relies on perfectly orchestrated differentiation events, triggered by environmental changes such as nutrients, pH and temperature. Protein kinases are fundamental to sensing these environmental cues and signalling differentiation. In the sand fly, procyclic promastigotes undergo several differentiation events, resulting in either the infective metacyclic form or the haptomonad form. Metacyclic cells can be enriched by growth in Graces media, but the haptomonad form has low abundance in these conditions. Here we investigate a *L. mexicana* haptomonad differentiation protein kinase (HDK1) null mutant, producing cultures enriched in haptomonad parasites, triggered by reduction in pH from 7.5 to 5.5, but not to a change in temperature. This HDK1 null mutant can infect the sand fly midgut but cannot colonise the stomodeal valve, indicating an impaired onwards transmission. Proteomic and RNAseq analysis, comparing expression levels between the procyclic and haptomonad life cycles stage of this HDK1 mutant have identified markers of the haptomonad stage and are aiding to unravel this signalling pathway.

12:00 (10 mins) - A25549 - Optimisation of in vitro feeding and long-term storage of the hematophagous mite *Dermanyssus gallinae*.

Authors
F Nunn[1]; K Bartley[1]; AJ Nisbet[1];
[1] Moredun Research institute, UK

Abstract
Poultry red mites (PRM) are blood feeding ectoparasites that live off-host, only seeking a bird to rapidly engorge every few days. Three of the five lifestages are hematophagous and are highly mobile making them difficult to contain in a controlled experimental environment. In vitro feeding techniques have been previously been devised to overcome containment issues (e.g. McDevitt et al., 2006; Bartley et al., 2015) for the preliminary screening of PRM vaccines. Mite feeding rates can be highly variable after storage and here we describe steps towards optimising egg laying and evaluating feeding rates of mites stored in different conditions.
Recently we described utilising Baudruche membrane in an in vitro device to feed adult females (Nunn et al 2020) using goose blood, which led to improved and reproducible feeding rates and fewer animal procedures due to the increased blood volume per procedure. We evaluated the device to feed the hematophagous nymph stages of PRM and demonstrated significant correlation between the feeding rates of deutonymph stages and

adult females (Spearman rs. =0.54, p = 0.0008, n=48) and protonymphs and adult females (Spearman rs. = 0.60, p = 0.00001,=48). A highly significant correlation was demonstrated between the proportion of the two nymph stages (Spearman rs. = 0.919,p = >0.00001,n=48) fed adults with no significant feeding of nymph stages in the absence of adult females. To obtain reasonable in vitro feeding rates, mites are generally starved (conditioned) to allow digestion of their last blood meal, moulting and egg laying. Traditionally, mites are conditioned at room temperature(RT) before being stored at 4-8 C,briefly brought to RT before a feeding assay. Mite feeding can be variable under these conditions and are generally only useful for 2-4 weeks post conditioning at RT. Based on a study by Wang et al (2020) we performed a study to establish feeding rates of mites kept over longer periods of time after two different conditioning regimes. Three collections of mites (1-3) were each divided into two, with one cohort stored at 5C until conditioning at RT for one week prior to feeding (1a-3a) and with the other cohort (1b-3b) conditioned at RT for one week followed by storing at 5C until feeding. The percentage feeding rate of adult females in cohorts 1a-3a, fell by less than 10% over the course of the study (10 weeks), demonstrating feeding rates of 2.6, 1.26 and 1.4 times that of cohorts 1b-3b at 6weeks and 6.75, 2.1 and 4.75 respectively at 8weeks.To see if egg laying of mites was improved by being kept in groups of fed females, we compared numbers of offspring per fed mite on two occasions. Fed mites were incubated individually (n=60,n=41 respectively)and 5 replicate groups of 5, 15 and 30 mites and counting offspring after incubation for 7 days. No difference in offspring/fed mite was demonstrated across the different experimental groups. Repeated feeding of adult female mites was then performed.

12:10 (10 mins) - A26203 - The MISP family of surface glycoproteins from *Trypanosoma brucei* is co-expressed with VSG and BARP in the metacyclic trypomastigote stage, adopts a triple helical bundle structure, and is not essential for the colonization of the tsetse salivary glands

Authors

A Casas-Sanchez[1]; S Perally[1]; R Ramaswamy[4]; LR Haines[1]; C Rose[1]; C Yunta-Yanes[1]; M Aguilera-Flores[5]; L Smithson[3]; S Vaughan[3]; M Lehane[1]; IC Almeida[5]; J Van Den Abbeele[2]; M Boulanger[4]; A Acosta-Serrano[1];

[1] Liverpool School of Tropical Medicine, UK; [2] Institute of Tropical Medicine, Antwerp, Belgium; [3] Department of Biological and Medical Sciences, Oxford Brookes University, Oxford, UK; [4] University of Victoria, Canada; [5] Department of Biological Sciences, The University of Texas at El Paso, United States

Discussion

Trypanosoma brucei spp. develop into mammalian-infectious metacyclic trypomastigotes inside the tsetse salivary glands. Besides acquiring a variant surface glycoprotein (VSG) coat, little is known about the expression of invariant surface antigens by the metacyclic stage. Proteomics analyses of saliva from *T. brucei*-infected flies identified, in addition to VSG and *Brucei* Alanine-Rich Protein (BARP) peptides, a family of GPI-anchored surface proteins herein named Metacyclic Invariant Surface Proteins (MISP). The MISP family is encoded by five paralog genes with >80% protein identity, which are exclusively expressed by salivary gland stages of the parasite, and peaks in metacyclic stage as shown by confocal microscopy and immuno-high resolution scanning electron microscopy. Crystallographic analysis of MISP and a high confidence model of BARP reveal a triple helical bundle architecture commonly found in other trypanosome surface proteins. Molecular modelling combined with live fluorescent microscopy suggests that MISP expose immunogenic N-terminal epitopes above the VSG coat, although vaccination with a recombinant MISP isoform did not protect mice against a *T. brucei* infectious bite. Lastly, both using RNAi and CRISPR-Cas9-driven knock out of all MISP paralogues suggests they are not essential for parasite development in the tsetse vector.

12:20 (10 mins) - A25683 - CTL4 controls TEP1-independent melanization of human malaria parasites

Authors

M L Simoes[1]; G Dimopoulos[2];

[1] London School of Hygiene and Tropical Medicine, UK; [2] Johns Hopkins University, United States

Abstract - Melanization is one of the most effective innate defense mechanisms in mosquito vectors. Numerous studies have shown that the *Anopheles* TEP1-controlled complement-like system is essential for melanization of the rodent model malaria parasite *Plasmodium berghei*, which evades this defense by recruiting C-type lectins. But the role of TEP1 has not been sufficiently addressed in the context of malaria infection with the clinically relevant human malaria parasite, *Plasmodium falciparum*. Using CRISPR/Cas9 genome editing, we show that the melanization of P. *falciparum* is independent of the TEP1-controlled complement-like system, and a small proportion of P. *falciparum* ookinetes are capable of evading this defense mechanism in the midgut tissue of CTL4null mosquitoes, in contrast to the complete melanization of rodent P. *berghei*. Furthermore, we discovered that the major anti-*Plasmodium* pathway Imd does not influence *Plasmodium* melanization. Our study proves CTL4 as one of the most potent malaria transmission-blocking targets.

Session 7 – Wed 23rd Mar 14:00 - 15.20

Day 3 - Parasite Biochemistry *(Lecture Theatre K/018)*

23-March-2022, at 14:00 to 15:00

Chairs - Prof Anthony Wilkinson & Dr Michael Plevin

14:00 (20 mins) - A26226 - TBD

Dr Chi-Min Ho

Columbia University

TBD

14:20 (10 mins) - A26204 - Invariant surface glycoprotein 65 of *Trypanosoma brucei* is a complement C3 receptor important for virulence

Authors

A Cook[1]; O Macleod[2]; M Crow[2]; H Webb[2]; R Burns[2]; M Redpath[2]; S Seisenberger[2]; C Trevor[2]; L Peacock[4]; A Schwede[2]; N Kimblin[2]; AF Francisco[6]; J Pepperl[2]; S Rust[5]; P Voorheis[3]; W Gibson[4]; MC Taylor[6]; M Higgins[1]; M Carrington[2];
[1] University of Oxford, Department of Biochemistry, UK; [2] University of Cambridge, Department of Biochemistry, UK; [3] Trinity College Dublin, Ireland; [4] School of Biological Sciences & Bristol Veterinary School, University of Bristol, UK; [5] AstraZeneca, UK; [6] Department of Pathogen Molecular Biology, London School of Hygiene and Tropical Medicine, UK

Discussion

African Trypanosomes replicate in the blood of mammalian hosts yet are completely exposed to the adaptive and innate immune systems. Despite this, Trypanosomes can sustain long-term infections - how can this parasite survive constant host immune-surveillance? Trypanosomes evade the adaptive immune response through antigenic variation of a surface coat consisting of a dense layer of variable surface glycoprotein. However, very little is known about how they negate the innate system, including the blood circulating complement system. We have discovered that an invariant surface glycoprotein, ISG65, is a receptor for Complement Component 3 (C3). We show how ISG65 binds to the thioester domain of C3b. We also show that knockout of the ISG65 locus greatly decreases the pathogenicity of trypanosomes in a mouse model. Deposition of C3b on pathogen surfaces is a central point in activation of the complement system and C3b has been observed on trypanosome surfaces. Our findings therefore suggest that trypanosomes have a C3 receptor distributed across their surfaces that greatly decreases their susceptibility to complement-mediated killing.

14:30 (10 mins) - A25538 - A *Toxoplasma gondii* oxopurine transporter binds nucleobases and nucleosides using different binding modes.

Authors

GD Campagnaro[1]; HA Elati[1]; S Balaska[1]; ME Martin Abril[1]; F Hulpia[2]; MJ Natto[1]; K Lee[1]; L Sheiner[1]; S Van Calenbergh[2]; **HP De Koning**[1];
[1] University of Glasgow, Institute of Infection, Immunity & Inflammation,, UK; [2] Universiteit van Gent, Belgium

Abstract

Toxoplasma gondii are unable to synthesize purines de novo, and instead salvages them from its environment, for which they need high affinity carriers because the concentration of free unphosphorylated nucleosides and nucleobases is very low inside the host cell. Apart from the essential role in purine salvage, nucleoside analogues have shown promise as potential therapeutics against *T. gondii*. In order to optimise the targeting of such analogues, the characterization of the purine transporters is required.

Here, we report the expression of one of the four *T. gondii* Equilibrative Nucleoside Transporter genes, *Tg244440*, in a *Trypanosoma brucei* strain from which nucleobase transporters had been deleted. Tg244440 transported nucleobases hypoxanthine and guanine with similar affinity (K_m ~1 µM), while inosine and guanosine displayed K_i values of 4.05 and 3.30 µM, respectively. Low affinity was observed for adenosine, adenine, and pyrimidines, classifying Tg244440 as a high affinity oxopurine transporter.

A large number of purine analogues were used to probe the substrate-transporter binding interactions, mostly through competitive inhibition of [^{3}H]-guanine transport, culminating in quantitative models showing different binding modes for oxopurine bases, oxopurine nucleosides, and adenosine. Hypoxanthine and guanine interacted through protonated N1 and N9, and through unprotonated N3 and N7 of the purine ring, whereas inosine and guanosine mostly employed the ribose hydroxy groups for binding, in addition to N1H of the nucleobase. Conversely, the ribose moiety of adenosine barely made any contribution to binding. *Tg244440* is the first gene identified to encode a high affinity oxopurine transporter in *T. gondii* and, to the best of our knowledge, the first purine transporter to employ different binding modes for nucleosides and nucleobases. The unprecedented flexibility of the binding pocket, apparently allowing oxopurine bases and nucleosides to bind with very similar affinity but different orientations, shows the potential of *T. gondii* purine transporters for the uptake of cytotoxic purine analogues.

Day 3 - Parasite ImmunoPathology *(Lecture Theatre T/005)*

23-March-2022, at 14:00 to 15:00

Chairs - Dr Damian Perez Mazliah & Dr Jillian Barlow

14:00 (20 mins) - A25409 - Multi-omic approaches reveal a dynamic crosstalk between plasma cells and Cx3cr1+ microglia in the murine brain during chronic *Trypanosoma brucei* infection

Authors

J Quintana Alcala[3]; P Chandrasegaran[3]; M Sinton[3]; R Heslop[3]; E Briggs[2]; T Otto[3]; NA Mabbott[1]; A MacLeod[3];
[1] The Royal (Dick) School of Veterinary Studies and the Roslin Institute, The University of Edinburgh, UK; [2] Centre for Immunity, Infection and Evolution, School of Biological Sciences. University of Edinburgh, UK; [3] Wellcome Centre for Integrative Parasitology (WCIP), UK

Abstract

Chronic infections with the parasite *Trypanosoma brucei*, the causative agent of Human African trypanosomiasis, lead to severe neuroinflammation and death if left untreated. However, a detailed understanding of the cellular and molecular interactions that mediate this severe pathology is lacking. Using single cell and spatial transcriptomics, we have identified for the first time, a unique population of CD138$^+$plasma cells in the brain ventricles of infected animals compared to naïve controls. These plasma cells express a robust innate-like, regulatory transcriptional profile, characterised by the expression of pathogen-sensing molecules (*Tlr4),* anti-inflammatory cytokines *(Il10)* and pro-survival receptor molecules such as*Tnfrsf17 (*B cell maturation antigen, BCMA). Additionally, we detected a subpopulation of *Cx3cr1*$^+$ microglia that express a wide range of factors associated with B cell recruitment and survival, such as *Cxcl12* and *Tnfsf13b* (B cell activating factor).

Interestingly, *Cx3cr1*$^+$ microglia are the only cells in our dataset expressing both *Il10ra* and *Il10rb*, suggesting that they are primed to respond to IL-10. Further *in vitro* studies demonstrated that these regulatory, innate-like plasma cells can stimulate microglia polarisation towards an anti-inflammatory state *via* IL-10 signalling. We propose a model in which unresolved brain infections induce the activation of *Cx3cr1*$^+$ microglia, leading to the recruitment and survival of plasma cells mediated by CXCL12 and BAFF-BCAM signalling, respectively. In turn, these regulatory plasma cells alleviate inflammation by dampening *Cx3cr1*$^+$ activation *via* IL-10 signalling, limiting pathology. This work provides novel insights into the mechanisms of B cell-stromal interactions in the brain during infection.

14:20 (10 mins) - A26131 - Dissecting side-by-side *Trypanosoma cruzi*-specific and cardiac-specific B cell responses in Chagas disease

Authors

D Perez Mazliah[1];

[1] University of York, UK

Discussion

Background

Chagas disease, caused by *Trypanosoma cruzi* parasites, is the most frequent cause of infectious cardiomyopathy in the world and the highest-impact parasitic disease in the Americas. No vaccines are available, and current medicines are toxic and limited. Historically a neglected tropical disease of the Americas, it is now spreading globally. Chagas heart disease (CHD) is an inflammatory cardiomyopathy that develops in approximately one-third of infected people. The reasons why some infected individuals develop cardiomyopathy while others remain asymptomatic for life remain largely unknown. In response to *T. cruzi* infection, and driven by poorly understood triggers, the immune system produces both antibodies against the parasite and against the host's heart tissues.

Methods

We used magnetic enrichment and novel B-cell tetramers combined with mouse models of bioluminescent *T. cruzi* infection to track, by flow cytometry and side-by-side, the development of *T. cruzi*-specific and cardiac-specific B-cell responses in spleen and heart.

Results

We observed a striking accumulation of plasma cell/plasmablast B cell subsets in the heart of *T. cruzi*-infected mice during early chronic infection. While *T. cruzi*-specific B cells gave rise to robust germinal centre responses in the spleen, cardiac-specific B-cell responses were primarily extrafollicular.

Conclusions

Our data suggest that activation of *T. cruzi*-specific and autoreactive cardiac-specific B cell responses are driven by drastically different mechanisms. Future work with our newly developed tools will allow us to identify the main drivers of pathogen vs autoreactive cardiac-specific B cell responses, as well as exploring the role of different B-cell subsets in the pathogenesis of CHD.

14:30 (10 mins) - A26107 - Experimental digestive Chagas disease: spatio-temporal infection dynamics, immunopathological mechanisms and the prospect of functional cure with trypanocidal benznidazole chemotherapy.

Authors

A Khan[1]; H Langston[1]; AF Francisco[1]; L Walsh[1]; FC Costa[1]; F Olmo[1]; S Jayawardhana[1]; J Penning-Lambert[1]; MC Taylor[1]; CJ McCann[2]; JM Kelly[1]; MD Lewis[1];

[1] London School of Hygiene and Tropical Medicine, UK; [2] University College London, UK

Discussion

Digestive Chagas disease (DCD) is an enteric neuropathy caused by infection with the protozoan pathogen *Trypanosoma cruzi* that affects ~1 million people. Parasitism of the GI tract provokes a type 1 inflammatory response, which causes collateral damage to the enteric nervous system (ENS) and dysfunctional peristalsis, progressing to digestive megasyndromes in severe cases. Beyond this, little is known about the kinetics, underlying molecular/cellular mechanisms or determinants of susceptibility. The lack of a robust, predictive animal model has held back research in these areas. We screened a series of mouse models using gastrointestinal tracer assays and *in vivo* infection imaging systems to discover a subset exhibiting chronic digestive transit dysfunction and significant retention of faeces in both sated and fasted conditions. The colon was a specific GI region of tissue parasite persistence, delayed transit and dramatic loss of myenteric neurons, as revealed by whole-mount immunofluorescence analysis. Immune transcriptome profiling of colon tissue from DCD resistant BALB/c and susceptible C3H mice identified the macrophage scavenger receptor MARCO as significantly associated with disease severity. Next, we tested the hypothesis that benznidazole-mediated cure of infection translates into alleviation of DCD pathology. Sterilisation of infection by early treatment (6 weeks post-infection) resulted in sustained and complete reversal of GI transit delay, accompanied by an ENS tissue repair transcriptional profile dominated by glial cell markers. However, late treatment (24 weeks post-infection) only led to partial reversal of the DCD phenotype, suggesting the accumulation of permanent tissue damage during chronic infections. Importantly, benznidazole treatment failed to cure 40% of mice and parasite relapse coincided with a worsening of disease, but not to the level seen in untreated controls. These data prove that DCD pathogenesis is sustained by enduring *T. cruzi* infection and can be interrupted by sterilising anti-parasitic chemotherapy. However, they also show that if treatment is delayed there is a risk that parasite-induced enteric neuromuscular tissue damage and dysfunction will become irreversible.

14:40 (10 mins) - A25934 - A library of cell-surface and secreted *Schistosoma mansoni* proteins to investigate host:parasite interactions

Authors

C Crosnier[1];

[1] Department of Biology, University of York, UK

AbstractSchistosomiasis is a major global health burden in terms of its incidence and socio-economic impact. Despite its importance, no licenced vaccine is available and schistosomiasis control in endemic populations mostly relies on mass-administration of praziquantel. To investigate the biology of the parasite, we have recently developed a library of 120 recombinant cell-surface and secreted *Schistosoma mansoni* proteins, which we produce in mammalian cells to try and preserve conformational epitopes. These full-length extracellular proteins show good immunoreactivity when exposed to sera from individuals living in endemic areas, and contain heat-labile epitopes. We have used this protein library in the context of controlled human infections to identify early serological markers of infection, and in vaccine challenge studies in a murine model of schistosomiasis. The prolonged survival of schistosomes into their human hosts involves multiple mechanisms of immune suppression and modulation. To identify protein:protein interactions involved in host immune regulation, we are currently using our recombinant *S. mansoni* protein collection to interrogate a unique library of over 700 human immune receptors. We hope this study will shed light on novel

immunomodulatory mechanisms that will constitute new avenues in the development of a vaccine against schistosomiasis, and more broadly in the control of allergic and auto-immune responses.

Day 3 – Parasite Genetic Architecture *(Lecture Theatre P/X001)*
23-March-2022, at 14:00 to 15:00
Chairs - Dr Nathaniel Jones & Dr Joana Faria

14:00 (20 mins) - A26192 - Thousands of messenger RNA untranslated regions reprogram post-transcriptional gene expression profiles in trypanosomes

Prof David Horn
University of Dundee

Discussion

Abstract Cells express thousands of proteins that differ in abundance over a wide range. In nucleated cells, messenger RNA untranslated regions (UTRs) can contribute to post-transcriptional expression control, but relatively few UTRs have been shown to exert these controls, and cis-regulatory sequences have remained largely uncharacterised. In the *Trypanosoma*tids, genome-wide polycistronic transcription places a particular emphasis on post-transcriptional controls. We used a massive parallel reporter assay coupled with UTR-seq profiling in the African trypanosome, revealing post-transcriptional reprogramming by thousands of 3'-UTRs. Genome-scale UTR-seq screening identified an abundance of regulatory fragments that either increased or reduced reporter expression. Analysis of regulatory fragments and native UTRs yielded a correlation between gene expression and 3'-UTR sequence composition. A machine learning approach guided by these findings effectively predicted observed measures of translation efficiency at a transcriptomic scale; R2 was 0.69. This approach also provided quantitative measures of the relative contribution of sequences within native 3'-UTRs and revealed similarly predictive sequences within 5'-UTRs. Thus, UTR-seq reveals the cis-regulatory UTR sequences that control gene expression in the context of a genome that lacks promoter-based transcription control. Thus, gene expression profiles in trypanosomes are post-transcriptionally reprogramed by thousands of regulatory UTRs.

14:20 (10 mins) - A25669 - Heterogeneous elongation of RNA polymerase I transcription at the active VSG expression site in *Trypanosoma brucei*

Authors
J Budzak[1]; G Rudenko[1];
[1] Imperial College London, UK

AbstractA Variant Surface Glycoprotein (VSG) coat protects bloodstream form *Trypanosoma brucei* within the mammalian host. Prodigious amounts of VSG mRNA (~7-10% total) are generated by RNA polymerase I (Pol I), from a single VSG gene located within one of ~15 VSG expression sites (ES). Transcription of VSG-ESs occurs in an extra-nucleolar Pol I body called the Expression Site Body (ESB). Importantly, T. *brucei* must maintain very high levels of VSG expression, as VSG is essential for proliferation both in vitro and in vivo. Trypanosomes use Pol I to transcribe their ESs, presumably as Pol I has a higher rate of initiation than Pol II. It has been assumed that ESs are either fully transcribed or silent. However using a variety of different microscopy approaches, we have consistently observed that not all cells contain an ESB, indicating that transcription of the active ES may be heterogeneous. We therefore investigated this by generating cell lines where constructs containing MS2 sequences were inserted at less than 2, 6, 10, 20, 40 and 60 kb downstream of the active ES promoter. Using single molecule RNA-FISH experiments, we now show that Pol I transcription elongation is not always fully processive at the active ES in individual cells. We find that transcription elongation is gradually attenuated as distance from the promoter increases. Interestingly, we find a significant reduction in transcription elongation after the first ~10kb of the active ES. In addition, across the length of the active ES, we find the highest pre-mRNA levels at the active ES promoter and the lowest pre-mRNA levels immediately upstream of the telomeric VSG. We have recently discovered that a number of nuclear splicing condensates are positioned in close proximity to the VSG at the active ES telomere. Possibly these boost VSG mRNA expression even in the absence of fully processive Pol I transcription. These unexpected observations challenge the paradigm that RNA Polymerase I provides continuously high levels of transcription throughout the active ES in T. *brucei*.

14:30 (10 mins) - A25922 - The *Trypanosoma brucei* RNA/DNA hybrid interactome reveals a role for RAD51 in R-loop homeostasis and repair of VSG-localised DNA breaks during antigenic variation

Authors
M Girasol[3]; J Damasceno[1]; C Marques[1]; M Krasilnikova[1]; C Lapsley[1]; R Carruthers[4]; D Beraldi[1]; P Rivera[2]; E Briggs[5]; R McCulloch[1];
[1] Wellcome Centre for Integrative Parasitology, Institute of Infection, Immunity and Inflammation, University of Glasgow, UK; [2] College of Public Health, University of the Philippines Manila, Philippines; [3] College of Medicine, University of the Philippines Manila, Philippines; [4] Institute of Cancer Sciences, University of Glasgow, UK; [5] School of Biological Sciences, University of Edinburgh, UK

Abstract
The process of cell division requires the coordination of two often conflicting events that access the genome simultaneously: DNA replication and transcription. R-loops, which are three-stranded nucleic acid structures comprised of an RNA/DNA hybrid in the context of a displaced single-stranded DNA, usually arise when an elongating transcript reinvades the template DNA, but can also occur in *trans*. They can serve as obstacles during replication and can be sources of DNA damage. Because of this, R-loops are involved in many crucial processes in all organisms and are therefore under tight regulatory control. Mapping R-loops in the unicellular protozoan parasite *Trypanosoma brucei* revealed widespread enrichment, including in subtelomeric Variant Surface Glycoprotein (VSG) expression sites, linking them to DNA damage and antigenic variation (Briggs et al., 2018a; Briggs et al., 2018b). However, the mechanisms that control many aspects of R-loop biology in the *T. brucei* genome remain unclear. Using RNA/DNA hybrid immunoprecipitation coupled with mass spectrometry, 616 putative R-loop–interacting proteins were identified, including interactors with activities linked to RNA processing and DNA replication, repair, and recombination. To search for R-loop interactors with roles in antigenic variation, interactomes from bloodstream form and procyclic form parasites were compared. Of these proteins, four putative interactors were further investigated: two recombinases with known roles in VSG switching, RAD51 and RAD51-3, and two other proteins with unreported functions, a putative SNF2 chromatin remodeler and an ATP-dependent DEAD/H RNA helicase (DH). Loss of all proteins

each led to nuclear genome damage and alterations in VSG expression dynamics. RNA/DNA hybrid immunofluorescence analysis revealed that loss of only RAD51 resulted in a global decrease in R-loop abundance, while depletion of SNF2, DH, and RAD51-3 led to a global increase. Genome-wide mapping of R-loop distribution in RAD51 mutants using DRIP-seq indicated depletions in R-loops at genomic sites including VSG-associated 70-bp repeats. Moreover, using Breaks Labelling *In Situ* and Sequencing (BLISS) we found pronounced levels of DNA breaks that localise to the 3' end of the expressed VSG and become more abundant in RAD51 mutants. Our data reveal multiple unexplored activities that may influence R-loop function in the *T. brucei* genome and provide a mechanistic link between R-loops and the parasite's ability to evade host immunity through VSG switching.

Briggs, E., Crouch, K., Lemgruber, L., Lapsley, C., & McCulloch, R. (2018a). Ribonuclease H1-targeted R-loops in surface antigen gene expression sites can direct trypanosome immune evasion. *PLoS Genetics, 14*(12). doi:10.1371/journal.pgen.1007729

Briggs, E., Hamilton, G., Crouch, K., Lapsley, C., & McCulloch, R. (2018b). Genome-wide mapping reveals conserved and diverged R-loop activities in the unusual genetic landscape of the African trypanosome genome. *Nucleic Acids Research, 46*(22), 11789-11805. doi:10.1093/nar/gky928

Session 8 – Wed 23rd Mar 15:50 - 17.00

Day 3 - Parasite Biochemistry *(Lecture Theatre K/018)*

23-March-2022, at 15:50 to 16:40

Chairs - Prof Anthony Wilkinson & Dr Michael Plevin

15:50 (10 mins) - A26146 - Biochemical investigations revealed the inhibitory mechanisms of novel inhibitors of Trypanosome Alternative Oxidase active against human and animal African trypanosomiasis

Authors

GU Ebiloma[3]; H de Koning[1]; C Dardonville,[2];

[1] Institute of Infection, Immunity and Inflammation, University of Glasgow, UK; [2] Instituto de Química Medica, IQM-CSIC, Spain; [3] School of Health and Life Sciences, Teesside University, UK

Discussion

Abstract

African trypanosomiasis caused by the protozoan parasite, *Trypanosoma brucei* is a neglected parasitic disease of public health importance and undermine food security in endemic areas, and parasites' resistance to the available drugs is widespread. Nevertheless, the pathogens possess certain unique metabolic features amenable to developing new efficient drugs. Particularly, they rely on an indispensable, mitochondrially-localized enzyme, Trypanosome Alternative Oxidase (TAO), which is involved in the respiration of the bloodstream form trypomastigotes of the parasite. Interestingly, TAO is absent in the mammalian hosts and hence an attractive target for designing safe trypanocides. We recently cloned, expressed, and purified the physiologically relevant form of TAO, which is devoid of the N-terminal 25 amino acid mitochondrial targeting sequence (ΔMTS-TAO). A newly designed and synthesized class of cationic and non-cationic 4-hydroxybenzoate and 4-alkoxybenzaldehyde inhibitors enabled the first structure-activity relationship (SAR) studies on ΔMTS-TAO. Remarkably, we obtained compounds with *in vitro T. brucei* inhibition of up to 1.4 nM and enzyme inhibition values (IC50) as low as 2 nM, which were also active against multidrug-resistant strains of *T. brucei* and *T. congolense*. The inhibitors designed with a mitochondrion-targeting lipophilic cation tail displayed trypanocidal potencies comparable to the reference drugs diminazene and pentamidine and exhibited no cross-resistance with the critical diamidine and melaminophenyl arsenical based trypanocides. The cationic inhibitors were also much more selective over human cells than the non-targeted neutral derivatives. A preliminary *in vivo* study showed that modest doses of the inhibitors were effective against parasitaemia of mice infected with *T. b. rhodesiense* (STIB900).

Conclusion

We have successfully developed a new class of potent and selective hits active against veterinary (*T. congolense*) and human (*T. brucei spp*.) African trypanosomes and confirmed their designed mode of action as inhibition of TAO using a combination of chemical and biochemical tools. This was achieved by efficiently targeting the compounds to the parasite's mitochondrion, thus increasing the potency of the original small molecule inhibitors against *T. brucei*. These compounds represent a promising new class of potent and selective hits against African trypanosomes.

Key words: SHAM, Triphenylphosphonium salt (TPP), Quinolinium salt, Lipophilic cation, Trypanosomiasis, Trypanocide, Mitochondrial targeting, Parasite respiration, Trypanosome alternative oxidase (TAO), *Trypanosoma brucei*, T. b. rhodesiense, T. *congolense*.

16:00 (10 mins) - A25983 - Investigating the role of glycosylation in *Toxoplasma gondii* protein homeostasis

Authors

G Bandini[2]; RW Meek[3]; JC Samuelson[1]; CM West[4]; GJ Davies[3];

[1] Department of Molecular and Cell Biology, Boston University, United States; [2] York Biomedical Research Institute, Department of Biology, University of York, UK; [3] York Structural Biology Lab, Department of Chemistry, University of York, UK; [4] Department of Biochemistry and Molecular Biology, University of Georgia, UK

Discussion

Protein glycosylation is one of the most abundant and widespread post-translational modifications (PTMs). This PTM class is involved in many biological processes including host-pathogen interactions and protein quality control and therefore plays a role in many key aspects of parasite biology.

Toxoplasma gondii is an opportunistic pathogen of humans that is estimated to infect up to 30% of the world population. Different glycosylation pathways have been shown to affect the kinetics of protein folding and stabilisation in the parasite. *O*-fucosylation of nucleocytoplasmic proteins by *Tg*SPY, a paralog of host *O*-GlcNAc transferase (OGT), is one of these pathways. This modification affects protein steady state levels, resulting in slower parasite replication and differentiation to the chronic stage *in vitro*.

Additionally, complementation of *spy*-deficient parasites with specifically selected mutants allows the study of the biochemistry and evolution of this family of glycosyltransferases in a cell system, highlighting the importance of divergent protozoa as model organisms.

16:10 (10 mins) - A26309 - The disorderly behaviour of Leishmania hydrophilic acylated surface proteins (HASPs)

Dr Michael Plevin
University of York
TBD

Turbo Talks – 5 min each talk –
23-March-2022, at 16:55 to 17:05

16:55 (5 mins) - A26116 - Biophysical and biochemical characterisation of the interaction between *Leishmania braziliensis* PRMT1 and PRMT3

Authors
E Nay[1]; PB Walrad[2]; MJ Plevin[1];
[1] University of York, UK; [2] University of York, Centre for Immunology and Infection, UK
Discussion
Arginine methylation is a key post-translational modification that can alter the structure, dynamics and interaction profiles of proteins. Protein arginine methyltransferases (PRMTs) catalyse the transfer of a methyl group from a *S*-adenosylmethionine molecule onto the arginine side chain guanidino group. Mammalian PRMTs are classified into subtypes – PRMT1, 2, 3, 4, 6 and 8 catalyse asymmetric dimethylation (ADMA); PRMT5 and 9 catalyse symmetric dimethylation (SDMA); and PRMT7 catalyses monomethylation (MMA). Kinetoplastids possess five homologues: PRMT1, 3, 5, 6 and 7. *T. brucei* PRMT3 has been shown to be a pro-enzyme (prozyme) which lacks key conserved motifs including in the catalytic double E loop. *T. brucei* PRMT1 is only active in complex with the PRMT3 prozyme. In *Leishmania*, however, PRMT3 retains the conserved double E loop, which raises questions about its role in this organism. Here we use recombinant protein samples to investigate *L. braziliensis* (*Lbr*) PRMT1 and 3 *in vitro*. Activity assays show that methylation of a substrate peptide only occurs when PRMT1 and 3 are both presence. Analytical size exclusion chromatography (SEC) and SEC-MALLS show that *Lbr*PRMT1 and 3 form a heterotetrameric complex in solution. Mutation of double E loop residues revealed that *Lbr*PRMT1 is the active component of the complex. Previous work suggested *Lbr*PRMT3 could interact with and modulate the activity of other *Lbr*PRMTs, however methyltransferase assays showed that *Lbr*PRMT3 had no effect on the activities of *Lbr*PRMT5 and 7 *in vitro*. Moreover, *Lbr*PRMT3 could not methylate a peptide substrate previously monomethylated with PRMT7. Our data suggests that *Lbr*PRMT1 and 3 form a similar complex to *T. brucei* PRMT1-3. However, the retention of the conserved double E loop in *Leishmania* PRMT3 enzymes suggests an as of yet undiscovered functional difference between the two trypanosmatids.

17:00 (5 mins) - A26002 - Investigating a galactokinase orthologue from *Leishmania donovani*

Authors
H Baber[1]; E King[1]; M Maciej-Hulme[2]; H Price[1]; **A Winter**[1];
[1] Keele University, UK; [2] Radboudumc, Netherlands
Discussion
Leishmania donovani is the causative organism of visceral leishmaniasis. We have identified an enzyme in *L. donovani* known as a galactokinase-like protein (*Ld*GalK), which could be a novel target for drug development. In the human host, two GalK paralogues are expressed, GalK1 and GalK2, which metabolise galactose in the Leloir pathway and N-acetyl galactosamine pathway, respectively. Both kinases instigate the first committed step in their respective pathways and phosphorylate the carbon-1 position in their carbohydrate ligand.
We have expressed recombinant *Ld*GalK in *E. coli* and purified the protein to high purity and yield. The recombinant enzyme is catalytically active with a substate affinity to galactose in the low micromolar range. Interestingly, size exclusion chromatography of *Ld*GalK suggests either an open/closed conformation of the enzyme or dimerization. Future research will test potential inhibitors against recombinant *Ld*GalK in vitro and against *L. donovani* parasites in vivo.

Day 3 - Parasite ImmunoPathology *(Lecture Theatre T/005)*
23-March-2022, at 15:50 to 16:40
Chairs - Dr Damian Perez Mazliah & Jillian Barlow

15:50 (10 mins) - A26163 - Investigating the roles of the cell regulator TRIM24 during visceral leishmaniasis

Authors
E Muscutt[1]; E Myburgh[1]; PM Kaye[1];
[1] University of York, UK
Discussion
Visceral leishmaniasis (VL) is a neglected tropical disease caused by infection with protozoan parasites *Leishmania donovani* and *L. infantum*. More than 95% cases of VL are fatal if left untreated, and current treatments are limited, expensive, and toxic, and there are currently no vaccines available. Macrophages are essential for the pathogenesis of VL, as *Leishmania* parasites modulate signalling pathways within macrophages to switch off their anti-parasite phenotypes, thereby avoiding their destruction while persisting in this niche. However, the mechanisms by which this occurs remain poorly understood. Recently TRIM24, a member of the tripartite motif protein family and a previously identified regulator of interferon STAT signalling, was predicted to be differentially expressed during VL. In this study we investigate the immune roles of TRIM24 in the steady state and during *L. donovani* infection by utilising TRIM24 knockout (KO) C57BL/6 mice. Immune characterisation of KO mice revealed TRIM24 to be dispensable for immune cell development *in vivo*, however generation of 50:50 mixed bone marrow chimeric mice led to a significant skew in favour of KO cells most notably in the bone marrow. Neutrophils, monocytes, macrophages, and B cells (but notably not T cells) were responsible for this selective advantage of KO cells, pointing to an interesting advantage specific to the bone marrow chimera system. *L. donovani* infection of bone marrow chimeras had little effect on this skew, and flow cytometry-based analysis of immune cells revealed little change in the release of the cytokines TNF, IFN-γ, IL-6 and IL-10 from T cells and macrophages/monocytes. This was reflected in *in vitro* studies

using bone marrow-derived macrophages (BMMs), where loss of TRIM24 did not affect release of TNF or IL-6. However, an increase in iNOS+ cells and release of nitric oxide (a pro-inflammatory mediator important for macrophage-mediated parasite clearance) was observed from KO BMMs. Furthermore, these BMMs released more interferon-beta, providing a potential mechanism for increased iNOS expression. Interestingly, no change in *Leishmania* parasite burden was seen between infected WT and KO mice. Further transcriptomic analysis on BMMs infected *in vitro*, and bone marrow from chimeric and total WT and KO mice will provide a deeper understanding of the roles of TRIM24 during *L. donovani* infection.

16:00 (10 mins) - A26155 - Mapping the immune response in schistosomiasis – insights from controlled human infection models.

Authors
E Houlder[1]; JP Koopman[1]; KA Stam[1]; MH König[1]; P Niewold[1]; JC Sijtsma[1]; JJ Janse[1]; A Diepen[1]; A Ozir-Fazalalikhan[1]; CH Hokke[1]; M Yazdanbakhsh[1]; M Casacuberta-Partal[1]; AS MacDonald[2]; M Roestenberg[1];
[1] Leiden University Medical Centre, Netherlands; [2] University of Manchester, UK

Abstract
Infection with the parasitic worm *Schistosoma mansoni* causes considerable global morbidity, affecting over 200 million people, particularly in sub-Saharan Africa. Due to difficulties with diagnosing and tracking early infections, often asymptomatic in endemic individuals, prior studies have not been able to precisely define immune responses in the initial weeks of schistosome infection. Here, we have utilised a pioneering controlled human schistosome infection model to investigate the early immune responses to single-sex male or female schistosomes. At 2 weeks post infection, we used flow cytometry to reveal a change in the myeloid compartment, with an increase in pulmonary dendritic cells during schistosome lung migration. Next, and coinciding with the first moderate or severe clinical manifestations of acute schistosomiasis syndrome, at week 4 post infection we saw evidence of an inflammatory immune response. Proteomic assessment of the serum revealed increases in type-1 messengers such as IFNγ and CXCL10, as well as activation of monocytes and $CD4^+$ effector memory T cells seen by mass cytometry (CyTOF). This inflammatory response had lessened by week 8, when we saw evidence of regulation, with intracellular cytokine staining showing an increase in IL-10 expressing $CD4^-CD8^-$ T cells, and a reduction in expression of the pro-inflammatory cytokine TNFα by T and B cells. Alterations in the B cell compartment at week 8 with an increase in $CD11c^+$ atypical memory B cells, observed by CyTOF, coincided with the appearance of anti-schistosome antibodies in the circulation, measured by antigen-specific ELISA. Notably, initial cytokine and antibody results suggest broad similarity in the clinical and immunological profiles of infection with single-sex male or female cercariae. Current and future work will use refined techniques (CyTOF, RNA-seq) to better understand responses post infection with female cercariae, to compare to the male infection model. This novel data elevates fundamental understanding of the development of immune responses during *S. mansoni* infection, providing clinically relevant insight into the pathology of acute schistosomiasis, and setting an immunological baseline to assess changes in future vaccine studies.

16:10 (10 mins) - A26183 - Chronic schistosome infection remodels bone marrow haematopoiesis

Authors
J Hewitson[1];
[1] University of York, UK

Discussion
Joanna Greenman, Grace Boyd, Jack Fisher, Moses Egesa, Alison Elliott, David Kent, James Hewitson People living in schistosome-endemic regions undergo repeated cycles of reinfection and drug clearance. To determine how chronic *Schistosoma mansoni* infection modifies responses to reinfection, we compared skin immune responses to S. *mansoni* challenge in mice that were chronically infected (12wks) with animals that were exposed for the first time. We found chronic infected leads to weak skin immune responses after rechallenge with marked reductions in macrophages, and that this is maintained after curative praziquantel treatment. As bone marrow (BM) chimera experiments revealed skin macrophages originate from infiltrating monocytes, we performed RNAseq on BM monocytes and found worm infection strongly impacts on gene expression in this site. We hypothesised distal worm infection impacts on bone marrow haematopoiesis by modifying the BM cytokine microenvironment. Cytokine Bead Array analysis showed chronic infection leads to elevated IL-4 (but not other type 2 cytokines) in BM aspirates. We next tested whether IL-4 can alter haematopoiesis and found haematopoietic stem cells (HSC) express Il4ra and in vitro assays (single cell cultures, colony formation assays) revealed HSC directly respond to this cytokine. Surprisingly then, the dominant signature of infection on immune progenitors (BM LSK cells) in vivo was instead interferon-related (type I and II). Using competitive bone marrow transplants, we found HSC from infected mice to be functionally impaired. We are now assessing the extent to which infection-induced changes to haematopoiesis persist after parasite clearance, and how this impacts on heterologous immune challenges in mice and humans.

Day 3 – Parasite Genetic Architecture *(Lecture Theatre P/X001)*

23-March-2022, at 15:50 to 16:40
Chairs - Dr Nathaniel Jones & Dr Joana Faria

15:50 (10 mins) - A26167 - RNase H1, a R-loop resolving enzyme, acts to suppress R-loop mediated DNA replication and limit genome instability in *Leishmania*

Authors
J Damasceno[3]; E Briggs[2]; JL Reis-Cunha[5]; K CrouchC Lapsley[3]; D Bartholomeu[4]; R McCulloch[3];
[1] University of Glasgow , UK; [2] University of Edinburgh, UK; [3] Wellcome Centre for Integrative Parasitology, Institute of Infection, Immunity and Inflammation, University of Glasgow, UK; [4] Federal University of Minas Gerais - UFMG, Brazil; [5] York Biomedical Research Institute, Department of Biology, University of York, UK

Discussion
Stable RNA-DNA hybrids (R-loops) act in several genomic processes, but their roles in DNA replication are unclear. By using DRIP-seq, we show that R-loop distribution within each chromosome parallels the spatial, temporal and functional compartments of the unconventional DNA replication programme in *Leishmania*. Strikingly, R-loop levels correlate with chromosome size, which in turn correlate with replication timing. MFA-seq analyses shows that DiCre-mediated *RNase H1* KO results in origin-independent DNA replication initiation, profoundly changing the DNA

replication programme. Such alteration leads to genome-wide, chromosome-size dependent instability, including aneuploidy, SNPs and InDels, as revealed by whole genome sequencing. Therefore, our data reveal a crucial role of RNase H1 in controlling R-loop-mediated DNA replication initiation, favouring conventional origin-directed initiation, and places the hybrids as a pivotal player in *Leishmania* global genome instability.

16:00 (10 mins) - A26143 - The open chromatin profile changes at genome compartments and at tDNA loci in *Trypanosoma cruzi* life formstract

Authors

A Lima[2]; S Poubel[2]; J Roson[2]; L de Lima[2]; H Costa-Silva[2]; HG Silva[2]; PC de Lima[2]; CS gonçalves[2]; PA Galante[6]; FB Holetz[4]; MC Motta[3]; AM Silber[1]; MC Elias[2]; **JP Cunha**[5];

[1] Biomedical Science Institute - USP, Brazil; [2] Instituto Butantan, Cell Cycle Special Lab, São Paulo, Brazil; [3] Laboratorio de Ultraestrutura Celular Hertha Meyer, Instituto de Biofısica Carlos Chagas Filho, Universidade Federal do Rio de Janeiro, Brazil; [4] Laboratório de Regulação da Expressão Gênica, Instituto Carlos Chagas, Fiocruz, Brazil; [5] BUTANTAN, Brazil; [6] Hospital Sirio Libanes, Brazil

AbstractGenomic organization and gene expression regulation in trypanosomes are remarkable because protein-coding genes are organized into codirectional gene clusters with unrelated functions. Moreover, there is no dedicated promoter for each gene, resulting in polycistronic gene transcription, with posttranscriptional control playing a major role. Nonetheless, these parasites harbor epigenetic modifications at critical regulatory genome features that dynamically change among parasite stages, which are not fully understood.Here, we investigated the impact of chromatin changes in a scenario commanded by posttranscriptional control exploring the parasite *Trypanosoma cruzi* and its differentiation program using genome-wide approaches supported by transmission electron microscopy. The integration of FAIRE and MNase-seq data, two complementary epigenomic approaches, enabled us to identify differences in *T. cruzi* genome compartments, putative transcriptional start regions, and virulence factors. In addition, we also detected developmental chromatin regulation at tRNA loci (tDNA), which seems to be linked to the translation regulatory mechanism required for parasite differentiation. Strikingly, a positive correlation was observed between active chromatin and steady-state and nascent transcription levels. Taken together, our results indicate that chromatin changes reflect the unusual gene expression regulation of trypanosomes and the differences among parasite developmental stages, even in the context of a lack of canonical transcriptional control of protein-coding genes. Supported by FAPESP, Serrapilheira, CAPES.

16:10 (10 mins) - A25926 - *Leishmania* Bromodomain Factor 5 is an Essential Transcriptional Regulator

Authors

NG Jones[3]; V Geoghegan[3]; G Moore[1]; J Carnielli[3]; K Newling[1]; F Calderon[2]; T Wilkinson[1]; J Mottram[3];

[1] University of York, UK; [2] GSK, Spain; [3] York Biomedical Research Institute, UK

Abstract*Leishmania* are unicellular parasites that cause human and animal disease. Alongside other organisms in kinetoplastida, they have evolved an unusual genome architecture that requires all RNA polymerase II transcribed genes to be expressed constitutively, with transcriptional start regions denoted by histone variants and histone lysine acetylation. However, the way these chromatin marks are interpreted by the cell is not understood. Seven predicted bromodomain factors (BDF1-7), the reader modules for acetyl-lysine, were identified across *Leishmania* genomes. Using *L. mexicana* as a model, Cas9-driven gene deletions indicate that BDF1-5 are essential for promastigote survival, whilst DiCre inducible gene deletion of the dual bromodomain factor *BDF5* identified it to be essential for both promastigotes and amastigotes. ChIP-seq assessment of BDF5s genomic distribution revealed it as highly enriched at transcriptional start sites. Using an optimised proximity proteomic and phosphoproteomic technique, XL-BioID, we defined the BDF5-proximal environment to be enriched for other bromodomain factors, histone acetyltransferase 2, and proteins essential for transcriptional activity and RNA processing. Inducible deletion of BDF5, led to a disruption of pol II transcriptional activity and global defects in gene expression. Our results indicate the requirement of *Leishmania* to interpret histone acetylation marks for normal levels of gene expression and thus cellular viability.

16:20 (10 mins) - A25994 - A kinetoplastid-specific subunit of the Origin Recognition Complex?

Authors

CA Marques[2]; MS Da Silva[3]; JM Batista[2]; C Tiengwe[1]; R Burchmore[4]; R McCulloch[2];

[1] Dept. Life Sciences, Imperial College, University of London, UK; [2] Wellcome Centre for Integrative Parasitology, Institute of Infection, Immunity and Inflammation, University of Glasgow, UK; [3] Department of Chemical and Biological Sciences, São Paulo State University, Brazil; [4] Institute of Infection, Immunity and Inflammation and Glasgow Polyomics, University of Glasgow, UK

Discussion

Nuclear DNA replication is initiated at specific genomic loci termed origins. In eukaryotes, these sites are defined by the binding of the six-subunit Origin Recognition Complex (ORC). Frequently described as a well-conserved complex, phylogenetic studies querying a wide variety of species from across the eukaryotic tree suggest that many may have lost one or more ORC subunits. In *Trypanosoma brucei*, four ORC subunits (ORC1/CDC6, ORC4, ORC5 and ORC2) have been identified to date, most of which diverge considerably from their eukaryotic orthologues. The two remaining canonical subunits, Orc3 and Orc6, remain unidentified, and it is unclear whether a six-subunit ORC exists in *T. brucei*.

To elucidate the composition of TbORC, we have resorted to two main approaches: bioinformatics and experimental. Using publicly available genome and transcriptome datasets, we have run extensive sequence-based homology searches. Orc3 orthologues were detected in most analysed Discoba lineages but not in kinetoplastids, suggesting that this subunit might be missing in this group. Immunoprecipitation of multiple TbORC subunits resulted in the common identification of Tb1120 (Tb927.6.1120), and gel filtration analysis showed that Tb1120 elutes in fractions of similar molecular weight to those of other TbORC subunits. Moreover, RNAi depletion resulted in severe growth arrest, cell cycle de-regulation, and almost complete abrogation of DNA replication, akin to the effects of depletion of other TbORC subunits. Protein domain searches and *de novo* structure predictions suggest that the only similarity between Tb1120 and canonical ORC subunits is a putative C-terminal winged helix-turn-helix domain (commonly associated with DNA binding and/or protein-protein interactions). Sequence-based homology searches failed to detect evidence for a relationship between Tb1120 and any known ORC subunit, and were unable to find any orthologues of Tb1120 outside of the kinetoplastids, raising the hypothesis of it replacing Orc3 in TbORC. If correct, incorporation of a kinetoplastid-specific ORC subunit may have necessitated wider reorganisation of the kinetoplastid ORC, consistent with the considerable divergence in ORC2 and ORC5 as described

previously. The biological reasons behind kinetoplastids incorporating a putative novel subunit into their ORC are unknown, but are the subject of ongoing studies.

Day 4 – Thu 24th Mar 2022 -

Session 9 – Thu 24th Mar
10:00 - 11.20 for '*Combative Strategies: Drug Discovery*', '*Parasite Cell Biology II', and CHIM workshop: Controlled Human Infection Models;*
10:30 – 11:20 for *'Diversity in Science II: Conversations toward inclusion and equityI'.*

Day 4 - Combative Strategies: Drug Discovery *(Lecture Theatre K/018)*
24-March-2022, at 10:00 to 11:00
Chair - Dr Elmarie Myburgh

10:00 (20 mins) - A26231 - A platform for drug target deconvolution and exploitation

Susan Wyllie
University of Dundee

Discussion

The development of new drugs to treat kinetoplastid and many other infectious diseases has been hampered by a severe lack of robustly validated drug targets. This has left drug discovery programs heavily reliant upon phenotypic screening to identify suitable chemical start points. Development and optimisation of phenotypically-active compounds is also hindered by lack of information regarding mechanism(s) of action (MoA) and molecular target(s). Specifically, knowledge of molecular targets is often crucial in developing strategies to overcome issues such as poor pharmacokinetics and toxicity. When targets of phenotypically-active compounds are identified, target- and structure-based drug discovery programmes can be initiated allowing optimisation based on selectivity over human orthologues. Thus, MoA studies can effectively integrate these two, often disconnected, approaches to drug discovery. Furthermore, understanding the MoA is critical in developing appropriate combination therapies of the future. Historically, MoA studies have been of secondary consideration for drugs being developed for NTD. If these studies were carried out at all, they were initiated after the development of pre-clinical or clinical candidates. Over the last 6 years, my group have developed an integrated drug target deconvolution platform, employing a range of established and new methodologies encompassing high-throughput genetics, cell biology and chemical proteomics. Using this platform our aim is to provide MoA input and information in real-time for on-going drug discovery programmes. This enables the data we provide to guide and indeed drive the evolution of the best possible drug candidates. Details of my group's integrated approach to drug target identification in kinetoplastids will be provided and I will describe how we are transitioning this approach to study MoA in Cryptosporidium, *Plasmodium* and *Schistosoma*.

10:20 (10 mins) - A26109 - SMGBs as novel in vitro and in vivo anti-infective agents for *Acanthamoeba* spp. infections.

Authors
A Carpinteyro Sanchez[2]; FJ Scott[1]; CW Roberts[2];
[1] University of Strathclyde, UK; [2] Sipbs University Of Strathclyde, UK

Discussion

Acanthamoeba spp. are causative agents of a painful and severe sight-threatening corneal infection that can lead to blindness known as *Acanthamoeba* keratitis and a subacute disease in the brain which is usually fatal known as granulomatous amoebic encephalitis. Over the last few years, there has been a notorious increase in the number of infections due to *Acanthamoeba* spp. Poor diagnosis, problems of side effects and toxicity of the current drug treatment contribute to a high mortality rate. Strathclyde Minor Groove Binders (S-MGBs), compounds that bind to the minor groove of the DNA that designed and synthesised at University of Strathclyde were evaluated as potential alternative inhibitors against *Acanthamoeba* infections. Through cell viability microplate alamarBlue assays 42 S-MGBs were screened from which a library of 6 showed potent active inhibitory effect with half maximal inhibitory concentration (IC_{50}) below 1 µM. S-MGB 235 showed the most potent inhibitory effect with IC_{50} in the nanomolar range against five *Acanthamoeba* isolates after 24 h and 96 h incubation. Confocal microscopy of trophozoites labelled with fluorescent S-MGB 363 (analogue of S-MGB 235) showed this compound in the nucleus, nucleolus and distributed over the granuloplasm causing cell lysis, supporting the potent effect observed *in vitro* by S-MGB 235. Furthermore, conditions were standardised to establish *Galleria mellonella* larvae as a new *in vivo* infection model for *A. castellanii* Neff infections to assess the efficacy and toxicity of voriconazole, miltefosine and S-MGB 235. Voriconazole and miltefosine did not protect larvae from trophozoite infection, however S-MGB 235 significantly protected larvae when compared with the negative control. Preliminary results show S-MGB 235 as cysticidal with minimum inhibitory concentration (MIC) ranging 0.5-1 µM in most of the strains, which is a potent inhibitory effect in low concentrations compared to hexamidine and PHMB, both drugs widely used in the current clinical treatment. Given the above, we suggest S-MGB-235 as a relevant novel compound that might complement aromatic diamidines in the current combination therapies for *Acanthamoeba* spp. diseases.

10:30 (10 mins) - A26085 - First Insights into the Autophagy Machinery and its Induction by Imatinib in *Schistosoma mansoni*

Authors
MN Mughal[1]; CG Grevelding[1]; S Haeberlein[1];
[1] Institute of Parasitology, Biomedizinisches Forschungszentrum Seltersberg, Justus Liebig University, 35392, Giessen, Germany, Germany

Discussion

Schistosomiasis is a neglected tropical disease caused by blood flukes (schistosomes) of the genus *Schistosoma*. This debilitating and chronic disease is a significant veterinary and public health problem with > 200 million people infected. The main pathogenicity is caused by eggs laid by the female worm, which requires the female to be in a constant pairing with a male partner. We hypothesize a role for a fundamental cellular process, autophagy, in the regulation of key processes in schistosome biology.

Autophagy is activated during starvation or cellular stress and contributes to maintain homeostasis. Although autophagy is known as essential pathway involved in regulating cell survival, reproduction, organ and body reshaping in various organisms, autophagy has been basically neglected in schistosome research. Here, for the first time, we shed light on the autophagy machinery, its involvement in reproduction of *Schistosoma mansoni*,and its suitability as anti*Schistosoma*l therapeutic target. We identified autophagy genes by *in-silico* analyses and quantified their transcript level by qRT-PCR in female and male worms prior and after *in vitro* culture. Furthermore, worms were treated with autophagy inhibitors (bafilomycin A1, wortmannin and spautin-1) or an autophagy inducer (rapamycin) to evaluate effects on worm vitality and reproduction as well as autophagy protein expression. Among the identified autophagy genes were Beclin, Ambra1, Vps34, Dram, DAP1, and LC3B. The damage-regulated autophagy modulator DRAM was significantly higher transcribed in males compared to females, while for the death-associated protein DAP1 it was the opposite. The conversion of the autophagy protein LC3B, a key marker for autophagic activity, was impaired by bafilomycin A1 but induced by rapamycin. All autophagy inhibitors negatively affected worm fitness and egg production as well as the morphology of gonads and intestine.
An anticancer drug, imatinib (Gleevec), drastically affected intestinal morphology and caused death of adult worms within 3-4 days *in vitro* (already published). We present first evidence that imatinib induces autophagy in adult *S. mansoni*, which was evident from significant increase of LC3B. Interestingly, the drastic effects induced by imatinib on pairing stability, egg production, and gut dilatation were mitigated by autophagy inhibition using bafilomycin A1.
Our study demonstrates that autophagy genes in *S. mansoni* not only show an interesting sex-dependent expression pattern, which might point to developmental or reproductive roles, but when disrupted by chemical inhibition, can affect parasite viability and reproduction.

10:40 (10 mins) - A26156 - How do Schistosomes Breathe?

Authors
AM Burgess[1]; I Chalmers[1]; K Hoffmann[1];
[1] Aberystwyth University - IBERS, UK

Discussion

Schistosoma mansoni is a blood fluke species responsible for schistosomiasis. Human infection is currently treated by a single drug, praziquantel. While praziquantel's mechanism of action has recently been revealed to operate through a transient receptor potential melastatin ion channel, drug resistant fears remain and fuel the search for alternative chemotherapies. As basic investigations of S. *mansoni* biology and biological processes could reveal vulnerabilities suitable for drug discovery, we present a functional genomics characterisation of two S. *mansoni* gene products putatively involved in energy metabolism. One gene product (SmFHII) is homologous to human class II fumarase while the second (SmFHI) is more closely related to the distinct class I fumarase commonly associated with prokaryotes. These enzymes are active in the mitochondria (TCA cycle), cytosol (urea cycle) and nucleus (DNA-damage repair). BLASTp analyses of SmFHI and SmFHII homologues revealed that only the lophotrochozoans possess a class I fumarase, while class II fumarases can be found in all other representative metazoan groups examined. The predicted localisation of SmFHI and SmFHII were determined using TargetP and revealed that SmFHI is predicted to possess a mitochondrial targeting peptide and likely responsible for mitochondrial activity. Relative expression of these genes across the human infective lifecycle stages was determined by qRT-PCR and showed that both genes are highly expressed within this crucial part of the lifecycle. Finally, RNA interference (RNAi) studies revealed that knockdown of both genes (SmFHI AND SmFHII) led to a defect in adult worm motility; single knockdown of SmFHI OR SmFHII did not significantly reduce this phenotype. These results suggest that S. *mansoni* fumarases can perform compensatory functions/activities and that these (e.g. respiration, DNA-damage repair or urea cycle) are involved in adult worm motility. Total oxygen consumption in RNAi treated worm cultures is currently being measured to quantify how loss of fumarase function affects respiration. RNAi of SmfhI and SmfhII will be performed on schistosomula to determine how these genes affect in vitro development, phenotype and motility. Additionally, the use of known fumarase class II inhibitor 2,3-Dicarboxyaziridine in tandem with predicted class I inhibitors are to be tested on adult worms and schistosomula. Together, these results will reveal the importance of SmFHII and SmFHI to schistosome viability, respiration and development.

Day 4 - Parasite Cell Biology II *(Lecture Theatre P/X001)*

24-March-2022, at 10:00 to 11:00
Chairs - Dr James LaCourse & Dr Eden Ramalho Ferreira

10:00 (10 mins) - A26093 - The unique mRNA decapping enzyme ALPH1 of trypanosomes

Authors
PA Castañeda Londoño[1]; N Banholzer[1]; J Odenwald[1]; C Moreira[1]; M Zoltner[2]; SK Kramer[1];
[1] Biocentre, Department of Cell and Developmental Biology, University of Würzburg, Germany; [2] BIOCEV, Department of Parasitology, Charles University, Czech Republic

Discussion

The 5′ ends of eukaryotic mRNAs are co-transcriptionally modified with a protective m^7G cap structure. In the course of RNA turnover, this cap is removed by the major eukaryotic mRNA decay enzyme, the nudix hydrolase Dcp2, that is part of the decapping complex. Trypanosomes lack homologs to all proteins of the decapping complex but instead use the ApaH-like phosphatase TbALPH1: this is unique to Kinetoplastida. ApaH like phosphatases (ALPHs) are a bacterial-derived class of enzymes present in all eukaryotic super-groups, but absent in mammals.
TbALPH1 is essential for cultured trypanosome cells and consists of the catalytic domain and unique C- and N-terminal extensions. We found that ALPH1 has *in vitro* decapping activity in a wide range of conditions without cap-type preference and, surprisingly, even in the absence of its C- and N-terminal domains. We found no evidence for RNA binding activity, consistent with the absence of any known RNA binding domains and the fact that ALPH1 accepts cap analogues as a substrate too. The N-terminal domain is dispensable even *in vivo*, but needed for localization of ALPH1 to the posterior pole of the cell. BioID data suggest that the C-terminus is required for interaction with the 5′ to 3′ exoribonuclease XRNA, the enzyme that acts downstream of ALPH1.
ALPH1 is one of only two eukaryotic ApaH like phosphatases with a known function and the only mRNA decapping enzyme that does not belong to the nudix hydrolase family. A better understanding of its mechanism and regulation is essential and our data are an important contribution. Moreover, the absence of ApaH like phosphatases in mammals rises the possibility that ALPH1 can be exploited as a drug target and we are currently investigating this option.

10:00 (20 mins) - A26264 - Intrabody induced cell killing by targeting an essential cytoskeletal protein in *T. brucei*

Abstract*Trypanosoma brucei* is a protist parasite. It belongs to a genus of pathogens that cause life-threatening and economically important diseases of human and animals in Sub-Saharan Africa. Exo-/endocytotic trafficking in these cells occurs *via* a single copy organelle called the flagellar pocket (FP). The FP is maintained and enclosed around the flagellum by the flagellar pocket collar (FPC). To date, the most important cytoskeletal component of the FPC is an essential, calcium-binding, polymer-forming protein called TbBILBO1. In searching for novel immune-tools to study this protein, we raised nanobodies (Nb), against TbBILBO1. Nanobodies were selected according to their binding properties to TbBILBO1, and were tested as immunofluorescence tools and expressed as intrabodies (INb). Nb48, proved to be the most robust nanobody and intrabody. We further demonstrate that inducible, cytoplasmic expression of INb48 was lethal to these parasites, producing abnormal phenotypes resembling those of TbBILBO1 RNAi knockdown. Our results validate the feasibility of generating functional single-domain antibody derived intrabodies to target trypanosome cytoskeleton proteins and potentially other non-trypanosome proteins.

10:20 (10 mins) - A26265 - Characterisation of the essential trypanosome protein TbSmee1 reveals that endocytosis is required for flagellar pocket access of surface-bound cargo (Brooke Morriswood)

Abstract

All endo- and exocytosis in the African trypanosome *Trypanosoma brucei* occurs at a single point on its plasma membrane. This plasma membrane subdomain, the flagellar pocket, is a small vase-shaped invagination containing the root of the cell's single flagellum. A number of cytoskeleton-associated multiprotein complexes are coiled around the neck of the flagellar pocket on its cytoplasmic face. One of these, the hook complex, has been proposed to affect the entry of cargo into the flagellar pocket. Previous characterisation of the hook complex component TbMORN1 showed that its depletion resulted in an apparent size exclusion effect, with larger cargo now unable to enter the flagellar pocket. In this study, the hook complex component TbSmee1 was characterised in bloodstream form *Trypanosoma brucei*.TbSmee1 localised to both the hook complex and the flagellum attachment zone tip of replicating cells, with different parts of its primary structure mediating targeting to each. Depletion of TbSmee1 by RNAi was lethal, and resulted in an enlargement of the flagellar pocket. Like TbMORN1, depletion of TbSmee1 was found to affect the entry of cargo, with small fluid-phase cargo still able to enter the flagellar pocket lumen, but larger surface-bound molecules trapped outside. Unexpectedly, the same effect was also produced by depletion of clathrin, suggesting that endocytic activity is a prerequisite for the entry of surface-bound cargo into the flagellar pocket.

Day 4 - Diversity in Science II: Conversations toward inclusion and equity *(Lecture Theatre T/005)*
24-March-2022, at 10:30 to 11:20
Chair - Dr Giulia Bandini

10:30 (20 mins) - A26330 - Women in science: a picture of the Brazilian scenery

Prof Angela Kaysel Cruz
University São Paulo, Ribeirao Preto

Discussion

Gender equity in academia is a worldwide issue. In Brazil, women are underrepresented in science, and their participation decreases as the career progresses. Different explanations for the observed decrease include maternity or lower scientific abilities. I will present studies conducted by others and some numbers associated with the gender inequities in the Brazilian science and technology system. These studies point to a combination of barriers leading to the decrease of women as the career advances. The diagnoses may vary, and so possible improvements in the system, but it is urgent to face the problems and difficulties to overcome them. Academic institutions and individuals must embrace concrete solutions to promote equity in academia, and some of the activities towards improving the current situation will be discussed.

10:50 (20 mins) - A26331 - Is Open Access Inclusive?

Prof Ariel Silber
University of São Paulo

Discussion

A reflection about the OA policies by Alicia Kowaltowski, Marcus Oliveira and Ariel M. Silber (alphabetical order) Open Access emerged as an idea by the first time in the decade of 1970. However, it was not structurally implemented until 2001, when several editorial groups embraced its main principles. In fact, these principles were idealistic and looked in line with a more equalitarian world. Open access is based on two main principles: i. all the published scientific information should be available for everyone regardless he/her status of pertaining to institutions subscribed to the venues where these information is published; ii. the **Authors** should keep the copyrights of their intellectual production. To make economically possible this new paradigm, it was broadly agreed that **Authors** (or their funders) should pay to the editors the Article Processing Charges (APCs) to cover the costs of publishing and making the information available for anyone for free. However, most APCs are increasing beyond inflation indexes, and more **Authors** excluded from publishing their own work in reputed journals due to overpricing and unfair policies of waivers. We are witnessing a process that is dividing the academics all over the world among those that have the resources to freely publish their research in the most reputed journals regardless the APC costs, and those that are not able to publish in reputed journals because they are not able to afford the (many often abusive) APCs. Despite the initial good intentions of the Open Access movement, in its current format, it is making science less (and no more) inclusive.

Session 10 – Thu 24th Mar 11:50 - 13.00.
Day 4 - Combative Strategies: Drug Discovery *(Lecture Theatre K/018)*
24-March-2022, at 11:50 to 12:40
Chair - Dr Elmarie Myburgh

11:50 (10 mins) - A25951 - Investigating Bromodomain Proteins as Targets for Anti-Leishmanial Drug Discovery

Authors

C Russell[1]; NG Jones[2]; AJ Wilkinson[1]; J Mottram[1];

[1] University of York, UK; [2] York Biomedical Research Institute, UK

Abstract

A key element within the drug discovery pipeline for anti-leishmanial therapies is the identification and validation of new target proteins in *Leishmania*. Bromodomains (BDs) are epigenetic protein domains that recognise acetylated lysine residues in histone tails and regulate gene transcription through this interaction. BDs have been implicated in various diseases, which has opened up a field of BD inhibitor research. These efforts have been particularly rewarding with regard to cancer research, with several BD inhibitors entering clinical trials. These encouraging results provide rationale for the selective targeting of BDs in other diseases, such as leishmaniasis. Genetic target validation has identified *Leishmania* bromodomain factor 5 (BDF5) as essential for parasite survival. BDF5 contains two BDs, BDF5.1 and 5.2, and is hypothesised to play a role in global transcriptional regulation. Using recombinant protein production, BDF5 bromodomains have been expressed at York and screened by the pharmaceutical company GSK against small molecules and reactive compounds. This yielded numerous hit compounds which have been distributed to York for validation and characterisation in biophysical assays alongside various other compounds. Thermal shift assays (TSA), NMR and fluorescence polarization (FP) assays have been developed to validate these hits for on-target engagement using recombinant BDs. These assays have also been used to test binding of histone peptides to the *Leishmania* BDs, to investigate the acetylated histone sequence with which *Ld*BDF5 interacts. Together, this work provides a platform for the validation and characterisation of hit compounds against the *Leishmania* BDs. With the support of genetic evidence of BDF protein essentiality, these approaches aim to establish these epigenetic proteins as targets for further anti-leishmanial drug discovery and development.

12:00 (10 mins) - A26178 - The development of an oral oleylphosphocholine treatment for cutaneous leishmaniasis

Authors

K Van Bocxlaer[1]; D Van Den Heuvel[4]; H Platteeuw[4]; K McArthur[5]; A Harris[5]; M Alavijeh[5]; SL Croft[2]; V Yardley[3];

[1] University of York, UK; [2] London School of Hygiene & Tropical Medicine, UK; [3] London Schhool of Hygiene & Tropical Medicine, UK; [4] Avivia Bv, UK; [5] Pharmidex Pharmaceuticals, UK

Abstract

Introduction

With an estimated 0.7 to 1 million new infections a year globally, cutaneous leishmaniasis (CL) is the most prevalent form of leishmaniasis; it clinically manifests as a variety of skin lesions ranging from closed nodules, to plaques and ulcers. Currently recommended drugs have proved to be clinically unsatisfactory indicating the urgent need for novel safe and efficacious drugs. Oleylphosphocholine, an alkylphoshoplipid structurally similar to miltefosine, demonstrated potent activity against *Leishmania* species causing visceral leishmaniasis (VL) both *in vitro* and *in vivo*. Given the discrepancies between the target product profiles of VL and CL, we here report the *in vitro* and *in vivo* efficacy of orally administered oleylphosphocholine-based formulations (two with a fast-release and two with a slow release profile) against CL-causing *Leishmania* species.

Materials and methods

The antileishmanial activities of OLPC and miltefosine were evaluated against intracellular amastigotes of six *Leishmania* species (*L. major*, *L. tropica*, *L. aethiopica*, *L. mexicana*, *L. braziliensis*, *L. panamensis*). Following promising results, the *in vivo* efficacies of both drugs were investigated in two stages. First, the performance of the efficacious dose for OLPC for VL was evaluated using an experimental CL model. Secondly, the antileishmanial activity of various formulations of OLPC with diverse release profiles was investigated alongside a dose response using bioluminescent *L. major* parasites. Tissue concentrations in skin of OLPC were determined using LC-MS/MS.

Results and Discussion

The *in vitro* activities of OLPC against CL-causing species ranged from 0.74 to 31.06 uM and are similar to those obtained for miltefosine. In the experimental CL models, OLPC administered orally at a dose of 35 mg/kg once daily for ten days was able to significantly reduce the lesion size to a similar extend as the positive control (paromomycin sulphate, ip, 50 mg/kg/day – repeated-measures ANOVA, post-hoc Tukey, $p < 0.05$). In contrast, the administration of miltefosine (same dose and regimen as OLPC) only resulted in a halt of the lesion size progression but was unable to decrease the lesion diameter. The second *in vivo* study was able to confirm these results and demonstrated a superior activity of the fast- (OLPC with lactose or cellulose carrier) over the slow-release (OLPC absorbed into a diffusion-controlled silica carrier) test formulations as measured by a significantly greater bioluminescence signal (~ parasite load) decrease when compared to the untreated controls. Extraction of the drugs from the infected skin site 24 hours after the oral administration of 1 dose (35 mg/kg) demonstrated higher concentrations of OLPC versus miltefosine (t-test). This difference was no longer present at the end of the 10-day treatment period even though OLPC blood concentrations at the end of treatment were 2-fold higher than for miltefosine.

Conclusions

OLPC demonstrated potent activity in the intracellular macrophage model using a range of CL-causing species and was able to reduce the parasite load in an experimental *L. major* CL model after ten days of treatment. In a next step, the drug delivery profile into *Leishmania*-infected and uninfected mouse skin will be compared using skin microdialysis.

Funding

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12:10 (10 mins) - A26102 - CPSF3 and beyond…a Systematic View of the Mode-of-Action of Benzoxaboroles in African Trypanosomes

Authors

N Zhang[3]; M Zoltner[2]; Y Freund[4]; M Alley[4]; MP Barrett[1]; D Horn[3]; MC Field[3];

[1] Wellcome Centre for Molecular Parasitology, Institute of Infection, Immunity and Inflammation, University of Glasgow, UK; [2] BIOCEV, Department of Parasitology, Charles University, Czech Republic; [3] Wellcome Centre for Anti-Infectives Research, Division of Biological Chemistry and Drug, University of Dundee, Dundee, UK, UK; [4] Anacor Pharmaceuticals, Inc., Palo Alto, California, U. S. A., United States

Discussion

Benzoxaboroles (BOBs) offer new opportunities for the pharmaceutical invention against a broad spectrum of infectious diseases including human and animal African trypanosomiasis. CPSF3/CPSF73, the sole catalytic component of the pre-mRNA 3'-end cleavage and polyadenylation specific factor complex (CPSF), has been proposed as the primary target of BOBs in various protozoan pathogens, raising a possibility of common targeting mechanism. We demonstrated a rapid increase in the global protein SUMOylation upon BOBs exposure as the key element of this potential common mode-of-action (MoA). *Trypanosoma brucei* developed cross-resistance to structurally diverse BOBs derivatives when the functions of the SUMOylation conjugation machinery were compromised through RNA*i*. This is in line with the result from our high throughput genetic screen (RIT-seq) uncovering the functional network underlying BOBs MoA. Importantly, the hyperSUMOylation triggered by BOBs leads to the destabilization of the TbCPSF complex in a proteosome-dependent manner, in parallel to changes in a cohort of surface proteins as well as a set of RNA-binding proteins (RBPs). Furthermore, we observed catastrophic nucleolar alterations specific to BOBs that may also lead to certain new insights into the common MoA of BOBs.

12:20 (10 mins) - A26078 - Oligo targeting for profiling drug resistance mutations in the parasitic *Trypanosoma*tids

Authors

M Ridgway[1]; S Altmann[1]; E Rico[1]; S Carvalho[1]; A Trenaman[1]; H Donnelly[1]; M Tinti[1]; S Wyllie[1]; D Horn[1];

[1] University of Dundee, UK

Discussion

*Trypanosoma*tids cause the neglected tropical diseases sleeping sickness, Chagas disease and the leishmaniases. New and improved genetic technologies would facilitate studies on these lethal parasites. Scalable precision editing methods, for example, could be used to improve our understanding of potential mutations associated with drug resistance. This is a current priority given that several new anti-*Trypanosoma*l drugs, with known targets, are currently in clinical development. We report the development of a simple oligo targeting method for rapid and precision editing of priority drug targets in otherwise wild type *Trypanosoma*tids. In *Trypanosoma brucei*, approx. 50-b single-stranded oligodeoxynucleotides were found to be optimal, multiple base edits could be incorporated, and editing efficiency was substantially increased when mismatch repair was suppressed. Resistance-associated edits were introduced in *T. brucei* cyclin dependent kinase 12 (CRK12) and cleavage and polyadenylation specificity factor 3, in the *Trypanosoma cruzi* proteasome b5 subunit, and in *Leishmania donovani* CRK12. We further implemented oligo targeting for site saturation mutagenesis, targeting a specific codon in *T. brucei* CRK12, which revealed fourteen resistance-conferring edits encoding six distinct amino acids. The outputs confirm on-target drug activity, reveal a variety of resistance-associated mutations, and facilitate rapid assessment of potential impacts on drug efficacy.

Day 4 - Parasite Cell Biology II *(Lecture Theatre P/X001)*

24-March-2022, at 11:50 to 12:40

Chairs - Dr James LaCourse & Dr Eden Ramalho Ferreira

11:50 (10 mins) - A25970 - Single-cell RNA-Sequencing Analysis of Life and Cell Cycle Progression in *L. mexicana*

Authors

FS Warren[1]; E Briggs[2]; T Otto[1]; MS Llewellyn[3]; R McCulloch[1];

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AbstractA crucial life-cycle development stage of Leishmania takes place in the macrophage vacuole, where metacyclic promastigotes differentiate into amastigotes. Dynamic changes in transcription underlying these complex cellular transitions through life cycle stages have not yet been described in detail. To evaluate these changes and identify putative differentiation regulators, we employed single-cell RNA sequencing (scRNA-seq) at five time-points as *L. mexicana* differentiated from promastigotes to axenic-amastigotes in culture. Clustering analysis of over 16,500 parasites across three experiments revealed five developmental stages, including promastigote and amastigote forms, as well as populations transitioning between these. From these data, we identified over 800 differentially expressed markers for a metacyclic-like cluster. Our analysis additionally suggests we can transcriptionally distinguish different replicative promastigote forms going through cell-cycle stages, which have previously been associated with changes in morphology. We also explored one transitional group of cells between the metacyclic and axenic amastigotes, revealing genes possibly associated with survival strategies required during the switch from the insect vector to mammalian host. The many novel genes detected include some that have been found to be essential for life-cycle progression in *Trypanosoma brucei* but have not yet been investigated in *L. mexicana*, as well as cell-cycle progression markers. Trajectory inference analysis further revealed phasic expression of transcripts of interest within the transitional and cell cycling clusters. To explore this transitional population in more detail, we performed scRNA-seq of *L. mexicana*-infected human macrophages, allowing us to ask if the transcriptional processes seen during differentiation to axenic amastigotes in culture are also seen as amastigotes form in a host cell. Ultimately, our aim is to find and validate potential new targets for halting the development of the parasite within the mammal, and thereby limiting infection and disease.

12:00 (10 mins) - A25679 - *Schistosoma mansoni* α-N-acetylgalactosaminidase (SmNAGAL) regulates coordinated parasite movement and egg production

Authors

B Hulme[2]; K Geyer[2]; JE Forde-Thomas[2]; G Padalino[2]; D Phillips[2]; W Ittiprasert[1]; S Karinshak[1]; V Mann[1]; IW Chalmers[2]; P Brindley[1]; CH Hokke[3]; KF Hoffmann[2];

[1] George Washington University, United States; [2] Aberystwyth University, UK; [3] Leiden University Medical Centre, Netherlands

Abstractα-galactosidase (α-GAL) and α-N-acetylgalactosaminidase (α-NAGAL) are two glycosyl hydrolases responsible for maintaining cellular homeostasis by regulating glycan substrates on proteins and lipids. Mutations in the human genes encoding either enzyme lead to neurological and neuromuscular impairments seen in both Fabry- and Schindler/Kanzaki- diseases. Here, we investigate whether the parasitic blood fluke *Schistosoma mansoni*, responsible for the neglected tropical disease schistosomiasis, also contains functionally important α-GAL and α-NAGAL proteins. As infection, parasite maturation and host interactions are all governed by carefully-regulated glycosylation processes, inhibiting *S. mansoni'*s α-GAL and α-NAGAL activities could lead to the development of novel chemotherapeutics. Sequence and phylogenetic analyses of putative α-GAL/α-NAGAL protein types showed Smp_089290 to be the only *S. mansoni* protein to contain the functional amino acid residues necessary for α-GAL/α-NAGAL substrate cleavage. Both α-GAL and α-NAGAL enzymatic activities were higher in females compared to males (p<0.05; α-NAGAL > α-GAL), which was consistent with *smp_089290*'s female biased expression. Spatial localisation of *smp_089290* revealed accumulation in parenchymal cells, neuronal cells, and the vitellaria and mature vitellocytes of the adult schistosome. siRNA-mediated knockdown (>90%) of *smp_089290* in adult worms significantly inhibited α-NAGAL activity when compared to control worms (*siLuc* treated males, p<0.01; *siLuc* treated females, p<0.05). No significant reductions in α-GAL activities were observed in the same extracts. Despite this, decreases in α-NAGAL activities correlated with a significant inhibition in adult worm motility as well as in egg production. Programmed CRISPR/Cas9 editing of *smp_089290* in adult worms confirmed the egg reduction phenotype. Based on these results, Smp_089290 was determined to act predominantly as an α-NAGAL (hereafter termed SmNAGAL) in schistosome parasites where it participates in coordinating movement and oviposition processes. Further characterisation of SmNAGAL and other functionally important glycosyl hydrolases may lead to the development of a novel anthelmintic class of compounds.

12:10 (10 mins) - A25958 - Mistargeting of aggregation-prone mitochondrial proteins activates a nucleus-mediated posttranscriptional quality control pathway in trypanosomes

Authors

C Dewar[3]; S Oeljeklaus[1]; J Mani[3]; W Mühlhäuser[1]; C von Känel[3]; T OchsenreiterB Warscheid[1]; A Schneider[3];

[1] Department of Biochemistry and Functional Proteomics, Universität Freiburg, Germany; [2] Institute of Cell Biology, University of Bern, Switzerland; [3] Department of Chemistry, Biochemistry and Pharmaceutical Sciences, Universität Bern, Switzerland

Discussion

Mitochondrial quality control (MQC) is the network of pathways by which eukaryotic cells monitor and maintain the function of their mitochondria. *Trypanosoma brucei* has a large single mitochondrion, which prevents the elimination of dysfunctional mitochondria as in some other organisms. When this essential mitochondrion is not functioning correctly, for example when mitochondrial protein import is defective, cell viability suffers. The processes by which this parasite regulates its mitochondrial function are of great interest, particularly in relation to its life cycle, where the mitochondrion undergoes massive programmed morphological and functional alterations.

MQC pathways in yeast and metazoa are regulated on the transcriptional level. However, in *T. brucei*, due to polycistronic transcription, MQC regulation in this way is not possible. Additionally, other than ubiquitin and the proteasome, orthologues of most common MQC factors found in yeast and metazoa are absent in *T. brucei.* Mitochondrial biogenesis in *T. brucei* has been shown to be greatly impacted by convergent evolution, and we expect the same to be the case for mechanisms governing MQC.

95% of mitochondrial proteins in *T. brucei* are encoded in the nuclear DNA. The multisubunit ATOM complex is the mediator of protein import through the mitochondrial outer membrane in trypanosomes (Pusnik et al., 2011, Mani et al., 2015). We show data demonstrating the existence of a MQC pathway in *T. brucei* triggered when the import of aggregation-prone proteins is blocked, specifically. Using a variety of proteomic and biochemical approaches, we show that the proteasome and putative components of a ubiquitin-driven pathway are recruited to the mitochondrion upon the induction of this import defect. *Trypanosoma*tid-specific candidates were investigated as to their roles within this MQC pathway. Of particular interest is a nuclearly-localised protein with a ubiquitin-like domain which is released into the cytoplasm upon the induction of a mitochondrial import defect. Nuclear release of this protein is required for this MQC mechanism to function.

12:20 (10 mins) - A25547 - Characterisation of a host receptor for *Plasmodium falciparum*-infected erythrocyte rosette formation

Authors

M Carlier[1]; L Bruce[2]; JA Rowe[1];

[1] Institute of Immunology & Infection Research, University of Edinburgh, UK; [2] Bristol Institute for Transfusion Sciences, NHS Blood and Transplant, Bristol, UK

Abstract

P. falciparum rosetting, the binding of two or more uninfected erythrocytes to an infected erythrocyte, is a key virulence factor associated with severe malaria. Rosette formation is mediated by *P. falciparum* erythrocyte membrane protein 1 (PfEMP1) expressed on the surface of infected erythrocytes. Several molecules have been proposed as host rosetting receptors (such as Blood Group A antigen and Complement Receptor 1), but none of these can account for rosetting interactions across all parasite strains, suggesting that major host receptors remain unidentified. The Wright^b blood group antigen, which is formed by a physical interaction between Band 3 and Glycophorin A, has been identified by the Rowe lab as a potential novel rosetting receptor. Antibody fragments targeting Wright^b, disrupt rosettes in several *P. falciparum* strains, but the mechanism of action of this antibody is largely unknown. This study aimed to characterise this key receptor-ligand interaction implicated in rosetting and determine if a rosette-disrupting antibody to Wright^b is active against a range of *P. falciparum* strains, suggesting therapeutic potential.

To determine whether the Wright^b antigen or Band 3 is a rosetting receptor, we tested the ability of Band 3-transfected K562 cells and naturally occurring Glycophorin null cells to form rosettes with purified *P. falciparum*-infected erythrocytes. Our results showed that Band 3-transfected

cells (expressing the Wright^b antigen) form rosettes with two parasite strains, whereas wild type K562 cells (expressing Glycophorin A alone) do not. Rosette frequency and rosette size did not vary across the glycophorin genotypes examined, showing that Glycophorin A is not essential for rosetting for the parasite strains tested. Further, we revealed that anti-Wright^b antibody fragments show variable activity against a panel of rosetting culture-adapted *P. falciparum* strains and clinical isolates. Thus, our data suggest that Wright^b could be a potential anti-rosetting therapeutic target, but that, due to the complexity of rosetting mechanisms, multiple host receptors may need to be targeted to obtain an intervention effective against all P. *falciparum* strains.

Day 4 - Diversity in Science II: Conversations toward inclusion and equity *(Lecture Theatre T/005)*
24-March-2022, at 11:50 to 12:15
Chair - Dr Giulia Bandini

11:50 (20 mins) - A26332 - Autism: myths and realities: a disability, a neurotype, and challenges for inclusion and equity.

Dr Mariana De Niz

Discussion

Around 1% of the global population is autistic. Autism is not a medical condition and has no cure – moreover, it is for life (not just a 'condition' of childhood). Autism has been defined in many ways: some consider it a neurotype – it is a different way of understanding and interacting with the world. Some define it as a developmental disability, because of how it affects the way autistic people communicate and interact with others, and perceive the world. The media has played an important role in the social perception of autism: this has come most often in vaccination-related debates that saw a surge in fear towards autism; and also in the form of film and television, whereby autistic people are often depicted as either incapable of communication (often as children), or extremely bright geniuses (yet awkward and alienated). However, autism is a spectrum. With 75 million people being autistic, it is likely that most, if not all professions, have autistic personnel with all levels of skills and abilities. These professions include science. In this presentation I will discuss in a conversation, barriers faced in terms of inclusion and equity. I will also address my experience on effective actions that can promote inclusion and equity. This is with the hope of raising awareness to create a more empathic environment for current autistic scientists; of addressing taboos surrounding autism; of addressing prejudices regarding whether one can be a successful scientist "despite" being autistic; and with the hope of facilitating integration and acceptance of young autistic scientists to scientific life.

Day 5 – Fri 25th Mar 2022 -

Session 15 – Fri 25th Mar 10:00 - 11.20
Day 4 - Parasite Evolutionary Genomics *(Lecture Theatre K/018)*
Chairs - Dr Daniel Jeffares & Dr Joao Cunha

10:00 (20 mins) - A26223 - Comparative Genomics in *Trypanosoma*tids: genetic diversity of isolates in endemic regions in Brazil and identification of virulence factors

Daniella Bartholomeu
University Federal Minas Gerais

Discussion

Comparative genomics in *Trypanosoma*tids has opened myriad possibilities that have contributed to a more comprehensive understanding of these parasite population dynamics in endemic regions, their biology, and interactions with hosts. In the first part of this talk, we will present studies on the genomic variability of Leishmania infantum isolates in different endemic regions in Brazil. Our findings reveal intra- and inter-population variability shaped by spatial and temporal components. In the second part, we will present a new methodology for the comparative study of repetitive genomic sequences among *Trypanosoma cruzi* isolates based on unassembled short reads. Its application on the identification of biomarkers for diagnosis and on the study of multigene families encoding virulence factors will be discussed.

10:20 (10 mins) - A26060 - Population genomics and geographic dispersal in Chagas disease vectors: Landscape drivers and evidence of possible adaptation to the domestic setting.

Authors

LE Hernandez-Castro[5]; B Cheaib[4]; **MS Llewellyn**A Bacigalupo[4]; MJ Grijalva[3]; AG Villacís[2];
[1] University of Glasgow , UK; [2] Pontifical Catholic University of Ecuador, Ecuador; [3] Ohio University, United States; [4] Institute of Biodiversity, Animal Health and comparative Medicine, University of Glasgow, UK; [5] The Roslin Institute, University of Edinburgh, UK

Discussion

Accurate prediction of vectors dispersal, as well as identification of adaptations that allow blood-feeding vectors to thrive in built environments, are a basis for effective disease control. Here we adopted a landscape genomics approach to assay gene flow, possible local adaptation, and drivers of population structure in *Rhodnius ecuadoriensis*, an important vector of Chagas disease. We used a reduced-representation sequencing technique (2b-RADseq) to obtain 2,552 SNP markers across 272 *R. ecuadoriensis* samples from 25 collection sites in southern Ecuador. Evidence of high and directional gene flow between seven wild and domestic population pairs across our study site indicates insecticide-based control will be hindered by repeated re-infestation of houses from the forest. Preliminary genome scans across multiple population pairs revealed shared outlier loci potentially consistent with local adaptation to the domestic setting, which we mapped to genes involved with embryogenesis and saliva production. Landscape genomic models showed elevation is a key barrier to *R. ecuadoriensis* dispersal. Together our results shed early light on the genomic adaptation in triatomine vectors and facilitate vector control by predicting that spatially-targeted, proactive interventions would be more efficacious than current, reactive approaches.

10:30 (10 mins) - A26159 - Aneuploidies are an ancestral feature in *Trypanosoma*tids and could be related to parasite adaptation

Samuel Carvalho

Authors

SA Pimenta-Carvalho[5]; LV Almeida[5]; A Coqueiro-dos-Santos[5]; RP Baptista[3]; GF Rodrigues-Luiz[4]; CN Costa[7]; MS Cardoso[5]; CA Grace[6]; DC Jeffares[1]; R McCulloch[2]; D Bartholomeu[5]; JL Reis-Cunha[6];

[1] University of York, UK; [2] Wellcome Trust Centre for Molecular Parasitology, University of Glasgow, UK; [3] University of Georgia, United States; [4] Campinas State University (UNICAMP), Brazil; [5] Federal University of Minas Gerais - UFMG, Brazil; [6] York Biomedical Research Institute, Department of Biology, University of York, UK; [7] Federal University of Piauí, Teresina, Brazil

AbstractAneuploidy, the presence of an aberrant number of chromosomes in a cell, usually results in severe abnormalities in multicellular eukaryotes as humans. However, some unicellular eukaryotes rely on aneuploidy as a mechanism to allow rapid adaptation to changing environments, having a positive fitness in stress conditions and promoting drug resistance. Aneuploidies have been largely described in protozoan parasites as *Leishmania* and *Trypanosoma cruzi*, where duplicated chromosomes vary in different hosts and can promote drug resistance. Interestingly, the closely related parasite *Trypanosoma brucei* is mainly euploid. Hence, to evaluate if aneuploidies are an ancestral or a recent feature in *Trypanosoma*tids we estimated the chromosome copy number variation in 13 *Trypanosoma*tidae species, including *Angomonas*, *Crithidia*, *Leptomonas* and *T. vivax*, using whole genome sequencing and read depth coverage variations. Aside from *T. brucei*, *T. evansi* and *T. vivax*, all the remaining species have evidence of aneuploidies, including *ParaTrypanosoma confusum*, an early-branching *Trypanosoma*tid, indicating that it is an ancestral character in these parasites. The presence of aneuploidies could be detrimental in *T. brucei* clade, as their genome is packed in a lower number of larger chromosomes. Next, we evaluated if there were consistent chromosomal duplications in the evaluated species. *Leishmania* chromosome 31 is constantly supernumerary, a fact reassured by our analysis of ~200 isolates from *L. donovani* and *L. infantum* populations in Africa, Asia and Brazil. This chromosome had an increased nucleotide diversity (π), which is expected, as having extra copies per cell results in more sites to be randomly mutated. In addition, redundant copies of genes could allow a rapid adaptation and diversification without loss of function. Regarding the other *Trypanosoma*tid species, the chromosomes that have most of its genes orthologous to *Leishmania* chromosome 31 were also consistently supernumerary, even in the euploid *T. brucei* clade where regions of this chromosome are duplicated into two sequences, chromosomes 4 and 8. This suggests that there are important genes enrolled in the parasite survival in these sequences. We evaluated the function of these shared duplicated genes and we found genes involved in parasite's osmoregulation and response to stress, as well as diverse cytoskeleton mediated processes such as cell morphogenesis, flagellar motility and cell division. We evaluated the function of these shared duplicated genes and we found genes involved in housekeeping functions as osmoregulation and response to stress, diverse cytoskeleton mediated processes such as cell morphogenesis, flagellar motility and cell division, energy obtaining pathways, host immune system evasion, infectivity and intracellular trafficking. We are now evaluating species-specific genes that were inserted in these duplicated regions specifically in each protozoan, as those can be important to each parasite adaptation.

10:40 (10 mins) - A26110 - Assessing LRV1 role as risk factor for mucosal leishmaniasis occurrence and its relationship with TRL3 polymorphism

Authors

F Pazmino[1]; C Vargas[1]; D Parra[1]; C Saavedra[1]; L Cadavid[1]; S Muvdi[2]; C Ovalle-Bracho[2]; **M Echeverry**[1];

[1] Universidad Nacional de Colombia, Colombia; [2] Hospital Universitario Centro Dermatológico Federico Lleras Acosta, Colombia

Discussion

Mucosal leishmaniasis (ML) is a serious clinical form of leishmaniasis that is characterised by destruction of the nasal and/or the oral mucosae and due to unknown reasons appears as a late complication in 5%–10% of cutaneous leishmaniasis (CL) cases produced by species belonging to *Leishmania Viannia* subgenus. Experimental data suggests that strains of *Leishmania* spp. carrying an RNA virus known as Leishmania RNA virus type 1(LRV1) triggers an immunological response that involves the endosomal Toll-like receptor 3 (TLR3) and has been associated with persistence and dissemination of *Leishmania* (V.) *guyanensis*. Moreover, TLR3 gene displays several single nucleotide polymorphisms (SNPs) associated with resistance or susceptibility to viral infectious diseases. The present work evaluated LRV1 and TLR3 gene polymorphisms as risk factors for the occurrence of ML throughout a retrospective case-control study involving 102 patients. Cases were defined as patients with ML (n=33) and controls corresponded to patients who had CL without mucosal lesions (n=69). A subgroup of controls (n=19) was followed up for a median time of 16 years to rule out ML occurrence. Clinical data were recorded from the patients' medical records and cryopreserved biopsies were used for *Leishmania* species identification, LRV type-1 (LRV1) detection and TLR3 (exons 2, 3, and 4) genotypification. Bivariate and logistic regression analyses were applied to estimate the risk factors associated with ML occurrence. The predominant *Leishmania* species in both groups was *L. (V.) braziliensis.* Multivariate logistic regression indicated that the unique factor linked with the occurrence of ML was the infection with *Leishmania* spp. carrying LRV1 [OR,8.81, 95%CI 1.72–45.76 and p = 0.009]. Four SNPs on TLR3 gene were identified and showed no association to ML development. Therefore, LRV1 presence is an independent risk factor for developing ML.

Day 4 - Parasite Gene Expression *(Lecture Theatre T/005)*

25-March-2022, at 10:00 to 11:00

Chairs - Dr Nathaniel Jones & Dr Joana Faria

10:00 (20 mins) - A25990 - Roles and interactions of the specialized initiation factors EIF4E2, EIF4E5 and EIF4E6 in *Trypanosoma brucei*: EIF4E2 maintains the abundances of S-phase mRNAs

Authors

F Falk[1]; R Palhares[1]; C Clayton[1];

[1] ZMBH - Heidelberg University, Germany

Discussion

Trypanosoma brucei has six versions of the cap-binding translation initiation factor EIF4E. We here investigated the functions of EIF4E2, EIF4E5 and EIF4E6 in bloodstream forms. Purification and mass spectrometry of non-RNAse treated extracts, and comparison with EIF4E3, confirmed the specific associations previously found in the procyclic form, which infects the Tsetse fly host. Some co-purification of RNA-binding proteins, especially with EIF4Es 3 and 6 was also observed. Bloodstream forms lacking EIF4E5 grew normally and could differentiate to replication-incompetent procyclic forms. Depletion of EIF4E6, in contrast, inhibited bloodstream-form trypanosome growth and translation.
EIF4E2 complexes with a putative RNA stem-loop binding protein, SLBP2, but has no EIF4G partner, and no association with general translation factors, suggesting that it is not an active translation initiation factor. Bloodstream forms lacking EIF4E2 multiplied slowly, with a slight increase in G2 cells at the expense of G1. They had a low maximal cell density, with expression of the stumpy-form marker PAD1 but no evidence for enhanced stumpy-form signalling. RNAi targeting SLBP2 had similar effects. The EIF4E2 knock-out cells differentiated readily to procyclic forms, which grew normally. mRNA pull-downs revealed strong association of EIF4E2 with mRNAs that are maximally abundant in S phase, three of which were previously shown to be bound and stabilised by the pumilio domain protein PUF9. The same mRNAs had decreased abundances in EIF4E2 knock-out cells. Yeast 2-hybrid results suggest that PUF9 interacts directly with SLBP2, but PUF9 was not present inEIF4E2 pull-downs. A possible interpretation is that the EIF4E2-SLBP2 complex interacts with PUF9, and its bound RNAs, only early during G1/S, stabilising the mRNAs in preparation for translation later in S phase or in early G2.

10:20 (10 mins) - A26176 - RNA binding proteins as trans-regulators impacting surveillance and infectivity in Leishmania

Authors
EP Parry[2]; NT Teles[2]; RN Neish[2]; K Newling[1]; J Mottram[2]; PB Walrad[2];
[1] University of York, UK; [2] University of York, Centre for Immunology and Infection, UK
AbstractLeishmania spp. protozoan Kinetoplastids present peculiar gene expression fundamentally dependent upon post-transcriptional control. This elevates the importance of RNA binding proteins for gene regulation in these parasites. Building upon the mRBPome we isolated previously (Pablos, Ferreira et al., MCP, 2019), 70 mRNA-bound RBPs were selected from the three main L.*mexicana* lifecycle stages. A trans-regulator knockout clone library was created through barcoded CRISPR and screened for essential roles in cellular differentiation and macrophage or mouse infections. Of the 70 RBPs screened, 40 are essential to cell viability and 18 contribute to lifecycle progression to human-infective stages and/or parasite infectivity. Examination of individual knockout lines for amastigote-specific mRBPs showed normal promastigote growth dynamics, whereas infection of peritoneal macrophages was inhibited or ablated, suggesting essential roles of RBPs for amastigote viability and virulence. Immunoprecipitation of multiple mRBPs will identify associated transcript targets that may represent novel virulence factors. Key words: Leishmania, RBP, RNA, Knockout, infection.

10:30 (10 mins) - A25988 - Characterisation of a new Apicomplexa-specific zinc-finger protein family in *Plasmodium* with a key role across different stages of the life cycle.

Authors
LV Carruthers[1]; I Fitzmaurice-O'Neill[1]; R Morton[1]; L Tavernelli[1]; D Beraldi[1]; KK Modrzynska[1];
[1] University of Glasgow, Institute of Infection, Immunity & Inflammation,, UK
Abstract
The lifecycle of *Plasmodium*, causative agent of malaria, is composed of several stages, each with its own form and environment. Transitions between these stages requires dramatic modification of the gene expression profile, but the molecular factors regulating these transitions remain poorly understood. CCCH zinc-finger domains are proportionally enriched in the *Plasmodium* proteome, however little is known about their biological role in the parasite. We analysed 3xCCCH zinc-finger domain distribution in the *Plasmodium* genome revealing they congregate within a new *Apicomplexa*-specific protein family with predicted RNA-binding function. We characterised one of these proteins, Zn3_3, using the rodent malaria model *Plasmodium berghei*. C-terminal epitope tagging of Zn3_3 demonstrated it has a stage-specific expression profile detectable with a cytoplasmic distribution in developing mosquito stages ~6 h post-transmission, and remains visible through to the mature ookinete form. Zn3_3 KO had no impact on production of the mammalian forms (asexual blood stages and gametocytes), but completely abolished formation of normally-shaped zygotes/ookinetes and transmission to female anopheline mosquitoes. RNA sequencing profiles of the Zn3_3 KO line generated at 8 h post-fertilisation did not reveal any transcriptomic changes. However, RNA immunoprecipitation experiments performed in ookinetes reveal for the first time that Zn3_3 is involved in RNA-binding. Other members of the 3xCCCH family have been identified to play key roles in other life-stages (being crucial for asexual growth and gametocytogenesis), and their targets and effect on the gene expression are being explored. In summary we confirm the 3xCCCH protein family plays key roles during *Plasmodium* lifecycle progression. Further analysis of their function may reveal new ways of the gene expression regulation in *Plasmodium* and related parasites.

10:40 (10 mins) - A26160 - Post-transcriptional iron regulatory mechanisms in *Trypanosoma brucei*

Authors
C Gilabert Carbajo[2]; M Tinti[1]; P Yates[3]; **C Tiengwe**[2];
[1] University of Dundee, UK; [2] Imperial College London, UK; [3] Oregon Health and Science University, Portland OR, 97239, USA, UK
Discussion
Iron is essential for many enzymatic reactions but also potentially toxic when in excess. Consequently, cells exert a tight control of intracellular iron levels. Despite its central role in the virulence of protozoan parasites, our understanding of iron homeostasis in T. *brucei* remains limited.
Our transcriptomic analyses of bloodstream form (BSF) *T. brucei* identified a cohort of iron regulated mRNAs that include the main iron carrier protein (the transferrin receptor, TfR), an RNA binding protein (RBP5), genes involved endocytosis (Phosphatidic Acid Phosphatase, PAP2), ESAG3, and a variety of membrane proteins. RBP5 and TfR are upregulated within one hour of iron depletion, suggesting that they are part of the primary starvation response. In contrast, PAP2 upregulation begins after RBP5 levels have nearly peaked. We propose that PAP2 upregulation is a secondary response to iron deficiency that may be mediated by RBP5. In support of this notion, ectopic overexpression of RBP5 results in upregulation of PAP2 transcripts while knockdown of RBP5 reduces PAP2 levels. In addition, RNA immunoprecipitation (RIP) analysis indicates that

RBP5 binds to PAP2 mRNA. Current efforts are focused on identifying sequence element(s) in the PAP2 RNA bound by RBP5 and determining the full cohort of mRNAs bound and potentially regulated by RBP5.
Interestingly, RIP shows that RBP5 also binds its own mRNA but not to TfR mRNAs, suggesting that RBP5 does not mediate the primary response to iron depletion. To gain insights into the factors directing the initial response to iron depletion, we have begun to define the cis-acting elements in the RBP5 mRNA that regulate RBP5 expression. By employing a dual luciferase system and deletion analyses we defined a 94-nucleotide iron responsive element (IRE) within the RBP 3'UTR that represses expression under iron replete conditions. A second 99 nucleotide element appears to constitutively down-modulate RBP5 expression, though it does not appear to be essential for iron-responsive regulation. We are in the process of refining the boundaries of the IRE to define the minimal sequences required for iron responsiveness, with the long-term goal of identifying associating RBP(s).
In support of the biological relevance of RBP5 regulation by iron levels, available transcriptome data show that RBP5 is upregulated in relatively iron-poor niches: in salivary glands relative to midgut, in adipose tissues relative to blood, and in metabolically quiescent stumpy forms. We propose a model in which early upregulation of RBP5 upon iron starvation facilitates the subsequent regulation of a variety of iron responsive genes. Insights gained from these studies will help to reveal the complexities of iron homeostasis in BSF *T. brucei*.

Day 4 - Trafficking, Signaling *(Lecture Theatre P/X001)*
25-March-2022, at 10:00 to 11:00
Chairs - Dr Romina Nievas & Dr Nicola Baker

10:00 (20 mins) - A26206 - Heavy Metal: The role of iron storage in *Toxoplasma gondii*

Clare Harding
University of Glasgow

Discussion

Iron is an essential element which plays central roles in eukaryotic metabolism. However, in the wrong place it poses a danger to cells, forming dangerous reactive oxygen species. For parasites, iron uptake and storage presents different challenges, as it must subvert host pathways to acquire the iron needed for multiplication. The ubiquitous pathogen *Toxoplasma gondii* is able to infect and replicate within almost all tissues, in very different nutritional environments. Recently, the role of iron in *Toxoplasma* biology has come under renewed scrutiny, however the basis of iron handling of these parasites remains largely unknown. Our recent work focusing on iron storage revealed that iron compartmentalization and storage is important to the parasite in a number of ways. We also find that iron storage has an important role in mediating virulence in vivo, possibly linked to survival in immune cells. The importance of uptake and trafficking of iron in apicomplexan parasites suggests iron as an attractive target for therapeutic interventions.

10:20 (10 mins) - A25985 - *Ascaris suum* Pseudocoelomic Fluid: A Peptide-rich Biofluid that Modulates Nematode Motility

Authors
D McKenzie[1]; C McCoy[1]; C Graham[1]; NJ Marks[1]; A Maule[1]; B Graham[1]; L Atkinson[1]; A Mousley[1];
[1] Queen's University Belfast, UK

Discussion

Bioactive peptides, including Neuropeptide-like Proteins (NLPs), FMRFamide-like Peptides (FLPs) and Insulin-like Peptides (ILPs), are known to influence key nematode neuromuscular functions such as locomotion, feeding and egg laying. The wired component of the nematode neuropeptide signalling system is well characterised, however non-synaptic routes of communication are less well known. Nematode pseudocoelomic fluid (PCF) may serve as an alternative, non-synaptic, route of peptide transmission. Unfortunately, the size-related intractability of most nematodes has prevented the characterisation of the PCF peptidome, however the large size of the pig parasite *Ascaris suum* offers an opportunity to achieve this in a nematode pathogen. A recent LC-MS/MS study detected 76 peptides (FLPs, NLPs and AMPs) in *Ascaris*-PCF (As-PCF) and demonstrated that As-PCF is bioactive on nematode muscle. In this study we have employed *in silico* bioinformatics (HMM/BLASTp), Liquid-Chromatography Tandem Mass Spectrometry (LC-MS/MS), Reverse-Phase High-Performance Liquid Chromatography (RP-HPLC) and nematode bioassays to expand on these analyses by examining the ILP component of As-PCF and the bioactivity of As-PCF on nematode behaviour. The data demonstrate that: (i) 409 Insulin-like Peptide (ILP) sequelogs are present in 109 nematode species (262 novel ILPs); (ii) 9 ILP sequelogs are present in *A. suum*; (iii) As-PCF contains 90 peptides including 7 NLPs, 4 FLPs, 76 AMPs and 3 ILPs (ILP-1, -18 and -31); (iv) As-PCF significantly reduces *Caenorhabditis elegans* motility. These data have expanded the peptide library used to mine As-PCF LC-MS/MS data, documented the presence of ILPs in As-PCF, and move towards the identification of the bioactive peptide components of As-PCF. Further characterisation of the As-PCF peptidome will enhance our understanding of nematode biology, including non-synaptic peptide transmission, and may inform drug development strategies for parasite control.

10:30 (10 mins) - A25761 - Decoding heat shock signalling in the African trypanosome

Authors
CP Ooi[3]; C Benz[1]; C Dewar[2]; M Aelmans[2]; **MD Urbaniak**[2];
[1] Institute of Parasitology, Biology Centre of the Czech Academy of Sciences, České Budějovice, Czech Republic, Czech Republic; [2] Division of Biomedical and Life Sciences, Faculty of Health and Medicine, Lancaster University, UK; [3] Department of Biology, Edge Hill University, Lancashire L39 4QP, UK

Discussion

The African trypanosomes *Trypanosoma brucei* and *Trypanosoma congolense* are vector borne parasites of domestic cattle and are a major cause of economic hardship in sub-Saharan Africa, with two sub-species of *T. brucei* also causing fatal infections in humans. Their ability to sense and respond to their host environment is critical for their survival and virulence, and is achieved despite a near complete lack of transcriptional control that results in a reliance on RNA binding proteins. Symptoms of human and animal African trypanosomiasis include periods of fever as high as 41 ^{0}C, eliciting a heat shock response in the parasites that is essential for their survival. Eukaryotic cells respond to heat shock by triggering a

general arrest in protein translation through phosphorylation of eIF2alpha and transcriptional up-regulation of heat shock protein (HSP) expression to aid protein folding and degradation. This general response appears to be conserved in trypanosomes, but the mechanisms mediating the response are divergent and post-transcriptional in nature. To capture the molecular events involved in sensing and responding to heat shock in the mammalian infective form we have conducted an initial SILAC-based quantitative proteomic and phosphoproteomic analysis of *T. brucei* cells treated at 41 ^{0}C for 1h [1]. Our analysis indicates that protein abundance does not rapidly respond (≤1 h) to heat shock, and that the changes observed in phosphorylation site abundance are larger and more widespread. The heat shock responsive phosphorylation sites included RNA binding proteins with putative roles in heat shock response such as P-body / stress granules components and the eukaryotic translation initiation 4F complex, but no phosphorylation of eIF2alpha occurred. Dynamic phosphorylation of zinc finger protein ZC3H11 was observed for the first time, a key regulatory RNA binding protein that stabilises heat shock responsive mRNAs and up-regulates HSP expression [2]. We have demonstrated that heat shock causes a specific and reversible cell cycle arrest, and are exploiting temporal quantitative proteomic and phosphoproteomics to reveal the molecular mechanism of heat shock response and recovery. We are also examining the conservation of heat shock response and recovery in *T. congolense*, the major cause of animal African trypanosomiasis.

References:

1. Ooi, C.P., C. Benz, and M.D. Urbaniak, *Phosphoproteomic analysis of mammalian infective Trypanosoma brucei subjected to heat shock suggests atypical mechanisms for thermotolerance.* J. Proteomics, 2020. **219**: p. e103735. 2. Droll, D., et al., *Post-transcriptional regulation of the trypanosome heat shock response by a zinc finger protein.* PLoS Pathog, 2013. **9**(4): p. e1003286.

10:40 (10 mins) - A25839 - Exploiting Omics Approaches to Unravel Endocannabinoid Biology in *Strongyloides* Parasites

Authors

LC Cadd[1]; L Atkinson[1]; NJ Marks[1]; AG Maule[1]; A Mousley[1]; B Crooks[1];

[1] Queen's University Belfast, UK

Abstract

Parasitic nematodes infect >1.5 billion of the world's population, significantly affecting LMIC populations, where many of the most problematic pathogens are the soil-transmitted helminths. Overreliance on a limited number of anthelmintics exacerbates drug resistance pressures, thus there is a pressing need to identify novel therapies for parasite control ahead of a resistance crisis. The optimisation of appropriate end-to-end strategies, in relevant and tractable parasitic nematodes, is required for the identification and validation of novel anthelmintic targets from therapeutically unexploited facets of nematode biology. The nematode endocannabinoid (EC) system remains a relatively uncharacterised aspect of nematode neurosignalling. EC-signalling has been linked to key roles including cholesterol mobilisation, ageing, axon regeneration, locomotion, feeding and nociception in the free-living nematode *Caenorhabditis elegans* however, its role and importance in parasites is unclear. Key facets of the EC-system are broadly conserved across the nematode phylum including in key parasite species (unpublished), and that EC-signalling may play a putative role in parasitic nematode host immune modulation. This project will develop a drug target prioritisation pipeline in the tractable parasite *Strongyloides ratti* by interfacing *in silico* bioinformatics, optimised *in vitro* bioassays, and functional genomics approaches and, in tandem, exploit this to explore the drug target potential of parasitic nematode EC-signalling. Our data: (i) reveal an extensive EC-signalling network in 30 lifestyle-diverse nematodes representing 7 clades; (ii) provide an optimised bioassay toolkit for the elucidation of pre- and post- functional genomics phenotypes in *Strongyloides* spp. and, (iii) begin to probe NPR-19 (EC-receptor) function using functional genomics (RNAi) in *S. ratti*. Characterisation of the EC-system using the *Strongyloides* functional genomics pipeline will enhance our understanding of parasitic nematode biology, and may reveal novel anthelmintic targets for the control of medically and agriculturally importance parasites.

Session 16 – Fri 25th Mar 11:50 - 13.00

Day 4 - Parasite Evolutionary Genomics *(Lecture Theatre K/018)*

25-March-2022, at 11:50 to 12:40

Chairs - Dr Daniel Jeffares & Dr Joao Cunha

11:50 (10 mins) - A25502 - Morphological and PCR Screening of *Schistosoma* Hybrid infecting humans in communities around the Oyan River Dam Area, Ogun State, Nigeria

Authors

AA Bayegun[2]; FA Adebayo[2]; KO Ademolu[2]; OP Akinwale[3]; PV Gyang[3]; JR Stothard[1]; **UF Ekpo**[2];

[1] Liverpool School of Tropical Medicine, UK; [2] Federal University of Agriculture Abeokuta, Nigeria; [3] Nigerian Institute of Medical Research, Yaba, Lagos, Nigeria

AbstractThere is a growing concern of hybridization between *Schistosoma haematobium* and *Schistosoma bovis*, where humans and livestock shared same surface water in schistosomiasis endemic communities. The implication of livestock as reservoir for human infection is worrying. This study investigated the possible occurrence of *Schistosoma* hybrids using egg morphology and PCR screening. *Schistosoma* eggs were collected from human urine in Imala-Odo, Abule-Titun, Apojula and Ibaro-Oyan communities, Oyan Rivers dam area, Ogun State, Nigeria. The eggs were counted, photographed, and measured for Total Length (TL), Maximum Width (MW), and Egg Shape (L/W) with IC Measure™. Unusual eggs shapes were characterized by PCR amplification of *Schistosoma* specific *Dra1* gene. Positive amplicons for the *Dra1* gene were further subjected to PCR amplification of an ITS-2 rDNA. A total of 1,984 *Schistosoma* eggs were collected. The total length ranges from 70.90 - 262.30µm and Maximum Width ranges from 30.10 - 102.60µm. Egg shape ranges from 1.60 - 4.06µm. There was significant variation ($p < 0.05$) in egg morphology, where 67.8% of the eggs had typical round-to-oval shape of *S. haematobium*, 32.2% were atypical spindle-shaped of hybrid *S. haematobium / S. bovis* and 1.1% of the eggs were spineless. PCR screening revealed 54 (62.1%) and 33 (61.1%) of the eggs were positive for *Dra1* and ITS-2 genes respectively and of *Schistosoma* origin. DNA sequencing of spindle-shaped eggs is currently ongoing. These observations suggest that *Schistosoma* hybrids may be circulating in human population in the study area.

12:00 (10 mins) - A26059 - The clonal dynamics and molecular epidemiology of Amoebic Gill Disease in *Salmo salar* via multiplex amplicon sequencing

Authors

B Cheaib[3]; P Schwabl[3]; W Liu[4]; A Bacigalupo[2]; L Ryder[5]; J Kaufmann[5]; P McGinnity[5]; F Hernandez[6]; M Barret[1]; MS Llewellyn[3];

[1] Institute of Infection, Immunity and Inflammation, University of Glasgow, UK; [2] Institute of Biodiversity, Animal Health and comparative Medicine, University of Glasgow, UK; [3] Institute of Biodiversity, Animal Health and Comparative Medicine, University of Glasgow, UK, UK; [4] Wellcome Centre for Integrative Parasitology, Institute of Infection, Immunity and Inflammation, University of Glasgow, UK; [5] Marine Institute, Newport, Ireland; [6] Institute of Biomedical and Environmental Health Research, School of Health and Life Science, University of West Scotland, UK, UK

Discussion

Amoebic gill disease (AGD), caused by various strains of *Neoparamoeba perurans*, is as a worldwide disease of salmonids aquaculture associated with sustained and intensive year-on-year losses (≥20%). *N. perurans* hosts in the vicinity of its nucleus an endosymbiotic aflagelate kinetoplastid called *Perkinsela spp known as Perkinsela-like organism* (PLO) or *ihctyobodo Necator* related organism (IRO), similar tokinetoplastid pathogens of human and domestic livestock (*Trypanosoma brucei* sp., *T. cruzi*, *Leishmania sp.*). As well as a kinetoplastid, the *N. perurans* cytoplasm is also home to many intracellular prokaryotes. Genome sequencing efforts have been delayed and direct sequencing of parasite genomic material is frustrated by microbial contamination.Rapid, low cost- characterization of *N. perurans* genetic polymorphisms strains at once would be a powerful epidemiological surveillance management for AGD. To quantify the genetic diversity associated with AGD, we developed a simple, cost-effective, multiplexamplicon sequencing protocol tool based on massive parallel amplification of >400 information hotspots throughout the target genomes of *N.* perurans and its endosymbionts. These markers will be applied to daily samples across four sites of Salmon farms, two sites in west Scotland, and two sites in western Ireland over two summers seasons, 2019 in Ireland and 2021 in Scotland to establish the colonisation and re-infection dynamics of this pathogen. To sequence the AGD agent, more than one Terra bp of genomic and transcriptomic data isolated for pure *N. perurans* culture treated with antibiotics were generated with Nanopore and Novaseq Illumina. The data were processed, curated, and assembled to characterize the functional associations between the host and its endosymbiont. A candidate list of housekeeping, metabolic and accessories genes will be amplified with the GLST method and will provide a unique level of genomics, spatial and temporal resolution to understand the dynamics of parasite.

Keywords: Endosymbiosis, Kinetoplastid, Amoeba, Amoebic gill disease, Omics, Salmonids

12:10 (10 mins) - A26045 - Parasite genotype strongly influences mortality risk in visceral leishmaniasis.

Authors

CA Grace[1]; KS Carvalho[4]; MI Lima[3]; VC Silva[2]; JL Reis-Cunha[1]; MJ Brune[1]; SJ Forrester[1]; CM Silva de Azevedo[3]; D Speed[5]; JC Mottram[1]; DC Jeffares[1]; DL Costa[4];

[1] York Biomedical Research Institute, Department of Biology, University of York, UK; [2] Federal University of Piauí, Teresina, Brazil, UK; [3] Federal University of Maranhão, São Luís, Brazil; [4] Federal University of Piauí, Teresina, Brazil; [5] Aarhus University, Denmark

Abstract

Background:Visceral leishmaniasis (VL) is a potentially fatal disease mainly caused by Leishmania infantum in South America and L. infantum and L. *donovani* in Asia and Africa. In Brazil, L. infantum causes 4200-6500 cases per year, 90% of the cases registered in the Americas. Screening of healthy blood donors in Brazil indicates that asymptomatic L. infantum infections are common, occuring in ~1-6% of the general population. However, the fatality rate of patients who receive treatment in Brazil is almost 10%, one of the highest in the world. This high proportion of asymptomatic patients, but also high mortality suggests that there are parasites with varying virulence in Brazil. Since case fatality is getting worse, it is imperative to understand the factors that lead to this increased mortality in Brazil. Visceral leishmaniasis disease severity and outcomes have been associated with several host traits, as patient genotype, nutrition, age, sex, co-morbidities, and co-infections. However, the impact of the parasite genetic variability in the disease severity is poorly understood. In this study, we examine for the first time the effects of population-wide parasite genetic variation on VL disease severity in Brazil using genome-scale methods.

This study:We quantified the effects of L. infantum parasite genotype on disease severity and mortality. We collected and sequenced the genomes of 109 L. infantum isolates from patients in northeast Brazil. We also retrieved matching patient clinical data from medical records, including mortality, sex, HIV co-infection and laboratory data (creatinine, haemoglobin, leukocyte and platelet counts). We identified genetic differences between parasite isolates, including single nucleotide polymorphisms (SNPs), small insertions/deletions (indels), and variations in genic, intergenic, and chromosome copy numbers (copy number variants, CNVs). To describe associations between the parasite genotypes and clinical outcomes, we applied quantitative genetics methods of heritability and genome-wide association studies (GWAS), treating clinical outcomes as traits that may be influenced by parasite genotype.

Findings:

Parasite genotype explains 83% chance of mortality (narrow sense heritability h2 = 0·83±0·17), and has a significant relationship with patient sex (h2 = 0·60±0·27). Impacts of parasite genotype on other clinical traits are lower (h2 ≤0·34). GWAS identified 17 CNVs that were significantly associated with mortality, two with creatinine and two with bacterial co-infection, one jaundice and HIV co-infection; and two SNPs/indels that associate with age and jaundice, HIV.

Implications:

We have shown that parasite genotype is an important factor in VL disease severity in Brazil. Since the genetic diversity of L. infantum in Brazil is lower than that of L. *donovani* which causes VL in Africa and the Indian subcontinent, we can reasonably expect similar effects of parasite genot

Day 4 - Parasite Gene Expression *(Lecture Theatre T/005)*

25-March-2022, at 11:50 to 12:40

Chairs - Dr Nathaniel Jones & Dr Joana Faria

11:50 (10 mins) - A25919 - Control of variant surface glycoprotein expression by CFB2 in African trypanosomes and quantitative proteomic connections to translation and cytokinesis

Authors

G Bravo Ruiz[1]; M Tinti[1]; M Ridgway[1]; D Horn[1];

[1] The Wellcome Trust Centre for Anti-Infectives Research, School of Life Sciences, University of Dundee, UK

AbstractVariant Surface Glycoproteins (VSG) coat parasitic African trypanosomes and underpin antigenic variation and immune evasion. These VSGs are super-abundant virulence factors that are subject to post-transcriptional gene expression controls mediated via the *VSG* 3'-untranslated region (3'-UTR). To identify positive VSG regulators in bloodstream form cells, we used genome-scale screening data to prioritise mRNA binding protein (mRBPs) knockdowns that phenocopy VSG mRNA knockdown, displaying loss-of-fitness and pre-cytokinesis accumulation. The top three candidates were CFB2 (cyclin F-box protein 2), MKT1 and PBP1 (polyadenylate binding-protein binding-protein). Notably, CFB2 was recently found to regulate VSG transcript stability, and all three proteins were found to associate. We used data-independent acquisition for accurate label-free quantification and deep proteome coverage to quantify expression profiles following depletion of each mRBP. Only CFB2 knockdown significantly reduced VSG expression and the expression of a reporter under the control of a *VSG* 3'-untranslated region (3'-UTR). CFB2 knockdown also triggered depletion of cytoplasmic ribosomal proteins, consistent with translation arrest observed when VSG synthesis is blocked. In contrast, PBP1 knockdown triggered depletion of CFB2, MKT1, and other components of the PBP1-complex. Finally, all three knockdowns triggered depletion of cytokinesis initiation factors, consistent with a cytokinesis defect, confirmed here for all three knockdowns. Thus, genome-scale knockdown datasets facilitate the triage and prioritisation of candidate regulators. Quantitative proteomic analysis confirms 3'-UTR dependent positive control of VSG expression by CFB2 and interactions with additional mRBPs. Our results also reveal connections between VSG expression control by CFB2, ribosomal protein expression, and cytokinesis.

12:00 (10 mins) - A26179 - The mobile genome – transposable elements in *Schistosoma mansoni* and *Fasciola hepatica*

Authors

A Bilat[3]; T Brann[1]; J Tort[3]; C Chaparro[2]; **AV Protasio**[1];

[1] Department of Pathology, University of Cambridge, UK; [2] University of Perpignan via Domitia, France; [3] Facultad de Medicina, Universidad de la Republica, Uruguay

Abstract

The genomes of parasitic flatworms are occupied to a great extent by transposable elements. These mobile genetic entities have the capacity to replicate and/or move from one location to another in the genome. During this process, some of these transposable elements multiply in number and if left unchecked, they can cause significant and dramatic changes to the genomes they parasitise. Hence, an arms race between transposable elements and the host genome evolves. In this talk I will present data illustrating our current understanding of the diversity and genomic ecology of transposable elements in two trematodes, *Schistosoma mansoni* and *Fasciola hepatica*. Despite their close phylogenetic relationship, their transposable element complement is diverse, presenting an opportunity for the comparative study of their contribution to the host's genome architecture and function.

12:10 (10 mins) - A25379 - Using nanopore sequencing to identify transcript variation between different life cycle stages of the parasitic nematode *Strongyloides ratti*

Authors

D Lastik[1]; B Murcott[1]; V Hunt[1];

[1] University of Bath, UK

Abstract

Parasitic infections account for a large burden of disease in animals, plants, and humans. The gastrointestinal parasitic nematode genus *Strongyloides* has a unique and complex life cycle that alternates between genetically identical parasitic and free-living generations, making them a great model to study parasitism. Identification and characterization of life cycle specific gene expression is important for understanding the fundamental principles of parasitic mechanisms, its evolution, and the control/treatment of the parasite infections. Previous research has shown that parasitic and free-living adults both express a unique set of mRNA and sRNAs, respectively, implying an underlining mechanism of post-transcriptional gene regulation. Most of our knowledge about mRNA transcripts in parasites is incomplete as it relies on short read sequencing which often results in missing information about alternative splicing, 5'UTRs, 3'UTRs and polyA tails. These play an important role in gene regulation and currently need to be predicted computationally. In this study, using nanopore sequencing, we have identified full-length transcripts for the gastrointestinal parasite *Strongyloides ratti*; a species that has one of the best assembled genomes among nematodes (Hunt et al., 2016). We have obtained 50 million reads across 4 life cycle stages with a minimum of 76% of reads representing full-length transcripts. The sequenced life cycle stagesof *S. ratti* include genetically identical parasitic and free-living adults, both with at least 2 million full-length transcripts per replicate. **10,000** novel isoforms were identified across the 4 life cycle stages. In at least **80%** and almost **50%** of full-length mRNA we have established a 3'UTR* or a 5'UTR, respectively. To better understand the mechanisms of gene regulation in parasitism, we investigate how alternatively spliced transcripts, UTRs and polyA tails vary between life cycle stages. This new data will help us recognise the role of isoforms and UTR variation in parasitism and show how long read sequencing can improve our current genome annotations.

12:20 (10 mins) - A26095 - A single-cell atlas of the free-living miracidium larva of *Schistosoma mansoni*

Authors

T Attenborough[1]; **K Rawlinson**[1]; CL Diaz Soria[1]; G Sankaranaranayan[1]; G Rinaldi[1]; M Berriman[1];

[1] Wellcome Trust Sanger Institute, UK

Discussion

The neglected tropical disease schistosomiasis affects millions of people annually, and is caused by infection with *Schistosoma*, a genus of parasitic flatworms. More than 230 million people were estimated to need treatment in 2019, and over 700 million people live in areas where they are at

risk of infection. There is one currently available treatment, praziquantel, which is both safe and relatively effective. However, it has varying efficacy against different life stages, does not provide protection against re-infection, and drug resistance is of particular concern given the paucity of other treatment options. Thus, other potential drug treatments, vaccine candidates, and non-pharmaceutical interventions are all areas of active research. A key strategy to advance these goals is to develop a deeper understanding of the causative agent: *Schistosoma. Schistosoma mansoni* has a complex lifecycle involving multiple hosts, and goes through multiple developmental transitions before reaching maturity, where it is sexually dimorphic. *S. mansoni* eggs hatch in fresh water, where the free-living miracidia emerge. These miracidia seek out snail hosts, inside which they transform into mother sporocysts, before asexually producing large numbers of daughter sporocysts that generate human-infective cercariae to continue the life-cycle. In this project we seek to build a greater understanding of the cells present in miracidia, and their transcriptional activity, through single-cell sequencing and analysis. We've established that the miracidium is composed of just 365 cells and we have sequenced enough cells to achieve >10x theoretical cover of each cell. We have identified transcriptional profiles which indicate stem, muscle, protonephridia, neural, tegumental, and parenchymal tissues, and work is in progress validating these findings with *in-situ* hybridisations. Within these tissue types, we have detected subclusters of cells. These subclusters indicate functional heterogeneity, particularly within neural tissues, and we have identified miracidia-specific cell types such as the ciliary plates for swimming. Additionally, we have identified sex-specific transcriptional activity that is particularly striking in the stem cell populations of this sexually immature developmental stage. Our focus on single-cell detail in the miracidium provides the foundation for understanding the cell types and their transcriptomes that make up *Schistosoma mansoni.* Furthermore, identifying the cellular composition of this simple and short-lived larval stage will lead to a greater understanding of its infective behaviour that leads to the propagation of the life cycle.

Day 4 - Trafficking, Signalling *(Lecture Theatre P/X001)*
25-March-2022, at 11:50 to 12:40
Chairs - Dr Romina Nievas & Dr Nicola Baker

11:50 (10 mins) - A26027 - K13-associated endocytic structures in *Toxoplasma* are required for plasma membrane homeostasis rather than parasite growth

Authors
BN Mercado-Saavedra[1]; L Koreny[1]; **RF Waller**[1];
[1] University of Cambridge, Department of Biochemistry, UK

Discussion
Toxoplasma is a parasite that depends on nutrients provided by its host, but the processes through which this is achieved are largely unknown. We found a ring-like structure in *Toxoplasma* composed of proteins known to be important for endocytosis (DrpC, EPS15, the AP2 complex) and others that have not been implicated in this process before (K13, UBP1, Arf-GAP). This structure is present at the inner membrane complex (IMC) of the parasite in both mother and forming daughter cells, indicating that this structure is formed early in the development of the IMC and only makes contact with the plasma membrane during mature parasite formation. Live-cell imaging shows that this structure and its constituent proteins are stable features of the IMC. Depleting most of these proteins results in strong growth phenotypes, manifesting as extra-parasite cytosolic extensions within the parasitophorous vacuole, swollen parasites, and a failure of the rosette-like organisation of parasites. However, the replication rate was not significantly affected upon suppression of the structure's proteins. Our results show that while endocytosis is essential for the parasite's intracellular survival, its greater role in this parasite stage might be plasma membrane homeostasis rather than parasite nutrition.

12:00 (10 mins) - A25929 - Protein Kinase Involvement in *Leishmania* Cell Cycle Regulation Revealed Using Chemical Genetics

Authors
J Brambilla Trindade Carnielli[1]; J Brannigan[1]; M Saldivia[2]; T Wilkinson[1]; JC Mottram[1];
[1] University of York, UK; [2] Novartis Institute for Tropical Diseases, United States

AbstractCell division is a core biological process for both multicellular and unicellular organisms. It is a conserved process in eukaryotes, but *Trypanosoma*tids are evolutionary diverse, consequently they have some unique aspects to their cell division that is reflected in the repertoire of proteins involved in the process. Here, we used genetic and chemical approaches to explore the role of some essential protein kinases in cell cycle progression. We used CRISPR-Cas9 to perform precision editing of the *L. mexicana* genome to generate analogue sensitive mutants suitable for chemical genetic inhibition. For the kinetochore protein kinase KKT2, the cyclin-dependent kinase CRK9 and a CMGC family proteins kinase CMGCa, a replacement of the bulky gatekeeper methionine residue with a glycine in the ATP-binding site makes the enzymes sensitive to the bulky inhibitor 1NM-PP1. For the kinetochore protein kinases CLK1 and CLK2 (also known as KKT10 and KKT19, respectively) replacement of a cysteine near to the ATP-binding domain prevents binding of the c*ovale*nt Michael-acceptor in the inhibitor AB1, validating the specificity of this compound against CLK1/CLK2. The chemical validation demonstrated that these protein kinases are essential for the promastigote and intracellular amastigote stages of the parasite. The specific inhibition of CLK1/CLK2, KKT2 and CMGCa caused a cell cycle arrest in G2/M stage of the promastigote. A further investigation, by fluorescence microscopy labelling the mitotic spindle, revealed that KKT2 inhibition is followed by a significant accumulation of cells in early mitosis, where mitotic spindle coordination in the nucleus failed. Furthermore, it was observed that CMGCa inhibition also impaired chromosome segregation, but mitosis reaches a more advanced stage, suggesting CMGCa activity is required later in mitosis than KKT2. In addition, CLK1/CLK2 inhibition doesn't affect the coordination of the mitotic spindle, but it blocks cell cycle progression in cytokinesis. These studies bring new insights into the essential biological process of cell division in *Leishmania* and provide a source of new potential therapeutic targets.

12:10 (10 mins) - A26114 - Investigating the role of unique kinesin-2 motors in intraflagellar transport in trypanosomes

Authors - A A Alves[2]; P Bastin[1];
[1] Institut Pasteur, Paris, France; [2] Institut Pasteur, France

Discussion - Trypanosomes have a single flagellum essential for survival, motility, life cycle and host-parasite interaction. These parasites are an excellent model to study flagella assembly, as they build a new flagellum while conserving the old one. Flagella assembly requires a specific transport system called intraflagellar transport (IFT). IFT is the bidirectional movement of multiprotein complexes, or IFT trains trafficking along flagellar microtubules. The transport towards the ciliary tip is called anterograde, driven by kinesin-2. These motors comprise two subfamilies: the heterotrimeric kinesin-2, with two different motor subunits and a non-motor subunit, and the homodimeric kinesin-2, with two identical motor subunits. *Trypanosoma brucei* is unique since it lacks the heterotrimeric version but contains two kinesin-2 genes – KIN2A and KIN2B. KIN2A and KIN2B involvement on anterograde IFT transport was shown by RNAi knockdown of the two motors together. Still, how each motor contributes individually to the anterograde IFT remains unclear. To investigate KIN2A and KIN2B roles, we generated cell lines expressing the kinesins fused to the fluorescent protein mNeonGreen (mNG) and followed IFT by confocal spinning-disk microscopy. Compared to the IFT protein IFT81, KIN2A-containing particles move at a similar speed but have a lower frequency, suggesting KIN2A is not present in every anterograde train. KIN2B-containing particles present a unique pattern: only a minor part of them can reach the flagellar tip. Most KIN2B particles are restricted to the proximal region of the flagellum and are slower than IFT anterograde trains. Double-tagged cells expressing KIN2B and the IFT protein IFT140 confirmed that KIN2B proximal particles do not colocalise with the IFT140, showing they are not associated with IFT trains. In contrast, the distal KIN2B-containing particles have the expected IFT speed and colocalise with IFT140 but show a low frequency. The frequencies of KIN2A and KIN2B distal particles together reach the expected frequency of anterograde IFT trains. Together, our data suggest KIN2A and KIN2B carry different trains and function as homodimers.

Posters – by Number

Presenter: **Prof Somaia Abouakkada**, *Proessor, Alexandria University*

Poster 1 : Molecular identification of drug-resistance in *Trypanosoma evansi* of camels in Egypt

Authors - SS Abouakkada[2]; A Deweir[3]; HP Price[1]; N Labn[2]; SA henidy[2];
[1] School of Life Sciences, Keele University, UK; [2] Faculty of Veterinary Medicine, Alexandria University, Egypt; [3] Faculty of Veterinary Medicine, Alexandria University, Egypt, Egypt

Objective - *Trypanosoma evansi* is the causative agent of Surra, one of the most economically important veterinary diseases in the world. Emergence of drug resistance represents a great barrier to chemotherapeutic control against trypanosomosis. Drug resistance is usually caused by changes to the drug transporters of the parasites. In Egypt, the prevalence of drug-resistant trypanosomes in endemic regions remains poorly understood. In the present study, we aim to investigate molecular markers of drug resistance in the form of mutations in adenosine transporter P2 (TbAT1) and Aquaglyceroporin (AQP2) genes encoding drug transporters in different strains of *T.evansi* collected in Egypt. We have sequenced the TbAT1 and the AQP2 genes from six *T.evansi* field strains collected from camels from different localities in Egypt and one laboratory UK strain. Blood samples were collected from the animals for extraction of total genomic DNA was performed for amplification of 164 bp TBR1/2 primers. The seven TBR1/2 positive DNA samples were used for amplification of 1600 and 1416 bp encoding for adenosine transporter P2 gene and Aquaglyceroporin transporter gene, respectively. Purified PCR products were sequenced and BLAST analysis was performed to establish sequence identity to sequences accessed from GenBank.Results revealed that the seven tested DNA sampleswere positive to TBR1/2 primers. In addition, three strains representing South Sinai, Matrouh and Halaib had lost both AQP2 and TbAT1 genes. The other four strains were positive for AQP2 and TbAT1 genes and gave specific bands at 1416 and 1600 bp, respectively. Alignment results of AQP2 gene revealed no polymorphism at the nucleotide or amino acid levels at any of the tested strains. Alignment of AT1 gene sequences revealed substitution of TG nucleotide by CT at the position of 972 and 973 laboratory and Cairo1 strains, with insertion of two nucleotides, GC at the position of 974 and 975 that resulted in insertion of a new amino acid, Cysteine. Both strains carried the same point mutations. Cairo2 strain showed a substitution of one nucleotide T by C at the position of 972 and insertion of the two nucleotides GC at the same positions of laboratory and Cairo1 strains. This resulted in insertion of the amino acid, Glycine at this site. Moreover, substitution of nucleotide G by T at the position of 710 resulted in substitution of glycine amino acid at the position 237 with Valine. A Kom-hamada strain displayed no polymorphism neither at nucleotide nor amino acid levels. In conclusion, the data presented involved the detection of TbAT1 gene in 4 out of 7 strains of

Presenter: **Dr Zouhour El Mouna Ayadi**, *Maitre de conferences B, Université des Sciences et de la Technologie Houari Boum*

Poster 2 : Diversity of *monogenea* (Platyhelminths) gill parasites of Scombrid fishes off the Algerian coast

Authors - Z Ayadi[1]; K Benmeslem[1]; F Tazerouti[1];
[1] University of Sciences and Technology Houari Boumediene, Faculty of Biological Sciences, Laboratory of Biodiversity and Environment : Interactions and Genomes, Algiers, Algeria

Objective - The fish parasite diversity in the Algerian coast still incompletely explored. During a survay on gill parasites of two Scombrid fishes: *Euthynnus alletteratus* (Rafinesque, 1810) and *Sarda sarda* (Bloch, 1793) off Algerian waters we have collected 3 species of monogenean belonging to the sub-class Monopisthocotylea Odhner, 1912 and Polyopisthocotylea Odhner, 1912. Among Monopisthocotylea, we have identified one species of the family Capsalidae Baird, 1853: *Capsala manteri* Price, 1951 from *Euthynnus alletteratus*. This monogenean is characterized by a septate haptor provided of 7 septa and two pair of anchors. The sub-class Polyopisthocotylea is represented by 2 species belonging to the family Hexostomatidae Price, 1936: *Hexostoma lintoni* Price, 1936 and *Neohexostoma euthynni* (Meserve, 1938) Price, 1961 collected from *Sarda sarda* and *Euthynnus alletteratus* respectively. The most distinctive features between the two genera are the disposition of clamps and the presence of waist-like constriction between the testicular region and the haptor. In the genus *Hexostoma,* the clamps are disposed in a more or less straight transverse row (Rohde, 1978) and there is no waist-like constriction in the testicular region, whereas in *Neohexostoma*, the clamps are disposed in a more or less vertical row with waist-like constriction in the testicular region.

The present study is the first record of *Capsala manteri, Hexostoma lintoni* and *Neohexostoma euthynni* from Scombrid fishes of Algerian waters. In addition, we report for the first time *Hexostoma lintoni* from *Sarda sarda* off Mediterranean.

Key-words: Monogenea, parasites, Capsalidae, Hexostomatidae, Scombridae, Algerian coast

Presenter: **Miss Supannee Taweechai**, *PhD student, University of Leeds*

Poster 3 : Biochemical characterization and inhibitor screening of UMP-CMP kinase from malaria parasites

Authors - **S Taweechai**[1]; G McConkey[1];
[1] School of Biology, Faculty of Biological Sciences, University of Leeds, UK

Objective - In this research, we are investigating UMP-CMP kinase, a pivotal enzyme in the pyrimidine metabolism for DNA and RNA synthesis that was identified by in silico modelling as an antimalarial target and found essential based on negative evidence from genome-wide *Plasmodium falciparum* saturation mutagenesis [1,2]. To dissect the functions of UMP-CMP kinase from *P. falciparum* (PfUMP-CMP kinase), the recombinant protein was produced and its kinetic properties investigated. By using ATP as phosphate donor, the enzyme activity assay demonstrated that the ribonucleoside monophosphates CMP and UMP are the preferred substrates of PfUMP-CMP kinase compared to deoxyribonucleoside monophosphates (ie. dCMP). CMP showed the lowest K_m of 28 µM whereas UMP and dCMP had 3.9-fold and 17-fold higher K_m values, respectively. In addition, the role of cysteines in PfUMP-CMP kinase was investigated through iodoacetamide (IA) alkylation. The kinase was found to be sensitive to alkylation with significantly decreased enzyme activity that was dependent on IA concentration. This implies that the cysteines are necessary for PfUMP-CMP kinase activity and accessible, that could lead to development of an irreversible Cys-interacting inhibitor. Inhibitors targeting the cysteineswere modelled and selected in collaboration with medicinal chemists, and tested for inhibitory activity. Compound 7 exhibited the lowest inhibition constant (K_i value) for PfUMP-CMP kinase; at the low micromolar range, making it a promising hit compound for further development of candidate compounds.

1. Totanes, F.I.G., Docteral dissertation, 2017, University of Leeds

2. Zhang. M., et al., 2018, 360(6388)

Presenter: **Miss Laura Filipe**, *Doctoral researcher, Durham University*

Poster 4* : Deconvoluting the Mode of Action of a Suite of Novel Antileishmanials

Authors - **L Filipe**[1]; E Alpizar-Sosa[1]; PW Denny[1];
[1] Durham University, UK

Objective - Cutaneous leishmaniasis (CL) is a human disease caused by species of the insect-borne protozoan parasite *Leishmania*. CL presents with skin lesions, which can produce lifelong scarring or serious complications. While rarely fatal, CL causes a huge global burden of morbidity and social stigma. Its chronic debilitating nature and link to poverty classify CL as a Neglected Tropical Disease. There is no vaccine, and all available drugs are toxic and increasingly ineffective with rising drug resistance. The lack of known therapeutic targets in *Leishmania* species hinders drug development, so their discovery is of utmost importance.

Our current work examines 17 anti-kinetoplastid compounds identified at GSK via high throughput phenotypic screening and not currently being pursued in any drug discovery effort. These compounds were triaged by GSK according to anti-kinetoplastid activity in *Leishmania (L.) donovani* (visceral leishmaniasis) or *Trypanosoma cruzi* (Chagas Disease) ($EC_{50} \leq 10$ µM), availability (≥ 10 mg), chemical druggability and selectivity (Peña et al., 2015). Using a pipeline of cutting-edge 'omic' tools and industrial partnerships, which builds on our previous work (Mina et al., 2021), we aim to deconvolve the modes of action of these compounds in *L. major* and *L. mexicana* (causative agents of CL), in the hope of discovering novel therapeutic targets.

To date, we have demonstrated high potency of some of these compounds in *L. major* and/or *L. mexicana* and for these produced resistant cell lines by *in vitro* evolution, which are currently under investigation following whole genome sequencing. Following the identification of potential targets further validation will be sought utilising metabolomics, lipidomics and proteomics, alongside genetic validation of targets. Using CRISPR Cas9 technology the proposed target of the recently identified antileishmanial tamoxifen (a antineoplastic drug; Trinconi et al., 2018), inositol phosphorylceramide synthase, was ablated. The data generated phenotyping these mutant strains demonstrated the importance of this approach in target validation.

Presenter: **Prof Uwemedimo Ekpo**, *Professor, Federal University of Agriculture Abeokuta*

Poster 5* : Occurrence and Geographical distribution of *Microsporidia* in tick population in Ogun State, Nigeria

Authors - DO Ajagbe[1]; OO Omitola[1]; FA Akande[1]; AA Aladeshida[1]; UF Ekpo[1];
[1] Federal University of Agriculture, Abeokuta, Nigeria

Objective - *Microsporidia* endosymbionts are promising microorganisms for the biological control of arthropod vectors. We mapped the occurrence and geographical distribution of *Microsporidia* in ticks, considered as the second most important arthropod vector of public health concern after mosquitoes. Tick species were collected from 68 cattle and 13 dogs from 4 locations viz, Gbonagun, Asero, Lafenwa and Ita-Eko in Ogun State, Nigeria. The ticks were morphologically identified and characterized into sex and developmental stage. Each tick was then homogenized, and an aliquot was used to prepare a thin smear on a microscope slide, air-dried, fixed with 70% methanol, stained with Giemsa, and examined microscopically for microsporidia spores. A total of 880 ticks were collected and identified as *Amblyomma variegatum* (8.07%), *Rhipicephalus* (*Boophilus*) *decoloratus, (6.93%), Rh.* (*Boophilus*) *microplus* (47.61%)*, Rh.* (*Boophilus*) *annulatus* (37.05%) *and Hyalomma marginatum* (0.34%). 277 (31.4%) of ticks were infected with *Microsporidia.* The prevalence of *Microsporidia* infection by tick species was as follows: *Amblyomma variegatum* (23.94%), *Rhipicephalus (Boophilus*) *decoloratus* (24.59%)*, Rhipicephalus* (*Boophilus*) *microplus* (37.47%)*, Rhipicephalus* (*Boophilus*) *annulatus* (26.99%) and *Hyalomma marginatum* (0%). The occurrence of *Microsporidia* infection in the species of the

different tick collected was significant (p=0.005). This study shows that *Microsporidia* are widely distributed in major tick species populations in Ogun State. PCR screening and characterization of the detected *Microsporidia* is ongoing.

Presenter: **Mr Jose Carlos Paredes-Franco**, *PhD candidate, University of Dundee*

Poster 6* : Expression and characterization of a mitochondrial fucosyltransferase from *Trypanosoma cruzi* and use of monoxenous parasite *Crithidia fasciculata* as an enzymatic source for synthesis of radioactive GDP-Fucose.

Authors - **JC Paredes-Franco**[1]; MA J Ferguson[1];
[1] Wellcome Centre for Anti-Infectives Research, Division of Biological Chemistry and Drug Discovery, University of Dundee, Dundee, UK, UK

Objective - For all eukaryotes, surface glycoproteins and glycolipids are made in the secretory pathway (i.e., the endoplasmic reticulum and the Golgi apparatus) and, therefore, this is where the vast majority of glycosyltransferases reside.

However, previous research has identified a specific glycosyltransferase enzyme, a fucosyltransferase, in the single mitochondrion of *Trypanosoma brucei* and of *Leishmania major* (Bandini G, et al., 2021, eLife; Guo H, et al., 2021, PNAS). This enzyme, called FUT1, not only has an unusual mitochondrial localization, but also it is essential for both parasites. The presence of FUT1 suggests that some novel type of glycoprotein or glycolipid glycosylation occurs in the mitochondria of *Trypanosoma*tid parasites. Nevertheless, nothing is known about the orthologous enzyme from *Trypanosoma cruzi* (TcFUT1) and so we decided to perform expression and characterization studies with it.

Based on the work done with TbFUT1 (Bandini G, et al., 2021, eLife), initial expression and purification attempts were done using different *E. coli* strains as expression systems. When eventually recombinant TcFUT1 was purified, although in low yields, we performed fucosyltransferase activity assays involving a panel of synthetic glycosidic acceptor substrates and radioactive guanosine diphosphate fucose (GDP-[^{3}H]Fuc) as the donor substrate. However, no activity was detected, and new trials were done using eukaryotic Expi293F cells as the expression system, since HEK293-derived cells have been reported as useful for expressing many human glycosyltransferases. At the same time, we have established a procedure to synthesize our own GDP-[^{3}H]Fuc based on previous knowledge from our group on using the monoxenous parasite *Crithidia fasciculata* as an enzyme source for the generation of this and other GDP nucleotide sugars (Schneider P, et al., 1995, Biochem. J.; Mengeling B J, et al., 1999, Analytical Biochem.). Combining our home-made radioactive donor, and recombinant TcFUT1 obtained from the culture medium of our eukaryotic expression system, we aim to define the substrate specificity of this fucosyltransferase and potentially characterize the fine chemical structure of its product(s).

Presenter: **Miss Catherine Oke**, *PhD Student, University of Edinburgh*

Poster 7 : How will the response of mosquitoes to vector control shape malaria parasite evolution?

Authors - **C Oke**[1]; SE Reece[1];
[1] University of Edinburgh, Institute of Evolutionary Biology, UK

Objective - Malaria parasites (*Plasmodium* spp.) have a complex lifecycle, where transmission between human hosts relies on passage through female *Anopheles* mosquitoes. Vector control tools targeting mosquitoes, such as insecticide-treated bednets, reduce transmission opportunities for the parasite and have been key in decreasing the malaria burden over the past two decades. This widespread use of vector control tools has led to significant changes in mosquito ecology and their evolutionary responses are well documented (e.g. insecticide resistance). However, how vector control interventions alter the nature of vector-parasite interactions, and consequently the selection pressures on parasites, are unknown. The counter-evolution of parasites in response to interventions that act within the host (e.g. antimalarial drug resistance) has mitigated fitness losses for parasites. Therefore, there is potential for parasites to adapt to the consequences of vector control interventions, but these evolutionary responses have been overlooked. Using different combinations of mosquito strains and well-characterised *P. chabaudi* strains as a model system, we aim to investigate whether vector control interventions affect vector-parasite interactions and selection on parasite traits. This includes evolutionary responses of parasites to direct interactions with insecticides (e.g. via insecticide resistant mosquitoes) and indirect consequences (e.g. via bednets altering the biting time-of-day of mosquitoes). In particular, quantifying the genetic variation and phenotypic plasticity underpinning parasite traits involved in sporogony will provide insight into the evolutionary potential of parasites in response to changing vector ecology and altered within-vector environments. Understanding the short and long-term consequences of how vector control could affect malaria parasite evolution is key for the mitigation of any detrimental consequences of current control programmes. Furthermore, knowledge of factors that shape parasite evolution could inform development of novel control strategies by targeting evolutionarily constrained parasite traits.

Presenter: **Mr Shadrack Madu**, *Research Student, De Montfort University*

Poster 8* : Prediction of Miltefosine Exposure in Mouse Model using Physiologically Based Pharmacokinetic Modelling (PBPK) Approach

Authors - **SJ Madu**[1]; M Li[1];
[1] De Montfort University, UK

Objective - Leishmaniasis is an abandoned tropical illness caused by the protozoan[1]. Its forms include: cutaneous leishmaniasis (CL), mucocutaneous leishmaniasis (ML), and visceral leishmaniasis (VL). Among them, VL is the most severe form with higher chances of fatality without proper medical intervention. The available treatments for VL are limited, toxic, and expensive. Miltefosine (MT) is the first and only oral medication to be successfully utilised as antileishmanial agent. However, administration of MT is associated with many side effects, including gastrointestinal manifestations, hepatotoxicity and nephrotoxicity which requires monitoring thereby increasing the cost of therapy [2]. It is also associated with high chances to develop resistance as a result of its long half-life of approximately seven days[3]. Drug combinations have proved to be a successful strategy to shorten the course of therapy, reduce toxicities through lower dosage and reduce the selection of resistant mutations for several infectious diseases, most notably malaria and tuberculosis. Similarly MT combination therapies, such as fluconazole and MT [4], AmBisome and MT [5], and ketoconazole and MT [6], have demonstrated to have potential in delaying the development of resistance and shortening the duration of treatment for improving compliance and reducing cost. However, to develop an optimal MT combination therapy and dosing regimens, extensive preclinical in vitro and animal tests must be conducted to show its efficacy, safety and benefits, which are expensive and time consuming. Physiologically based pharmacokinetic (PBPK) modelling is widely used within the pharmaceutical industry to predict oral drug absorption [7]. It can be used to predict the plasma concentration–time profiles from preclinical in vitro and in vivo data, providing a valuable resource to support decisions at various stages of the drug development process. The aim of this work was to develop a PBPK model to predict the plasma concentrations of MT under different dosing regimens in mice. Simcyp (Mouse Simulator Version 20) was employed in the work. As the first step of the model development, simulations of the oral mouse plasma concentration−time profiles of MT were performed based on the relevant physicochemical, physiological, and pharmacokinetic input data of the drug which have been taken from the literature [8-12]. The PBPK model simulations were then compared to the in vivo plasma concentration−time profiles of MT from animal experiments, which provided a means to refine the model parameters through the parameter sensitivity analysis and parameter fittings and to verify the PBPK model's predictive ability of the in vivo performance. Finally, the model validation was conducted using experimental data reported in the literature [10], indicating that the developed PBPK model can accurately predict the concentration-time profiles of MT in mice following different single/multiple dose administration. Refer

Presenter: **Dr Thaise Teixeira**, *Postdoctoral Researcher, Universidade Federal de São Paulo*

Poster 9 : Deletion of the P21 gene triggers changes in the invasion and replication of *Trypanosoma cruzi*

Authors - T Teixeira[2]; MA Chiurillo[3]; N Lander[3]; CC Rodrigues[4]; TS Onofre[2]; ER Ferreira[1]; CM Yonamine[2]; JG Santos[4]; RA Mortara[2]; CV Silva[4]; JF Silveira[2];
[1] University of York, UK; [2] Federal University of Sao Paulo, Brazil; [3] University of Cincinnati, United States; [4] Federal University of Uberlandia, Brazil

Objective - P21 is a protein encoded by a single-copy gene and no orthologs are found in other *Trypanosoma*tids. Previous studies demonstrated the role of P21 during the infection of *Trypanosoma cruzi.* P21 operates as a signal transducer molecule, triggering a signaling cascade, which results in the alteration of the actin cytoskeleton of host cell, increasing the internalization of parasites. In addition, *T. cruzi* infected mice treated with recombinant P21 have shown increasing in leukocytes chemotaxis and fibrosis, but reduced angiogenesis and replication of intracellular amastigotes, indicating the role of P21 in pathogenesis of Chagas disease. However, the mechanisms underlying the role of P21 remain poorly understood. In this study, we generated P21 knockout parasites using CRISPR/Cas9 and analysed the phenotypic effects of the deletion of this gene. Our results showed that ablation of P21 reduced the growth rate of epimastigotes evaluated for 14 days. Furthermore, P21 knockout epimastigotes showed an increase in the length of the G1 phase and reduction in the S phase, resulting in a delay on cell cycle progression. Invasion assays performed with metacyclic trypomastigotes revealed that P21 knockout impairs parasites ability to invade HeLa cells when compared to the wild-type control. In contrast, intracellular replication rate of amastigotes is increased in P21 knockout parasites, observed after 72 hours of infection. Taken together, our data reveals the involvement of P21 during the different life stages of *T. cruzi*, demonstrating its importance throughout the parasite life cycle. Support: FAPESP 2016/15000-4, FAPESP 2019/05049-4, FAPEMIG APQ-00971-17, CNPq and CAPES.

Presenter: **Mr Benedict Davies**, *PhD student, St George's, University of London*

Poster 10 : Characterisation of a cation diffusion facilitator from the malaria parasite *Plasmodium falciparum*

Authors - BM Davies[1]; SJ Moss[1]; S Krishna[1]; HM Staines[1];
[1] St George's University of London, UK

Objective - Transition metals such as zinc and iron are essential micronutrients to the malaria parasite *Plasmodium falciparum,* yet become toxic at high concentrations. Hence, the careful regulation of transition metals is required as the parasite progresses through its complex life cycle. **C**ation **D**iffusion **F**acilitators (CDFs) are a family of membrane transporters that enable the detoxification of transition metal ions from cells. Here we study the sole predicted CDF from the malaria parasite, *Plasmodium falciparum*. Utilising the *Saccharomyces cerevisiae* (yeast) heterologous expression system, we show that expression of PfCDF in a zinc-sensitive yeast line (Dzrc1cot1) conferred partial zinc tolerance when cultured in zinc-replete conditions. This suggests that PfCDF transports Zn^{2+} out of the cytosol, thus potentially implicating PfCDF as an important mediator of zinc tolerance in the parasite. Additionally, comparisons with functionally characterised CDF homologues suggest that PfCDF features key amino acid residues that could enable the transport and therefore detoxification of iron, in addition to zinc. This hypothesis will form the basis of future study.

Presenter: **Ms Antonella Bacigalupo**, *PhD Student, University of Glasgow, BAHCM*

Poster 11 : Wild foci of the Chagas disease vectors *Triatoma infestans* and Mepraia spinolai in Chile, a country that has declared the interruption of *Trypanosoma cruzi* vectorial transmission.

Authors - A Bacigalupo[1]; P Arroyo[2]; RA Gacitúa-Gajardo[2]; D Gajardo-Canto[2]; R Muñoz-Ramos[2]; JA Segura[3]; MJ Caniullán[4]; C Adones[5]; A Parra-Garcés[6]; PE Cattan[2]; KR Elmer[1]; MS Llewellyn[1];
[1] Institute of Biodiversity, Animal Health and Comparative Medicine, University of Glasgow, UK, UK; [2] Facultad de Ciencias Veterinarias y Pecuarias, Universidad de Chile, Chile; [3] Seremi de Salud Región Metropolitana, Unidad de Zoonosis y Control de Vectores, Chile; [4] Seremi de Salud Región de Atacama, Unidad de Zoonosis y Control de Vectores, Chile; [5] Seremi de Salud Región de Coquimbo, Unidad de Zoonosis y Control de Vectores, Chile; [6] Ministerio de Salud, Oficina de Zoonosis y Control de Vectores, Chile

Objective - Chagas disease is a neglected illness caused by the protozoan parasite *Trypanosoma cruzi*. Nearly six million people are infected and about 10,000 die annually as a consequence of this chronic disease. Transmission includes several routes, including vectorial, which involves hematophagous insects of the Subfamily Triatominae (Hemiptera: Reduviidae) that are primarily distributed in the Americas. Some triatomines have adapted to living inside human dwellings, and these species represent a higher infection risk, due to their contact rate with the inhabitants. *Triatoma infestans* is one of the species highly adapted to live in houses and is distributed in Peru, Bolivia, Brazil, Paraguay, Uruguay, Argentina and Chile. In Chile, *T. infestans* domestic colonies have been eliminated by residual insecticide spraying, but adults continue to invade houses. The interruption of intra-domiciliary transmission is currently based on governmental spraying of the house for three consecutive years after a specimen of this species is reported. Triatomines of the genus *Mepraia* are considered mainly sylvatic; however, they are frequently found in peridomestic areas, and can also invade houses. Due to repeated findings of *T. infestans* in Chilean dwellings, our goal was to detect sylvatic foci of this species in six endemic areas, to compare them with the specimens found inside houses. We set ~60 georeferenced yeast-baited traps per night during 5 nights in each of the six sylvatic sites during the Southern hemisphere summer of 2020-2021, spanning a ~600 km latitudinal range that included arid, semiarid and Mediterranean climate. Triatomines were collected, classified, and placed in labelled flasks, protected from extreme environmental conditions. We captured 260 *T. infestans* and over 4,000 individuals of the non-target species, *M. spinolai*. The altitude of *T. infestans* trapping points ranged from 370 to 1476 m.a.s.l (738.2±189.1). Only one male adult *T. infestans* was captured manually at night. The nymphal instars I-II were most frequently found (72.8%), followed by III-IV (17.5%), and instar V (9.3%). The highest abundance of *T. infestans* was detected in the southernmost study site (n=132), where it was the only species recorded, while *M. spinolai* was widespread in several of the studied sites. Both species were found in terrestrial bromeliads and rock piles, whilst *M. spinolai* was also found in rocky outcrops, and this is the first report of *T. infestans* among rubble and vines' wood piles in Chile. With this study, we expand the knowledge of Chilean sylvatic foci for both vector species and show that wild triatomine populations remain a threat in endemic areas, even in countries with high levels of domiciliary vector control. The ecotope preferences of these species should be informed in future prevention campaigns displayed at endemic areas, targeted to inhabitants, workers and industries, to avoid risky behaviour that could expose people to vectors. To continue this work, we are performing DNA extraction, followed by sequencing, to analyse *T. infestans* population genomics, comparing these wild specimens with those found inside houses.
Funding: ANID/Programa Becas/Doctorado Becas Chile 2019 72200391; ANID/FONDECYT Regular 1180940.

Presenter: **Mr Oluwayomi Adeyemi**, *Assistant Lecturer/PhD Student, University of Lagos*

Poster 12 : Prevalence and Associated Risk Factors of *Eimeria* Species of Chicken in Lagos, Southwest, Nigeria.

Authors - OO Adeyemi[1]; ET Idowu[1]; B Akinsanya[1]; ID Jatau[3]; DP Blake[2];
[1] University of Lagos, Nigeria; [2] Royal Veterinary College, UK; [3] Ahmadu Bello University, Zaria, Nigeria

Objective - Coccidiosis is an intestinal disease caused by apicomplexans of the genus *Eimeria* that affects chicken health and productivity globally. Here, we determined the occurrence and diversity of *Eimeria* parasites of chicken, and accessed associated risk factors in Lagos, Nigeria. Pooled faecal/litter samples were collected from 245 chicken flocks selected at random from 137 chicken farms. Samples were screened by microscopy for *Eimeria* oocysts after salt floatation. Oocyst enumeration per gram (OPG) faeces was estimated and oocyst length/width measurements taken for species identification. Our findings revealed farm and flock prevalence of 56.2% and 45.3% respectively, with average oocyst count of $1.5x10^4±7.3x10^4$opg per flock. Broiler flocks had the highest mean oocyst counts ($4.8x10^4±1.6x10^5$opg). Birds less than five weeks (64.7%) and those between six and ten weeks (68.8%) were observed to have higher prevalence of infection when compared to older birds. Oocyst counts were highest in chicks up to five weeks ($2.8x10^4±4.7x10^4$opg). Larger flocks had a significantly higher number of oocysts ($4.1x10^4±2.8x10^4$opg) compared to smaller flocks ($6.6x10^3±1.3x10^4$opg) ($P<0.05$). Higher mean oocyst counts were found in deep litter systems ($3.0x10^4±1.2x10^5$opg) than battery cages ($6.8x10^3±1.9x10^4$opg). All seven species recognized to infect chicken were detected: *E. maxima* (35%), *E. brunetti* (34%), *E. mitis* (28%), *E. necatrix* (26%), *E. acervulina* (24%), *E. tenella* (18%) and *E. praecox* (11%). Mixed species infections were more common than single species. We show that *Eimeria* species are circulating among chicken reared in commercial poultry establishments in Lagos State and suggest that urgent steps be taken to address the situation. **Keywords:** Coccidiosis, Prevalence, Chicken, *Eimeria* species, Nigeria.

Presenter: **Dr Ana Menezes**, *Postdoctoral researcher , Butantan Institute*

Poster 13* : The impact of glucose in *Trypanosoma cruzi* viability, cell growth, cell cycle progression, differentiation, and histone post-translational modifications

Authors - AP Menezes[2]; AM Silber[1]; MC Elias[2]; JP Cunha[2];
[1] Universidade de São Paulo, Brazil; [2] Butantan Institute, Brazil

Objective - The *Trypanosoma cruzi*, the etiological agent of Chagas disease, is a digenetic flagellated parasite that infects vertebrate and invertebrate hosts. Metacyclogenesis involves the differentiation of epimastigote into metacyclic trypomastigote forms and is triggered by nutritional stress, cellular adhesion and by changes in the microenvironments such as temperature and pH. This differentiation induces morphological alterations as well as changes in nuclear position and shape, which occurs in parallel with an increasement in the heterochromatin and changes in histone post translation modifications (PTMs). Histone acetylation is associated with an open chromatin status and transcription activation. Importantly, the acetyl-CoA, a central metabolite, is the main source of acetylation indicating an important link between metabolism and epigenetics. As the glycolytic pathway is one of the main sources of acetylCoA, we aimed to evaluate the impact of glucose concentration on the parasite growth, viability, cell cycle progression, metacyclogenesis, and global histone acetylation levels. The *T. cruzi* CL Brener parasites were kept in the Liver Infusion Tryptose (LIT) medium, pH 7.4, supplemented with 10% inactivated fetal bovine serum with a glucose gradient from 0.0 to 0.4% (w/v), at 28°C. The epimastigotes growth curve was initiated at a density of 2.5x106 cells/mL, and the phenotypical analysis (growth, viability, and cell cycle progression) were evaluated at three-time points of the exponential phase, namely: early (at 24 h), middle (at 72 h), and late (at 120 h). As expected, parasite density was higher at glucose concentration of 0.2% and 0.4% compared to 0.0% and 0.1%. Furthermore, at late timepoint, the cell viability (MTT assay) was dropped by around 30% in parasites cultivated with 0-0.1% of glucose. . Flow cytometry analyses indicated that glucose concentration does not interfere in the cell cycle progression. Interestingly, the differentiation rate of epimastigotes to metacyclic trypomastigotes was higher in the parasites kept at 0.0 to 0.2% of glucose compared with those cultivated in higher glucose concentrations. Using an optimized workflow of MS-based data-independent acquisition analysis preliminary data showed no differences in the global histone acetylation levels in parasites cultivated with 0.0 and 0.4% of glucose. Ongoing research is being performed to associate glucose concentration and the generation of acetyl-CoA and global histone acetylation. The results proposed in this study will be pioneering to uncover the underestimated potential of glucose metabolism in epigenetic changes in parasite protozoa.

Presenter: **PhDStudent Affaf Boukadoum**, *Phd Student, University of Sciences and Technology Houari Boumdienne, Faculty of Biological Sciences, Laborratory Biodiversity and Environnement : Interactions an*

Poster 14* : Some species of the family *Microcotylidae* (*Polyopisthocotylea, Monogenea*) from Sparid fishes off the Algerian coast

Authors - A Boukadoum[1]; FZ Zedam[1]; F Tazerouti[1];
[1] University of Sciences and Technology Houari Boumdienne, Faculty of Biological Sciences, Laborratory Biodiversity and Environnement : Interactions and Genomes, BP 32, El Alia Bab Ezzouer, Algiers, Algeria

Objective - During the examination of four species of Teleostean fishes (*Diplodus sargus sargus* (Linnaeus, 1758), *Lithognathus mormyrus* (Linnaeus, 1758), *Sarpa salpa* (Linnaeus, 1758) and *Sparus aurata* Linnaeus, 1758) caught in the Algerian coast, five gill ectoparasitic monogenean species were found, which all belong to the family: Microcotylidae Taschenberg, 1879.

The species *Atriaster heterodus* Lebedev & Parukhin, 1969 and *Polylabris tubicirrus* (Paperna & Kohn, 1964) Mamaev & Parukhin, 1976 were both found in *Diplodus sargus sargus; Bychowskicotyla mormyri* (Lorenz, 1878) Unnithan, 1971 was collected from *Lithognathus mormyrus; Atrispinum salpae* (Parona & Perugia, 1890) Euzet & Maillard was obtained from *Sarpa salpa* and finally *Sparicotyle chrysophrii* (Van Beneden & Hesse, 1863) Mamaev, 1984 was extracted from *Sparus aurata*. The key to the identification of these monogeneans is based on the morpho-anatomical characteristics such as the shape and armature of the genital atrium.

This study has allowed us to have further insight into the biodiversity of the parasitic Monogenea in the Algerian Teleostean fishes.

Keywords: Biodiversity, Parasites, Monogenea, Microcotylidae, Sparid fishes, Algerian coast.

Presenter: **Ms Yasmine Kumordzi**, *PhD student, Durham University*

Poster 15 : An investigation into Leishmania genome plasticity in response to disruption of sphingolipid biosynthesis

Authors - Y Kumordzi[2]; E Alpizar-Sosa[2]; M Barrett, PW Denny[2];
[1] University of Glasgow , UK; [2] Durham University, UK

Objective - *Leishmania* parasites cause devastating diseases in tropical areas around the world. With a lack of vaccines, treatment relies entirely on drugs such as amphotericin B which have several limitations, including severe side effects and emerging resistance. These are major concerns

worldwide which have led to the use of genetic-based approaches for the identification of targets essential for parasite survival where can be exploited for drug development.
Genetic approaches in *Leishmania* species have relied heavily on homologous recombination, however whole genome sequencing of a serine palmitoyltransferase (SPT, the first enzyme in sphingolipid biosynthesis) knockout in *L. major* identified surprising non-targeted deletions. The putative functions of these encoded proteins of the deleted genes led us to consider that they may have compensated for the loss of SPT. Recent advancements in genetic technology include CRISPR-Cas9, this was employed to investigate this phenomenon further in the tractable *L. mexicana* model. Notably, deletion of SPT was not possible in the parental background suggesting an essential function. However, loss of the SPT locus was achieved when the genes encoding ceramide synthase (an upstream enzyme in biosynthesis) were knocked out first. This provided proof of principle that compensatory deletions may facilitate the loss of essential genes, a finding that was further investigated with respect to the non-targeted genes lost in the *L. major* SPT knockout. In an alternative mode of genome plasticity, CRISPR-Cas9 mediated deletion of sphingosine kinase (SK) in *L. mexicana* resulted in targeted deletion and a phenotype resembling that reported for homologous recombination driven SK knockout in *L. major* (*Zhang et al*, 2013). However, further analyses demonstrated that the gene encoding SK was maintained in a different chromosomal location, leading us to conclude that it was essential.
Taken together these data demonstrate that genetic knockouts, especially those obtained via homologous recombination, need reanalysis due to the genome plasticity *Leishmania.*

Presenter: **Miss Praveena R G Chandrasegaran**, *Research Assistant, University of Glasgow*

Poster 16* : Comparative single cell transcriptomic analysis of the murine CNS in response to *T. brucei brucei* and *T. brucei gambiense* infections

Authors - **P Chandrasegaran**[2]; M Sinton[2]; TD Otto[1]; A MacLeod[2]; JF Quintana[2]; A Girard[2]; M Kimuda[2];
[1] Institute of Infection, Immunity & Inflammation, University of Glasgow, UK; [2] Wellcome Centre for Integrative Parasitology (WCIP), UK

Objective - Chronic infection with *Trypanosoma brucei*, the causative agent of Human African trypanosomiasis (HAT), induces gliosis and neuroinflammation in the central nervous system (CNS). Disease outcome varies greatly depending on the parasite subspecies; rhodesiense HAT is considered more aggressive, whereas *gambiense* HAT often causes a milder, subclinical infection. In this study, we used single cell transcriptomics to investigate CNS responses in murine models of infection with trypanosome causing subclinical or clinical infections (*T. b. gambiense* and *T. b. brucei*, respectively). We analysed ~20, 000 cells from the hypothalamus of naïve and infected mice and identified a total of 10 cell clusters with an ~500 genes/ cell and ~1,000 transcripts/cell, including microglia, astrocytes, vascular-associated cells, T and B cells. Of these, B cell with a regulatory phenotype (*Ighm, Cd79a, Cd79b, Il10*) and microglia with a robust pro-inflammatory phenotype (*Il1a*, *Tgfbr1*, *Ifngr1*, *Il6ra*, *Il10ra*) were abundant in *T. b. brucei* infection compared to *T. b. gambiense*. These observations are consistent with a reduced gliosis and neuroinflammation in *T. b. gambiense* infections compared to *T. b. brucei* infections. Furthermore, *T. b. gambiense* infection induces a progressive change in circadian activity without changes in clinical scoring, whereas *T. b. brucei* infection induces clinical symptoms that precedes changes in circadian behaviour. Taken together, our data suggest that changes in circadian behaviour in *T. b. gambiense* infection may arise due to low-grade CNS inflammation. Given the lack of clinical symptoms and based on the transcriptional and behavioural findings presented here, we propose that infection with *T. b. gambiense* is an ideal model to study how trypanosomes interfere with circadian rhythms without the need for pharmacological interventions. Our work provides important insights into mechanisms underlying the immunological responses of the CNS to different parasite subspecies, opening new avenues to investigate the molecular basis of HAT-associated circadian disorders.

Presenter: **Dr Fernanda Coelho**, *Schistosome and Snail Resource, Instituto René Rachou (Fiocruz-Minas)*

Poster 17 : The Schistosome and Snail Resource (SSR) - supporting global schistosomiasis research

Authors - **FS Coelho**[2]; A Cieplinski[1]; V Yardley[2]; AM Emery[1]; A Bustinduy[2]; B Webster[1];
[1] Natural History Museum, UK; [2] London School of Hygiene & Tropical Medicine, UK

Objective - Schistosomiasis (bilharzia) is a chronic and debilitating tropical parasitic disease caused by schistosomes (*Schistosoma* spp.) transmitted by freshwater snails. It is a Neglected Tropical Disease (NTD) of both humans and animals, with considerable health and economic impacts. Endemicity is associated with low/middle-income countries with considerable disease burden within impoverished communities despite widespread control efforts. While substantial advances have been made in the control of schistosomiasis, the diversity and complexity of *Schistosoma* species and their specific fresh-water snail hosts warrants fundamental research requiring lifecycles, live material and diverse collections. Due to the complexity and financial burden of maintaining *Schistosoma* lifecycles currently, very few labs are able to maintain the parasites and/or the snail hosts and current long-term cultures/collections lack the genetic heterogeneity observed in natural populations. Without the availability of diverse *Schistosoma* lifecycles/ live material, future research faces substantial obstacles. The Schistosome and Snail Resource (SSR) is a Wellcome Trust funded (2021-2026) biomedical resource run through a partnership between the Natural History Museum (London) and the London School of Tropical Medicine and Hygiene. Its overall aim is for the creation and maintenance of live material (*Schistosoma* and Snail host species), lifecycles and collections that are currently limited or that do not exist elsewhere. The SSR aims to provide access to: 1) the "standard/model" *Schistosoma* and snailspecies; 2) key African *Schistosoma* species/strains; 3) cultures of diverse snail vectors, enhancing current research and capacity while enabling new research avenues. Our historical expertise in establishing and maintaining unique schistosome and snail isolates/collections from different endemic settings, together with the state-of-the art snail facility (NHM) and LSHTM rodent facility will facilitate the development of the resource. The SSR, is an open resource for the global research community and will add considerable value by facilitating priority research needed to support schistosomiasis control and elimination.

Presenter: **Miss Sarah Davey**, *PhD Student, IBERS Aberystwyth University*

Poster 18 : The *Fasciola hepatica* histone acetylation machinery is developmentally regulated and contains druggable candidates.

Authors - SD Davey[2]; G Padalino[2]; AG Maule[1]; IW Chalmers[2]; KF Hoffmann[2];
[1] Queen's University Belfast, UK; [2] Aberystwyth University, UK

Objective - Within the global livestock production trade, *Fasciola hepatica* remains a parasite of significant detriment in terms of economics, veterinary impact and food safety. The emergence of drug resistance within multiple distinct *F. hepatica* populations has led to the urgent search for alternative chemotherapies as a means of control. Of the drug discovery strategies available to parasitologists, drug repositioning reduces pre-clinical development time by taking advantage of existing research. Among *F. hepatica* candidates to enter such an approach are the histone modifying enzymes (HMEs). HMEs are a diverse class of proteins involved in catalysing (writers), reversing (erasers) and recognising (readers) post-translational modifications on proteins (including histones). Considering the significant developmental changes that occur during the *F. hepatica* life cycle, in addition to promising HME inhibition studies in the related blood fluke, *Schistosoma mansoni*, HMEs represent highly attractive targets for further study. Here, we present the first complete bioinformatic characterisation of the histone acetylation machinery (histone acetyl transferases – HATs (writers); histone deacetylases – HDACs (erasers) and bromodomain containing proteins – BCPs (readers)) in *F. hepatica.* Briefly, BLAST searches were performed against available *F. hepatica* genomic and transcriptomic datasets using known HAT, HDAC and BCP protein sequences with an *E*-value cut-off of $1e^{-10}$. BioMart searches were also performed against a *F. hepatica* genome in WormBase ParaSite using InterPro accessions for each domain to ensure retrieval of novel sequences which may be missed by homology-based methods. Matched sequences were subsequently annotated using InterPro domain scans and were manually checked in alignments against their reference sequences to confirm the presence of key functional residues according to existing literature and UniProt records. Of the proteins putatively identified, 9 were HAT orthologues, 13 were HDAC orthologues and 40 were BCP orthologues. Among the putatively identified BCP proteins, the simultaneous presence of multiple functional domains was observed, including 5 sequences which contained both HAT and BCP domains. Domain architecture annotation indicates the conservation of key functional motifs and residues present in the human and *S. mansoni* orthologues, some of which have been confirmed by RT-PCR and sequencing. Mining of existing RNA-Seq data revealed the differential expression of these HATs, HDACs and BCPs during aspects of *F. hepatica* development. The results of these and future functional genomics/whole organism compound screening investigations will provide evidence for the importance of HMEs in *F. hepatica* lifecycle transitions and highlight repositioned compounds suitable for further development as next-generation flukicides.

Presenter: **Mrs maha aloraini**, *PHD Student, university of Glasgow*

Poster 19 : Cloning and functional complementation of *Schistosoma mansoni* cyclic nucleotide phosphodiesterases in *Trypanosoma brucei*

Authors - M aloraini[1];
[1] University of Glasgow, UK

Objective - Schistosomiasis is a neglected tropical disease caused by the parasitic flatworm genus *Schistosoma*. The disease has the highest infection rates in the developing world. Currently, there is only one drug treatment available, an anthelmintic called Praziquantel. This reliance on one single medication has raised significant concerns over drug resistance in the worms and the continued effectiveness of the treatment. However, regulatory systems including cyclic nucleotide metabolism are emerging as primary candidates for drug discovery. Here, we report the cloning, and the complementation of *S. mansoni* cyclic nucleotide phosphodiesterases (SmPDEs), specifically SmPDE7var, SmPDE1, SmPDE8, and SmPDE9C in a specialised *Trypanosoma brucei* cell line constructed as a model for heterologous expression of PDEs, created for this purpose by the deletion both alleles of the essential locus TbrPDEB1/B2. The growth of the final transfection was dependent on tetracycline-induced expression of the *Schistosoma* PDE gene (functional complementation); removal of tetracycline from the medium, as a control, was designed to result in cell death. In addition to this, a construct of TbrPDEB1 with the catalytic domain of SmPDEs 4A or SmPDE7var, successfully complemented the PDEB1/B2 *null* strain of *T. brucei*. *SmPDE11* failed to complement the *T. brucei* cell line and was assessed in a *Schizosaccharomyces pombe* system. It was found to hydrolyse cGMP over cAMP. This explains its failure to complement in *T. brucei* system, as there is no known role for cGMP in these cells, but hydrolysis of cAMP is essential.

This project has established that *S. mansoni* PDEs 1, 7var, 8 and 9C are functional cAMP phosphodiesterases and that SmPDE 11 is a cGMP phosphodiesterase. The resulting cell lines are now employed to screen several classes of PDE inhibitors to establish the pharmacological profile of each PDE separately, in a cellular system.

Presenter: **Miss Rhiannon Heslop**, *Postgraduate, University of Glasgow*

Poster 20 : Development of a single molecule fluorescent in situ hybridisation (smFISH) pipeline for the detection of host-pathogen interactions in the murine CNS

Authors - **R Heslop***[1]; **C Bentley-Abbot***[1]; P Chandrasegaran[1]; M Sinton[1]; A MacLeod[1]; JF Quintana[1];
[1] Wellcome Centre for Integrative Parasitology (WCIP), UK

Objective - Inflammation is an essential protective response against invading pathogens, but its resolution is required to limit host tissue damage. Regulatory B cells (Bregs) play a critical role in inflammation resolution by the release of anti-inflammatory cytokines IL-10, TGFβ and IL-35. However, the molecular and cellular mechanisms by which Bregs contribute to the anti-inflammatory response in the brain is understudied. *Trypanasoma brucei* invades the CNS during chronic infection, causing gliosis and neuroinflammation. Using spatial and single cell transcriptomics, we have recently identified a bidirectional crosstalk between brain dwelling Bregs and microglia mediated by the anti-inflammatory cytokine *Il10* and the survival factor *Tnfsf13b*. Here, we validated the transcriptional profiles of these cells in the circumventricular organs (CVOs) of *T. brucei* infected mice using single molecule fluorescent in situ hybridisation (smFISH). We have developed a robust analytical pipeline using Qupath to count cells positive for multiple targets with >75% accuracy. We are able to quantify single sub-cellular "transcriptional spots", representing individual RNA transcripts, and clustering behaviour in host cells in a semi-automated manner and automatically partition cells by RNA expression. Furthermore, large datasets comprising multiple tissue sections can be analysed holistically and z-stacks reconstructed. Our pipeline accurately detected an increase in *Il10* expression and in *Cd79a+/Il10+* Bregs in the choroid plexus of infected mice compared to naïve controls, validating our multi-omics findings. We also visualised, for the first time, *Gapdh+/Pyk1+* slender forms and *Pad2+/Epi1+* stumpy forms of T. *brucei* in the choroid plexus of chronically infected mice using smFISH. Our pipeline offers efficient, automated data processing for multi-dimensional, multi-target QuPath generated smFISH datasets and can therefore be readily applied to a variety of biological questions. Integration of smFISH with this pipeline presents a means to investigate the molecular interactions between Bregs and microglia, or indeed other cell populations, in chronic CNS infection.

Presenter: **Mr Aleksander Goll**, *Student, Medical University of Gdansk*

Poster 21* : Biomonitoring of haemoparasites in sylvatic rodents from the Mazury Lake District region of Poland

Authors - **A Goll**[2]; J Nowicka[1]; M Krupińska[2]; K Baranowicz[2]; M Grzybek[2];
[1] Medical University of Gdańsk, Poland; [2] Medical University of Gdansk, Poland

Objective - Rodents, members of the most abundant and diversified mammalian order Rodentia, can pose a significant threat to the health of humans, livestock, and wildlife because they are hosts for a wide range of pathogens and, in some cases, constitute important reservoir hosts for life-threatening zoonoses. Haemoparasites are vector-borne pathogens (VBP) that may affect animal and human health. The study aimed to monitor the prevalence of haemoparasites in the six abundant vole species found in the region (*Myodes glareolus, Microtus arvalis, Microtus agrestis*, and *Alexandromys oeconomus, Apodemus sylvaticus, Apodemus agrarius, Apodemus flavicolis*). Rodents (n=78) were sampled in September 2020. Trapping was carried out for five consecutive days in study sites in the forest and grasslands in NE Poland. Blood samples were collected directly from the heart by cardiac puncture using a sterile 1.5 mL syringe immediately after death from over-exposure to an anaesthetic. Blood smears were air-dried, fixed in absolute methanol, stained for 45 min in Giemsa's stain(diluted 1 : 3) in buffer at pH 7.2. Each smear was examined under oil immersion using Zeiss Axio Lab 5 microscope). We found three blood parasites within the studied population. The most prevalent species was *Mycoplasma* spp. 35.9% [25.1-48.1], followed by *Trypanosoma* spp. = 6.4% [2.3-15.3] and *Babesia microti* = 1.3% [0.1-7.9]. We believe that identifying rodent species that can serve as reservoirs of pathogens and zoonotic diseases and predicting regions, where new outbreaks are most likely to happen are crucial steps in preventing and minimizing the extent of zoonotic diseases in humans.

Presenter: **Miss Sophia DonVito**, *PhD Candidate, London School of Hygiene and Tropical Medicine*

Poster 22* : Subclinical *Plasmodium falciparum* infections are missed by routine diagnostic methods in school-age Gambian children

Authors - JP Mooney[2]; **S DonVito**[2]; M Jahateh[5]; H Bittaye[5]; M Keith[2]; LJ Galloway[4]; M Ndow[5]; AJ Cunnington[1]; U D'Alessandro[5]; C Bottomley[3]; E Riley[2];
[1] Imperial College London, UK; [2] The University of Edinburgh, UK; [3] London School of Hygiene & Tropical Medicine, UK; [4] The University of Glasgow, UK; [5] Medical Research Council Unit in The Gambia at the London School of Hygiene and Tropical Medicine, UK

Objective - Recent episodes of malaria have been associated with an increased risk of developing systemic bacterial infections (sepsis). The aetiology of this association is unclear, but malaria-related haemolysis may be one contributing factor. Furthermore, a significant proportion of *Plasmodium falciparum* infections in endemic areas are maintained subclinically, acting as a reservoir for onward transmission and often going untreated. A pilot study in Burkina Faso demonstrated ongoing haemolysis and raised plasma haem, HO-1 and IL-10 concentrations in children with persistent, subclinical P. *falciparum* infections after a 35-day follow up, compared to those without detectable parasitaemia, so a larger study was designed to further characterise the physiological consequences of persistent and recently resolved P. *falciparum* infections. From December 2017 – January 2018 (the end of the annual malaria transmission season), 1650 apparently healthy children (ages 8 – 15) living in villages throughout the Upper River Region of The Gambia were screened for P. *falciparum* infections (by 18S PCR) and/or anaemia (by haematocrit). P. *falciparum* infected children and children with moderate to severe anaemia were age-matched to healthy, uninfected, non-anaemic controls and screened again two months later (later in the dry season). During analysis, all children were re-screened for P. *falciparum* by the more-sensitive varATS qPCR and divided into four groups: 'Controls', with no detectable infection and normal haemoglobin concentrations; 'Anaemic', with no detectable

infection but low haemoglobin concentrations at the start of the study; 'Chronic', who maintained infections throughout the two-month follow up period; and 'Resolved', who were P. *falciparum* positive at first assessment but tested negative at the two-month follow up. Compared to the pilot study where 95% of children maintained infections for the 35-day study period, here, only 23% remained positive until the two-month follow up. In this cohort, chronically infected children maintained stable parasitaemias and did not differ significantly in terms of haematological or proinflammatory markers compared to the healthy, uninfected controls. However, chronically infected children demonstrated a positive correlation between parasite density and IL-10 concentrations, suggesting a tolerogenic response to persistent, subclinical infection. Surprisingly, children who naturally resolved their infections during the study period exhibited mild erythrocytosis and higher concentrations of pro-inflammatory markers compared to the other groups. Erythrocytosis amongst children with recently-resolved infections was not associated with a reduction in iron handling markers or anaemia prevalence compared to healthy controls, suggesting that iron availability is not a limiting factor in restoring red cell homeostasis following parasite clearance. These findings shed light on a 'resetting' and potential overshoot of the homeostatic hae

Presenter: **Miss Shannan Summers**, *Student, London School of Hygiene and Tropical Medicine*

Poster 23* : Investigating the genetic diversity of *Schistosoma bovis* and its hybrids across Africa

Authors - S Summers[3]; J Archer[1]; M Rabone[2]; D Rollinson[2]; F Allen[2]; T Pennance[2]; AM Emery[2]; B Webster[2];
[1] Liverpool School of Tropical Medicine, UK; [2] Natural History Museum, UK; [3] London School of Hygiene and Tropical Medicine, UK

Objective - Hybridisation between species within the *Schistosoma* genus is an emerging public health problem. The hybridisation between *Schistosoma bovis*, a veterinary parasite, and *S. haematobium*, a human parasite is of great interest where it was responsible for the re-emergence of schistosomiasis in Southern Europe. The consequences of hybridisation and subsequent introgression can impact the host range, transmission, pathology and epidemiology of schistosomiasis. This study used available *cox1* data to investigate the geographical and host structuring of *S. bovis* and *S. haematobium-bovis* hybrids across Africa. The *cox1* dataset formed a complex and diverse haplotype network for *S. bovis* but, apart from two haplotypes, all (n=41) data from samples identified as *S. haematobium-bovis* hybrids formed two distinct genetic clusters. These clusters showed little diversity with the majority being identical mitotypes from different hosts and countries. Additionally, the clusters did not form part of the *S. bovis* network, which showed geographical structuring into West, Central and East African populations. The low genetic diversity observed within the *S. haematobium-bovis* hybrids suggests that selection or genetic bottlenecking may occur during hybridisation. There was also little apparent mixing between hybrid and *S. bovis* populations, suggesting that the hybrid genetic signatures seen in human hosts may be the result of past introgression. Findings in this study demonstrate the complexity and the subsequent challenges surrounding the origin, intermediate host range and host in which hybridisation occurs. Populations from different hosts are clearly mixing and there appears to be cross over between adjacent regions which is likely supported by livestock movement. The evolution of the *Schistosoma* genus and how they adapt to anthropogenic changes will aid in understanding the factors allowing the emergence and establishment of *S. haematobium-bovis* hybrids. Further studies are required to elucidate the risk factors associated with these hybrids populations, particularly the possibility of zoonotic transmission.

Presenter: **Dr Thomas Gasan**, *Post Doctoral Research Fellow , Queens University Belfast*

Poster 24 : Improving diagnostics for *Schistosoma bovis* infections in cattle across Africa.

Authors - T Gasan[2]; J Nzalawahe[3]; JR Stothard[1]; J LaCourse[1]; H Wei[4]; G Gobert[2];
[1] Liverpool School of Tropical Medicine, UK; [2] Queens University Belfast, UK; [3] Sokoine University of Agriculture, Tanzania; [4] Fudan University, UK

Objective - The platyhelminth *Schistosoma bovis* is responsible for significant pathology and reduction in productivity in cattle across large regions of Africa. In addition to its veterinary importance, *S. bovis* has been identified as a potential zoonotic threat, through hybridisation with *S. haematobium*. Diagnostics techniques for many helminth parasites currently rely on low sensitivity, low throughput, microscopic methods. To improve on these limitations, new diagnostic tools such as lateral flow assays (LFA) are available. Molecular tests such as LFAs represent a medium throughput technology, with excellent sensitivity for the detection of active disease.

Using a framework of technologies originally developed for the detection of *Schistosoma japonicum* specific antibodiesin humans, we aim to provide enhanced detection of *S. bovis* in cattle across Africa. We have shortlisted protein targets from *S. bovis* by identifyinghomologues of *S. japonicum* proteins with the highest diagnostic potential. So far, several proteins have been identified as diagnostic targets and have been recombinantly produced in *E. coli*. These diagnostic targets are involved in various biological processes within the parasite, including metabolic pathways, transcriptional regulation, glycolysis, phosphorylation, and cell signalling. Alongside these targets are a number of potential diagnostic candidates with currently incomplete coding sequence predictions. We intend to clarify these transcript sequences using cDNA derived from *S. bovis* worms originating from natural infections and include them in the recombinant production pipeline.

We aim to screen this small library of *S. bovis* proteins with infected cattle serum, incorporating any candidates with diagnostic potential into lateral flow technologies for point-of-care detection of *S. bovis* infection.

Presenter: **Dr Alexandra Juhasz**, *PDRA, LSTM*

Poster 25 : First insights into veterinary and zoonotic schistosomiasis in Malawi

Authors - A Juhasz[1]; J Musaya[4]; M Al-Harbi[1]; S Kayuni[1]; L Cunningham[1]; J Archer[1]; P Makaula[3]; S Jones[1]; EJ Lacourse[2]; JR Stothard[1];
[1] Liverpool School of Tropical Medicine, UK; [2] Liverpool School of Tropical Medicine / UoL, UK; [3] Research in Health, Environment and Development (RHED), Malawi; [4] MLW, UK

Objective - Despite ongoing control, rural African communities typically suffer from urogenital (*Schistosoma haematobium*) and intestinal (*S. mansoni*) schistosomiasis, a common snailborne parasitic worm disease. New unknowns are now emerging about schistosome worms along Lake Malawi and the Shire River Valley environments. Abrupt changes in the genetic makeup of human schistosome worms are known whichly likely demonstrate abilities to form viable hybrids with closely related *Schistosoma* species (e.g. S. *bovis* & *S. mattheei*) suspected present in local livestock. One of the **Objective**s of our Wellcome Trust funded, 4-year multidisciplinary investigation entitled "HUGS (hybridisation in urogenital schistosomiasis)" is to reveal hybrid environmental transmission by livestock tracking and abattoir surveillance. Collected schistosome worms, and larvae thereof, will be subjected to advanced molecular typing using real-time PCR assays and DNA sequencing of key gene targets. To ascertain the local prevalence of veterinary schistosomiasis in livestock, a combination of feacal sampling, carcass inspections and snail surveys has taken place as part of an initial pilot sub-study. In November 2021, a total of 66 cattle and goats were examined within Mangochi, Blantyre, Chikhwawa and Nsanje, inclusive of 166 feacal samples using classic sedimentation and filtration methods. In this presentation, we first report on the detection of *S. mattheei* and *S. bovis* in Malawian cattle.

Presenter: **Dr Alexandra Juhasz**, *PDRA, LSTM*

Poster 26 : Fascioliasis in captive vicuñas at Knowsley Safari Park in UK

Authors - A Juhasz[1]; E Chapman[1]; L Cunningham[1]; B Johnson[3]; S Jones[1]; J Quayle[3]; J Cracknell[3]; EJ Lacourse[2]; JR Stothard[1];
[1] Liverpool School of Tropical Medicine, UK; [2] Liverpool School of Tropical Medicine / UoL, UK; [3] Knowsley Safari, UK

Objective - Infections with the liver fluke *Fasciola hepatica* can be common in South American camelids, especially in llamas and alpacas, either in natural or conservation settings. Of note, the liver fluke, and certain intermediate host snails, are thought to have been introduced to South America with autochthonous transmission ensuing. Today, whilst South American camelids can be found throughout the world, typically in managed settings, parasitological surveys of such exotic animals are infrequently reported; no more so than in those areas of the UK and Europe where fasciolosis is a common burden in farmed livestock. Given sufficient epidemiological opportunity, fascioliosis could potentially damage imported or naturally reared camelids. Knowsley Safari Park is located near Prescot, Merseyside and is a member of the British and Irish Association of Zoos and Aquariums and European Association of Zoos and Aquaria. The park has links with various conservation projects across the world and houses a range of semi-captive camelids. A particular enclosure houses 5 vicuñas, with about 5 acres of grazing pasture with a lake edge fringe and marsh. As part of a general coprological and malacological survey for snail-borne diseases within the Knowsley Safari Park ungulates, we confirmed that all vicuña were shedding ova of *F. hepatica*. Although no *Lymnaea* (*Galba*) *truncatula* was found in this enclosure, this snail was present in other park enclosures. At the time of survey, the vicuñas appeared asymptomatic but closer inspections for fasciolosis will take place as part of a revised disease management plan. The detection of fascioliasis here is of concern for such captive animals could be more vulnerable infection(s) than their wild counterparts. Vicuñas are a particularly important exotic within the park and an attraction for park visitors. In this presentation, we discuss our most up-to-date information.

Presenter: **Mr John Archer**, *PhD candidate, Liverpool School of Tropical Medicine*

Poster 27 : Hybridisation in UroGenital Schistosomiasis (HUGS): Pilot study findings upon parasitological surveys in Mangochi and Nsanje Districts, Malawi

Authors - J Archer[1]; A Juhàsz[1]; L Cunningham[1]; S Jones[1]; S Kayuni[2]; M Al-Harbi[1]; P Makaula[2]; JE LaCourse[1]; J Musaya[2]; JR Stothard[1];
[1] Liverpool School of Tropical Medicine, UK; [2] College of Medicine, University of Malawi; Malawi-Liverpool-Wellcome Trust Clinical Research Programme, UK

Objective - The HUGS (Hybridisationin UroGenital Schistosomiasis) study is a 4-year Wellcome Trust funded collaboration between the Liverpool School of Tropical Medicine and the Malawi-Liverpool-Wellcome Trust Clinical Research Centre. In November of 2021, a pilot parasitological survey was carried out in eight primary schools in Mangochi District, along the shoreline of Lake Malawi, and in Nsanje District, along the Lower Shire River. The primary purpose of these surveys was to assess the prevalence of urogenital schistosomiasis, caused by infection with *Schistosoma haematobium* and collect various samples for molecular identification of schistosomes, specifically human-infecting (e.g. *S. haematobium*)and animal-infecting (e.g. *S. mattheei* and *bovis*)worms. A collection was also made of their associated intermediate freshwater snail hosts. In addition, the prevalence of intestinal schistosomiasis, caused by infection with *S. mansoni*, was surveyed following a recent outbreak of intestinal

schistosomiasis in Mangochi District. The intention of the pilot was to help in careful selection of two communities in each district for later in-depth disease surveillance, inclusive of assessments of Giardiasis and malaria in each area. Here, we report on initial epidemiological findings from the pilot study, as well as outline planned future work with respect to additional parasitological surveys implementing molecular genotyping/diagnostics assays. Collectively, we will explore *Schistosoma* hybrids and their molecular epidemiology concurrent alongside preventive chemotherapy campaigns.

Presenter: **Miss CAROLINA HERNANDEZ**, *PhD Student , UNIVERSIDAD DEL ROSARIO*

Poster 28* : Phylogenetic relationships and evolutionary patterns of the genus Psammolestes Bergroth, 1911 (Hemiptera: Reduviidae: Triatominae): tools for vector control of Chagas disease

Authors - **CC Hernandez**[2]; M Alvarado[3]; F Salgado-Roa[4]; N Ballesteros[3]; N Rueda[4]; J Oliveira[5]; K Alevi[6]; J da Rosa[6]; P Urbano[7]; C Salazar[4]; JD Ramírez[1];
[1] Facultad de Ciencias Naturales, Universidad del Rosario, Colombia; [2] UNIVERSIDAD DEL ROSARIO, Colombia; [3] Centro de Investigaciones en Microbiología y Biotecnología-UR (CIMIBIUR), Facultad de Ciencias Naturales, Universidad del Rosario, Bogotá, Colombia., Colombia; [4] Grupo de Genética Evolutiva y Filogeografía, Departamento de Biología, Facultad de Ciencias Naturales, Universidad del Rosario, Bogotá, Colombia., UK; [5] Universidade Estadual Paulista (UNESP), Faculdade de Ciências Farmacêuticas, Araraquara, Sao Paulo 01000, Brazil, UK; [6] Universidade Estadual Paulista (UNESP), Faculdade de Ciências Farmacêuticas, Araraquara, Sao Paulo 01000, Brazil., UK; [7] Grupo de Investigaciones Biológicas de la Orinoquia, Fundación Universitaria Internacional del Trópico Americano (Unitrópico), Yopal, Colombia., UK

Objective - The kissing bugs of the subfamily Triatominae distinguish themselves among the subfamilies of Reduviidae due to their hematophagous behavior, but especially for being vectors of *Trypanosoma cruzi*, which causes Chagas disease. This subfamily comprises 156 species and all the extant species are potential vectors of *T. cruzi*. Current vector control strategies could benefit from a deep understanding of the vector's biology, ecology, and evolution. For this reason, understanding factors that shape vector species diversity and their speciation processes are essential for establishing successful control strategies. The first step to accomplish this goal involves a complete characterization of the number of lineages inside of genera, their phylogenetic relationships, and its evolutionary patterns. The evolutionary history of biodiversity in South America has been poorly studied in the seasonal dry tropical forest (SDTF). Here we studied the diversification of *Psammolestes,* a genus endemic of the SDTF and naturally infected with *T. cruzi*. We collected 92 individuals of the three *Psammolestes* species, from 12 localities in Venezuela, Colombia, and Brazil. DNA extraction was performed using the DNeasy® Blood & Tissue kit. We performed PCR and sequencing of seven loci: 6 nuclear and 1 mitochondrial locus. Phylogenetic relationships were recovered using maximum likelihood and Bayesian inference. Divergence times were determined using CYTB locus. Population genetics analyses, tests of neutrality, haplotype networks and STRUCTURE were performed. Geographical diversification was explored testing for isolation by distance, linear regression between the genetic distance and the geographical distances and Monmonier's algorithm. Number of lineages were established with two methods: Bayesian Phylogenetics and Phylogeography method and the multi-rate Poisson Tree Processes method. Niche modelling were performed using BIOMOD2.

Our multilocus analyses recovered *P. coreodes* and *P. tertius* in a monophyletic clade sister to *P. arthuri*. Species delimitation tests recovered these lineages as different species despite the shared genetic variation observed between *P. coreodes* and *P. tertius* in five genes. Also, genetic variation of the genus clustered in three groups that were consistent with the three morphospecies. Our demographic model predicted a scenario of divergence in absence of gene flow, suggesting that mixed haplotypes may be the result of shared ancestral variation since the divergence of the subtropical-temperate species *P. coreodes* and *P. tertius*. In contrast, the tropical species *P. arthuri* was highly differentiated from the other two in all tests of genetic structure, and consistently, the Monmonier's algorithm identified a clear geographical barrier that separates this species from *P. coreodes* and *P. tertius*.

We found three genetically structured lineages within *Psammolestes* that diverged in absence of gene flow in the late Miocene. This result supports a scenario of species formation driven by geographical isolation rather than by divergence in the face of gene flow associated with climatic oscillations in the Pleistocene. Also, we identified the Amazon basin as a climatic barrier that separates tropical from subtropical-temperate species, thus promoting allopatric speciation after long range dispersion. Finally, each species of *Psammolestes* occupies different climatic niches, suggesting that niche conservatism is not crucial for species differentiation. These findings influence the current vector surveillance programs of Chagas disease in the region, and it could be used for the design the new strategies for vector control of triatomines.

Presenter: **Dr Caroline Dewar**, *Senior Postdoc, Lancaster University*

Poster 29 : Mistargeting of aggregation-prone mitochondrial proteins activates a nucleus-mediated posttranscriptional quality control pathway in trypanosomes

Authors - **C Dewar**[3]; S Oeljeklaus[1]; J Mani[3]; W Mühlhäuser[1]; C von Känel[3]; T OchsenreiterB Warscheid[1]; A Schneider[3];
[1] Department of Biochemistry and Functional Proteomics, Universität Freiburg, Germany; [2] Institute of Cell Biology, University of Bern, Switzerland; [3] Department of Chemistry, Biochemistry and Pharmaceutical Sciences, Universität Bern, Switzerland

Objective - Mitochondrial quality control (MQC) is the network of pathways by which eukaryotic cells monitor and maintain the function of their mitochondria. *Trypanosoma brucei* has a large single mitochondrion, which prevents the elimination of dysfunctional mitochondria as in some other organisms. When this essential mitochondrion is not functioning correctly, for example when mitochondrial protein import is defective, cell viability suffers. The processes by which this parasite regulates its mitochondrial function are of great interest, particularly in relation to its life cycle, where the mitochondrion undergoes massive programmed morphological and functional alterations.

MQC pathways in yeast and metazoa are regulated on the transcriptional level. However, in *T. brucei*, due to polycistronic transcription, MQC regulation in this way is not possible. Additionally, other than ubiquitin and the proteasome, orthologues of most common MQC factors found in yeast and metazoa are absent in *T. brucei.* Mitochondrial biogenesis in *T. brucei* has been shown to be greatly impacted by convergent evolution, and we expect the same to be the case for mechanisms governing MQC.

95% of mitochondrial proteins in *T. brucei* are encoded in the nuclear DNA. The multisubunit ATOM complex is the mediator of protein import through the mitochondrial outer membrane in trypanosomes (Pusnik et al., 2011, Mani et al., 2015). We show data demonstrating the existence of a MQC pathway in *T. brucei* triggered when the import of aggregation-prone proteins is blocked, specifically. Using a variety of proteomic and biochemical approaches, we show that the proteasome and putative components of a ubiquitin-driven pathway are recruited to the mitochondrion upon the induction of this import defect. *Trypanosoma*tid-specific candidates were investigated as to their roles within this MQC pathway. Of particular interest is a nuclearly-localised protein with a ubiquitin-like domain which is released into the cytoplasm upon the induction of a mitochondrial import defect. Nuclear release of this protein is required for this MQC mechanism to function.

Presenter: **Mr Richard Childs Hunt**, *PhD Student, London School of hygiene and tropical medicine*

Poster 30* : What's that twinkle in your eye? Mischief or a stellate opportunist?

Authors - R Childs Hunt, C Rogers[1]; CJ Sutherland[1]; FL Henriquez-Mui[2]; R Mooney[2]; D Nolder[1];
[1] London School of Hygiene and Tropical Medicine, UK; [2] University of the West of Scotland, UK

Objective - Background: *Acanthamoeba* is a genus of free-living amoebae that opportunistically infect humans, most commonly as an infection of the cornea known as Acanthamoeba Keratitis (AK). AK is a very painful and destructive infection that, if untreated or insufficiently treated can cause total blindness in one or both of a patient's eyes. AK is difficult to treat because of the environmentally resistant cyst stage of its life cycle, which is also more tolerant to drug treatment.

Aims: To characterise the *Acanthamoebae* isolated from AK infections using currently accepted and widely used microscopy and molecular methods. Including ASA.S1-fragment 18s RDNA based genotyping and riboprinting. The hopeful outcome of this work and planned further work, including whole genome sequencing, is to assess whether the clinical position of there being no difference in treatment success between AK cases due to causative species is still correct if, (potentially) more precise sub-genus diagnostics were available.

Methods: Sub-genus level diagnosis of AK cases 2019-2021 using multiple methods,18s RDNA PCR and subsequent RFLP of the gene & Sanger sequencing of ASA.S1 fragment. As well as using the Page 1988 dichotomous key with Inverted microscopy of acanthamoeba cysts.

Presenter: **Mr Sergios Antoniou**, *PhD student, University of York*

Poster 31 : Proteomics-based investigation of substrates for the Leishmania deubiquitinase DUB2

Authors - S Antoniou[1]; AA Dowle[1]; C MacDonald[1]; AJ Wilkinson[1]; JC Mottram[1];
[1] University of York, UK

Objective - The deubiquitinating enzyme (DUB)-mediated cleavage of ubiquitin plays a critical role in balancing protein synthesis and degradation. Twenty DUBs exist in the *Leishmania mexicana* parasite, of which four, includingDUB2, are essential for the viability of *L. mexicana* promastigotes. DUB2 has a broad ubiquitin linkage specificity, and it is known to be crucial in establishing infection in mice. However, the functional role of DUB2 is not clear. Thus, we aim to identify the substrates of DUB2 through a comprehensive proteome, ubiquitinome and interactome analysis using mass-spectrometry-based quantitative proteomics, affinity-based ubiquitinated peptide enrichment and proximity dependent biotinylation. For the latter approach, 84 proximal proteins to DUB2 were identified as being significantly enriched. Gene ontology enrichment analysis categorised these proximal proteins to 17 biological processes, with protein translation being the most significant, followed by RNA binding/processing, and microtubule-associated functions, suggesting that DUB2 might have a pleiotropic function. Furthermore, initial investigation of the total ubiquitinome in *L. mexicana* using a Data Dependent Acquisition (DDA) mass-spectrometry workflow revealed that 28 of the DUB2 proximal proteins are ubiquitinated, suggesting that these might be substrates of DUB2. Currently, we are investigating whether some of these proximal proteins are interacting partners of DUB2 via co-immunoprecipitation and we are characterising the total ubiquitinome of *L. mexicana* via an improved proteomics Data Independent Acquisition (DIA) methodology.

Presenter: **Miss Yeng Yi Abbey Chan**, *Student, Aberystwyth University*

Poster 32 : Exploring the secretome of *Schistocephalus solidus*: Extracellular vesicles for host manipulation

Authors - A Chan[1]; RM Morphew[1]; J Leonard[1]; P Wititkornkul[1]; S Grinsell[1]; PM Brophy[1];
[1] Aberystwyth University, UK

Objective - The Pseudophyllidian cestode *Schistocephalus solidus* has been observed manipulating the behaviour of their secondary intermediate hosts, the three-spined stickleback (*Gasterosteus aculeatus*) to promote parasite transmission. Recent research has demonstrated that some level of manipulation occurs at the molecular level, moving away from solely the physical presence of the parasite within the body cavity of the host. The excretory secretory proteins (ESP) produced by the plerocercoid stage has been examined in relative depth and has yielded novel putative host manipulating proteins. A crucial part of the secretome, so far neglected in *S. solidus*, is the presence, or indeed absence, of extracellular vesicles (EVs). EVs have been widely demonstrated to be secreted from a number of helminths including both trematodes and cestodes. Therefore, given that the molecular mechanism of how *S. solidus* manipulates its host is unknown, there exists the potential that *S. solidus* derived EVs may act as a crucial host manipulator. Therefore, this project is expanding the known secretome of *S. solidus* through the investigation of both the free secreted proteins and EVs released from *S. solidus* into the host itself using a GeLC proteomic approach. *S. solidus* parasites were collected from natural infections and maintained in vitro. ESPs and EVs were purified using size exclusion chromatography and subjected to 1D SDS PAGE analysis followed by mass spectrometry. Preliminary evidence suggests the presence, albeit low, of EVs within the *S. solidus* secretome providing the most complete *S. solidus* secretome to date. Exploring the impact of *S. solidus* derived EVs will further complement our understanding of putative host manipulation molecules.

Presenter: **Dr Rachel Neish**, *PDRA, University of York*

Poster 33 : Role of RDK2 and its interacting protein kinases in Leishmania *mexicana* differentiation.

Authors - RP Neish[1]; V Geoghegan[1]; K Newling[1]; K Hogg[1]; J Smith[1]; J Mottram[1];
[1] University of York, UK

Objective - The protein kinase RDK2 (Repressor of differentiation 2) has been proposed to be involved in differentiation of *L. mexicana*. To investigate this, we generated an *RDK2* null mutant (Δ*rdk2*)using CRISPR-Cas9 genome editing. In *T. brucei,* RNA inference of RDK2 promotes differentiation from bloodstream stumpy form to procyclic form. In *L. mexicana in vitro*, the Δ*rdk2* cells were able to undergo normal differentiation from promastigote to amastigote. To attempt to quantitatively measure this differentiation using flow cytometry, we generated a mNeonGreen-dynein line where the newly formed flagellum can be a measured fluorescently. We found Δ*rdk2* was able to transform from amastigote to promastigote in vitro, albeit at a slower rate compared to the control, during the initial phase of differentiation.

The Δ*rdk2* cell line was screened within a protein kinase knockout library using BarSeq to identify genes with a loss of fitness in differentiation from lesion-derived amastigotes to promastigotes. Like the *in vitro* assays, Δ*rdk2* had a delayed differentiation from amastigote to promastigote. We generated an N-terminal MYC tagged RDK2 and carried out immunoprecipitation and mass spectrometry to identify potential interacting proteins. From this we identified other protein kinases that may be involved in the same pathway as RDK2. From these screens, we are attempting to unravel the role of RDK2 and its interactome in *L. mexicana* differentiation.

Presenter: **Dr Romina Nievas**, *Research Associate, University of York*

Poster 34 : Quiescence in Leishmania *mexicana*

Authors - R Nievas[1]; K Newling[1]; J Mottram[1];
[1] University of York, UK

Objective - Microbes commonly employ cellular quiescence to survive environmental stresses such as starvation, immune surveillance, or chemotherapeutic interventions. Moreover, dormancy often underlies chronic infections that complicate the clinical management of infected patients. A few studies have identified quiescent and semi-quiescent population of amastigotes in leishmania-infected mice, characterized by low rates of transcription and protein turnover. However, since access to persistent amastigotes is a major logistical bottleneck, further mechanistic analysis on these cells are limited. Leishmania promastigotes also enter a non-replicating but viable state in culture when starved of purines that have hallmarks of persister-like cells. These cells can survive without the provision of purines for more than 3 months, during which, growth arrest is reversible by the addition of exogenous purine. The study of these growth arrested leishmania forms provides a model to unravel general mechanisms underpinning transition between quiescent and proliferative states and may also provide answers to the frequent drug treatment failures in the absence of genetically selected resistant parasites. We are using the adenine-starvation model and multi-omics approaches to identify markers for persistent parasites and to elucidate the protein kinase-dependent signaling mechanisms that regulate entry and exit from quiescence in Leishmania.

Presenter: **Miss Monique Johnson**, *PhD Student, University of Cambridge*

Poster 35 : Investigating DNA damage responses in Apicomplexan parasites

Authors - MK Johnson[2]; S Chelaghma[1]; L Koreny[1]; RF Waller[1]; CJ Merrick[2];
[1] University of Cambridge, Department of Biochemistry, UK; [2] University of Cambridge, Department of Pathology, UK

Objective - Across the Apicomplexan phylum, DNA damage response pathway(s) are not well characterised. Many anti-parasitic drugs rely on causing DNA damage or interrupting DNA replication as their mode of action, so it would be very useful to learn more about how these mechanisms work. In model eukaryotes, cell cycles are closely regulated and DNA damage is flagged for repair at a series of cell-cycle checkpoints. These checkpoints are signalled through phosphatidylinositol 3-kinase-related kinases (PIKKs). Interestingly, some apicomplexans have retained homologs of human PIKKs, such as ATM and ATR, whereas others have not. In *Toxoplasma gondii (T. gondii)*, there are putative homologs of ATM and ATR, whereas in close relatives such as *Plasmodium falciparum (P. falciparum)* these are missing and the closest remaining homolog to this family is the lipid kinase phosphoinositide 3-kinase (PI3K).

To investigate this, we created an inducible knockdown of the putative *ATM* gene in *T.gondii*. The knockdown was found to be non-lethal so the protein is probably not essential, which was unexpected because a recent genome-wide knockdown screen reported a severe fitness defect for this gene. By contrast, our knockdown grew normally. DNA damaging agents (e.g. the alkylating agent MMS) were added to see if the knockdown made cells more vulnerable to damage and whether this would affect growth, but knockdown parasites were not apparently sensitised to DNA damage. Nevertheless, knockdown parasites did fail to produce a marker of DNA damage, the phosphorylation of histone 2A, in response to the topoisomerase inhibitor camptothecin – a double-strand-break-inducing agent. Furthermore, when low levels of DNA damage were applied, *T. gondii* replication was slowed and the ATM knockdown reduced this effect, possibly by preventing initiation of a checkpoint. Overall, results thus far suggest that the putative *T. gondii* ATM does play a role in the DNA damage response and checkpoint signalling, but is non-essential.

Previously published work using an inhibitor of the human ATM, KU-55933 (usually used in cancer studies) showed that this inhibited histone phosphorylation after DNA damage in *T. gondii*. The **Authors** therefore concluded that this inhibitor acts on the *T. gondii* ATM homolog. However, our results call into question the specificity of this inhibitor: although it did prevent histone phosphorylation in response to damage, it did not phenocopy the genetic knockdown, and caused severe growth defects in parasites in the absence of DNA damage. Off-target cytotoxic effects are possible, as is an effect on the other PIKK homolog (putative ATR) in this parasite.

Future work will focus on the role(s) of *T. gondii* ATR versus ATM, and on cross-complementation studies to establish how *Plasmodium* responds to DNA damage in the apparent absence of either of these kinases.

Presenter: **Dr Rita Wassef** , *Lecturer of Parasitology, Helwan University*

Poster 36 : Evaluation of Mono and Combined Nitrofurantoin Therapy for Toxoplasmosis in Swiss Albino Mice

Authors - A Elkholy[2]; R Wassef [1];
[1] Helwan University , Egypt; [2] Banha University, Egypt

Objective - Toxoplasmosis is a commonly frequented disease with an estimated prevalence of more than one billion human cases worldwide and over one million new infections each year. It is classified as the second most common cause of deaths and the fourth leading cause of hospitalizations attributed to foodborne diseases. The disease may pass unnoticed in healthy individuals but could be fatal in the immunocompromised. Available anti-*Toxoplasma* drugs are associated with many side effects. Therefore, search for new more reliable, more efficient, and less toxic therapeutic agents is a continuous endeavor. This study assesses the potential use of nitrofurantoin, a compound with well-established antimicrobial properties, as a potential anti- *Toxoplasma* drug in vivo. It compares its efficacy to the commonly used anti-*Toxoplasma* agent spiramycin by molecular and histopathological methods in acute and chronic infection. The results demonstrate a significant ability to eliminate the parasite ($P<0.001$) whether used as mono- or combined therapy with spiramycin in the acute and chronic stages. When compared to the anti- *Toxoplasma* drug spiramycin, nitrofurantoin achieved similar efficacy in the acute and chronic infection ($P =0.65$ and $P =0.096$, respectively). However, better results were obtained when using a combination of both drugs ($P<0.001$). Additionally, nitrofurantoin showed good inhibitory effects on the inflammatory process in the liver, kidney, and uterus of the experimentally infected animals. Therefore, nitrofurantoin can be considered as a potential anti-*Toxoplasma* agent. Nevertheless, further studies are recommended before consideration for clinical trials.

Presenter: **Mr Felipe Miguel Nery Lunkes** , *Biologist , Instituto Rene Rachou/ Fiocruz Minas*

Poster 37* : *Schistosoma mansoni* phenotypic evaluation after aspartyl proteases cathepsin D-like knockdown

Authors - F Lunkes[3]; SG Gava[2]; NC Tavares[2]; MR Senger[1]; FP Silva-Júnior[1]; MM Mourão[2];
[1] Instituto Oswaldo Cruz, Rio de Janeiro, Brazil; [2] Instituto René Rachou - Fiocruz Minas, Brazil; [3] Instituto Rene Rachou/ Fiocruz Minas, Brazil

Objective - Praziquantel is the only commercially available drug for the treatment of schistosomiasis, although its mechanism of action remains unknown. Studies show the need to search for new targets, including different classes of peptidases that play an important role in the development of the parasite and the success and maintenance of the infection. An aspartyl protease (AP) similar to cathepsin D from *Schistosoma mansoni* (SmCD1) has been reported to be involved in the initial breakdown of hemoglobin in the host's erythrocytes, with

distinct hemoglobin cleavage points and 51% identity with the human ortholog. Subsequently, other SmAPs were identified (SmCD2 and SmCD3). This work aims to functionally characterize SmCD1 and SmCD2 and to validate these targets as potential therapeutic targets. For this, the RNA interference technique was used, with exposure of schistosomula and adult worms to specific SmCD-dsRNAs. First, by qPCR, it was observed that the targets are more expressed in female adult worms. Significant reductions in target transcripts were observed in schistosomula (~99.9%) on the fifth day of dsRNA exposure. Phenotypic changes were observed in female adult worms recovered by perfusion, with reduced body length and decreased formation of hemozoin pigment in the digestive tract. Confocal microscopy analyzes showed a reduction in the ovary area, absence of eggs in the reproductive tract, and a decrease in mature oocytes, possibly related to sexual immaturity and indicating possible participation of the targets in the worm development. However, no phenotypic changes were observed in males recovered and analyzed so far. Analyzes of adult worms' motility, exposed to dsRNA, showed a decrease in motility of male adult worms on the fifth day for both targets. The evidence presented here suggests that SmCD1 and SmCD2 are promising targets for inhibitor screening in search of a new therapy against schistosomiasis.

Keyword: *Schistosoma mansoni*; Aspartyl proteases; RNAi;

Presenter: **Mr Felipe Lunkes**, *Master's student, Fundação Oswaldo Cruz*

Poster 38* : *Schistosoma mansoni* phenotypic evaluation after aspartyl proteases cathepsin D-like knockdown

Authors - F Lunkes[3]; SG Gava[2]; NC Tavares[2]; MR Senger[1]; FP Silva-Júnior[1];
[1] Instituto Oswaldo Cruz, Rio de Janeiro, Brazil; [2] Instituto René Rachou - Fiocruz Minas, Brazil; [3] Fundação Oswaldo Cruz, Brazil

Objective - Praziquantel is the only commercially available drug for the treatment of schistosomiasis, although its mechanism of action remains unknown. Studies show the need to search for new targets, including different classes of peptidases that play an important role in the development of the parasite and the success and maintenance of the infection. An aspartyl protease (AP) similar to cathepsin D from *Schistosoma mansoni* (SmCD1) has been reported to be involved in the initial breakdown of hemoglobin in the host's erythrocytes, with distinct hemoglobin cleavage points and 51% identity with the human ortholog. Subsequently, other SmAPs were identified (SmCD2 and SmCD3). This work aims to functionally characterize SmCD1 and SmCD2 and to validate these targets as potential therapeutic targets. For this, the RNA interference technique was used, with exposure of schistosomula and adult worms to specific SmCD-dsRNAs. First, by qPCR, it was observed that the targets are more expressed in female adult worms. Significant reductions in target transcripts were observed in schistosomula (~99.9%) on the fifth day of dsRNA exposure. Phenotypic changes were observed in female adult worms recovered by perfusion, with reduced body length and decreased formation of hemozoin pigment in the digestive tract. Confocal microscopy analyzes showed a reduction in the ovary area, absence of eggs in the reproductive tract, and a decrease in mature oocytes, possibly related to sexual immaturity and indicating possible participation of the targets in the worm development. However, no phenotypic changes were observed in males recovered and analyzed so far. Analyzes of adult worms' motility, exposed to dsRNA, showed a decrease in motility of male adult worms on the fifth day for both targets. The evidence presented here suggests that SmCD1 and SmCD2 are promising targets for inhibitor screening in search of a new therapy against schistosomiasis.

Presenter: **Dr Aitor Casas-Sanchez**, *Postdoctoral Research Associate, Liverpool School of Tropical Medicine*

Poster 39 : The MISP family of surface glycoproteins from *Trypanosoma brucei* is co-expressed with VSG and BARP in the metacyclic trypomastigote stage, adopts a triple helical bundle structure, and is not essential for the colonization of the tsetse salivary glands

Authors - A Casas-Sanchez[1]; S Perally[1]; R Ramaswamy[4]; LR Haines[1]; C Rose[1]; C Yunta-Yanes[1]; M Aguilera-Flores[5]; L Smithson[3]; S Vaughan[3]; M Lehane[1]; IC Almeida[5]; J Van Den Abbeele[2]; M Boulanger[4]; A Acosta-Serrano[1];
[1] Liverpool School of Tropical Medicine, UK; [2] Institute of Tropical Medicine, Antwerp, Belgium; [3] Department of Biological and Medical Sciences, Oxford Brookes University, Oxford, UK; [4] University of Victoria, Canada; [5] Department of Biological Sciences, The University of Texas at El Paso, United States

Objective - *Trypanosoma brucei* spp. develop into mammalian-infectious metacyclic trypomastigotes inside the tsetse salivary glands. Besides acquiring a variant surface glycoprotein (VSG) coat, little is known about the expression of invariant surface antigens by the metacyclic stage. Proteomics analyses of saliva from *T. brucei*-infected flies identified, in addition to VSG and *Brucei* Alanine-Rich Protein (BARP) peptides, a family of GPI-anchored surface proteins herein named Metacyclic Invariant Surface Proteins (MISP). The MISP family is encoded by five paralog genes with >80% protein identity, which are exclusively expressed by salivary gland stages of the parasite, and peaks in metacyclic stage as shown by confocal microscopy and immuno-high resolution scanning electron microscopy. Crystallographic analysis of MISP and a high confidence model of BARP reveal a triple helical bundle architecture commonly found in other trypanosome surface proteins. Molecular modelling combined with live fluorescent microscopy suggests that MISP expose immunogenic N-terminal epitopes above the VSG coat, although vaccination with a recombinant MISP isoform did not protect mice against a *T. brucei* infectious bite. Lastly, both using RNAi and CRISPR-Cas9-driven knock out of all MISP paralogues suggests they are not essential for parasite development in the tsetse vector.

Presenter: **Dr Marzuq Ungogo**, *PhD student, University of Glasgow*

Poster 40* : Revisiting quinapyramine: mechanism of uptake, action and resistance

Authors - MA Ungogo[1]; L Lemgruber Soares[1]; HP De Koning[1];
[1] Institute of Infection, Immunity and Inflammation, College of Veterinary, Medical and Life Sciences, University of Glasgow, United Kingdom, UK

Objective - Quinapyramine has been used in the treatment of veterinary trypanosomiases since 1950's. The use of quinapyramine to treat African Animal Trypanosomiasis was stopped in sub-Saharan Africa due to cross-resistance to diminazene, homidium and isometamidium. However, the drug is still used in the treatment of *T. evansi*, *T. vivax* and *T. equiperdum* infections outside the tsetse belt of Africa. Nevertheless, the mechanism of action and resistance to the drug is largely unclear. We induced quinapyramine resistance in *T. equiperdum* and *T. evansi* bloodstream forms through adaptation to increasing concentration of the drug added to growth medium. *In vitro* development of resistance to quinapyramine was slow in both *T. equiperdum* and *T. evansi,* with parasites resistant to 50× EC_{50} generated in approximately 280 days.Low-level resistant clones, generated after 80 days, did not show cross-resistance to commonly used trypanocides. However, high-level resistant clones, obtained after further adaptation, showed cross resistance to diminazene, pentamidine, ethidium and isometamidium. To investigate the cross-resistance patterns observed, we tested quinapyramine against a panel of drug resistant *T. brucei* and *T. congolense* cell lines generated previously. We observed a higher EC_{50} for quinapyramine in isometamidium-resistant *T. brucei* and diminazene-resistant *T. congolense* strains with low mitochondrial membrane potential. Using confocal microscopy, we have observed colocalization of quinapyramine with a mitochondrial stain (Mitotracker). In addition, the EC_{50} of quinapyramine was 2-fold higher in the P2 adenosine transporter knock-out strain (TbAT1-KO) compared to wild type. Further investigation revealed that quinapyramine inhibits P2-type [^{3}H]-adenosine uptake through the P2 transporter in a dose-dependent manner with *Ki* = 12. 6 ± 0.7 (n=3). Likewise, an adenosine concentration of 10 µM or higher inhibited the fluorescence-monitored uptake of quinapyramine. The growth of trypanosomes was strongly inhibited in the presence of 5× and 10× EC_{50} quinapyramine. However, *Trypanosoma*l killing was slow, taking 96 – 120 h, compared to diminazene which kills the parasites within 24 – 48 h. In addition, parasite growth recovered when the drug was removed, as exposure for 8 h, 24 h or 48 h did not lead to the death of all the parasites. Cell cycle analysis showed that the majority of cells in cultures treated with 2× and 5× EC_{50} quinapyramine contained multiple nuclei and/or kinetoplasts from 48 h. Treated cells imaged by fluorescence microscopy and electron microscopy appeared disfigured, lost their slender shape, and contained multiple flagella. Thus, quinapyramine is trypanostatic, has minimal effects on G, S and M phase of cell cycle but inhibits t

Presenter: **Dr Alexander Cook**, *PDRA, University of Oxford*

Poster 41 : Invariant surface glycoprotein 65 of *Trypanosoma brucei* is a complement C3 receptor important for virulence

Authors - A Cook[1]; O Macleod[2]; M Crow[2]; H Webb[2]; R Burns[2]; M Redpath[2]; S Seisenberger[2]; C Trevor[2]; L Peacock[4]; A Schwede[2]; N Kimblin[2]; AF Francisco[6]; J Pepperl[2]; S Rust[5]; P Voorheis[3]; W Gibson[4]; MC Taylor[6]; M Higgins[1]; M Carrington[2];
[1] University of Oxford, Department of Biochemistry, UK; [2] University of Cambridge, Department of Biochemistry, UK; [3] Trinity College Dublin, Ireland; [4] School of Biological Sciences & Bristol Veterinary School, University of Bristol, UK; [5] AstraZeneca, UK; [6] Department of Pathogen Molecular Biology, London School of Hygiene and Tropical Medicine, UK

Objective - African Trypanosomes replicate in the blood of mammalian hosts yet are completely exposed to the adaptive and innate immune systems. Despite this, Trypanosomes can sustain long-term infections - how can this parasite survive constant host immune-surveillance? Trypanosomes evade the adaptive immune response through antigenic variation of a surface coat consisting of a dense layer of variable surface glycoprotein. However, very little is known about how they negate the innate system, including the blood circulating complement system. We have discovered that an invariant surface glycoprotein, ISG65, is a receptor for Complement Component 3 (C3). We show how ISG65 binds to the thioester domain of C3b. We also show that knockout of the ISG65 locus greatly decreases the pathogenicity of trypanosomes in a mouse model. Deposition of C3b on pathogen surfaces is a central point in activation of the complement system and C3b has been observed on trypanosome surfaces. Our findings therefore suggest that trypanosomes have a C3 receptor distributed across their surfaces that greatly decreases their susceptibility to complement-mediated killing.

Presenter: **Miss Caroline Ricce Espada**, *Postdoctoral researcher, University of Sao Paulo*

Poster 42* : The potential role of long non-coding RNAs in key steps of Leishmania (Viannia) braziliensis life cycle

Authors - CR Espada[1]; JC Quilles[1]; RM Magalhães[1]; TP Defina[1]; AK Cruz[1];
[1] University of São Paulo, Brazil

Objective - *Leishmania (Viannia) braziliensis* is an important causative agent of cutaneous and mucocutaneous leishmaniasis in Americas. During its life cycle, this parasite colonizes two different hosts facing dramatic changes in physiological conditions which requires a fast and dynamic gene expression modulation in order to survive. The genetic organization of *Leishmania*, where unrelated genes are transcribed by RNA Polymerase II as polycistronic units in the absence of canonical promoters, suggests that the regulation of gene expression in these organisms results mainly from other processes rather than transcription itself. Non-protein coding RNAs (ncRNAs) have been identified in different *Trypanosoma*tids parasites, including *L. braziliensis*. In the latter, the transcriptome of the main morphologies across the *Leishmania* lifecycle(procyclic promastigotes, metacyclic promastigotes and axenic amastigotes) were compared and revealed the presence of 11,372 putative ncRNAs of which at least 295

were differentially expressed in all three stages. Using CRISPR/Cas9, we are investigating the functional role of these elements in *L. braziliensis*. Up to date, 14 ncRNAs (including short and long ncRNAS) were successfully knocked out from *L. braziliensis* M2903 genome by our group and phenotypically screened for phenotypic alterations. Herein, we present the results obtained for three potential long ncRNAs (lncRNAs) each of them enrolled in different phenotypes of *L. braziliensis.* The fitness of each knocked out line was compared to the parental wild-type line (WT) in experiments mimicking important points of *Leishmania* life cycle such as parasite multiplication, survival to oxidative and nutritional stresses, metacyclogenesis and infectivity. We found that deletion of lncRNA66 led to a significant reduction in parasite growth as promastigotes whereas the deletion of lncRNA31 increased the doubling time of axenic amastigotes from 8.7 hours (WT) to 11.6 hours. Deletion of lncRNA52 resulted in a lower percentage of metacyclic parasites recovered after Ficoll enrichment, suggesting that this lncRNA may be enrolled in metacyclogenesis in *L. braziliensis*. Characterization of these transcripts by northern blotting, determination of ncRNA extremities and posttranscriptional processing (presence or absence of CAP and PolyA tail) by RNA circularization coupled with sequencing, and identification of possible ligands using a S1M tag-mediated pull down are currently ongoing to better understand the biogenesis and role of these putative regulatory ncRNAs in the parasite. Our results will help to understand the regulation of *Leishmania* gene expression and may result in the discovery of ncRNAs enrolled in parasite fitness and pathways essentials for parasite survival.

Presenter: **Dr James Hewitson**, *Lecturer, University of York*

Poster 43 : Chronic schistosome infection remodels bone marrow haematopoiesis

Authors - J Hewitson[1];
[1] University of York, UK

Objective - Joanna Greenman, Grace Boyd, Jack Fisher, Moses Egesa, Alison Elliott, David Kent, James Hewitson People living in schistosome-endemic regions undergo repeated cycles of reinfection and drug clearance. To determine how chronic *Schistosoma mansoni* infection modifies responses to reinfection, we compared skin immune responses to S. *mansoni* challenge in mice that were chronically infected (12wks) with animals that were exposed for the first time. We found chronic infected leads to weak skin immune responses after rechallenge with marked reductions in macrophages, and that this is maintained after curative praziquantel treatment. As bone marrow (BM) chimera experiments revealed skin macrophages originate from infiltrating monocytes, we performed RNAseq on BM monocytes and found worm infection strongly impacts on gene expression in this site. We hypothesised distal worm infection impacts on bone marrow haematopoiesis by modifying the BM cytokine microenvironment. Cytokine Bead Array analysis showed chronic infection leads to elevated IL-4 (but not other type 2 cytokines) in BM aspirates. We next tested whether IL-4 can alter haematopoiesis and found haematopoietic stem cells (HSC) express Il4ra and in vitro assays (single cell cultures, colony formation assays) revealed HSC directly respond to this cytokine. Surprisingly then, the dominant signature of infection on immune progenitors (BM LSK cells) in vivo was instead interferon-related (type I and II). Using competitive bone marrow transplants, we found HSC from infected mice to be functionally impaired. We are now assessing the extent to which infection-induced changes to haematopoiesis persist after parasite clearance, and how this impacts on heterologous immune challenges in mice and humans.

Presenter: **Miss Jessica Dagley**, *Research assistant, Liverpool School of Tropical Medicine*

Poster 44 : A dirofilariasis mouse model for heartworm preclinical drug testing

Authors - AE Marriott[1]; JL Dagley[1]; S Hegde[1]; A Steven[1]; MJ Taylor[1]; JD Turner[1];
[1] Liverpool School of Tropical Medicine, UK

Objective - The filarial nematode, *Dirofilaria immitis,* is the cause of veterinary heartworm disease; a potentially life-threatening infectious disease of companion animals, and the cause of zoonotic pathologies in humans. Heartworm preventative and curative drug R&D has been reinvigorated by the identification of macrocyclic lactone drug-resistant isolates and incidence of chemoprophylactic treatment failures in pets. With no small animal model available, cats and dogs are utilised in primary *in vivo* drug screening against *D. immitis*. We therefore assessed lymphopenic immunodeficient mice with ablation of the interleukin-2/7 common gamma chain (gc) as susceptible *D. immitis* hosts with utility to test heartworm preventative drug efficacy. Non-obese diabetic (NOD) Severe Combined ImmunoDeficient (SCID)gc-/-(NSG) mice produced consistent yields of viable *D. immitis* larvae at two weeks post-infection across multiple experiments and different batches of infectious larvae inoculates. Developing larvae were found in subcutaneous tissue, the natural site of this stage of the heartworm life cycle in dogs. Larvae retrieved from NSG mice were morphologically mid-L4 stage of development. Compared with age-matched *in vitro* propagated larvae, *in vivo* derived L4 were significantly larger and contained expanded intracellular *Wolbachia* titres, determined by QPCR of the single copy *Wolbachia* surface protein gene and Fluorescent *in situ* Hybridisation (FISH) of *Wolbachia* 16S ribosomal RNA. We validated the NSG mouse model and an *ex vivo* L4 screening system against the reference macrocyclic lactone, moxidectin and the anti-*Wolbachia* reference drug, doxycycline. We have subsequently adopted the mouse model screen to assess efficacy of novel, fast-acting azaquinazoline anti-*Wolbachia* compounds. We contend that future adoption of the mouse model of heartworm will benefit end-user laboratories conducting R&D of novel heartworm preventatives and provide reduction and refinement in long-term procedures requiring cats or dogs.

Presenter: **Dr El-Sayed El-Alfy**, *lecturer, Mansoura University*

Poster 45 : Global Epidemiology of *Taenia Multice*ps: A Comparative Meta-Analysis Study

Authors - **E El-Alfy**[2]; I Abbas[2]; S Saleh[1];
[1] Mansoura University, Egypt; [2] Faculty of Veterinary Medicine, Mansoura University, Egypt

Objective - *Taenia multiceps* infections are very common in canids and small ruminants all over the world, with human zoonotic instances reported. Despite the fact that a vast amount of data on *T. multiceps* infections from various geographical regions has been published, no retrospective study has been conducted. The present paper provides the first meta-analysis investigation for the published data on *T. multiceps* infections in definitive and intermediate hosts worldwide, aiming at better understanding of the global epidemiology of this ubiquitous taeniid.A total of 207 studies on prevalence, diagnosis, molecular and treatment of *T. multiceps* were retrieved (PubMed and Scopus, Science Direct and Google Scholar) of which, 154 articles were used for meta-analysis (prevalence studies) and analyzed using the software Open Meta[Analyst]. The pooled prevalence in various definitive and intermediate hosts as well as the heterogeneity between studies (I^2 test) were estimated based on a 95% confidence interval and employing the random effect model. Different subgroup analyses were conducted to detect the variabilities in coenuri locations as well as age-wise and gender-wise prevalence. The pooled global prevalence of *T. multiceps* infection in dogs was 5.8 % (95% CI, 4.7 – 6.9%). Surprisingly, grey wolves had the statistically (*p*-value = 0.0049) highest *T. multiceps* prevalence (21.6%, 15.7 – 27.5%) among all definitive hosts. From slaughterhouse surveys; the pooled global prevalence of cerebral coenuri of sheep was 8.4 % (95% CI, 7.0 – 9.9%) and in goats was 6.1 % (95% CI, 4.0 – 8.1%). Out of 1147 sheep that displayed neurological symptoms, cerebral coenuri were detected in 437 animals resulting in a high prevalence rate (52.9%, 27.4 – 78.4%), which indicates that coenurosis is a major contributor in the neurological disease of sheep. Contrastingly, the non-cerebral coenuri in sheep was very low prevalent globally (0.0 - 0.1%) and in goats was 0.3 % (95% CI, 0.2 – 0.4%). Understanding the epidemiology of *T. multiceps* should aid in the development and implementation of prevention and control methods for public health, veterinary care practitioners, and animal owners.

Presenter: **Mr Thomas M. Arme**, *Research Assistant, University of Glasgow*

Poster 46 : Molecular malacology and xenomonitoring schistosomiasis: Implication of *Bulinus africanus* as an intermediate host of *Schistosoma haematobium* in Lake Malawi.

Authors - **TM Arme**[1]; MH Al-Harbi[1]; L Cunningham[1]; JR Stothard[1];
[1] Liverpool School of Tropical Medicine, UK

Objective - The life-cycle of *Schistosoma haematobium,* a trematode parasite, involves both humans and *Bulinus* freshwater snail species. Recent reports have identified *Bulinus* spp. novel to Lake Malawi, but their role in *Schistosoma* epidemiology is unknown. Due to the emergence of hybrids of *S. haematobium* and *Schistosoma bovis* south of Lake Malawi, this study sought to investigate the intermediate hosts of these *Schistosoma* spp., to identify parasite transmission foci, and determine potential areas for their hybridization. Infection screening was conducted on 106 snails collected in 2017 from the shoreline of Lake Malawi, Magochi Disrict, using both conventional and quantitative PCR xenomonitoring methods. Snails were selected for species identification (n=10) by inspecting a 644bp fragment of the *cox1*, which was later aligned to entries on GenBank. The distribution of *Bulinus* spp. and *Schistosoma* spp. was then mapped onto Mangochi district. Four snails were matched to sequences of *Bulinus africanus* and another identified as a *Bulinus angolensis*-like specimen. Although no snails were infected with *S. bovis*, the qPCR cycle threshold values indicated that individuals from both snail species were developing pre-patent infections with *S. haematobium* across the shoreline, including some Mangochi tourist beaches. This study builds on recent surveys implicating the newly reported *B. africanus* and *B. angolensis*-like snails in the transmission of *S. haematobium* in Lake Malawi for the first time. There is a risk for introduction of *S. bovis* and subsequent hybridisation with the endemic *S. haematobium*, as *B. africanus* is a competent host of both parasites. The finding of snails infected with *S. haematobium* on tourist beaches poses a risk for its translocation to non-endemic areas. Xenomonitoring will continue in Malawi, utilising novel *Schistosoma* species-specific TaqMan probes to ascertain the species identities of pre-patent infections in snails.

Presenter: **Mrs Dalal Ardan**, *PhD researcher , University of East Anglia*

Poster 47* : First observation of Parasitic viruses in Trichomonas gallinae

Authors - **D Ardan**[1];
[1] University of East Anglia, UK

Objective - **First observation of Parasitic viruses in *Trichomonas gallinae***

[1]Dalal Ardan, [2]Marlene Benchimol, [3]Sally Warring, [3]Neil Hall, [1] Diana Bell, [1]Kevin M. Tyler

[1]University of East Anglia, Norwich, UK [2]Laboratório de Ultraestrutura Celular Hertha Meyer, Instituto de Biofísica Carlos Chagas Filho, UFRJ, Rio de Janeiro Brasil. [3]Earlham Institute, Norwich, UK.

Trichomonas gallinae is a single cell protozoan parasite that causes avian trichomonosis in a diverse array of birds especially pigeons and doves. Numerous studies show that viruses can reside within protozoan pathogens and contribute towards pathogen virulence and the closely related *Trichomonas vaginalis,* can be infected with a double-stranded RNA (dsRNA) virus which enhances its pathogenicity. However, the presence of *T. gallinae* has remained hitherto undiscovered. We screened a cryobank containing hundreds of UK passerine columbid and raptor isolates of *T. gallinae* with a wide range of genotypes for the presence or RNA virus. An initial Agarose gel-based screen of extracted RNA from different isolates of *T. gallinae* revealed an extra band of RNA in two isolates (C3 and C10). This band of RNA is consistent with the size of viral dsRNA and indicative of viral infection in *T. gallinae*. The presence of dsRNA was further verified using immune fluorescence monoclonal antibodies J2 specific to dsRNA viruses. Both these isolates were from infections which lacked demonstrable pathology and which were considered to be avirulent strains. To characterize the effect of the virus on *Trichomonas gallinae* we compared these strains with two virulent strains strains which lacked virus namely (A1 and C4). We observed that virus infected cells of *T. gallinae* were smaller and grew less well than non-infecting cells. Moreover, using (scanning and transmission) electron microscopic methods, we found evidence of plasma membrane disruption and granular structures which may be virus budding from the cell surface. Using negative staining of supernatants, we found icosahedral structures which may be virions. Using RNA transcriptomics, we were able to show expression of viral RNAs with 70% RNA identity to Trichomonas Virus 1. Overall our study offers new insight into parasitic pathogenesis of*T. gallinae* which in contrast to *Trichomonas vaginalis* correlates with low virulence of strains. It is to be hoped that knowledge of the virus may provide a route to novel intervention strategies for avian trichomonosis in birds.

Presenter: **Miss Caoimhe Herron**, *PhD Student, Queen's University Belfast*

Poster 48* : Developmental Regulation and Functional Prediction of microRNAs in an Expanded *Fasciola hepatica* miRNome

Authors - C Herron[1]; E Robb[1]; E McCammick[1]; C Hill[1]; NJ Marks[1]; AG Maule[1]; P McVeigh[1];
[1] Queen's University Belfast, UK

Objective - *Fasciola hepatica* is a parasite of human and veterinary importance. In humans, fascioliasis case estimates range from 1.6 to 2 million people infected with a further 90 million at risk. Closer to home *F. hepatica* costs the UK ruminant industry over £20 million annually through yield losses and liver condemnation. Triclabendazole is the only drug effective against both the juvenile and the adult stage of the parasite; resistance to this and the few other available drugs is a major issue for fluke control. Micro (mi)RNA were first discovered in the early 90s as non-coding RNAs responsible for post transcriptional regulation of gene expression. Recent years have seen reports that miRNAs are secreted by parasites into their host environment and can manipulate expression of host genes. Our goal is to better understand the intracellular and host interacting functions of *F. hepatica* miRNAs. To this end, we have used small RNA sequencing to expand the *F. hepatica* miRNome to 150 mature miRNAs, many of which appear unique to *Fasciola*. Our study is the first to measure transcriptional changes in these miRNAs through fluke development. We show, for the first time that certain miRNAs are life stage dependent suggesting a function specific to that stage with the highest number of miRNAs found in metacercariae. To examine host-interacting functions of secreted miRNAs, we examined interactions between extracellular vesicle (EV) miRNAs and cow/sheep transcripts through *in silico* analysis. Reactome pathway analysis of these mRNA targets revealed that the majority of those secreted miRNA were involved in general gene expression pathways and signal transduction and as such suggests the involvement of these within fluke induced immunosuppression or cellular pathology. This work will form the basis of future functional genomic work to confirm these *in silico* predictions. Research carried out here has vastly expanded on the knowledge base of host parasite interactions within *Fasciola hepatica* and will go a long way to potentially discovering novel control and drug targets that are desperately needed

Presenter: **Mr Manuel Saldivia** , *Mr., Novartis*

Poster 49 : In cauda venenum, CLK1 inhibitors for Trypanosomiasis

Authors - M Saldivia[1]; J Jiricek[1]; S Rao[1];
[1] Novartis Institute for Tropical Diseases, United States

Objective - The current therapeutic challenges for kinetoplastid neglected tropical diseases (NTDs) ranges from toxicity, poor efficacy, difficult administration, or prohibitive cost. From a phenotypic screening, we recently identified AB1, an Amidobenzimidazole compound active across various kinetoplastids parasites. Detailed target deconvolution studies using *Trypanosoma brucei* revealed CLK1 as AB1 primary target. Further optimization of AB1 through medicinal chemistry efforts resulted in the AB3, a novel compound with improved potency against not only *T. brucei* but also *Trypanosoma cruzi*. AB3 compound showed promising inhibitory activity against the *Trypanosoma* CLK1 enzyme with improved selectivity against host kinases. Incubation of AB3 compound with parasite resulted in cell cycle progression defects. AB3 had favorable pharmacokinetic properties, which led to an evaluation in mice models of sleeping sickness and Chagas disease. Treatment with AB3 resulted in the complete cure of mice in the hemolymphatic model of sleeping sickness at significantly lower doses than AB1. AB3 also showed partial cure in the mice model of Chagas disease after three rounds of immunosuppression. Together with in vitro CLK1 inhibition and the mitosis defects, our studies indicate that *Trypanosoma* cidality after treatment could result from inhibition of CLK1 kinase activity and triggers a new treatment venue for these diseases.

Presenter: **Miss Georgina Hurle**, *Masters Student, University of East Anglia*

Poster 50* : Counting Cryptosporidium: Development of Simple in vitro Drug and Culture Platform

Authors - GR Hurle[1]; D Steverding[1]; KM Tyler[1];
[1] University of East Anglia, UK

Objective - INTRODUCTION: Recently there has been the development of several culture platforms for the propagation of *Cryptosporidium*. The simplest of these being through the infection of mammalian COLO-680N monolayers.

AIMS: We aimed to optimise and adapt this culture system for the purpose of assessing the efficacy and selectivity of therapeutic agents against *Cryptosporidium*.

METHODS: To do this we established 5mL flask culture systems and monitored the number of motile parasites released from COLO-680N cells. From this, we have subsequently reduced the culture volume to increase parasite growth throughput. Recently we have evaluated the potential use of the cytochemical Alamar Blue to detect *Cryptosporidium* growth using this platform.

RESULTS: Initial results show evaluating the number of motile merezoites and meronts in flask and plate cultures is an effective measure for gauging selective toxicity of compounds towards *Cryptosporidium*. Alamar blue was not effective for discriminating merezoite growth, but shows potential in host cell protection from infection.

DISCUSSION: Straightforward culture of *Cryptosporidium* has permitted the development of merezoite and meront inhibitor assays, which are likely to lead to the identification of novel therapeutic agents in the near future.

Presenter: **Miss Boontarikaan Wititkornkul**, *PhD Student, IBERS, Aberystwyth University*

Poster 51* : **Control of equine tapeworms through praziquantel: The hidden impact on the equine microbiome**

Authors - B Wititkornkul[1]; SE Girdwood[1]; HJ Worgan[1]; RM Morphew[1]; RE Wonfor[1];
[1] IBERS, Aberystwyth University, UK

Objective - Horses are hindgut fermenters and microbes within the hindgut play important roles in enhancing the fermentation processes providing nutrients and energy to the host, as well as maintaining intestinal homeostasis. However, dysbiosis of the equine hindgut microbiota is common and can be influenced by a variety of factors, often associated with metabolic diseases and disorders. The equine tapeworm, *Anoplocephala perfoliata*, infects horses worldwide and are found primarily attached to the caecal wall, adjacent to the ileocaecal valve. Both *A. perfoliata* and praziquantel (PZQ), a commonly used anthelmintic for *A. perfoliata* treatment and control, represent a substantial threat to the fragile equine microbiome. Recent molecular work generating the first *A. perfoliata* transcriptome and characterising the proteome of the secretome has supported our understanding of the host-parasite interaction. However, the interaction between the infection and the exposure of PZQ on the equine gut microbiome has so far been neglected. The current work therefore aimed to initially determine the effect of PZQ on hindgut fermentation kinetics using an *in vitro* hindgut fermentation model. We investigated the gas production over a 72 hours fermentation period as well as, pH, volatile fatty acids (VFAs), ammonia and lactate concentrations and the metabolome after 24 hours incubation following exposure to PZQ at various dosage levels. Our findings demonstrate that at 24 hours post incubation, a high dose of PZQ leads to a trend in decrease of the production of total VFAs and specifically acetate levels (p<0.1). In addition, at 72 hours post incubation, the high dose of PZQ significantly increased the ammonia concentration (p<0.05). However, there was no effect on the metabolome fingerprint. Following this first investigation into the impact of PZQ on the equine hindgut microbiome fermentation, we suggest that PZQ treatment may alter the fermentation pathways in the equine hindgut, which could impact on the nutritional functioning of the caecum. To further characterise PZQ impact, collected microbial samples are undergoing both meta-proteomic and meta-taxonomy analysis for a complete assessment of PZQ induced alterations.
KEYWORDS:*Anoplocephala perfoliata,* Praziquantel, Faecal Microbiome, *In Vitro* Hindgut Fermentation, Interaction

Presenter: **Dr Samuel Duncan**, *Post doctoral researcher, University of Dundee*

Poster 52 : The curious origin and evolution of kinetoplastid glycosyltransferases

Authors - SM Duncan[2]; MA Ferguson[1];
[1] Wellcome Trust Centre for Anti-Infectives Research, School of Life Sciences, University of Dundee, UK; [2] The University of Dundee, Wellcome Trust Centre for Anti-Infectives Research, UK

Objective - The synthesis of glycoconjugates is a particularly common feature of protozoan parasites to facilitate interactions with, and provide protection from, their host environment. Many tenets of *N*- or GPI- anchor glycosylation shared amongst eukaryotes are highly conserved in kinetoplastids, but these parasites exhibit peculiarities in the process of glycosylation, including the evolution of a large GT67 glycosyltransferase (GT) family. This presentation will summarise our current knowledge into the process of glycosylation in these organisms, and discuss the evolutionary divergence of the GT67 family and their contribution to complex *N*-glycan and GPI-sidechain glycan branch synthesis. Phylogenetic analysis of the kinetoplastid specific GT67 gene family reveals their separation into two distinct clades for *Leishmania* and African trypanosomes, indicative of the different selective pressure exerted on their carbohydrate biosynthesis. Further, investigations of the GT67 family in *T. brucei* indicate the significant expansion and repurposing of this ancestral, eukaryotic β3-glycosyltransferase into seven distinct *TbGT* lineages, presumably to facilitate complex glycan biosynthesis. Despite their evolution from a shared ancestor, the enzymes encoded by these seven GT67 gene lineages display comparatively low amino acid sequence identity. Experimental characterisation of their glycosyltransferase activity reveals they unexpectedly catalyse β1-2, β1-3 and β1-6 glycosidic linkages, and utilise UDP-Galactose and UDP-*N*-acetylglucosamine as donor sugars. We

will also highlight the discovery of an entirely novel and essential process of mitochondrial fucosylation in kinetoplastea and present recent investigations in *T. brucei* to assess the role of the GT11 family fucosyltransferase, TbFUT1. We will discuss the curious origin of TbFUT1, where recent phylogenetic analysis implicates the acquisition of the GT11 family gene from bacteria by horizontal transfer from a large, cytoplasmic DNA virus early in the evolution of the kinetoplastea. We propose the further acquisition of a bacterial GT25 family glycosyltransferase gene which, unlike GT67 family sequences, is highly conserved amongst kinetoplastids. Subsequent gene duplication has occurred in the kinetoplastids to encode two GT25 family enzymes, termed GTX and GTZ, with mitochondrial localisation but likely different glycosyltransferase activities. We aim to further investigate the function of mitochondrial glycosylation by identifying the substrate(s) of TbFT and characterising the activity of these bacterial-like, mitochondrial GTs.

Presenter: **Dr Jose Mengel**, *Senior Researcher, Oswaldo Cruz Foundation*

Poster 53 : Chronic *Trypanosoma cruzi* infection in mice lacking B cells (muMT)

Authors - F Cardillo[1]; J Nihei[2]; **J Mengel**[3];
[1] Gonçalo Moniz Research Center, Fiocruz-Bahia, Brazil; [2] Gonçalo Moniz Research Center, Fiocruz, Bahia, Brazil; [3] Oswaldo Cruz Institute, Fiocruz, Rio de Janeiro, Brazil

Objective - Introduction: B cells are crucial players during the acute *T. cruzi* infection. However, very little is known about their role in the chronic phase of the disease. This study analyses the consequences of the B cell absence in C57BL/6 mice infected with low numbers of the Tulahuen *T. cruzi* strain. **Material and Methods:** C57BL/6 or C57BL/6 muMT mice were infected with the Tulahuen *T. cruzi* strain (50 blood trypomastigotes, *i.p.*). Mice that survived the acute challenge were used between 4 and 8 months after infection. The following experiments were carried out: 1- Spleen and mononuclear heart and striated muscle cells were studied by flow cytometry. 2- Cytokines were measured by ELISA in supernatants from spleen cells after *in vitro* cultures. Histopathological studies of the heart and muscle were performed. **Results:** Despite very high parasitemia and high mortality, muMT mice controlled the number of parasites in circulation. When infected with low parasite numbers, about 30% of the mice survived and progressed to the chronic phase of the disease. Four to eight months after the initial challenge, mice started to die due to infection re-aggravation. MuMT mice that succumbed to the chronic infection were all lymphopenic, and this picture was never observed in B-cell sufficient mice. Spleen cells from muMT chronically infected mice produced equivalent amounts of IL-4 and IL-10 than wt mice but lower amounts of IL-2, IL-12, and INF-g. However, the production of IL-18 was much higher than wt mice. Histopathological studies showed a more intense inflammatory infiltrate in the striate muscle and heart with higher numbers of CD4+ and CD8+ T cells infiltrating these tissues. Also, their activation state is different from wt mice as lower percentages of T cells expressing $CD45Rb^{low}$ were found in these tissues. **Conclusion:** The results presented in the present study demonstrate that B cells are crucial to maintaining T cell numbers and functions during the chronic phase of *T. cruzi* infection, regulating tissue inflammation.

Presenter: **Miss Sara Chelaghma**, *Research Assistant , Univeristy of Cambridge*

Poster 54 : Apical annuli are essential exocytic sites in *Toxoplasma*

Authors - S Chelaghma[1]; H Ke[1]; K Barylyuk[1]; L Koreny[1]; RF Waller[1];
[1] University of Cambridge, UK

Objective - Secretion of factors that mediate host cell invasion and manipulation are key to the success of apicomplexans. However, we still do not know where or how some of these factors are secreted, for example, those from dense granules. *Toxoplasma* 'apical annuli' are small ring structures in the inner membrane complex (IMC) at the lower boundary of the apical cap cisterna. The function and significance of these enigmatic structures have been unknown. Furthermore, the previously identified annuli proteins are associated with the IMC, so it was uncertain if this structure has any interaction with or relevance to the plasma membrane and extracellular environment. We used hyperLOPIT spatial proteomics and proximity labelling to search for plasma membrane-associated proteins at the apical annuli and found four with this conspicuous location. One is a polytopic membrane protein containing LMBR1 domains but of unknown function, and three are Q-SNAREs implicated in vesicle docking. Depletion of each of these annuli proteins results in strong lytic cycle growth phenotypes. Specifically, these mutations manifest as defects in intracellular replication and the delivery of secreted proteins, including GPI-anchored proteins. Our data show that the apical annuli are essential structures to *Toxoplasma*, that their composition and activity span the plasma membrane, and that they represent additional exocytic sites to the apical complex in tachyzoites. We speculate that the complexity and persistence of the IMC in *Toxoplasma* has driven the need for apical annuli as further specialised secretion sites in the cell periphery.

Presenter: **Mr RICARDO OBONAGA GOMEZ**, *Postdoctoral researcher , Butantan Institute*

Poster 55 : Function of the MRE11 and EXO1 nucleases and RECQ2 helicase in DNA end resection and double-strand breaks repair in *Trypanosoma brucei*

Authors - R Obonaga[1]; MC Elias[1];
[1] Butantan Institute, Brazil

Objective - DNA double strand breaks (DSBs) are one of the most toxic forms of DNA damage. These can arise accidentally during normal cell metabolism or after exposure of cells to DNA-damaging agents. Failure to repair them can result in genomic instability, a characteristic of cancer cells. In eukaryotes, DSBs are repaired by homologous recombination (HR) and non-homologous end-joining (NHEJ). In HR and microhomology-mediated end joining (MMEJ) (a type of NHEJ Ku heterodimer-independent), the 5´ DNA strands of the DSBs are nucleolytically degraded through a process termed DNA end resection. This process, critical for repair pathway choice by HR or MMEJ and checkpoint signaling, is driven by the MRE11, DNA2 and EXO1 nucleases and generate 3'-ended single-stranded DNA tails with different lengths of homology sequences. In most eukaryotes, repair by HR and MMEJ is conserved, including *Trypanosoma brucei*, the parasite responsible for African human trypanosomiasis, a fatal disease if left untreated. In this parasite, DNA end resection is not well understood (or is not completely understood). So, in this context, the present research project intends to characterize the functions of the major nucleases in DNA resection and repair of DSBs. Therefore, the *T. brucei* MRE11 and EXO1 nucleases and RECQ2 helicase were tagged with different epitopes using a CRISPR/Cas9 alternative editing system without selection marker, and then one or two genes were silenced in different configurations using RNA interference. Instead of DNA2, the RECQ2 gene was silenced, because we did not find a reliable candidate for the DNA2 protein using sequence alignment tools. Besides that, the choice of studying RECQ2 was made because it works together with DNA2 to resect the DNA end. With this approach, we will measure parameters such as nuclear localization, DNA-bound and proteins levels. Locally, the DNA end resection will be measured using qPCR from a single DSB generated by the activity of the *I-SceI* restriction enzyme fused to destabilization domain (DD), which will be controlled in space and time by stabilization with Shield and globally, by induction of multiple DSBs generated by the treatment with ionizing radiation using the single molecule analysis of resected tracks. Additionally, the functions of these nucleases in repair pathway choice (HR or MMEJ) will be evaluated using a reporter system that allows reconstitution of green or red fluorescent protein genes depending on the DNA repair pathway used. Furthermore, we will determine whether there are differences in the participation of these nucleases in the DNA end resection and repair of DSBs cell cycle dependent. With these results, we hope to contribute with the expansion of our understanding on how the first steps of DNA DSBs repair occur in *T. brucei*.

Presenter: **Mrs CHRISTINE Moore**, *PhD student, University of Ghana, Legon*

Poster 56 : In vitro effects and mode of action of phenolic compounds on leishmania *donovani*

Authors - C Moore[1]; CC Amisigo[1];
[1] University of Ghana, Legon, Ghana

Objective - Background: Leishmaniasis is a disease caused by the protozoan parasite, Leishmania. The disease remains a global threat to public health requiring effective chemotherapy for control and treatment. In this study, the effect of some selected phenolic compounds on Leishmania *donovani* was investigated. The compounds were screened for their anti-leishmanial activities against promastigote and intracellular amastigote forms of Leishmania *donovani*.

Methodology/principal findings: The dose dependent effect and cytotoxicity of the compounds were determined by the MTT assay. Flow cytometry was used to determine the effect of the compounds on the cell cycle. Parasite morphological analysis was done by microscopy and growth kinetic studies were conducted by culturing cells and counting at 24 hours intervals over 120 hours. The cellular levels of iron in promastigotes treated with compounds was determined by atomic absorption spectroscopy and the effect of compounds on the expression of iron dependent enzymes was investigated using RT-qPCR. The IC50 of the compounds ranged from 16.34 µM to 198 µM compared to amphotericin B and deferoxamine controls. Rosmarinic acid and apigenin were the most effective against the promastigote and the intracellular amastigote forms. Selectivity indexes (SI) of rosmarinic acid and apigenin were 15.03 and 10.45 respectively for promastigotes while the SI of 12.70 and 5.21 respectively was obtained for intracellular amastigotes. Morphologically, 70% of rosmarinic acid treated promastigotes showed rounded morphology similar to the deferoxamine control. About 30% of cells treated with apigenin showed distorted cell membrane. Rosmarinic acid and apigenin induced cell arrest in the G0/G1 phase in promastigotes. Elevated intracellular iron levels were observed in promastigotes when parasites were treated with rosmarinic acid and this correlated with the level of expression of iron dependent genes.

Conclusions/significance: The data suggests that rosmarinic acid exerts its anti-leishmanial effect via iron chelation resulting in variable morphological changes and cell cycle arrest.

Presenter: **Miss Marianne Aelmans**, *MRes Student , Lancaster University*

Poster 57* : Characterising Heat Shock in *Trypanosoma congolense*

Authors - M Aelmans[1]; C Dewar[1]; M Urbaniak[1];
[1] Division of Biomedical and Life Sciences, Faculty of Health and Medicine, Lancaster University, UK

Objective - *Trypanosoma congolense* causes significant economic burden across Sub-Saharan Africa, as it is the etiological agent of Animal African Trypanosomiasis (AAT), a wasting disease affecting cattle which currently has no pharmaceutical treatment. *T. congolense* is a close relative of *Trypanosoma brucei*, they co-infect the same hosts so have been exposed to similar evolutionary selective pressures, and it is expected they will show similarities in host interactions. While *T. brucei* is a well-studied model organism, very little experimental work has been performed using *T. congolense* as tools have only recently been developed for genetic manipulation, but now is the time to use them to investigate the survival and infection mechanisms of the parasite. One of the major symptoms of AAT is a high fever which *T. congolense* responds to by eliciting the heat shock (HS) response, an important virulence factor which allows the parasite to survive in the host. The aim of this project is to characterise the *T. congolense* HS response, as understanding the mechanisms involved could pave the way for discovering novel drug targets in this parasite.

A bioinformatic analysis looking into the conservation of proteins, phosphorylation sites and active sites involved in the HS response between *T. brucei* and *T. congolense* has been conducted. Key proteins involved in the HS response are being tagged and analysed with immunofluorescence microscopy to see if they localise in the same way in *T. brucei* and *T. congolense* during HS. Flow cytometry is being used to characterise cell cycle progression through HS. RNAi knockdown of ZC3H11 will be performed to see its effect on the cells ability to survive HS. Preliminary results of this work will be shown.

Presenter: **Mrs Aline Araujo Alves**, *Postdoc, Institut Pasteur*

Poster 58 : Investigating the role of unique kinesin-2 motors in intraflagellar transport in trypanosomes

Authors - A A Alves[2]; P Bastin[1];
[1] Institut Pasteur, Paris, France; [2] Institut Pasteur, France

Objective - Trypanosomes have a single flagellum essential for survival, motility, life cycle and host-parasite interaction. These parasites are an excellent model to study flagella assembly, as they build a new flagellum while conserving the old one. Flagella assembly requires a specific transport system called intraflagellar transport (IFT). IFT is the bidirectional movement of multiprotein complexes, or IFT trains trafficking along flagellar microtubules. The transport towards the ciliary tip is called anterograde, driven by kinesin-2. These motors comprise two subfamilies: the heterotrimeric kinesin-2, with two different motor subunits and a non-motor subunit, and the homodimeric kinesin-2, with two identical motor subunits. *Trypanosoma brucei* is unique since it lacks the heterotrimeric version but contains two kinesin-2 genes – KIN2A and KIN2B. KIN2A and KIN2B involvement on anterograde IFT transport was shown by RNAi knockdown of the two motors together. Still, how each motor contributes individually to the anterograde IFT remains unclear. To investigate KIN2A and KIN2B roles, we generated cell lines expressing the kinesins fused to the fluorescent protein mNeonGreen (mNG) and followed IFT by confocal spinning-disk microscopy. Compared to the IFT protein IFT81, KIN2A-containing particles move at a similar speed but have a lower frequency, suggesting KIN2A is not present in every anterograde train. KIN2B-containing particles present a unique pattern: only a minor part of them can reach the flagellar tip. Most KIN2B particles are restricted to the proximal region of the flagellum and are slower than IFT anterograde trains. Double-tagged cells expressing KIN2B and the IFT protein IFT140 confirmed that KIN2B proximal particles do not colocalise with the IFT140, showing they are not associated with IFT trains. In contrast, the distal KIN2B-containing particles have the expected IFT speed and colocalise with IFT140 but show a low frequency. The frequencies of KIN2A and KIN2B distal particles together reach the expected frequency of anterograde IFT trains. Together, our data suggest KIN2A and KIN2B carry different trains and function as homodimers.

Presenter: **Ms Juliana Nunes Rosón**, *PhD student, Butantan Institute*

Poster 59* : Histones variants and their interaction with the chromatin in *Trypanosoma cruzi* life forms

Authors - J Rosón[1]; MO Vitarelli[2]; H Costa-Silva[1]; DS Pires[1]; T Rodrigues[1]; B Cordeiro[1]; A Kraus[3]; S Calderano[1]; N Siegel[3]; MC Elias[1]; JP Cunha[2];
[1] INSTITUTO BUTANTAN, CELL CYCLE SPECIAL LAB, SÃO PAULO, Brazil; [2] Butantan Institute, Brazil; [3] Division of Experimental Parasitology, Faculty of Veterinary Medicine, Ludwig-Maximilians-Universita¨ t in Munich, Munich, Germany, Brazil

Objective - Introduction: Histones variant deposition and histone posttranslational modifications (PTMs) can affect chromatin structure and gene expression. For *Trypanosoma brucei*, it is known that the histone variant H2B.V dimerizes with the H2A.Z and generates unstable nucleosomes in divergent switch strand sites. When nucleosomes harboring these variants associate with protein factors (e.g. bromodomains) and histone acetylation, a scenario of transcription activation is suggested. H3.V and H4.V are other examples of histone variants that affect nuclear architecture. In *T. brucei*, they are frequently positioned at transcription termination regions, often surrounded by J bases. Parasites lacking the H3.V/H4.V have shown profound changes in chromatin structure. In *T. cruzi,* H2B.V was found enriched in infective mammalian tissue culture trypomastigotes (TCT) compared to epimastigote forms by using quantitative proteomics. To date, H4.V was not identified in *T. cruzi* genome database. **Objectives**: First, to identify H4.V sequence candidates and generate parasites expressing H4.V and H2B.V tagged by CRISPR Cas9. Second, to compare histone variants (H4.V and H2B.V) associated with chromatin in distinct *T. cruzi* life forms, through gradient salt extraction. Third, to evaluate H2B.V genome location comparing epimastigotes with TCT forms, by ChIP-seq. **Results:** To retrieve a putative H4.V in *T. cruzi* (CL Brener Esmeraldo-like) we have performed BLAST and phylogenetic analysis by using *T. brucei* H4.V sequence. A strong candidate for H4.V was revealed (TcCLB.511681.20), homologous to *T. brucei* and *Leishmania*. To further evaluate histone variant genome location and chromatin interaction, lineages expressing a tagged version of H2B.V and H4.V were obtained by CRISPR-Cas9 system. By comparing the association of the canonical histone H3 and the variants H2B.V and H4.V to the epimastigote chromatin by salt extraction, we observed that variants associated weakly compared to histone H3. Also, H2B.V was weakly bonded to the TCTs' chromatin. Moreover, the genomic location of H2B.V was performed by ChIP-seq assay. Our results showed H2B.V peak enrichment in dSSRs, tDNAs, and regions between conserved (mainly protein-coding genes) and disrupted (non-synthetic regions, mainly virulence factors) genome compartments. H2B.V peaks were more evident in epimastigotes than TCT forms in agreement with a weak chromatin association in the latter. **Discussion/Conclusion**: This ongoing research highlights the differential histone variants association with the chromatin and contributes unveil their function among the parasite life forms. The integration of methods explored in this work reinforces the role of the H2B.V and H4.V in the chromatin structure and suggests differences among the parasite life forms.

Presenter: **Ms Adriana Diaz**, *PhD Student, The Royal Veterinary College*

Poster 60* : Evaluating a novel test-and-treat control strategy for livestock schistosomiasis in sub-Saharan Africa

Authors - **AV Diaz**[1]; M Walker[1]; JP Webster[1];
[1] Royal Veterinary College, University of London, UK

Objective - Schistosomiasis is a widespread neglected tropical disease (NTD) found predominantly in sub-Saharan Africa, as well as in Asia, the Caribbean and South America. Species of the causative blood fluke can infect humans and other mammalian definitive hosts, furthermore, reports of hybrid parasites between human-specific and animal-specific schistosomes are increasing. Existing control programs rely on mass drug administration of praziquantel in affected human populations and at present, no formalized control measures targeting African animal reservoirs have been implemented. Nevertheless, within sub-Saharan Africa, emerging reports of use and misuse of praziquantel in livestock plus recognition of the role of bovines as maintenance hosts for schistosome species that spill over and hybridize within humans warrant attention. Thus, we evaluate the potential effectiveness of a test-and-treat strategy to control bovine schistosomiasis using a transmission modelling approach. We show that the simulated test-and-treat strategy can be highly effective in supressing infection by means of an imperfect rapid point-of-care diagnostic applied to a small number of animals per herd. The proposed targeted chemotherapy One Health control strategy could not only improve livestock productivity and subsistence livelihoods but also limit zoonotic transmission and the emergence of hybrids. As acknowledged in the recently launched WHO guidelines, veterinary public health interventions will be a necessary step towards achieving the NTD Roadmap goal of eliminating schistosomiasis as a public health problem by 2030.

Presenter: **Dr Thomas Crellen**, *Research Fellow, University of Glasgow*

Poster 61 : Relationship between the worm burden and egg output in liver fluke infections of humans

Authors - **T Crellen**[1]; P Sithithaworn[5]; M Haswell[6]; PH Lamberton[1]; SE Spencer[3]; TD Hollingsworth[4];
[1] University of Glasgow, UK; [2] University of Glasgow , UK; [3] University of Warwick, UK; [4] University of Oxford, Big Data Institute, UK; [5] Khon Kaen University, Thailand; [6] University of Sydney, Australia

Objective - A general problem in the epidemiology of human-infective parasitic helminths is the relationship between observed measures of infection, such as egg counts or antigen concentrations, and the worm burden within hosts. Here we quantify the relationship between the worm burden and observed egg counts for the carcinogenic liver fluke (*Opisthorchis viverrini*) through a re-analysis of worm expulsion and autopsy studies. Using a Bayesian inference framework, we estimate the density dependent relationship between worms and eggs. The results from our best fitting model show that a single worm expels around 27 eggs per gram of stool, and 100 worms expel around 2500 eggs per gram of stool. In addition we obtain estimates of key epidemiological parameters, such as the mean worm burden, the negative binomial dispersion of worms, and the proportion of worms recovered by expulsion. These results will allow an individual's worm burden to be estimated from routinely collected survey data and so provide an insight into the effect of public health interventions on mean worm burden and parasite transmission.

Presenter: **Mr Jacob Thompson**, *E/S vaccination , University of Manchester*

Poster 62 : The impact of chronic whipworm infection on vaccine mediated immunity

Authors - **J Thompson**[1]; J Derrick[1]; KJ Else[1];
[1] Lydia Becker Institute of Immunology & Inflammation, UK

Objective - *Trichuriasis* is a disease that affects ~465 million people worldwide resulting from infection by the intestinal dwelling parasitic nematode *T. trichiura*, colloquially known as the human whipworm. Whipworm infections can cause colitis, growth retardation, and *Trichuris* dysentery syndrome (TDS). The current treatment strategies for Trichuriasis and other soil transmitted helminths promoted by the World Health Organisation (WHO) involve mass drug administration (MDA) campaigns. However, the effectiveness of these MDA campaigns, particularly against *T. trichiura,* is poor. Low drug efficacy has fuelled the search for alternative treatments for Trichuriasis including the development of anti-*T. trichiura* vaccines which have the potential to provide long lasting immunity. However, these novel vaccines face many challenges in the field that must be addressed if they are to be considered as viable treatment options for Trichuriasis. Specifically, the infection status of the target population is a concern, with an emerging body of literature detailing the ability of some parasitic infections to dampen vaccine-mediated immune responses. Here we investigated the influence of pre-existing *Trichuris* infections on the effectiveness of vaccine-mediated immunity provided by anti-*Trichuris* vaccines via the *Trichuris* mouse model, *T. muris*. We employ a crude vaccine composed of excretory/secretory (E/S) products in alum. This vaccine is known to provide immunity to naïve C57BL/6 mice against subsequent *T. muris* infections. We demonstrate that a low dose infection of *T. muris,* administered to C57BL/6 mice prior to E/S vaccination, inhibits the protective immunity provided by E/S vaccination when delivered prior to a low dose infection. As the vast majority of individuals living in areas where *T. trichiura* is endemic harbour low-level chronic *T.*

trichiura infections by a very early age, this research provides insight into some of the challenges that anti-*T. trichiura* vaccines will face in the field.

Presenter: **Miss Marketa Novotna**, *PhD student , University of Dundee*

Poster 63* : Investigating the roles of divergent histone tails using gene editing in *Trypanosoma brucei*

Authors - **M Novotna**[1]; D Horn[1];
[1] University of Dundee, UK

Objective - *Trypanosoma*tid genomes lack sequences readily identifiable as promoters and replication origins and it remains unclear to what extent transcription, DNA replication and DNA repair rely upon chromatin-based controls. The *N*-terminal histone tails, and tail modifications, such as acetylation, play key roles in these processes in other eukaryotes. Genetic manipulation and subsequent study of histone functions have proven particularly challenging, however, because core histone genes are typically present as many copies of each gene. In *Trypanosoma*tids, these genes are present in long polycistronic transcription units containing approx. 40 copies. The *N*-terminal tails are also highly divergent relative to the usual model eukaryotes. I have used an inducible CRISPR/Cas9 system in *Trypanosoma brucei* to delete all native gene copies of histone H4, complementing the defect with a single, recoded and highly expressed ectopic copy. CRISPR/Cas9 was then used for site saturation mutagenesis of an *N*-terminal tail lysine residue in the ectopic gene (H4K14 initially). Current results suggest that amplicon-seq can be used to monitor the relative fitness of these mutants. I am also using these strains to determine whether the *N*-terminal tail of histone H4 can be removed altogether, which was surprisingly possible in budding yeast.

Presenter: **Mr Dionysis Grigoriadis**, *WormBase ParaSite , WormBase*

Poster 64 : WormBase ParaSite - 2022 update

Authors - **D Grigoriadis**[3]; F Rodgers[1]; T Le[3]; M Zarowiecki[3]; KL Howe[3]; M Berriman[2];
[1] Wellcome Trust Sanger Institute, UK; [2] University of Glasgow, UK, UK; [3] European Molecular Biology Laboratory, European Bioinformatics Institute (EMBL-EBI), UK

Objective - Since its creation in 2014, WormBase ParaSite (http://parasite.wormbase.org) has been serving the helminthology community by gathering, organising and presenting helminth genomes on a large scale. Aiming to interrogate data for nearly all nematode and platyhelminth genomes, we are now presenting data for 202 genomes from 163 species including all major human-infective helminths as well as many plant and animal infective species of major agricultural importance. The resource also includes free-living relatives of the parasitic helminths, providing critical insights into their evolutionary histories, host-adaptation, and invasion. WormBase ParaSite takes publicly available genomes and annotations from the European Nucleotide Archive (ENA), and uses scalable automated methods to add further value to the data by way of systematic and consistent functional annotation (e.g. repetitive elements, non-coding genes annotation, protein domains, Gene Ontology terms), and comparative analysis (e.g. orthologues and paralogues). The data is presented in a user-friendly format providing several ways to explore it including genome and gene summary pages, text search, sequence search, a query wizard, bulk downloads functionality, a choice of genome browsers, an Application Programming Interface (API) and a high-throughput expression analysis suite.

In our latest release, WBPS16 - launched in September 2021, we have developed a system for capturing and presenting published gene-phenotype associations for well-studied genomes and propagating these associations between orthologs to all our hosted species. A researcher is therefore able to view phenotypes observed for their gene of interest and/or its orthologues for every species in WormBase ParaSite. These associations have been curated from the literature over many years, from RNAi and variant data. Until today, the majority of these datasets have been for Caenorhabditis elegans. However, we have already included data from a *Schistosoma mansoni* study and we anticipate that more gene-phenotype association studies of this type for helminths in the future. Further recent updates also include improved assembly/annotation quality metrics reported on our genome pages, while we have also introduced an archiving service so older WormBase ParaSite releases will remain available for browsing. Driven by community demand and the availability of new datasets we supplement our in-house curation platform by hosting Web Apollo instances for an increasing number of genomes.

As ever, WormBase ParaSite looks to further improve by adapting to the rapidly evolving field of worm omics. We, therefore, encourage the community to describe your use cases and make suggestions for improvements to help us prioritise future work.

Presenter: **Mr Peter McCann**, *PhD Student, Queen's Univeristy Belfast*

Poster 65 : *Galba truncatula* and Helminths, the Importance of Microbes

Authors - **P McCann**[4]; C McFarland[3]; J Megaw[3]; C Cantacessi[2]; G Rinaldi[1]; G Gobert[3];
[1] Wellcome Trust Sanger Institute, UK; [2] Department of Veterinary Medicine, University of Cambridge, UK; [3] Queens University Belfast, UK; [4] Queen's Univeristy Belfast, UK

Objective - Liver fluke (*Fasciola hepatica*) and rumen fluke (*Calicophoron daubneyi*) are endemic in the UK. Liver fluke is estimated to cost the UK agriculture industry approximately £300 million per year, particularly due to lamb deaths and liver condemnations. Rumen fluke is fatal in severe infections and only one flukicide, oxyclozanide, has been shown to effectively reduce rumen fluke burdens. The desirable potency of triclabendazole has stimulated its overuse for liver fluke control resulting in widespread anthelmintic resistance. Therefore, there is an urgent need to develop new control strategies for fasciolosis.

The microbiome is defined as the combined genetic material of the microorganisms inhabiting a particular environment. A host's microbiome is known to play a key role in many aspects of health and disease, including susceptibility to parasitic infection. While most microbiome studies have focused on the mammalian hosts of helminths, their intermediate hosts should also be considered. Recent research of mosquitoes infected with *Wolbachia* shows they cannot transmit dengue fever. As a result, efforts to control dengue fever are being focused on bacterial symbionts that can aid disease elimination. The interaction between the snail microbiome and life stages of parasitic trematodes residing in their intermediate hosts has not been investigated to any large extent.

This project is in its earliest phase. We aim to profile the microbiome of snail species harbouring active helminth infections. We will compare host stress markers, and investigate the role played by bacterial symbionts. Finally, the functional roles played by snail microbiota will be considered using classical microbiological methods.

Presenter: **Miss Abigail Webb**, *PhD Student, Aberystwyth University*

Poster 66 : Sequence conservation of Cryptosporidium invasion proteins which have potential as vaccine candidates against cryptosporidiosis in cattle and sheep

Authors - AL Webb[1]; J Pachebat[1]; RM Morphew[1]; J Hamilton[1];
[1] Aberystwyth University, UK

Objective - Bioinformatic analysis of four *Cryptosporidium* invasion proteins to develop understanding of whether these proteins could be potential vaccine candidates against cryptosporidiosis in cattle and sheep

Cryptosporidium causes the gastroenteric disease, cryptosporidiosis, which is a leading health, welfare and economic concern within the livestock sector, with the major aetiological agent being the pathogenic species, *C. parvum*. Despite this, treatment options are limited and there is currently not a vaccine available for either animals or humans. Specific proteins, considered to be involved in the attachment and invasion of this parasite to host cells, have since been investigated as potential vaccine candidates, but knowledge remains limited. A detailed understanding of the sequence variation and structures of these proteins allows a basis for the investigation of their potential as vaccine candidates. Bioinformatic analysis was performed on four *C. parvum* invasion proteins, Cpa135, CP2, CP15 and P23, comparing gene and amino acid sequence conservation both between *C. parvum* isolates and isolates from different *Cryptosporidium* species known to infect cattle and/or sheep. For each protein the amino acid sequences were highly conserved in *C. parvum* isolates, with some differences identified between *Cryptosporidium* species. Analysis of the Cpa135, CP2, CP15 and P23 protein structures across the *Cryptosporidium* species identified several domains and regions of interest. Cpa135, CP2 and P23 were all suggested to contain a signal peptide. Cpa135 was also identified to potentially contain a ricin-B, galactose-binding, fibrinogen-like and a limulus factor C cochlear (LCCL) protein domain. CP2 was found to contain multiple regions of coiling and disorder predictions and a region within the CP15 protein was identified as being a sequence which matched that of the ribosomal S19 family. Furthering the knowledge of these proteins allows for a way to investigate the presence of such in *Cryptosporidium* isolates and theorise their functions in infection along with their potential association with pathogenicity. All of which could then be used to advance vaccine production. This research provides a basis for the use of these proteins or their specific domains as potential vaccine candidates against cryptosporidiosis which could be developed upon through the production and expression of recombinant proteins.

Presenter: **Dr Ashwaq Alnazawi**, *Technical Office Director in VBDs, Ministry of Health*

Poster 67 : Wind tunnel studies on the effect of insecticide treated materials on Ae.aegypti host location behaviour

Authors - A Alnazawi[1]; D Weetman[2]; P mccall[2];
[1] Preventive Medicine Department, Public Health Directorate, Ministry of Health, Saudi Arabia; [2] Department of Vector Biology, Liverpool School of Tropical Medicine, Pembroke Place, Liverpool, UK

Objective - Background Insecticide treated materials (ITMs), such as bed nets, curtains and doorway and window screens can reduce domestic infestations of *Ae. aegypti* and potentially impact dengue transmission. All ITMs depend on pyrethroids, but as shown elsewhere in this thesis, strong pyrethroid resistance exists in *Ae. aegypti* populations from Makkah and Jeddah. This chapter reports on investigations into the behaviour of host-seeking adult females of these resistant populations as they respond to a host behind an ITM barrier containing holes.

Methods Field-collected insecticide resistant mosquitoes from Jeddah and Makkah,

and a fully susceptible New Orleans strain were released individually into a wind tunnel to fly upwind towards holed nets (untreated/PermaNet 2.0). The behavioural events (Flying, Resting, Bouncing, Visiting) were digitally recorded for 20 min and analysed.

Results In a wind tunnel bioassay, 100% of Makkah females, 87.5% of Jeddah and 60% of New Orleans (control susceptible strain) mosquitoes successfully passed through the holes in an untreated net within the 20 min trial period. There was a significant reduction in the number of mosquitoes that passed through the treated net compared to untreated net (P<0.0005). This reduction was significantly greater for the New Orleans susceptible strain (85%) compared to the resistant strains, Jeddah (59%) and Makkah (42%) (P<0.01). All New Orleans were knocked down by the end of the assay, Jeddah and Makkah were not knocked down but 90% and 45% mortality, respectively was recorded in the 24h post assay assessment. Analysis of specific behavioural events showed an increase in resting on the wind tunnel walls post contact with insecticide treated net and a reduction in bouncing and visiting the net, suggesting an impact of contact irritancy or a sub-lethal effect from deltamethrin.

Conclusion These data indicate that a PermaNet 2.0 net might fail to protect against the resistant Makkah and Jeddah mosquitoes. However, further behavioural studies are needed to understand mosquito behaviour to ITNs alongside other vector control interventions. Overall, this indicates that physiological resistance enabled resistant mosquitoes to pass throught the holed treated nets better by surviving long enough to do so, rather than by changing behaviours with patterns either similar between strains or not varying in a manner consistent with resistance level.

Presenter: **Miss Mona Suleiman**, *PhD student, University of Bath*

Poster 68* : piRNA-like small RNAs target transposable elements in a Clade IV parasitic nematode.

Authors - **M Suleiman**[1]; A Kohnosu[2]; B Murcott[1]; M Dayi[2]; B Pawluk[1]; A Yoshida[2]; T Kikuchi[2]; V Hunt[1];
[1] University of Bath, UK; [2] University of Miyazaki, UK

Objective - The small RNA (sRNA) pathways identified in the model organism *Caenorhabditis elegans* are not widely conserved across nematodes. For example, the PIWI pathway and PIWI-interacting RNAs (piRNAs) are involved in regulating and silencing transposable elements (TE) in most animals but have been lost in nematodes outside of the *C. elegans* group (Clade V nematodes), and little is known about how nematodes regulate TEs in the absence of the PIWI pathway. Here, we investigated the role of sRNAs in the Clade IV parasitic nematode *Strongyloides ratti.* We compared two genetically identical adult stages (the parasitic female and free-living female) and identified putative small-interfering RNAs, microRNAs and tRNA-derived sRNA fragments that are differentially expressed between the two adult stages. A parasite-associated class of 21-22 nucleotide (nt) long sRNAs with a 5' uracil (21-22Us) and monophosphate modification were predicted to regulate TE activity. The 21-22Us show striking resemblance to the 21U PIWI-interacting RNAs found in *C. elegans*, including an AT rich upstream sequence, overlapping loci and physical clustering in the genome.

Presenter: **Dr Mohammad Hossein Feiz Haddad**, *Academic Board , Ahvaz Jundishapur University of Medical Sciences*

Poster 69 : Co- infection of parasites and fungal infection and COVID- 19

Authors - S Maraghi[2]; A Rafiei[1]; **MH Feiz Haddad**[1];
[1] Infectious and Tropical Diseases Research Center, Health Research Institute and Department of Parasitology, Medical School, Jundishapur University of Medical Sciences, Ahvaz, Iran; [2] Department of Parasitology, School of Medicine, Ahvaz Jundishapur University of Medical Sciences, Ahvaz, Iran

Objective - COVID- 19 is still the most serious worldwide public health crisis. However, the differences in susceptibility to COVID- 19 and the disease severity across nations continue to be concern. The number of cases and mortality of COVID- 19 is on the rise. The morbidity and mortality of COVID- 19 in low income with chronic helminthic infection is lower than developed countries. According to the studies in African countries, patients co- infected with parasites had not severe symptoms. Parasite co- infection with both protozoa and helminthes may protect against prognosis of sever COVID- 19. The most common fungal infections in patients with COVID- 19 include invasive pulmonary aspergylosis, candidiasis and mucormycosis, especially in high risk and immune- compromised or on long term pharmacotherapies' to develop fungal infection. In our Medical diagnostic laboratory (IRANZAMIN) in Ahvaz city, Southwest Iran, I examine the specimens referred to the lab for parasitology and fungal examination. Since the outbreak of COVID- 19 in 2019. I reported many cases of opportunistic fungi, including aspergylosis, candisiasis, mucormycosis in lavage, sinus discharge and sputum received from the COVID- 19 patients. All diagnosed cases were in ICU ward. Most of them were diabetes or receiving immune- suppressed medicine. In a research carried out in Kerman province, by Professor Iraj Sharifi and his colleagues on COVID- 19 patients with a history of previous cutaneous leishmaniasis scar indicated the significantly prevented incidence of morbidity and mortality. The crossprotection mediated by the CL cured cases would presumably retard COVID- 19 in endemic countries. Analyses showed that COVID-19 cases reduced with endemicity of malaria, schistosomiasis, or soil-transmitted helminth infections suggestive of a possible protective effect from COVID-19. Nioni and Napoli (2020) reported that individuals in malaria endemic settings seem to be protected from COVID-19. These preliminary findings from an ecological analysis, support the hypothesis of a possible immune-modulatory mechanism induced by parasitic infections, which is protective against COVID-19 and warrants further investigation (Ssbambulidd et al, 2020)

Presenter: **Dr Valeria Silvestri**, *Post Grad Msc Student , MUHAS University of Dar es Salaam*

Poster 70* : Blood flukes and arterial damage. A review of aneurysm cases in patients with Schistosomiasis.

Authors - V Silvestri[1]; MI Mshana[1]; W Bonavenuture[1]; J Nyanda[1]; D Sabas[2]; B Ngasala[1];
[1] Department of Parasitology and Medical Entomology, Muhimbili University of Health and Applied Science MUHAS P.O. Box 65001 Dar es Salaam Tanzania, Tanzania; [2] Directorate of Library Services, Muhimbili University of Health and Allied Sciences, Tanzania, Tanzania

Objective - Introduction - Schistosomiasis is a three-stage disease caused by trematode worms of the genus *Schistosoma*. Organ-specific morbidity, according to the infecting *Schistosoma* spp., can develop in chronic stages. Clinical manifestations are caused by inflammatory response of the host to the accumulation of parasite eggs in vessels and organs. Vascular complications of Schistosomiasis are less described than the involvement of other organs or systems. Damage can occur by direct lesion of vessels walls or to contiguity with a focus of inflammation in surrounding tissues. Among vessel lesions, aneurysms have been described.

Aims and methods - Aim of our study was to analyse the occurrence of aneurysm lesions in patients with Schistosomiasis, through a review of case reports in literature.

Results - 12 cases were included in our review. The mean age of patients was 41.3±14.9 years (range 18-66), 7 male and 5 females. More frequently reported comorbidities were cardiovascular, including hypertension, previous aortic dissection and pulmonary valvular steno-insufficiency in 3 cases. Oesophageal varices, previous smoke and alcohol consumption have also been reported.

A previous history of Schistosomiasis was known in 7 cases. In the majority of cases an intestinal or hepato-splenic involvement was reported (7 cases), followed by pulmonary Schistosomiasis (5 cases) and urinary or testicular (2 cases). Pain (refereed to chest, hypochondrium or lumbar region) was the most frequently reported symptom (6 cases), followed by dyspnoea (in 5 cases). Fever, hoarseness, hyper-eosinophilia, syncope/cardiogenic shock and anaemia were reported in a minority of cases.

Lesion in pulmonary artery was reported in 5 cases. Aortic arch, thoraco-abdominal aorta, hepatic, renal, splenic artery and portal involvement were also reported. Aortic rupture on previous aortic graft occurred once.

In four cases no surgical treatment was performed: in 2 because of patients refusal, in one because of exitus and in 1 for unspecified reason. Among performed treatments 1 splenectomy, 1 aneurysmectomy with renal reimplantation, in aneurysmectomy of aortic arch and one reintervention on aortic graft have been reported.

Follow-up data were available for 6 patients. 3 survived. In one case, post splenectomy portal vein partial thrombosis complicated occurred, but resolved at 4 years follow up. Exitus occurred in three cases, and was due to post-operatory cardiogenic shock in aortic reintervention for aortic rupture; pulmonary embolism in pulmonary aneurysm (refused treatment) and for ruptured pulmonary artery aneurysm with cardiac tamponade.

Conclusions - Aneurysms may occur in patients with Schistosomiasis. Given the high burden of this neglected tropical disease in endemic regions, and the high morbidity and mortality of vascular conditions there is a need for studies that will better define physiopathology and guide clinicians.

Presenter: **Miss Huanghehui Yu**, *Student, University of Glasgow*

Poster 71* : Association between *Schistosoma mansoni* infection intensity, praziquantel side effects, and drug efficacy, in Ugandan school-aged children

Authors - H YuJ ClarkM Arinaitwe[2]; M Adriko[2]; NB Kabateriene[2]; JM Prada[3]; E Janoušková[3]; DW Oguttu[2]; JP Webster[1]; PH Lamberton
[1] Royal Veterinary College, University of London, UK; [2] Vector Control Division, Ministry of Health, Uganda; [3] University of Surrey, UK; [4] Institute of Biodiversity, Animal Health and comparative Medicine, and Wellcome Centre for Integrative Parasitology, University of Glasgow, UK

Objective - Schistosomiasis is a debilitating parasitic disease infecting over 240-million people and is contracted through contact with contaminated freshwater. This project focuses on *Schistosoma mansoni*, the main cause of intestinal schistosomiasis. The World Health Organization's recommended control strategy is treatment with the anthelmintic praziquantel, administered by mass drug administration (MDA). However, praziquantel has possible side effects including abdominal pain, diarrhoea, dizziness and vomiting. Side effects have been reported as a reason for low treatment uptake during MDA campaigns. Epidemiological data from primary school children in a *S. mansoni* endemic area in Uganda,were analysed to investigate: a) whether being infected with *S. mansoni*, or b) infection intensity, could predict the occurrence of side effects, and c) whether side effects (except vomiting) were positively associated with successful clearance of infection after treatment and d) if vomiting was negatively associated with infection clearance post treatment. Infection intensity pre and post praziquantel treatment and side effect data were collected from three schools in Mayuge District, Uganda from 2004 to 2006. Children who vomited within an hour of treatment were retreated. Using generalized linear modelling I show that *S. mansoni* infection prevalence is positively associated with side effects, but the students who experienced side effects were more likely to clear infection. However, the infection intensity did not explain the number of side effects, and vomiting did not affect treatment efficacy. This research highlights that school-aged children with intestinal schistosomiasisinfection are at higher risk of experiencing side effects when given praziquantel, regardless of the intensity of infection, than uninfected children, but students who experienced side effects also had higher treatment efficacy, which indirectly supports a causal link between worm death and side effects.

Presenter: **Ms Antonia Konle**, *Student , Cell- and developmental Biology, University Würzbu*

Poster 72* : Limits of Flagellar Pocket Access in African Trypanosomes

Authors - **A Konle**[1]; K Niedermüller[1]; B Morriswood[1];
[1] Department of Cell & Developmental Biology, Biocentre, University of Würzburg, Würzburg, Germany, Germany

Objective - *Trypanosoma brucei* is an extracellular parasite which lives in the bloodstream of infected mammalian hosts and is in continuous exposure to the immune system. Its surface is covered in a dense glycoprotein coat which is continually endo- and exocytosed in order to remove any bound antibodies. Nutrients such as LDL (low density lipoprotein) and transferrin are scavenged by endocytosis. Remarkably, all endo- and exocytic activity is restricted to a single small subdomain of the plasma membrane - an invagination called the flagellar pocket. Even though trypanosomes are capable of internalising very large macromolecular complexes such as LDL, the exact size limit for macromolecule entry to the flagellar pocket is unknown. Cytoskeleton-associated protein complexes that are coiled around the neck of the flagellar pocket, such as the hook complex, are suspected to influence the size limit and endocytic activity. In this study, the size limits for fluid phase and surface-bound cargo entry were systematically measured. Depletion of specific protein components of the hook complex did not influence the defined size limit. Surprisingly, surface-bound cargo accumulated at the flagellar pocket entry after depletion, prompting a re-evaluation of the proposed function of these complexes.

Presenter: **Dr Douglas Escrivani de Oliveira**, *Postdoctoral research Assitant, University of Dundee*

Poster 73 : Competitive growth drives a hierarchy of antigenic variation in African trypanosomes.

Authors - **D O. Escrivani**[1]; V Scheidt[1]; M Tinti[1]; J Faria[1]; D Horn[1];
[1] The Wellcome Trust Centre for Anti-Infectives Research, School of Life Sciences, University of Dundee, UK

Objective - Several persistent pathogens employ antigenic variation to continually evade mammalian host adaptive immune responses. African trypanosomes use variant surface glycoproteins (VSGs) for this purpose, transcribing one telomeric *VSG* expression site (ES) at a time, with strict silencing of other telomeric ESs. VSG switching occurs at low frequency, primarily exploiting a reservoir of (sub)telomeric *VSG* templates, including >60 minichromosomal VSGs in *Trypanosoma brucei*, to replace the active *VSG*. It has been known for over fifty years that VSGs emerge in a hierarchical order, and *VSGs* from silent ESs (ES-*VSGs*) and from minichromosomes (MC-*VSGs*) are now known to dominate the early phase of infection. Here, we quantitatively assess factors that contribute to the hierarchy, independent of host immune selection. We triggered high-frequency *VSG* gene recombination and switching in *in vitro* culture using inducible CRISPR-Cas9 to target the active *VSG*. *VSG* dynamics were then assessed using RNA-seq. Activation rates were highly variable, but typically higher for polycistronic ES-*VSGs*. MC-*VSGs* subsequently displayed a competitive advantage over ES-*VSGs*, however, and came to dominate the population, a feature that was particularly pronounced when only longer *VSGs* were considered. Slower growth associated with ES-*VSG* activation was linked to widespread transcriptome differences, including increased expression of ES-associated genes. We conclude that VSG dynamics are underpinned by differential VSG activation, and subsequent competitive growth, both of which are *VSG* template location-dependent, and which is also *VSG* length-dependent in the latter case. These features may prolong parasite immune evasion using a limited set of variant antigens.

Presenter: **Mr Mohammad Alharbi**, *PhD student, Liverpool school of Tropical Medicine*

Poster 74 : Molecular characterisation of medically important freshwater snails in Saudi Arabia

Authors - **M Al-Harbi**[1]; EJ LaCourse[1] , JR Stothard
[1] LSTM, UK

Objective - Health authorities in the Kingdom of Saudi Arabia (KSA) face several national challenges to eliminate schistosomiasis transmission, particularly in the southern region where there is active transmission is known and application of molecular methods of snail identification has been limited. Better knowledge of snail-related aspects is needed, and this study sought to better characterise local populations *Biomphalaria* and *Bulinus*. Two targeted malacological surveys were conducted to collect snails from active transmission foci in Albahah and Abha. Water chemistry measurements were made with snails subjected to traditional surveillance methods e.g., cercarial shedding test and modern molecular methods of DNA sequencing, microsatellite DNA typing and qPCR analysis to detect *Schistosoma* DNA within snails. A total of 80 *Biomphalaria* specimens were examined. Size-fractionation of microsatellite alleles at six loci revealed very low intra-populational genetic diversity and only two *cox*1 haplotypes were found. Although no *Biomphalaria* were found shedding cercariae in the field, the qPCR assay screening for *S. mansoni* DNA detected ten positive samples from Albahah. In addition, molecular characterisation of snails collected from an active transmission location in Abha confirmed the presence of *Bulinus forskalii*, which has not been reported from this area before. Our results provide a novel epidemiological insight into increasing opportunities for *Schistosoma* transmission. This raises some concern that requires further holistic efforts in environmental control to stop further emergence and spread of medically important snails and parasites, particularly KSA regions close to the border with Yemen.

Presenter: **Dr Kehinde Sowunmi**, *Lecturer, Enugu State University of Science and Technology*

Poster 75 : A preliminary survey of freshwater snails in Ugbawka, Nkanu East Local Government Area (LGA) of Enugu State, Nigeria.

Authors - K Sowunmi[1];
[1] Enugu State University of Science and Technology, Nigeria

Objective - Freshwater snails are an important target for the integrated approach to control of snail-borne human and animal trematode infections such as schistosomiasis and fascioliasis. This preliminary survey was conducted to determine the species variety and abundance of freshwater snails present in six streams/rivers of Ugbawka, Nkanu East LGA. A total of 128 snails were collected randomly from the six locations using a scoop net or hand-picked from vegetation and were identified morphologically. Five species of snails identified and their overall percentage frequencies were *Bulinus truncatus* (12.5%), *Bulinus forskalii* (10.2%), *Lymnaea (Radix) natalensis* (8.6%), *Lanistes libycus* (59.4%) and *Melanoides tuberculata* (9.4%). The highest number of snails was collected from the Ojolowo River (38.3%) and least species abundance was recorded in Umukwasiri Stream where only one species, *Lanistes libycus* was found. Snails screening revealed no cercariae in the snails collected. The importance of regular surveillance and monitoring of freshwater snail populations in rivers and streams used by residents of the community for farming, recreation and domestic purposes is discussed.

Presenter: **Miss Yee Wan Liu**, *Phd student, University of Glasgow, IBAHCM*

Poster 76 : Repurposing trypanocidal drugs to tackle amoebic gill disease in Atlantic Salmon

Authors - Y Liu[4]; B Cheaib[3]; MS Llewellyn[2]; M Barret[1]; P McGinnity[5]; N Ruane[5]; J Archibald[6]; R Williams[7]; F Hernandez[7];
[1] Institute of Infection, Immunity and Inflammation, University of Glasgow, UK; [2] Institute of Biodiversity, Animal Health and comparative Medicine, University of Glasgow, UK; [3] Institute of Biodiversity, Animal Health and Comparative Medicine, University of Glasgow, UK, UK; [4] Wellcome Centre for Integrative Parasitology, Institute of Infection, Immunity and Inflammation, University of Glasgow, UK; [5] Marine Institute, Newport, Ireland; [6] Dalhousie University, Halifax, Canada; [7] Institute of Biomedical and Environmental Health Research, School of Health and Life Science, University of West Scotland, UK, UK

Objective - Amoebic gill disease (AGD) is a devastating disease that causes multi-million-dollar loss annually in the salmonid fish farming business. The causative agent of AGD in Atlantic Salmon is *Paramoeba perurans* which belongs to the *paramoebidae* family. An interesting feature of most of the *paramoebidae* family members is the symbiotic relationship they have with the *perkinsela*-like organism (PLO). As there is a high level of metabolic interdependence between host and symbiont, elimination of the PLO, which resides adjacent to the nucleus of its symbiotic host, will hypothetically kill the parasite

The PLO, although losing its flagellum, belongs to the kinetoplastid group and contains many biochemical features similar to those of disease-causing parasites such as Leishmania and trypanosome. The use of anti-leishmanial and anti-*Trypanosoma*l may be able to kill the symbiont of *P. perurans*. In this study, we attempt to identify a candidate drug that targets PLO with via various drug assays and microscopy. Once identified the drug effect on *P. perurans* will also be explored using the omics approach (transcriptomics and metabolomics) to explore its mode of toxicity as well as to further probe the nature of this unique endosymbiosis. In addition to seeking treatment for AGD, repurposing drugs currently licensed for human and veterinary trypanomatids may be able to reduce their cost which is, in many cases, otherwise too expensive to implement to enable large scale treatment of neglected tropical diseases.

Presenter: **Mr Mahbod Entezami**, *PhD Student, University of Surrey*

Poster 77* : Evaluating control and elimination methods of cystic echinococcosis in South America – Beyond the 2030 goals

Authors - M Entezami[2]; J Widdicombe[2]; G Mujica[4]; E Larrieu[3]; MG Basáñez[1]; A Casulli[5]; G Lo Iacono[2]; JM Prada[2];
[1] Imperial College London, UK; [2] University of Surrey, UK; [3] Universidad Nacional de Rio Negro, UK; [4] Ministerio de Salud de Río Negro, UK; [5] Istituto Superiore di Sanitá, UK

Objective - Aim: The World Health Organization 2021–2030 roadmap on neglected tropical diseases has proposed that intensified control be implemented for cystic echinococcosis (caused by infection with the cestode *Echinococcus granulosus sensu lato)* in highly endemic areas of 17 countries by 2030. We aim to evaluate the effectiveness of different interventions in South America, which can be quantified with a transmission model for *E. granulosus* between sheep and dogs.

Methods: We developed a multi-host, individual-based transmission model that captures the parasite population dynamics processes across intermediate hosts (sheep)—which develop infective cysts; definitive hosts (dogs) that acquire the infection from ingestion of infected offal (and harbour adult worms), and the environment, contaminated with parasite eggs. Humans are accidental dead-end hosts that can develop cysts (hydatid disease). We simulated several interventions to assess their effectiveness in reducing CE prevalence in sheep and dogs.

Results: Local control of CE can be difficult using deworming drugs alone. However, the EG95 sheep vaccine could potentially be a game changer. Management practices play a large role in shaping transmission events and can have a substantial impact on human health.

Conclusions and Future work: Part of the challenge in controlling and eliminating CE is the costs of such a programme. As hydatid disease prevalence decreases in human communities, it becomes harder to justify the cost of elimination in the zoonotic reservoirs. We will conduct a cost-effectiveness analysis on each combination of interventions to evaluate their cost against the health benefits gained.

Presenter: **Mr Zihao Chen**, *student, University of Edinburgh*

Poster 78 : Assembly and annotation of *Trypanosoma congolense* kinetoplast DNA and comparison with *T. brucei*

Authors - Z Chen *[2]; E Wadsworth *[2]; P Buscher[1]; F Van den Broeck[4]; N Savill &[3]; A Schnaufer &[3];
[1] Institute Tropical Medicine, Antwerp, Belgium; [2] University of Edinburgh, UK; [3] Institute of Immunology & Infection Research, University of Edinburgh, UK; [4] Department of Microbiology, Immunology and Transplantation, Rega Institute, Belgium
*Shared first **Authors** & Shared senior **Authors**

Objective - The eponymous mitochondrial DNA in kinetoplastid organisms (also called kDNA) is unique in structure and gene content. Kinetoplastea include the *Trypanosoma*tid parasites, where kDNA is organised as a massive network of concatenated circular DNA molecules. *Trypanosoma brucei brucei* (Tbb) kDNA contains 20-50 practically identical copies of ~23-kb maxicircles and 5-10k highly heterogeneous 1-kb minicircles. Homologous to other eukaryotic mitochondrial genomes, maxicircles encode genes essential to oxidative phosphorylation and mitochondrial translation, including 2 rRNA genes and 18 protein coding genes. Minicircles encode guide RNAs (gRNAs) that direct post-transcriptional editing of the pre-mRNA products from 12 encrypted maxicircle genes by insertion and deletion of uridines by virtue of their complementarity to the fully edited version. Although numerous maxicircle genes are essential to the parasites' survival in the tsetse fly vector, only the A6 gene, encoding a subunit of the F1FO-ATP synthase, along with the maxicircle-encoded subunits of the mitoribosome required to translate its mRNA, are essential in the mammalian bloodstream stage.

T. b. equiperdum (Tbeq) and T. b. evansi (Tbev; the subspecies designation adopted here is under debate) evolved from Tbb on multiple occasions by switching from transmission via tsetse flies, with obligate development in the vector, to direct transmission between mammals, either sexually in horses (Tbeq) or mechanically between many mammalian species via biting flies or vampire bats. Thus, Tbeq and Tbev are polyphyletic taxa and, to complicate things further, monophyletic subgroups of Tbev include strains historically designated as Tbeq and vice versa. As Tbeq and Tbev are generally considered to have evolved kDNA independence and have undergone partial (and sometimes complete) loss of the organellar genome, key molecular differences between them and Tbb concerns kDNA . Previous studies of three distinct monophyletic groups of Tbeq and Tbev isolates have suggested that (i) Tbev isolates generally lack a maxicircle and (ii), where present, kDNA is dominated by one of three minicircle classes, type A, B, or C. Both observations are being used for diagnostic purposes. However, an in-depth comparative analysis of Tbeq/Tbev kDNA is lacking.

We have used next-generation sequencing data and a bespoke assembly pipeline to compare kDNA from 56 Tbeq/Tbev samples to each other and to a Tbb reference. Here, we report three main findings:

1) For three groups of Tbeq/Tbev, we confirm that the minicircle genomes consist of thousands of copies of a single type A, B, or C minicircle class specific and therefore diagnostic for each group.

2) Unexpectedly, one type A Tbev isolate (Vietnam strain, Gillingwater et al. 2007) had a complete maxicircle.

3) A fourth group of isolates has a higher minicircle diversity of 43-45classes per network. These minicircles encode gRNA genes sufficient for directing complete editing of the A6 mRNA. However, editing of the RPS12 mRNA, encoding a subunit of the mitoribosome, appears to be incomplete, suggesting that kDNA independence in these isolates evolved relatively recently in these strains.

Altogether, the project sheds light on how the complex evolutionary history of non-tsetse transmitted T. *brucei* strains has shaped their

Presenter: **Miss Safaa Elhassan Bashir** , *Medical laboratory scientist , Self employed*

Poster 79* : Seroprevelance and Risk Factors of *Toxoplasma Gondii* Infection Among Healthy Blood Donors in AL-Ribat Teaching Hospital Khartoum State, Sudan.

Authors - S Bashir[1];
[1] Sudan University of science and technology, Sudan

Objective - Toxoplasmosis is an opportunistic, zoonotic disease with a worldwide distribution. There are large variations in the seroprevalence of T. *gondii* infection in different regions of the world. Toxoplasmosis is a common parasitic disease can be transmitted to human through variety of routes including blood and there is risk of exposure to this parasite in blood donors during the periods of life. Nowadays, there is no laboratory screening of blood donors for T.*gondii* is not routinely available. This cross sectional study aimed to evaluate the seroprevalence and risk factors associated with *Toxoplasma* infection among healthy blood donors. Between March to August 2016 at Alribat teaching hospital in Khartoum state, a serum samples were taken from 100 blood donors with age range between 10-50 years old. The sera were examined for anti-*Toxoplasma* antibodies (IgG & IgM) by the ELISA test. The overall rate of anti-*Toxoplasma* antibodies determined by ELISA was (32%) (IgG) and (3%) (IgM). The results showed that the highest prevalence rate was reported among the 31-40 age group (47.8%) when examined by ELISA test. Drinking milk and

meat consuming were found to be of no significance in the transmission cycle. Contacts with cats have been shown to be of great importance in the transmission cycle. The present study indicates that prevalence of toxoplasmosis is high in the study area.

Presenter: **Dr Denis Dacheux**, *Associate Professor, Fundamental Microbiology and Pathogenicity ,UMR 5234 CNRS-Bordeaux University-Bordeaux INP*

Poster 80 : TFK1, a basal body transition fibre protein that is essential for cytokinesis in *Trypanosoma brucei*

Authors - **D Dacheux**[1];
[1] UMR-5234, Université de Bordeaux, CNRS, Bordeaux INP, Microbiologie Fondamentale et Pathogénicité , F-33000 Bordeaux, France., France

Objective - TFK1 is a kinetoplastid specific protein and a mature and maturing BB marker, localized on the transitional fibre. TFK1 is the third component of the transition fibres region, with TbRP2 and CEP164C, in T. *brucei*. Our high-resolution ultrastructure expansion microscopy data demonstrate that TFK1 is displayed in a typical radial arrangement in the distal appendage matrix, as nine dense points between the molecules of CEP164C. TFK1 is essential for BSF, unlike PCF. Its depletion induces, on one hand, previously undescribed cytokinesis defects by the absence of furrow associated with segregation of BBs similar to that of PCFs (KNKN) and on the other hand, leads to the blockage of abscission during cytokinesis in BSFs

Presenter: **Dr Okpala Michael**, *Lecturer, University of Nigeria, Nsukka*

Poster 81 : Efficacy of Combined Therapy Of Diminaveto® And Intromidium® In Albino Rats Experimentally Infected With *Trypanosoma brucei brucei*

Authors - CF Obi[2]; IO Ezeh[1]; **MI Okpala**[1]; LG Nwobi[1]; TA Nzeakor[1]; GE Aneru[1]; RC Ezeokonkwo[1];
[1] University of Nigeria, Nsukka, Nigeria; [2] University of Nigeria, Nsukka Nigeria, Nigeria

Objective - The effect of Diaminaveto®(diminazene aceturate) and Intromidium®(isomethamidium chloride) combination in rats infected with *Trypanosoma brucei* was evaluated. 40 albino rats were used for this study. The rats were grouped into 8 groups of 5 rats each. Rats in group 1 were not infected while groups 2-8 were inoculated intraperitoneally (IP) with 1 x 10^6 trypanosomes. Rats in group 2 were untreated. Rats in groups 3-8 were treated on day 17 post infection. Groups 3 and 4 rats were treated with 3.5 mg/kg Diaminaveto®(DM) and 0.5 mg/kg Intromidium®(IM) respectively. 7 mg/kg DM and 1 mg/kg IM were administered to rats in groups 5 and 6 respectively. Groups 7 and 8 were treated with 7 mg/kg DM and 1.0 mg/kg IM respectively and these treatments were reversed after two weeks. Level of parasitaemia, clinical signs, survivability, body weight changes, rectal temperature (RT), haematological indices (packed cell volume, hemoglobin concentration, total leucocytes count and differential leucocyte count) and rate of parasite clearance were used to evaluate the efficacies of the drugs and their combination. There was a significant ($P < 0.05$) reduction in PCV, HbC, TLC and weight of rats post infection (PI). These indices were reversed post treatment (PT), though this reversal was faster and lasted longer in rats in groups 7 and 8. The results of the study showed that the combined therapy of DM and IM at different time intervals was more efficacious than single treatment regimen of either DM or IM.

Keywords: *Trypanosoma brucei*; Isometamidium chloride; Diminazene aceturate;Rats

Presenter: **Dr Okpala Michael**, *Lecturer, University of Nigeria, Nsukka*

Poster 82 : Comparative antibody response of albino rats infected with *Trypanosoma brucei brucei* via intraperitoneal and intradermal routes

Authors - CF Obi[2]; IO Ezeh[1]; **MI Okpala**[1]; LG Nwobi[1]; DN Onah[1];
[1] University of Nigeria, Nsukka, Nigeria; [2] University of Nigeria, Nsukka Nigeria, Nigeria

Objective - The antibody responses of albino rats infected with *Trypanosoma brucei* via the intraperitoneal (IP) and intradermal (ID) routes was investigated and compared. A total of twenty-five adult male albino rats were used. The rats were randomly assigned to three groups (A, B, and C) comprising five rats in group A, and ten rats each in groups B and C respectively. Group A (uninfected control) were sensitized with washed sheep Red Blood Cells (wSRBC) at day 0. Group Bwas divided further into groups B^1 and $B^2$and infected intraperitoneally (IP) and intradermally (ID) with 4 x 10^5*Trypanosoma brucei* respectively, and then sensitized with wSRBC at day 0, and monitored. Group C was also divided further into groups C^1 and $C^2$and infected IP and ID with 4 x 10^5*Trypanosoma brucei* respectively at day 0, and then sensitized with wSRBC at day 2 and boosted at day 14 post sensitization and monitored. Level of parasitaemia (LOP), haematological indices (PCV, total WBC and differential WBC counts), rectal temperature and antibody response to wSRBC were used to compare the antibody responses via these routes of infections. Parasitaemia was recorded 3- and 5-days post infection via IP and ID routes respectively in rats sensitized at different times. There was a significantly higher ($P <$

0.05) parasitaemia in rats infected IP when compared to rats infected via ID route. The haematological indices of rats infected via ID routes were better than that of those infected via IP routes. In conclusion, the ID route of infection showed a higher antibody response which was maintained over a longer time when compared to the IP route.

Keywords: antibody response, intradermal/intraperitoneal infection, *Trypanosoma brucei brucei*, rats

Presenter: **Dr Chidinma Amuzie**, *Lecturer, Rivers State University*

Poster 83 : Intestinal helminth parasites of grasscutters (*Thryonomys swinderianus*) sold at Omagwa Bushmeat Market, Omagwa, Rivers State, Nigeria

Authors - C Amuzie[1]; B Robert[1]; GC Akani[1];
[1] Rivers State University, Nigeria

Objective - Grass-cutters (*Thryonomys swinderianus*) are important sources of bush-meat in southern Nigeria. Here, we examined intestinal helminth parasites of grass-cutters sold at Omagwa bushmeat market, Rivers State, Nigeria. Intestines of twelve specimens were bought from the bush-meat sellers from December, 2020 – February, 2021. They were fixed in 10% formalin to curtail decay and transported to the Entomology and Parasitology Laboratory, Department of Animal and Environmental Biology, Rivers State University, Port Harcourt, Nigeria. Each sample was incised and its contents examined in sections in 0.9% normal saline solution. Parasites encountered were washed in 0.9% normal saline solution, fixed in 70% ethanol and subsequently identified using taxonomical keys. Prevalence of infection was computed using standard formula. Nematode parasites were isolated from ten infected hosts: *Oesophagostomum venulosum* from the large intestine, *Strongylus* sp., *Toxocara vitulorum* and *Trichuris paravispicularis* from the small intestine. Each of *O. venulosum* and *Strongylus* sp. infected six hosts accounting for a prevalence of 50.0%. Mean intensity of infection was 12 parasites per infected host for *O. venulosum* and 7 parasites per infected hosts for *Strongylus* sp. *Trichuris paravispicularis* infected four hosts accounting for a prevalence of 33.3% and mean intensity of two parasites per infected host, while *Toxocara vitulorum* was recovered as a single individual from only one host at a prevalence of 8.3%. It is concluded that *T. swinderianus* is actively involved in the life cycle of nematode parasites and its conservation by domestication or in the wild would require deworming regimens using appropriate anthelminthic drugs and hygienic practices to ensure their health.

Presenter: **Mr Stanley Otoboh**, *PhD researcher, University of Ediburgh*

Poster 84* : Genetic validation of the function of PfEMP1 in *Plasmodium falciparum* rosette formation.

Authors - SE Otoboh[2]; HM Abkallo[1]; AJ Rowe[2];
[1] International Livestock Research Institute, Kenya; [2] Institute of Immunology and Infection Research, University of Edinburgh., UK

Objective - Rosetting, the binding of *P. falciparum* infected erythrocytes to uninfected erythrocytes to form clusters (rosettes), is thought to contribute to severe malaria. This adhesion phenotype is mediated through the adhesive properties of *P. falciparum* erythrocyte membrane protein 1 (PfEMP1), encoded by the var gene family (~60 distinct copies per parasite genome expressed in a mutually exclusive manner). The N-terminal domain (NTS-DBLα) of PfEMP1 has been implicated as the functional erythrocyte binding domain. However, the role of PfEMP1 in adhesion in live infected erythrocytes remains poorly understood, mainly due to the difficulty in genetically manipulating *P. falciparum*. Here, we aim to test a strategy to generate a population of parasites expressing a single *var* gene and use CRISPR-Cas9 genome editing technology to investigate whether specific motifs within DBLα of the rosetting PfEMP1 variant "IT4var60" are sufficient to mediate the rosetting phenotype in *P. falciparum*. We have shown that a *var* gene co-expressed with a drug resistance gene via 2A peptide can be inducibly and exclusively expressed under drug pressure. Presently, we are generating transgenic parasites to test the role of specific PfEMP1 domains/motifs/residues in rosette formation. This work has potential to contribute to the rational design of anti-rosetting interventions, with the ultimate goal of reducing deaths from severe malaria.

Presenter: **Miss Hasana Baber**, *PhD Student , Keele University*

Poster 85* : Investigating a galactokinase orthologue from *Leishmania donovani*

Authors - H Baber[1]; E King[1]; M Maciej-Hulme[2]; H Price[1]; **A Winter**[1];
[1] Keele University, UK; [2] Radboudumc, Netherlands

Objective - *Leishmania donovani* is the causative organism of visceral leishmaniasis. We have identified an enzyme in *L. donovani* known as a galactokinase-like protein (*Ld*GalK), which could be a novel target for drug development. In the human host, two GalK paralogues are expressed, GalK1 and GalK2, which metabolise galactose in the Leloir pathway and N-acetyl galactosamine pathway, respectively. Both kinases instigate the first committed step in their respective pathways and phosphorylate the carbon-1 position in their carbohydrate ligand.

We have expressed recombinant *Ld*GalK in *E. coli* and purified the protein to high purity and yield. The recombinant enzyme is catalytically active with a substate affinity to galactose in the low micromolar range. Interestingly, size exclusion chromatography of *Ld*GalK suggests either an

open/closed conformation of the enzyme or dimerization. Future research will test potential inhibitors against recombinant *Ld*GalK in vitro and against *L. donovani* parasites in vivo.

Presenter: **Dr Jennifer Ann Black**, *Pos-Doc, University of Sao Paulo*

Poster 86 : Monitoring replication stress response dynamics in the Kinetoplastid parasite *Leishmania major* links transcription derived instability to genome plasticity

Authors - J Black[1]; **S Virgilio**[1]; MS Bastos[1]; J Damasceno[2]; G La Silva[1]; K Crouch[2]; R McCulloch[2]; LR Tosi[1];
[1] University of São Paulo Medical School, Brazil; [2] Wellcome Centre for Integrative Parasitology, Institute of Infection, Immunity and Inflammation, University of Glasgow, UK

Objective - Leishmania parasites collectively cause the neglected tropical infection Leishmaniasis. Leishmania have extremely plastic genomes which may promote adaptability in harsh environments and allow the rapid acquisition of drug resistance, but we understand little of how they co-ordinate these processes. Recently, proteins involved in tackling stress during DNA replication have been implicated as key plasticity factors, but little is known about the Leishmania replication stress response (RSR). In most eukaryotes, exposed single-stranded DNA (ssDNA) accumulates at stalled or collapsed replication forks (i.e replication stress; RS), activating the RSR. The atypical kinase ATR is recruited and activated by the ssDNA binding tripartite complex RPA (RPA1-RPA2-RPA3) and the heterotrimeric complex, 9-1-1 (RAD9-RAD1-HUS1). Activated ATR then aids repair factor co-ordination promotes cell cycle stalling, DNA replication fork protection and DNA repair. Here, we examined the distribution of three core RSR factors, γH2A (a marker of DNA damage), LmRPA1 (RPA complex) and LmHUS1 (9-1-1 complex), throughout the Leishmania genome during a replication stress challenge by ChIP-seq. We show that known sites of variability are often marked by replication stress factors, which become enriched under replication stress. Many of these sites are also transcription hubs suggesting interactions between the processes of replication and transcription may drive plasticity. Furthermore, we show LmRPA1 mapping can be exploited to follow DNA replication under synchronising conditions.

Presenter: **Dr Chukwunonso Obi**, *Lecturer , University of Nigeria, Nsukka*

Poster 87* : Comparative pathogenicity of single and mixed drug-resistant *Trypanosoma brucei* and *Trypanosoma congolense* infections in rats: Clinico-haematological findings.

Authors - CF Obi[1]; MI Okpala[1]; DC Anyogu[1]; GE Aneru[1]; A Onyeabo[2]; IO Ezeh[1]; RC Ezeokonkwo[1];
[1] University of Nigeria, Nsukka, Enugu State,, Nigeria; [2] Michael Okpara University of Agriculture, Umudike, Abia State, Nigeria, Nigeria

Objective - Drug-resistant trypanosomes are widespread in the sub-Saharan Africa and in conjunction with the drug-sensitive phenotypes cause an important wasting and endemic protozoan disease in humans and animals. Using thirty-five female albino rats randomly divided into seven groups (1 – 7) of five rats each, pathogenicity of single and mixed drug-resistant *Trypanosoma brucei* and *Trypanosoma congolense* isolated from dogs was assessed. Group 1 served as the uninfected control group. Groups 2 and 3 rats were infected with 10^6 drug-sensitive *T. brucei* and *T. congolense* respectively while groups 4 and 5 rats were infected with 10^6 multidrug-resistant *T. brucei* and *T. congolense* respectively. Group 6 rats were infected with drug-sensitive *T. brucei* and *T. congolense* (5×10^5 each)while group 7 rats were infected with multidrug-resistant *T. brucei* and *T. congolense* (5×10^5 each). Pre-patent period (PPP), parasitaemic period, first peak parasitaemia, days to first peak parasitaemia (DTFPP), level of parasitaemia (LOP), clinical signs, body weight, rectal temperature, packed cell volume, haemoglobin concentration, red blood cell count, mean corpuscular volume, mean corpuscular haemoglobin, mean corpuscular haemoglobin concentration, total leucocyte count and survivability, gross and histopathological changes in the spleen were assessed. Parasitaemia occurred in all the infected groups between days 3 and 9 post infection. Groups 4 and 7 rats had longer (p<0.05) PPP, DTFPP, survivial time and lower (p<0.05) LOP than groups 2 and 6 rats while for groups 3 and 5 rats, these parameter were comparable. Anaemia was observed in the infected groups but the severity did not vary amongst the infected groups. Severe clinical signs and splenic lesions were observed in drug-sensitive trypanosome species infected groups of rats compared to the multidrug-resistant species. Therefore, we conclude that the trypanosome isolates were pathogenic. However, drug-sensitive *T. brucei* andmixed drug-sensitive *T. brucei* and *T. congolense* infections were more pathogenic than their multidrug-resistant counterparts.

Keywords: Drug-resistant trypanosomes; *Trypanosoma brucei; Trypanosoma congolense;* Pathogenicity; Clinical findings; Haematology; Rats.

Presenter: **Dr Chukwunonso Obi**, *Lecturer , University of Nigeria, Nsukka*

Poster 88* : **Prevalence and molecular identification of trypanosomes in dogs in Enugu North Senatorial Zone, South East Nigeria**

Authors - CF Obi[2]; IO Ezeh[2]; MI Okpala[2]; O Agina[2]; PU Umeakuana[3]; GA Essuman[1]; TM Gwira[1]; RC Ezeokonkwo[2];
[1] West Africa Centre for Cell Biology of Infectious Pathogens, University of Ghana, Ghana; [2] University of Nigeria, Nsukka, Enugu State,, Nigeria; [3] University of Abuja, Abuja, Nigeria, Nigeria

Objective - A cross-sectional survey was carried out to determine the prevalence and molecular identification of trypanosomes in dogs in Enugu North Senatorial Zone (ENSZ), South East Nigeria. Dogs (n = 450) were randomly sampled, their blood collected and some characteristics such as sex, breed, sampling location, season and age duly noted. The blood samples were screened for trypanosomosis using standard trypanosome detection techniques. Trypanosome-positive blood samples were spotted on FTA® cards for molecular identification using nested Tubulin-PCR, ITS-PCR, TgsGP-PCR and DNA sequencing. Some haematological parameters of the dogs such as packed cell volume (PCV), total leucocyte count (TLC), red blood cell count (RBC) were also determined. Of the 450 dogs sampled, 51 dogs were positive for trypanosomes with a prevalence rate of 11.3% (95% CI = 0.087 - 0.146). *Trypanosoma brucei* was the predominant trypanosome species infecting dogs in the study area. *T. congolense, T. evansi* and *T. vivax* were also identified. The prevalence of canine trypanosomosis was significantly associated with season (χ^2 = 13.821, *df* = 1,P = 0.0001) and the sampling location (χ^2 = 6.900, *df* = 2,P = 0.032) while sex, breed and age were not. The PCV and RBC of the infected dogs were significantly lower (p < 0.0001) than those of the uninfected dogs. We therefore conclude that CAT due to *T. brucei* is very prevalent in Enugu North Senatorial Zone, South East Nigeria and is associated with haematological changes. Our study also detected *T. vivax* in dogs in South East Nigeria which appears to be the first report of *T. vivax* in a dog in Nigeria.

Keywords: Prevalence; Molecular identification; Canine African trypanosomosis; Trypanosomes; Dogs; South East Nigeria.

Presenter: **Ms Isabel Saldanha**, *Research Associate & PhD Student, Liverpool School of Tropical Medicine*

Poster 89* : Detection of *Trypanosoma brucei* DNA in faeces of experimentally-infected cattle

Authors - I Saldanha[2]; M Betson[4]; KR Matthews[3]; E Paxton[1]; C Vrettou[1]; LJ Morrison[1]; SJ Torr[2]; LJ Cunningham[2];
[1] Roslin Institute, UK; [2] Liverpool School of Tropical Medicine, UK; [3] University of Edinburgh, UK; [4] University of Surrey, UK

Objective - species of *Trypanosoma* transmitted by the tsetse fly (*Glossina*) vector are responsible for clinically significant diseases in both human and animal populations. Although significant advances have been made in the control of human African trypanosomiasis (HAT), animal African trypanosomiasis (AAT) remains a disease of significant economic burden and livestock mortality in sub-Saharan Africa. Current AAT surveillance tools suffer from poor sensitivity and specificity, with serological methods also requiring animal restraint and blood collection by trained personnel. Faecal sampling is an attractive potential option for more accessible sample collection and screening. Therefore, this study set out to determine in the first instance whether it is possible to detect DNA of an AAT aetiological agent (*T. brucei*) in the faeces of experimentally-infected cattle. Five male Holstein-Friesian calves of post-weaning age were inoculated with *T. brucei* AnTat 1.1 and the infection course was followed for a total of 68 days. A total of 146 faecal samples (12 pre-infection, 134 post-infection) were passively collected and screened using PCR and a novel probe-based qPCR assay targeting a *Trypanozoon*-specific repeat region in kinetoplast minicircle DNA. Target DNA was successfully detected in 85% (n=114) of post-inoculation faecal samples by qPCR and 50% (n=67) by PCR. Target DNA was detected in samples collected between four days post-inoculation (dpi) to 66 dpi by both qPCR and PCR. Amplification of target DNA was confirmed by Sanger sequencing of PCR products, which revealed significant homology to the target sequence. These results confirm, for the first time, the ability to consistently detect *Trypanosoma* DNA from the faeces of infected cattle. This opens up the potential to use faeces as an easily accessible sample to screen for active AAT infection in cattle and potentially wild mammalian hosts. Future research should be directed at this novel diagnostic approach as a potential tool to improve AAT surveillance.

Presenter: **Mr Alexander Bailey**, *PhD Student, Imperial College London*

Poster 90 : Characterisation of *Plasmodium falciparum* oocyst development and transcriptome responses to dynamic nutrient stresses in the mosquito *Anopheles gambiae*

Authors - A Bailey[1]; [2]; D Vlachou[1]; [2]; GK Christophides[1]; [2];
[1] Imperial College London, UK; [2] Imperial College London, UK

Objective - Malaria remains a substantial global health burden. Recent years have seen a significant slowdown in the drop of malaria cases followed by a sharp rise in both cases and deaths in 2020. Inside the mosquito vector, the malaria parasite encounters a significant bottleneck in population size arriving at the sessile oocyst stage, during which many rounds of endomitotic replication leads to a recovery of parasite numbers at the sporozoite stage. The oocyst is therefore an attractive intervention target, but much about its physiology and interaction with the vector remains uncharacterised. Here, we used high throughput RNA-seq to shed light on how *Plasmodium falciparum* oocysts respond to dynamic nutrient stresses in the mosquito *Anopheles gambiae*. Analysis of the parasite and mosquito transcriptomes across time has uncovered a parasite developmental trajectory that is remarkably preserved between dramatically different vector mosquito environments; the main outcome of nutrient abundance or stress in the mosquito is to accelerate or slow oocyst development, respectively. Two main developmental transcription programmes are identified in the oocyst, one directing growth and endomitotic replication, and the other orchestrating sporogony, the process of sporozoite production. These programmes appear to be regulated by an array of AP2-domain transcription factors expressed at different stages of oocyst development. These results shed new light on the gene expression networks directing parasite development at this vulnerable life stage and may guide future efforts to better understand and target malaria parasites in the mosquito.

Presenter: **Dr Artur De Castro Neto**, *Postdoctoral Research Fellow, Federal University of São Paulo*

Poster 91 : Identification of protein content released by the G and Y strains of the extracellular amastigote form of *Trypanosoma cruzi*

Authors - A De Castro Neto[1]; NG Rizzo[1]; PT Florentino[1]; I Almeida[2]; RA Mortara[3];
[1] Federal University of São Paulo, Brazil; [2] Department of Biological Sciences, The University of Texas at El Paso, United States; [3] Federal University of Sao Paulo, Brazil

Objective - *Trypanosoma cruzi* possesses a complex life cycle, with different infective and invasive forms in different hosts. Among these infective forms, extracellular amastigotes (EAs), present in mammalian hosts, have been shown to be able to invade host cells without the need to differentiate back into trypomastigotes. Previous studies showed that these same parasite forms release vesicles and other molecules, in the presence of the host cell, which may be associated with the process of invasion and modulation of infectivity. These events were observed in previous results, in which vesicles and other released molecules from the G strain (highly infective) positively modulated the Y strain (low infective) host cell invasion *in vitro* and vice-versa. Therefore, this study aims to identify molecules released by extracellular vesicles and investigate their importance for the invasion of different strains of *T. cruzi* with different levels of infectivity in host cells. First, extracellular vesicles and proteins free of vesicles released from the G and Y strain were obtained by ultracentrifugation in three fractions, named V2, V16 and vesicle free (VF). The fractions were assessed relative to the vesicles sizes by NanoSight NS300 (Malvern Panalytical), which showed that V2 fractions (obtained after 2 hours of centrifugation) had an average size of 192 nm while V16 (obtained after 16 hours of centrifugation) had an average size of 90 nm in both strains. Following this analysis, the same fractions were assessed relative to their protein content by mass spectrometry. Preliminary gene onthology analysis showed that the V2, V16 and VF of G strain had a higher percentage of proteins related to *T. cruzi* virulence (18%, 12% and 56%, respectively), when compared to the Y strain (7%, 26% and 11%, respectively). The virulence proteins found in both strains comprise trans-sialidases (TS), mucins and mucin-associated surface proteins (MASPS) and gp63 (only present on Y strain). Notably, TS showed to be the most present in G strain with 78% of the total of virulence proteins, while in the Y strain it was identified 41% of TS, which was the same amount of MASPs for that strain. Furthermore, in the next steps of this study, the most frequent protein among the total of TS, MASP and mucins, will be chosen to be deleted through the CRISPR/CAS9 method. The parasite will then be evaluated for their capacity to invade host cells and modulate the invasion of other strains. MicroRNA searches are also being performed to identify other molecules that might also be involved in modulating the host cell during the infection process. The results from these analyzes will contribute for the identification of the main components released by the parasite during the invasion process and will lead to a better understanding of the invasion mechanism of *T. cruzi* extracellular amastigotes.

Presenter: **Mr Edward Nay**, *PhD student, University of York*

Poster 92 : Biophysical and biochemical characterisation of the interaction between *Leishmania braziliensis* PRMT1 and PRMT3

Authors - E Nay[1]; PB Walrad[2]; MJ Plevin[1];
[1] University of York, UK; [2] University of York, Centre for Immunology and Infection, UK

Objective - Arginine methylation is a key post-translational modification that can alter the structure, dynamics and interaction profiles of proteins. Protein arginine methyltransferases (PRMTs) catalyse the transfer of a methyl group from a *S*-adenosylmethionine molecule onto the arginine side chain guanidino group. Mammalian PRMTs are classified into subtypes – PRMT1, 2, 3, 4, 6 and 8 catalyse asymmetric dimethylation (ADMA); PRMT5 and 9 catalyse symmetric dimethylation (SDMA); and PRMT7 catalyses monomethylation (MMA). Kinetoplastids possess five homologues: PRMT1, 3, 5, 6 and 7. *T. brucei* PRMT3 has been shown to be a pro-enzyme (prozyme) which lacks key conserved motifs including in the catalytic double E loop. *T. brucei* PRMT1 is only active in complex with the PRMT3 prozyme. In *Leishmania*, however, PRMT3 retains the conserved double E loop, which raises questions about its role in this organism. Here we use recombinant protein samples to investigate *L. braziliensis* (*Lbr*) PRMT1 and 3 *in vitro*. Activity assays show that methylation of a substrate peptide only occurs when PRMT1 and 3 are both presence. Analytical size exclusion chromatography (SEC) and SEC-MALLS show that *Lbr*PRMT1 and 3 form a heterotetrameric complex in solution. Mutation of double E loop residues revealed that *Lbr*PRMT1 is the active component of the complex. Previous work suggested *Lbr*PRMT3 could interact with and modulate the activity of other *Lbr*PRMTs, however methyltransferase assays showed that *Lbr*PRMT3 had no effect on the activities of *Lbr*PRMT5 and 7 *in vitro*. Moreover, *Lbr*PRMT3 could not methylate a peptide substrate previously monomethylated with PRMT7. Our data suggests that *Lbr*PRMT1 and 3 form a similar complex to *T. brucei* PRMT1-3. However, the retention of the conserved double E loop in *Leishmania* PRMT3 enzymes suggests an as of yet undiscovered functional difference between the two trypanosmatids.

Presenter: **Mr John Ogunsola**, *PhD student, Biodiversity, Animal Health & Comparative Medicine*

Poster 93* : RNA-Seq reveals distinct renal responses to murine trypanosomiasis in susceptible and tolerant mice

Authors - J Ogunsola[3]; JF Quintana[2]; A CooperA MacLeod[2];
[1] University of Glasgow , UK; [2] Wellcome Centre for Integrative Parasitology (WCIP), UK; [3] University of Glasgow, UK, UK

Objective - Host factors play a key role in the outcome of infection with African trypanosomes. Despite the well-documented anaemia characteristic of trypanosome infection, little is known about the response of the kidney; an organ that is important in the response to dwindling oxygen levels and blood volumes. Using BALB/C (susceptible) and C57BL/6 (tolerant) mice infected with T. *brucei*, we profiled the transcriptional responses and tissue architecture of the mouse kidney at early (acute) and late (chronic) time points of infection. At a tissue level, early infection is characterised by tubular necrosis accompanied by mononuclear cell infiltration and vascular congestion in both strains. As the infection progresses, BALB/c mice displayed a worsening tubular necrosis whereas C57BL/6 mice showed signs of reparative tubular regeneration, correlating with a reduced parasite burden in these mice. Bulk transcriptomics and gene ontology analysis of the mouse kidney detected both mouse strain- and time-dependent transcriptional responses upon infection. Common immune signatures were upregulated in both strains at early time points, (TNF, B cell receptor, and C-type lectin signalling pathways), with IL-17 signalling a significant feature in susceptible BALB/C, coinciding with the onset of an inflammatory response. The transcriptome of C57BL/6 was dominated by genes associated with cell cycle, DNA replication and JAK-STAT signalling pathways. During the chronic stage of the infection, gene pathways associated with complement and coagulation cascades were preferentially upregulated in BALB/C. Taken together, trypanosome infection induced similar lesions and responses in the kidneys of both strains early in the infection. However, reparative, and proliferative mechanisms were uniquely upregulated in tolerant C57BL/6 but not susceptible BALB/C, coinciding with the histological findings. The increased susceptibility of BALB/C may be related to increased parasite burden, a reduced ability to repair damaged organs or a combination of both features.

Presenter: **Ms Gabriella Torres**, *Estudante , Instituto René Rachou/ Fiocruz Minas*

Poster 94* : The protein kinases SmFES and SmRAF may influence *Schistosoma mansoni* development, egg maturation, and hepatic granulomas progression in the mammalian host

Authors - GP Torres[1]; NC Tavares[1]; SG Gava[1]; MM Mourao[1];
[1] Instituto René Rachou - Fiocruz Minas, Brazil

Objective - Schistosomiasis has a high rate of morbidity induced, mainly, by the granulomas' formation by eggs deposited in mammalian host tissues, being correlated with the viability of mature eggs. Schistosomiasis treatment relies only on praziquantel administration. However, due to praziquantel limitations, functional studies to elucidate parasite's biology are needed to find alternative therapies. Studies suggest that protein kinases (PKs) have an essential role in *Schistosoma mansoni* development and survival. The PK SmFES is important in signal transduction pathways involved in larval transformation after penetration into hosts. SmRAF can influence the development and reproduction of *S. mansoni*. Therefore, this study aimed to investigate the functions of SmFES and SmRAF PKs in *S. mansoni* development and during mammalian infection establishment, using knockdown by RNA interference. Thus, mice were infected with schistosomula knocked-down for Sm*fes* and Sm*raf*. The number of eggs in the mice intestine was quantified and their maturation was evaluated. The square lobe of the liver was separated and stained for histological slides. The granulomas present on the slides were photographed under an inverted microscope, the area was measured using the *Image J* software and the granulomas were classified. Afterward, the number of granulomas was counted according to the tissue area. The development and reproductive system of adult worms recovered from mice infected with knocked-down schistosomula were analyzed by confocal microscopy. Mice inoculated with Sm*raf*-knocked-down parasites presented a 23% reduction in the egg number recovered from the intestine, whereas the SmFES group showed 57% more immature eggs in this tissue. The liver from mice infected with Sm*fes*-knocked-down parasites presented 166% more granulomatous lesions from the necrotic-exudative evolutionary phase than exudative-productive. Besides, granulomas from the SmFES group showed a 44.4% area reduction. Mice from the SmRAF group showed 6X more hepatic granulomas and 96% more granulomas in necrotic-exudative stage than in exudative-productive phase. On the other hand, in the analysis of recovered adult worm phenotypes under a confocal microscope, it was found that the females of the SmFES group showed a significant decrease in the ovarian area of 29.7% to the area of the negative control group and of 19.3% to GFP. This study reveals that SmFES influences egg maturation and exudate components, which affects granuloma formation. Accordingly, SmFES should be explored as a target to assist schistosomiasis morbidity control. Lastly, SmRAF may participate in egg production and perhaps, consequently, in granulomatous lesions formation, so more studies should be conducted to elucidate its role.

Presenter: **Ms Laurine Brouck**, *PhD student, University of Edinburgh*

Poster 95* : RNA editing ligase 1 as a drug target: on the road to lead generation

Authors - L Brouck[2]; Z Nare[2]; M Sardis[2]; J Smith[1]; M Wear[4]; A Morrison[3]; M Speake[3]; S McElroy[3]; AG Cook[4]; E Gluenz[1]; A Schnaufer[2];
[1] Institute of Infection, Immunity and Inflammation, University of Glasgow, UK; [2] Institute of Immunology & Infection Research, University of Edinburgh, UK; [3] European Screening Centre Newhouse, UK; [4] Institute of Quantitative Biology, Biochemistry and Biotechnology, University of Edinburgh, UK

Objective - RNA editing ligase 1 (REL1) plays a crucial role in uridylyl insertion/deletion mRNA editing, a unique and extensive form of post-transcriptional RNA modification in the mitochondria of *Trypanosoma*tid parasites. Previous gene knockdown and knockout studies showed that REL1 is essential for the survival of *Trypanosoma brucei*, the causative agent of sleeping sickness (Schnaufer *et al.*, 2001; Gao & Simpson, 2003). The crystal structure of *Tb*REL1 revealed a well-defined ATP-binding pocket with striking differences to mammalian DNA and RNA ligases but high conservation among *Trypanosoma*tid REL1 orthologues (Deng *et al.*, 2004). This offers exciting potential for the development of specific REL1 inhibitors with broad anti-*Trypanosoma*tid properties. We developed a high-throughput activity assay for REL1 (Zimmermann *et al.*, 2016) and

tested over 600,000 compounds in four independent screening campaigns. Promising REL1 small molecule inhibitors were identified with an average hit rate of ~1%, and interesting structure-activity relationships emerged for some hit series. Several compounds inhibited REL1 from different *Trypanosoma*tid species, including *Leishmania donovani* and *Trypanosoma cruzi*, but are much less potent against a related bacteriophage RNA ligase, suggesting high specificity *in vitro*. As part of ongoing hit-to-lead development efforts, we are investigating specificity *in vivo*, optimising the expression of recombinant REL1 orthologues with the help of differential scanning fluorimetry, and using crystallography and surface plasmon resonance as tools to study REL1-inhibitor interactions. In addition, we will present results from CRISPR-Cas9-based knockout studies which suggest that REL1 is essential in *Leishmania mexicana*. This confirms the expectation that RNA editing is a promising drug target beyond African trypanosomes. References:Schnaufer,A., Panigrahi, A. K., Panicucci, B., Igo, R. P., Wirtz, E., Salavati, R., Stuart, K. (2001). An RNA ligase essential for RNA editing and survival of the bloodstream form of *Trypanosoma brucei*, *Science*, 291:2159-62. Gao, G. and Simpson, L. (2003). Is the *Trypanosoma brucei* REL1 RNA ligase specific for U-deletion RNA editing, and is the REL2 RNA ligase specific for U-insertion editing?, *J Biol Chem*, 278:27570-4.Deng, J., Schnaufer, A., Salavati, R., Stuart, K., Hol, W. G. (2004). High resolution crystal structure of a key editosome enzyme from *Trypanosoma brucei*: RNA editing ligase 1, *J Mol Biol*, 343:601-13.*Trypanosoma brucei editosome enzyme REL1 and other RNA ligases, Nucleic Acids Res, 44:e24.*

Presenter: **Mr Jacob Leonard**, Student, Aberystwyth University

Poster 96 : Rumen Fluke-Microbiome Interactions; an Exploration into Understanding the Extracellular Vesicles of *Calicophoron daubneyi*

Authors - JL Leonard[2]; RM Morphew[2]; MF Fisher[4]; CC Cantacessi[5]; SA Huws[1]; PM Brophy[3];
[1] Queen's University Belfast, UK; [2] Aberystwyth University, UK; [3] Aberystwyth University - IBERS, UK; [4] Ridgeway Research Ltd, UK; [5] Department of Veterinary Medicine, University of Cambridge, UK

Objective - Recent work has demonstrated a substantial contribution of parasite-mediated changes in the ruminant gut microbiota following investigation into the rumen fluke, Calicophoron daubneyi, within an in vitro rumen model. Extracellular vesicles (EV) were identified as vital in shaping bacterial communities within the host rumen, yet the direct effects are not fully understood. Utilising purified C. daubneyi EVs via size exclusion chromatography (SEC), our initial focus was to identify the antimicrobial effects on the microbiome itself. EVs were initially purified from excretory secretory products using SEC as previously demonstrated. SEC purified EVs were confirmed using a global proteomics approach revealing common markers associated with EVs. Confirmed *C. daubneyi* derived EVs were then incorporated into bacterial cell lysis and bacterial optical density assays to assess the inhibition of bacterial growth against strains of Escherichia coli, Pseudomonas aeruginosa and Streptococcus aureus. Following antimicrobial assessment, the specific release of C. daubneyi EVs within an in vitro fermentation model, simulating the rumen environment, will be assessed to replicate the in vivo scenario. Adult C. daubneyi, will be maintained to release EVs into the in vitro rumen milieu. Following culture, all EVs, including fluke and microbiome derived, will be purified again using and SEC approach. In order to identify C. daubneyi specific EVs, fluke specific antibodies, such as Anti-FhGST-S1 known to bind to the surface of fluke EVs, will be used to identify EVs secreted into the in vitro rumen simulation through transmission electron microscopy analysis. In addition, a metaproteomic approach will be used to confirm C. daubneyi EV release using a GeLC approach. The final aim of this research is to utilise metataxonomy and metaproteomics, as key tools, to identify the mechanisms and the effects of EVs in the rumen in experimental infections. Once these interactions are understood and characterised novel approaches to control involving the interaction with the ruminant microbiome may be investigated.

Presenter: **Dr Federica Giordani**, Research Associate, University of Glasgow

Poster 97 : Analysis of the resistance profile of benzoxaborole AN11736 analogues in animal trypanosomes

Authors - F GiordaniD PaapeG HamiltonJM WilkesK CrouchR RitchieLJ Morrison[1]; H AutyMP Barrett
[1] Roslin Institute, UK; [2] University of Glasgow , UK

Objective - Benzoxaboroles are a class of bioactive compounds with a broad antimicrobial range. Their high activity against trypanosomes has led to the development of acoziborole, set to become the first single-dose oral treatment for human African trypanosomiasis.

Some compounds belonging to this class are also under evaluation for the treatment of animal African trypanosomosis, or nagana. This livestock disease remains a major cause of poverty and rural underdevelopment in African areas where it is endemic, causing significant economic losses to small-farm holders.

As a novel compound class, unrelated to current veterinary trypanocides, the risk of cross-resistance between benzoxaboroles and commercial trypanocides is low. Selection of resistance to an initial veterinary lead, AN11736, was obtained in vitro. We discovered that trypanosomes become resistant by losing the activity of serine carboxypeptidases (CBPs), which in wild type cells cleave the parental prodrug to a carboxylate derivative that accumulates at high levels.

We then assessed *Trypanosoma brucei* and T. *congolense* resistance and cross-resistance to a series of three benzoxaboroles, all analogues of AN11736. We found that the deletion of the CBP genes was a common feature in the development of resistance in vitro. However, one analogue did not share this profile in T. *congolense*, suggesting that other molecular mechanisms must be at play for development of resistance to this compound.

Presenter: **Ms Lauren Wilburn**, PhD student , Oxford Brookes University

Poster 98* : IFT train numbers drop rapidly after initiation from *Leishmania* promastigotes to amastigotes

Authors - L Wilburn[2]; JD Sunter[1];
[1] Department of Biological and Medical Sciences, Oxford Brookes University, Oxford, UK; [2] Oxford Brookes University, UK

Objective - During their life cycle, Leishmania parasites dramatically change their morphology in response to the different environments they encounter. This is exemplified by the flagellum, which is long and motile in the promastigote form in the insect but extremely short in the amastigote form found inside the macrophage. Flagellum assembly occurs at its distal tip with material transported there by intraflagellar transport (IFT) trains. These trains are composed of multiple components, which form two main complexes – IFTA and IFTB that are associated with retrograde and anterograde transport respectively. The IFT system is required to assemble the long promastigote flagellum yet its role in flagellum disassembly is unclear.

We fluorescently tagged different IFT proteins with mNeonGreen and examined their movement during the differentiation from promastigotes to amastigotes by microscopy. We found that the number of IFT trains per micron dropped by nearly 50% within the first two hours of the initiation of differentiation and then remained stable for the rest of the time course. The onset of differentiation also resulted in a slight decrease in the speed of the trains in both directions, with an increase in the number of slow trains. Overall, this indicates that flagellum disassembly likely occurs due to a decrease in delivery of components by the IFT system coupled to the natural turnover rate at the tip.

Presenter: **Ms Lauren Wilburn**, PhD student , Oxford Brookes University

Poster 99 : IFT train numbers drop rapidly after initiation from *Leishmania* promastigotes to amastigotes

Authors - L Wilburn
[1] Oxford Brookes University, UK

Presenter: **Dr Joao Cunha**, Research Associate, University of York

Poster 100 : Estimating complex and multiclonal infections in Leishmania infected patients and reservoirs

Authors - JL Reis-Cunha[1]; CA Grace[1]; KS Carvalho[2]; CN Costa[2]; DC Jeffares[1];
[1] University of York, UK; [2] Federal University of Piauí, Teresina, Brazil

Objective - Leishmaniasis a complex disease, comprising several parasite species, hosts, reservoirs, vectors and clinical symptoms. The clinical manifestations vary from mild cutaneous lesions to severe visceral damage, and genetic polymorphisms from both the host and the parasite have been associated with disease severity. Hybrids and genetic exchange between species and strains have already been reported for numerous parasite groups, including the L. *donovani* complex, where several species can cause visceral leishmaniasis. This has relevant implications in disease epidemiology, as it can fasten the spread of virulence and drug resistance genes. The presence of multiclonal infections, required for hybridization, could also aid in parasite adaptation to different hosts and stress conditions, as different sub-populations of the parasite could be selected in different scenarios. The presence of multiclonal infections have already been reported in some L *donovani* isolates from the Indian subcontinent and Africa using WGS and in L. infantum dogs in Brazil using multilocus microsatellite typing. However, the extent of multiclonal infection across several geographic locations, parasite species and hosts have not been yet estimated. In the present work, we are exploring fluctuations in allele frequency of heterozygous SNPs positions in genome sequencing of Leishmania isolates as a measure of multiclonal infection. The main premise is that while in clonal infections heterozygous SNPs are expected to have similar read depths in both alleles, complex multiclonal infections will disturb this proportion. We are correcting SNP calls for several confounding factors as chromosomal somy, mapping quality, call quality and read depth variations. As different Leishmania species/populations have different levels of heterozygosity and were sequenced in different depths, clonal simulated isolates with the same characteristics as each evaluated population were generated and used as a control. Preliminary results using ~450 whole genome sequencing of L. infantum and L. *donovani* isolates, from dogs and humans from Africa, Asia and Brazil have shown that a significant proportion of isolates from all sites and hosts appears to be multiclonal. We are planning on expanding this analysis to species from the L. viannia subspecies, to also evaluate multiclonal infections in cutaneous leishmaniasis. Finally, the proposed analysis will be packed in a framework, which could easily be adapted to other organisms.

Presenter: **Miss Madeleine Oakland**, Student, Aberystwyth University

Poster 101 : Discovery Transcriptomics to Support Future Control of *Nematodirus helvetianus*

Authors - ME Oakland[1]; M Hegarty[1]; PM Brophy[1]; E Scott-Baird[2]; RM Morphew[1];
[1] Aberystwyth University, UK; [2] Drayton Animal Health, UK

Objective - Gastrointestinal (GI) nematodes are a worldwide threat to sustainable farming and animal production. Specifically, the GI nematode Nematodirus helvetianus is a parasite of veterinary importance with cattle infected following the ingestion of infective L3 larvae found in pasture. The life cycle of N. helvetianus is direct. The larvae develop into mature adult worms in the small intestine excreting eggs in the faeces into the environment. Calves with naïve immunity are susceptible, and infection results in scouring, dehydration, and malnutrition. Thus, infection negatively impacts animal welfare and economic production. Furthermore, age-related resistance has been observed yet coinfection with other GI nematode populations can have detrimental effects. N. helvetianus is particularly problematic for pasture-based herds, as the larvae moults occur within the ensheathed eggs. This makes them resistant to harsh climates and desiccation, leading to a mass hatch and an accumulation of infectious L3 larvae in the spring. In addition, as the larvae and eggs can survive on the pasture for months to years, this renders control difficult for pasture-based herds. Currently, anthelmintics and alternate grazing strategies are used in control. However, the increasing challenge of resistance due to the misuse of anthelmintics, and the pressure of climate change, there is a need for novel control strategies. Recently, the use of omic technologies, such as genomics, transcriptomics and proteomics, in the control of related GI helminths offers hope for the alternative control of Nematodirus. At present, knowledge and thus omic resources focused on N. helvetianus are extremely limited; emphasised with the lack of an available genome or transcriptome datasets. Therefore, this project aims to generate the first discovery transcriptome to support future functional genomics in this important GI nematode. Samples of L3 larvae N. helvetianus were purified using a Baermann funnel and snap frozen prior to tRNA extraction. tRNA extractions were completed using Trizol and mechanical bead beating followed by purification using the Zymo spin column methodology. Transcriptome sequencing is currently underway. In addition, N. helvetianus are currently undergoing protein extraction to perform preliminary proteomic screening. The whole proteome is to be subjected to global proteomic analysis using a GeLC approach to provide the first evidence of the N. helvetianus proteome. Finally, of particular interest are the glutathione transferases (GSTs). GSTs have key roles involved in immunomodulation, detoxification, and critical house-keeping roles in cell signalling. These integral roles in helminths make them ideal targets in future control strategies.Initial investigations will focus on a transcriptome-based bioinformatics approach to reveal potential GST members produced by N. h

Presenter: **Dr Owain Donnelly**, Clinical Research Fellow, Francis Crick Institute

Poster 102 : Further optimisation of the *Schistosoma haematobium* Recombinase Polymerase Amplification assay: moving towards point of care use in endemic settings

Authors - O Donnelly[1]; Z Bartonicek[1]; E Lugli[1]; S Mesquita[1]; B Webster[1];
[1] Natural History Museum, UK

Objective – Background - Schistosomiasis is a neglected tropical disease (NTD) affecting millions of people globally, estimated to cost 1.6 million disability-adjusted life years (DALYs) with its combined morbidity and mortality (1). *Schistosoma haematobium* causes urogenital schistosomiasis, and improvements to existing diagnostics are essential to move beyond disease morbidity control and towards the goal of elimination as a public health problem (2), a WHO NTD roadmap priority (3). Recombinase Polymerase Amplification (RPA) offers significant promise as a sensitive, specific, and portable point-of-need diagnostic for S. *haematobium* (4), but needs further optimisation before clinical use in the field.

Methods - S. *haematobium* RPA (Sh-RPA) was performed on synthetic DNA standards, *Schistosoma* adult worm and egg genomic DNA, and clinical urine samples containing S. *haematobium* eggs. Key aspects of the reaction, such as betaine content (which reduces secondary structure formation) and sample volume, were altered to ascertain conditions for maximal sensitivity, specificity, and reaction efficiency. Advanced primer/probe combinations were designed and tested, with a view to improving analytical specificity. Additionally, four simple and field applicable sample preparation kits were compared for use in Sh-RPA by extracting DNA from a single S. *haematobium* egg, to fill a critical gap in knowledge for the potential use of DNA-based diagnostics in low resource settings.

Results - Overall, Sh-RPA performed best in reactions containing 0.5µL of betaine and with the addition of the maximal sample volume possible, which did not hinder assay performance. The advanced primer and probe design proved robust, with 100% analytical specificity, obviating the need for betaine and thereby simplifying reaction set up. The limits of detection were $1x10^2$ copies of synthetic Dra1 DNA and 1pg of genomic DNA. Two simple and rapid DNA extraction methods proved optimal for the preparation of DNA from single S. *haematobium* eggs, resulting in 100% sensitivity and specificity with Sh-RPA. These required only 1-2 steps using simple lysis or magnetic bead methodology, without the need for lab-based equipment. Of note, addition of S. *haematobium* eggs directly to the reaction yielded strongly positive results, suggesting promise for direct addition of unextracted urine in the field, although this requires evaluation with clinical samples.

Conclusions - Previous studies have shown that the Sh-RPA is a promising diagnostic for field use, but further optimisation was needed. Here, Sh-RPA's optimal analytical specificity has been achieved, using a simpler assay with no loss of sensitivity. Additionally, sample preparation methods with minimal equipment/resources have proven suitable for the Sh-RPA assay, with a robust lower limit of detection of a single S. *haematobium* egg. This research has greatly advanced the Sh-RPA towards field use, with clinical sample testing the next vital step.

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3.World Health Organization. Ending the neglect to attain the Sustainable Development Goals: A road map for neglected tropical diseases 2021–2030. WHO; 2020.

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Presenter: **Miss Michelle Clark**, PhD Candidate, The Walter and Elisa Hall Institute

Poster 103* : Unleishing' cell death: Cellular inhibitors of apoptosis are an effective therapeutic target for the treatment of *Leishmania donovani*

Authors - **MP Clark**[1]; L Mackiewicz[1]; M Doerflinger[1]; M Pellegrini[1];
[1] The Walter and Elisa Hall Institute, Australia

Objective - Leishmaniasis, a disease caused by the Leishmania spp. parasite, affects approximately 1 million people annually worldwide. Specific Leishmania species such as Leishmania *donovani* cause chronic spleen, liver and bone marrow infection which if left untreated leads to a fatal visceral infection resulting in 20,000-30,000 deaths globally each year. The rise of resistance among many of the current therapeutics for visceral leishmaniasis and the severe toxicity profiles of these anti-leishmanials suggest there is a desperate need for new, safer therapeutics for the treatment of leishmaniasis. Re-purposing already established therapeutics that target the host are an attractive target as they can be more rapidly deployed clinically than completely novel compounds and are less likely to elicit a selective pressure for resistance in the parasite as successful evasion would require considerable mutational changes.

As intracellular pathogens such as Leishmania spp. need to manipulate host cell death signaling pathways to survive, replicate and disseminate, we hypothesise that therapeutics originally designed for the treatment of various cancers that target these cell death pathways may provide effective new treatments for visceral leishmaniasis.

We used L. *donovani* in vitro and in vivo infection models along with microscopy, live-cell imaging, ELISAs, flow cytometry and immunohistochemistry to determine the effectiveness of therapeutic compounds targeting host cell apoptotic machinery for the treatment of visceral leishmaniasis.

Our results explored the impact of therapeutically inducing extrinsic apoptosis in Leishmania *donovani* infection. We showed that tumour necrosis factor (TNF), the instigator of extrinsic apoptosis, is upregulated in Leishmania *donovani* infection, however only upon the addition of inhibitor of apoptosis proteins (IAPs) inhibitors does cell death of infected cells occur. Infected mice treated with IAP inhibitors showed a decrease in parasite burden as well as a corresponding decrease in hepatosplenomegaly. More importantly, there is an additive effect when this therapeutic is combined with anti-leishmanial amphotericin-B, indicating IAP inhibitors could be used to enhance amphotericin-B therapy or enable the reduction of standard amphotericin-B dosage to decrease toxicity.

Collectively, these data showed that targeting host extrinsic apoptotic pathways using clinical stage IAP inhibitors may be a valid therapeutic option for the treatment of visceral leishmaniasis.

Presenter: **Mr Brian Suarez Mantilla**, Research Fellow, University of Durham

Poster 104 : Analysis of Nucleobases, Nucleotides and cyclic Nucleotides in *Leishmania*

Authors - **BA Suarez Mantilla**[1]; RE Dack[1]; DR Hodgson[2]; PW Denny[1];
[1] Department of Biosciences, University of Durham, DH1 3LE, UK; [2] Department of Chemistry, University of Durham, DH1 3LE, UK

Objective - Nucleotides are core macromolecules of bioenergetics, redox processes, nucleic acids metabolism, and cell signalling. Kinetoplastid parasites are reliant on salvage pathways to fulfil their requirements of purine and pyrimidine derived nucleotides. These protozoa have also evolved to uptake nucleobases and nucleosides from the extracellular milieu at their distinct host environments. Nucleotide biosynthesis requires a nucleobase linked to a pentose sugar (nucleoside) that can thus be mono, di or triphosphorylated (nucleotides) harbouring high-energy phosphoanhydride bonds. This latter can also give rise to cyclic nucleotides such as cAMP and the magic spot nucleotide pppGpp (guanosine pentaphosphate). Here, we used an HPLC-based method enabling the identification of the most abundant nucleobases, purine and pyrimidine nucleotides, and cAMP in promastigote forms of Leishmania *mexicana*. A pppGpp synthase identified in the free-living protist Bodo saltans was expressed in L. *mexicana* and its metabolic product was also detected using this method. The role of inositol pyrophosphate (5-InsP_7) as a key metabolite controlling the pool of purine nucleotides in these organisms will also be discussed. This method can be useful to analyse the nucleotide response to pharmacological or genetic perturbations in different cell systems.

Presenter: **Prof Russell Stothard**, Medical Parasitologist, Biomedical Parasitology Division

Poster 105 : Back in action for fieldwork: Update on schistosomiasis research and control in Malawi

Authors - R Stothard[3]; S Kayuni[1]; M Al-Harbi[3]; A Juhasz[1]; L Cunningham[1]; S Jones[1]; J Archer[1]; P Makaula[4]; EJ LaCourse[2]; J Musaya[5];
[1] Liverpool School of Tropical Medicine, UK; [2] Liverpool School of Tropical Medicine / UoL, UK; [3] LSTM, UK; [4] Research in Health, Environment and Development (RHED), Malawi; [5] MLW, UK

Objective - In April 2021, a new 4-year investigation officially started exploring the importance of hybrid schistosomes in Malawi. The Wellcome Trust funded study entitled "HUGS: Hybridisation in urogenital schistosomiasis" is a collaboration between LSTM and MLW/CoM. Owing to COVID, initial fieldwork planned for July was postponed but an appropriate window of opportunity later opened in October. This was also coordinated around newly re-started mass drug administration campaigns within the country. In this presentation, I briefly review previous studies on male genital schistosomiasis and medical malacology along Lake Malawi, alongside newly updated WHO guidelines for schistosomiasis control. I present preliminary findings of ongoing HUGS work ,along the lake's shoreline and that of the Lower Shire River, to highlight the growing importance of hybrid schistosomes within transmission of urogenital (and intestinal) schistosomiasis in Central Africa.

Presenter: **Dr Joana Correia Faria**, Sir Henry Dale Fellow, Department of Biology, University of York

Poster 106 : Multi-allelic exclusion by an allele-selective RNA-DNA helicase in African trypanosomes

Authors - J Correia Faria[1]; H Hashimoto[2]; M Tinti[1]; CA Marques[1]; E Debler[2]; D Horn[1];
[1] Wellcome Trust Centre for Anti-Infectives Research, School of Life Sciences, University of Dundee, UK; [2] Thomas Jefferson University, Philadelphia, United States

Objective - Allelic-exclusion mechanisms, including those that underpin antigenic variation in parasites and olfaction in mammals, are not fully understood, remaining one of the greatest outstanding mysteries in eukaryotic biology. In African trypanosomes, immune evasion involves expression of a single Variant-Surface-Glycoprotein (VSG) gene in a dedicated sub-nuclear expression factory. VSG-exclusion-1 (VEX1) and VEX2 are concentrated at the splicing and transcription compartments, respectively (PMID: 33432154).

Here, using ChIP-Seq, we show that VEX2 is associated with the active-VSG expression-site, forming an allele-specific bridge, via VEX1, to a trans-splicing locus on another chromosome. Further, single cell RNA-Seq analysis following VEX2 depletion revealed the simultaneous expression of several VSGs and showed: 1) the number of simultaneously active VSGs that can be tolerated; 2) a hierarchy of VSG transcriptional derepression.

Moreover, VEX2 is a large protein that forms a native multimeric complex of approximately 1 MDa. In vitro data using a recombinant helicase core showed that it is an RNA-DNA helicase. To assess whether this helicase activity was required for VSG-monogenic-expression, we established a FACS-based CRISPR-Cas9-mediated saturation mutagenesis assay combined with amplicon-Seq profiling. Replacement of a critical amino acid in the helicase core by any other amino acid disrupted allelic exclusion. This phenotype was only rescued by synonymous mutations.

Finally, we found that VEX1 and VEX2 interact via their N- and C-termini, respectively and reciprocal turnover control limits the abundance and maintains sequestration of the complex.

This work begins to reveal the mechanism by which the VEX complex sustains VSG-monogenic-expression in African trypanosomes.

Presenter: **Mr Juan Vicente Hernández Villena**, Research assistant , IZET, Universidad Central de Venezuela

Poster 107 : Venezuela´s health crisis: the resurgence of malaria, Leishmaniasis, and Chagas disease.

Authors - J Hernández Villena[2]; MS Llewellyn[1]; ME Grillet[2];
[1] Institute of Biodiversity, Animal Health and comparative Medicine, University of Glasgow, UK; [2] Instituto de Zoología y Ecología Tropical, Venezuela

Objective - Over the last twenty years, Venezuela´s public health quality has been declining due to political and socioeconomic factors, causing an ongoing humanitarian crisis. With a decaying healthcare infrastructure, a mass departure of trained medical personnel, and the decline of all public health programs, including disease surveillance and reporting, Venezuela is experiencing a surge and expansion of vector-borne diseases. Once recognized as a regional leader for public health and vector-control policies and programming, the country is facing a significant increase in incidence of malaria, Chagas, and leishmaniasis, among others.

In fifteen years (2000-2015) Venezuela reported a 365% increase in malaria cases and it has contributed with 53% (2017) and 51% (2018) of the almost 1 million per year of reported cases in the Latin American region. In 2017 *Plasmodium vivax* accounted for the majority of reported cases (76%) followed by *Plasmodium falciparum* (17.7%), mixed P. *vivax*/P. *falciparum* infections (6%) and *Plasmodium* malariae (<1%).

Recent focal seroprevalence estimates for Chagas disease are 15.7% compared with Colombia's estimates (0.2% in Santander in 2013-2014) indicating resurgence. Due to consumption of food and beverages contaminated with infected triatomines or feces, oral Chagas disease transmission has also become an issue of great concern with 16 outbreaks (321 cases) reported nationwide between 2007-2018, half of them occurred in peri-urban and urban areas.

Finally, cutaneous and mucocutaneous leishmaniasis (CL-MCL) is dispersed throughout the country and visceral leishmaniasis (LV) in three foci. In spite of a report of almost 61600 CL cases between 1990-2016 and the substantial expansion of its endemic areas, nothing in the available data suggests the clinical cases have been a consequence of the crisis. However, migratory trends might be contributing to the spread of the disease.

Overview of the Poster Programme by Session

ID	Track	No.	Delivery	Title	Presenter First Name	Presenter Last Name	Institution
	Tuesday						
P26201	BES: Parasite Ecology: Coinfections	Poster 69	Inperson	Co- infection of parasites and fungal infection and COVID- 19	Mohammad Hossein	Feiz Haddad	Ahvaz Jundishapur University of Medical Sciences
P26025	BES: Parasite Ecology: Coinfections	Poster 81	Inperson	Efficacy Of Combined Therapy Of Diminaveto® And Intromidium® In Albino Rats Experimentally	Okpala	Michael	University of Nigeria, Nsukka
P26150	BES: Wild Parasitology: into the field	Poster 75	Inperson	A preliminary survey of freshwater snails in Ugbawka, Nkanu East Local Government Area (LGA) of Enugu State, Nigeria.	Kehinde	Sowunmi	Enugu State University of Science and Technology
P26113	BES: Wild Parasitology: into the field	Poster 71	Inperson	Association between Schistosoma mansoni infection intensity, praziquantel side effects, and drug efficacy, in Ugandan school-aged children	Huanghehui	Yu	University of Glasgow
P25682	BES: Wild Parasitology: into the field	Poster 24	Inperson	Improving diagnostics for Schistosoma bovis infections in cattle across Africa.	Thomas	Gasan	Queens University Belfast
P26015	BES: Wild Parasitology: into the field	Poster 83	Inperson	Intestinal helminth parasites of grasscutters (Thryonomys swinderianus) sold at Omagwa Bushmeat Market, Omagwa, Rivers State, Nigeria	Chidimma	Amuzie	Rivers State University
P26021	BES: Wild Parasitology: into the field	Poster 88	Inperson	Prevalence and molecular identification of trypanosomes in dogs in Enugu North Senatorial Zone, South East Nigeria	Chukwunonso	Obi	University of Nigeria, Nsukka
P25995	BES: Wild Parasitology: into the field	Poster 14		Some species of the family Microcotylidae (Polyopisthocotylea, Monogenea) from Sparid fishes off the Algerian coast	Affaf	BOUKADOUM	University of Sciences and Technology Houari Boumdienne
P25539	BES: Wild Parasitology: into the field	Poster 11	Inperson	Wild foci of the Chagas disease vectors Triatoma infestans and Mepraia spinolai in Chile, a country that has declared the interruption of Trypanosoma cruzi vectorial transmission.	Antonella	Bacigalupo	University of Glasgow, BAHCM
P25938	BES: Wild Parasitology: into the field	Poster 2		Diversity of monogenea (Platyhelminths) gill parasites of Scombrid fishes off the Algerian coast	Zouhour El Mouna	Ayadi	University of Sciences and Technologie Houari Boum
P25728	BES: Wild Parasitology: into the field	Poster 26	**Remote**	Fascioliasis in captive vicuñas at Knowsley Safari Park in UK	Alexandra	Juhasz	LSTM
P25719	BES: Wild Parasitology: into the field	Poster 25	**Remote**	First insights into veterinary and zoonotic schistosomiasis in Malawi	Alexandra	Juhasz	LSTM
P25911	BES: Wild Parasitology: into the field	Poster 27	Inperson	Hybridisation in UroGenital Schistosomiasis (HUGS): Pilot study findings upon parasitological surveys in Mangochi and Nsanje Districts, Malawi	John	Archer	Liverpool School of Tropical Medicine
P25659	BES: Wild Parasitology: into the field	Poster 12	Inperson	Prevalence and Associated Risk Factors of Eimeria Species of Chicken in Lagos, Southwest, Nigeria.	Oluwayomi	Adeyemi	University of Lagos
P26137	Combative Strategies: Vaccines	Poster 66	Inperson	Sequence conservation of Cryptosporidium invasion proteins which have potential as vaccine candidates against cryptosporidiosis in cattle and sheep	Abigail	Webb	Aberystwyth University
	Combative Strategies: Vaccines		Inperson		Adam	Roberts	York
P25550	Molecular Genetics	Poster 15	Inperson	An investigation into Leishmania genome plasticity in response to disruption of sphingolipid biosynthesis	Yasmine	Kumordzi	Durham University
P25986	Molecular Genetics	Poster 89	Inperson	Detection of Trypanosoma brucei DNA in faeces of experimentally-infected cattle	Isabel	Saldanha	Liverpool School of Tropical Medicine
P26086	Molecular Genetics	Poster 101	Inperson	Discovery Transcriptomics to Support Future Control of Nematodirus helvetianus	Madeleine	Oakland	Aberystwyth University
P25764	Molecular Genetics	Poster 23	Inperson	Investigating the genetic diversity of Schistosoma bovis and its hybrids across Africa	Shannan	Summers	London School of Hygiene and Tropical Medicine
P26067	Molecular Genetics	Poster 74	Inperson	Molecular characterisation of medically important freshwater snails in Saudi Arabia	Mohammad	Alharbi	Liverpool school of tropical medicine

ID	Track	No.	Delivery	Title	Presenter First Name	Presenter Last Name	Institution
	Tuesday						
P26138	Molecular Genetics	Poster 68	Inperson	piRNA-like small RNAs target transposable elements in a Clade IV parasitic nematode.	Mona	Suleiman	University of Bath
P25918	Molecular Genetics	Poster 30	Inperson	What's that twinkle in your eye? Mischief or a stellate opportunist?	Richard	Childs Hunt	London School of hygiene and tropical medicine
P25670	Cell Biology	Poster 10	Inperson	Characterisation of a cation diffusion facilitator from the malaria parasite Plasmodium falciparum	Benedict	Davies	St George's, University of London
P26149	Parasite Cell Biology	Poster 54	**Remote**	Apical annuli are essential exocytic sites in Toxoplasma	Sara	Chelaghma	Univeristy of Cambridge
P26166	Parasite Cell Biology	Poster 52	Inperson	Barriers to growth: disrupting development in juvenile liver fluke by targeting fibroblast growth factor receptors	Paul	McCusker	Queen's University Belfast
A26115	Parasite Cell Biology	Oral/Poster 57	Inperson	Characterising Heat Shock in Trypanosoma congolense	Marianne	Aelmans	Lancaster University
A25984	Parasite Cell Biology	Oral/Poster 9	Inperson	Deletion of the P21 gene triggers changes in the invasion and replication of Trypanosoma cruzi	Thaise	Teixeira	Universidade Federal de São Paulo
A26105	Parasite Cell Biology	Oral/Poster 6	Inperson	Expression/characterization of mitochondrial fucosyltransferase from T. cruzi; use of monoxenous parasite Crithidia fasciculata as enzymatic source for synthesis of radioactive GDP-Fucose.	Jose Carlos	Paredes-Franco	University of Dundee - School of Life Sciences
A26165	Parasite Cell Biology	Oral/Poster 47	Inperson	First observation of Parasitic viruses in Trichomonas gallinae	Dalal	Ardan	University of East Anglia
P26125	Parasite Cell Biology	Poster 55	Inperson	Function of the MRE11 and EXO1 nucleases and RECQ2 helicase in DNA end resection and double-strand breaks repair in Trypanosoma brucei	RICARDO	OBONAGA GOMEZ	Butantan Institute
A26134	Parasite Cell Biology	Oral/Poster 65	Inperson	Galba truncatula and Helminths, the Importance of Microbes	Peter	McCann	Queen's Univeristy Belfast
P25980	Parasite Cell Biology	Poster 99	Inperson	IFT train numbers drop rapidly after initiation from Leishmania promastigotes to amastigotes	Lauren	Wilburn	Oxford Brookes University
P25965	Parasite Cell Biology	Poster 35	Inperson	Investigating DNA damage responses in Apicomplexan parasites	Monique	Johnson	University of Cambridge
P26065	Parasite Cell Biology	Poster 72	**Remote**	Limits of Flagellar Pocket Access in African Trypanosomes	Antonia	Konle	Cell- and developmental Biology, University Würzbu
P25944	Parasite Cell Biology	Poster 31	Inperson	Proteomics-based investigation of substrates for the Leishmania deubiquitinase DUB2	Sergios	Antoniou	University of York
A26184	Parasite Cell Biology	Oral/Poster 33	Inperson	Role of RDK2 and its interacting protein kinases in Leishmania mexicana differentiation.	Rachel	Neish	University of York
P26028	Parasite Cell Biology	Poster 79	**Remote**	Seroprevelance and Risk Factors of Toxoplasma Gondii Infection Among Healthy Blood Donors in AL-Ribat Teaching Hospital Khartoum State, Sudan.	Safaa Elhassan	Bashir	Self employed
P26141	Parasite Cell Biology	Poster 50		Straightforward assessment of drug efficacy using COLO-680N culture of Cryptosporidium	Georgina	Hurle	University of East Anglia
P26136	Parasite Cell Biology	Poster 80	**Remote**	TFK1, a basal body transition fibre protein that is essential for cytokinesis in Trypanosoma brucei	Denis	Dacheux	CNRS-Bordeaux University-Bordeaux INP
P25543	Parasite Cell Biology	Poster 13	Inperson	The impact of glucose in Trypanosoma cruzi viability, cell growth, cell cycle progression, differentiation, and histone post-translational modifications	Ana	Menezes	Butantan Institute
P26039	Evolutionary Genomics	Poster 78	Inperson	Assembly and annotation of Trypanosoma congolense kinetoplast DNA and comparison with T. brucei	Zihao	Chen	University of Edinburgh
P25961	Evolutionary Genomics	Poster 100	Inperson	Estimating complex and multiclonal infections in Leishmania infected patients and reservoirs	Joao	Cunha	University of York
P25967	Trafficking, Signaling	Poster 96	Inperson	Rumen Fluke-Microbiome Interactions; an Exploration into Understanding the Extracellular Vesicles of Calicophoron daubneyi	Jacob	Leonard	Aberystwyth University

ID	Track	No.	Delivery	Title	Presenter First Name	Presenter Last Name	Institution
	Wednesday						
A26116	Biophysics/Structures	Oral/Poster 92	Inperson	Biophysical and biochemical characterisation of the interaction between Leishmania braziliensis PRMT1 and PRMT3	Edward	Nay	University of York
P26002	Biophysics/Structures	Poster 85	Inperson	Investigating a galactokinase orthologue from Leishmania donovani	Hasana	Baber	Keele University
P25282	Drugs	Poster 1	Inperson	Molecular identification of drug-resistance in Trypanosoma evansi of camels in Egypt	Somaia	Abouakkada	Alexandria University
P26175	Combative Strategies: Drug Discovery	Poster 44	Inperson	A dirofilariasis mouse model for heartworm preclinical drug testing	Jessica	Dagley	Liverpool School of Tropical Medicine
P26122	Combative Strategies: Drug Discovery	Poster 97	Inperson	Analysis of the resistance profile of benzoxaborole AN11736 analogues in animal trypanosomes	Federica	Giordani	University of Glasgow
P25540	Combative Strategies: Drug Discovery	Poster 3	Inperson	Biochemical characterization and inhibitor screening of UMP-CMP kinase from malaria parasites	Supannee	Taweechai	University of Leeds
P25541	Combative Strategies: Drug Discovery	Poster 4	Inperson	Deconvoluting the Mode of Action of a Suite of Novel Antileishmanials	Laura	Filipe	Durham University
P26210	Combative Strategies: Drug Discovery	Poster 36	Inperson	Evaluation of Mono and Combined Nitrofurantoin Therapy for Toxoplasmosis in Swiss Albino Mice	Rita	Wassef	Helwan University
P26193	Combative Strategies: Drug Discovery	Poster 49	**Remote**	In cauda venenum, CLK1 inhibitors for Trypanosomiasis	Manuel	Saldivia	Novartis
P26119	Combative Strategies: Drug Discovery	Poster 56	Inperson	In vitro effects and mode of action of phenolic compounds on leishmania donovani	CHRISTINE	Moore	University of Ghana, Legon
P26127	Combative Strategies: Drug Discovery	Poster 76	Inperson	Repurposing trypanocidal drugs to tackle amoebic gill disease in Atlantic Salmon	Yee Wan	Liu	University of Glasgow, IBAHCM
P26200	Combative Strategies: Drug Discovery	Poster 40	**Remote**	Revisiting quinapyramine: mechanism of uptake, action and resistance	Marzuq	Ungogo	University of Glasgow
P26077	Combative Strategies: Drug Discovery	Poster 95	Inperson	RNA editing ligase 1 as a drug target: on the road to lead generation	Laurine	Brouck	University of Edinburgh
P25554	Combative Strategies: Drug Discovery	Poster 18	Inperson	The Fasciola hepatica histone acetylation machinery is developmentally regulated and contains druggable candidates.	Sarah	Davey	IBERS Aberystwyth University
P25676	Gene Expression, Genetic Architecture	Poster 19	**Remote**	Cloning and functional complementation of Schistosoma mansoni cyclic nucleotide phosphodiesterases in Trypanosoma brucei	maha	aloraini	university of Glasgow
P26117	Gene Expression, Genetic Architecture	Poster 73	Inperson	Competitive growth drives a hierarchy of antigenic variation in African trypanosomes.	Douglas	Escrivani de Oliveira	University of Dundee
P26158	Gene Expression, Genetic Architecture	Poster 48	Inperson	Developmental Regulation and Functional Prediction of microRNAs in an Expanded Fasciola hepatica miRNome	Caoimhe	Herron	Queen's University Belfast
P26147	Gene Expression, Genetic Architecture	Poster 59	Inperson	Histones variants and their interaction with the chromatin in Trypanosoma cruzi life forms	Juliana	Nunes Rosón	Butantan Institute
P26164	Gene Expression, Genetic Architecture	Poster 63	Inperson	Investigating the roles of divergent histone tails using gene editing in Trypanosoma brucei	Marketa	Novotna	University of Dundee
P26208	Gene Expression, Genetic Architecture	Poster 38	**Remote**	Schistosoma mansoni phenotypic evaluation after aspartyl proteases cathepsin D-like knockdown	Felipe	Lunkes	Fundação Oswaldo Cruz
P26185	Gene Expression, Genetic Architecture	Poster 42	Inperson	The potential role of long non-coding RNAs in key steps of Leishmania (Viannia) braziliensis life cycle	Caroline	Ricce Espada	University of Sao Paulo
P26100	Gene Expression, Genetic Architecture	Poster 64		WormBase ParaSite - 2022 update	Dionysis	Grigoriadis	WormBase
P26076	Host:Parasite interactions : Tissue Tropism	Poster 70		Blood flukes and arterial damage. A review of aneurysm cases in patients with Schistosomiasis.	Valeria	Silvestri	MUHAS University of Dar es Salaam
P26022	Host:Parasite interactions : Tissue Tropism	Poster 87	Inperson	Comparative pathogenicity of single and mixed drug-resistant Trypanosoma brucei and Trypanosoma congolense infections in rats: Clinico-haematological findings.	Chukwunonso	Obi	University of Nigeria, Nsukka
P25553	Host:Parasite interactions : Tissue Tropism	Poster 16	Inperson	Comparative single cell transcriptomic analysis of the murine CNS in response to T. brucei brucei and T. brucei gambiense infections	Praveena	R G Chandrasegaran	University of Glasgow

ID	Track	No.	Delivery	Title	Presenter First Name	Presenter Last Name	Institution
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P26139	Host:Parasite interactions : Tissue Tropism	Poster 51	Inperson	Control of equine tapeworms through praziquantel: The hidden impact on the equine microbiome	Boontarikaan	Wititkornkul	IBERS, Aberystwyth University
P25681	Host:Parasite interactions : Tissue Tropism	Poster 20	Inperson	Development of a single molecule fluorescent in situ hybridisation (smFISH) pipeline for the detection of host-pathogen interactions in the murine CNS	Rhiannon	Heslop	University of Glasgow
P25945	Host:Parasite interactions : Tissue Tropism	Poster 32	Inperson	Exploring the secretome of Schistocephalus solidus: Extracellular vesicles for host manipulation	Yeng Yi Abbey	Chan	Aberystwyth University
P25999	Host:Parasite interactions : Tissue Tropism	Poster 84		Genetic validation of the function of PfEMP1 in Plasmodium falciparum rosette formation.	Stanley	Otoboh	University of Ediburgh
P26170	Host:Parasite interactions : Tissue Tropism	Poster 45	Inperson	GLOBAL EPIDEMIOLOGY OF TAENIA MULTICEPS: A COMPARATIVE META-ANALYSIS STUDY	El-Sayed	El-Alfy	Mansoura University
P25982	Host:Parasite interactions : Tissue Tropism	Poster 91	Inperson	Identification of protein content released by the G and Y strains of the extracellular amastigote form of Trypanosoma cruzi	Artur	De Castro Neto	Federal University of São Paulo
P25977	Host:Parasite interactions : Tissue Tropism	Poster 93	Inperson	RNA-Seq reveals distinct renal responses to murine trypanosomiasis in susceptible and tolerant mice	John	Ogunsola	Biodiversity, Animal Health & Comparative Medicine
P26103	Mathematical Modeling	Poster 60		Evaluating a novel test-and-treat control strategy for livestock schistosomiasis in sub-Saharan Africa	Adriana	Diaz	The Royal Veterinary College
P26042	Mathematical Modeling	Poster 77	Inperson	Evaluating control and elimination methods of cystic echinococcosis in South America – Beyond the 2030 goals	Mahbod	Entezami	University of Surrey
P25528	Mathematical Modeling	Poster 8	Inperson	Prediction of Miltefosine Exposure in Mouse Model using Physiologically Based Pharmacokinetic Modelling (PBPK) Approach	Shadrack	Madu	De Montfort University
P26104	Mathematical Modeling	Poster 61	Inperson	Relationship between the worm burden and egg output in liver fluke infections of humans	Thomas	Crellen	University of Glasgow
P26026	Parasite ImmunoPathology	Poster 21	Inperson	Antibody responses associated with protection from clinical malaria pathology during P. falciparum infection, in two distinct cohorts in Apac, Uganda and Rourkela, India.	Sophia	DonVito	London School of Hygiene and Tropical Medicine
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P26157	Parasite ImmunoPathology	Poster 53	Inperson	Chronic Trypanosoma cruzi infection in mice lacking B cells (muMT)	Jose	Mengel	Oswaldo Cruz Foundation
P26024	Parasite ImmunoPathology	Poster 82	Inperson	Comparative antibody response of albino rats infected with Trypanosoma brucei brucei via intraperitoneal and intradermal routes	Okpala	Michael	University of Nigeria, Nsukka
P26120	Parasite ImmunoPathology	Poster 62	Inperson	The impact of chronic whipworm infection on vaccine mediated immunity	Jacob	Thompson	University of Manchester
P26209	Parasite ImmunoPathology	Poster 94	Inperson	The protein kinases SmFES and SmRAF may influence Schistosoma mansoni development, egg maturation, and hepatic granulomas progression in the mammalian host	Gabriella	Torres	Instituto René Rachou/ Fiocruz Minas
P25989	Vector:Parasite interactions	Poster 90	Inperson	Characterisation of Plasmodium falciparum oocyst development and transcriptome responses to dynamic nutrient stresses in the mosquito Anopheles gambiae	Alexander	Bailey	Imperial College London
P26098	Vector:Parasite interactions	Poster 7	Inperson	How will the response of mosquitoes to vector control shape malaria parasite evolution?	Catherine	Oke	University of Edinburgh
P26169	Vector:Parasite interactions	Poster 46	Inperson	Molecular malacology and xenomonitoring schistosomiasis: Implication of Bulinus africanus as an intermediate host of Schistosoma haematobium in Lake Malawi.	Thomas M.	Arme	University of Glasgow
P26006	Vector:Parasite interactions	Poster 5	Inperson	Occurrence and Geographical distribution of Microsporidia in tick population in Ogun State, Nigeria	Uwemedimo	Ekpo	Federal University of Agriculture Abeokuta
P25896	Vector:Parasite interactions	Poster 28	Inperson	Phylogenetic relationships and evolutionary patterns of the genus Psammolestes Bergroth, 1911 (Hemiptera: Reduviidae: Triatominae): tools for vector control of Chagas disease	CAROLINA	HERNANDEZ	UNIVERSIDAD DEL ROSARIO
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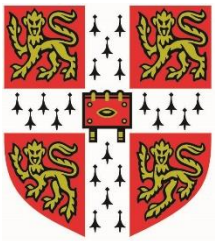

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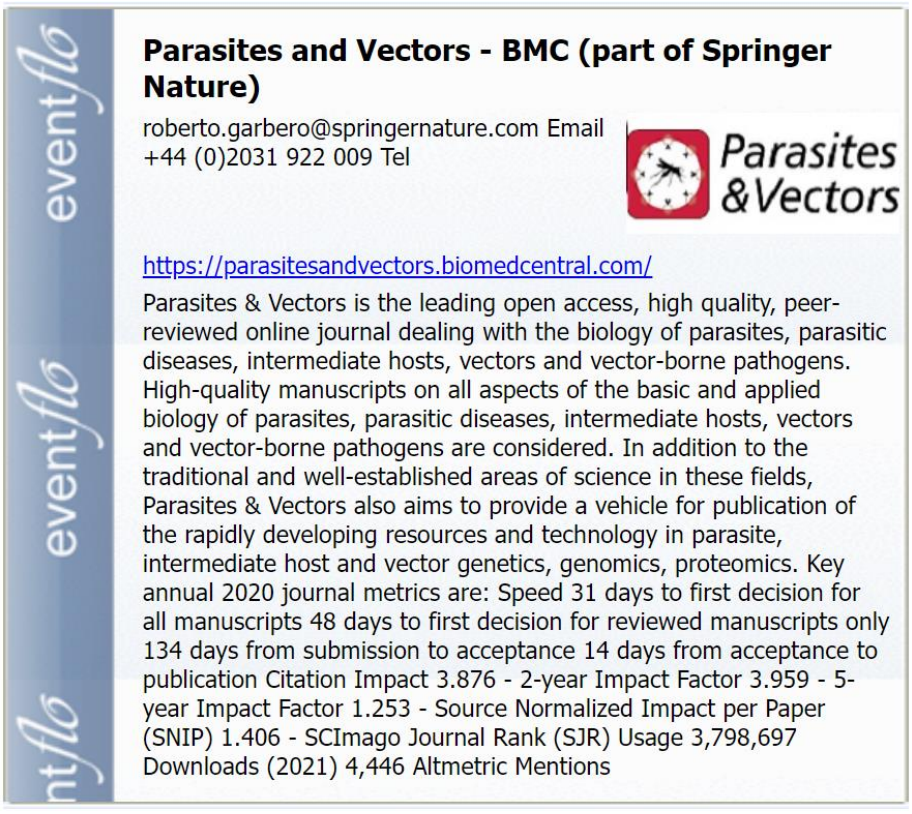
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