PARASITOLOGICAL SOCIETY OF SOUTHERN AFRICA · PARASITOLOGIESE VERENIGING VAN SUIDELIKE AFRIKA

PARSA

47th PARSA CONFERENCE

16-18 September 2018

Tshipise Forever Resort,
Limpopo, South Africa

University of Venda

Abstracts & programme

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Limpopo, South Africa

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Welcome note from PARSA Chairperson

Dear PARSA members

It is my privilege to welcome you all here at Tshipise Forever Resort. This is our 47th annual conference and I want to thank Prof Samie Amidou and his team from the University of Venda who volunteered to host this year's conference here at this beautiful venue. For participants who are attending PARSA for the first time, you are all very welcome and I hope that you will enjoy the conference and also realise the value of a meeting like this where you can discuss your interests with other people. To the "old" friends, you are also very welcome and it is always good seeing you again. This year's conference is fairly small with 67 registered participants which means that we can have quality time catching up, exchanging ideas and using this platform to forge new dimensions and collaborations in our research.

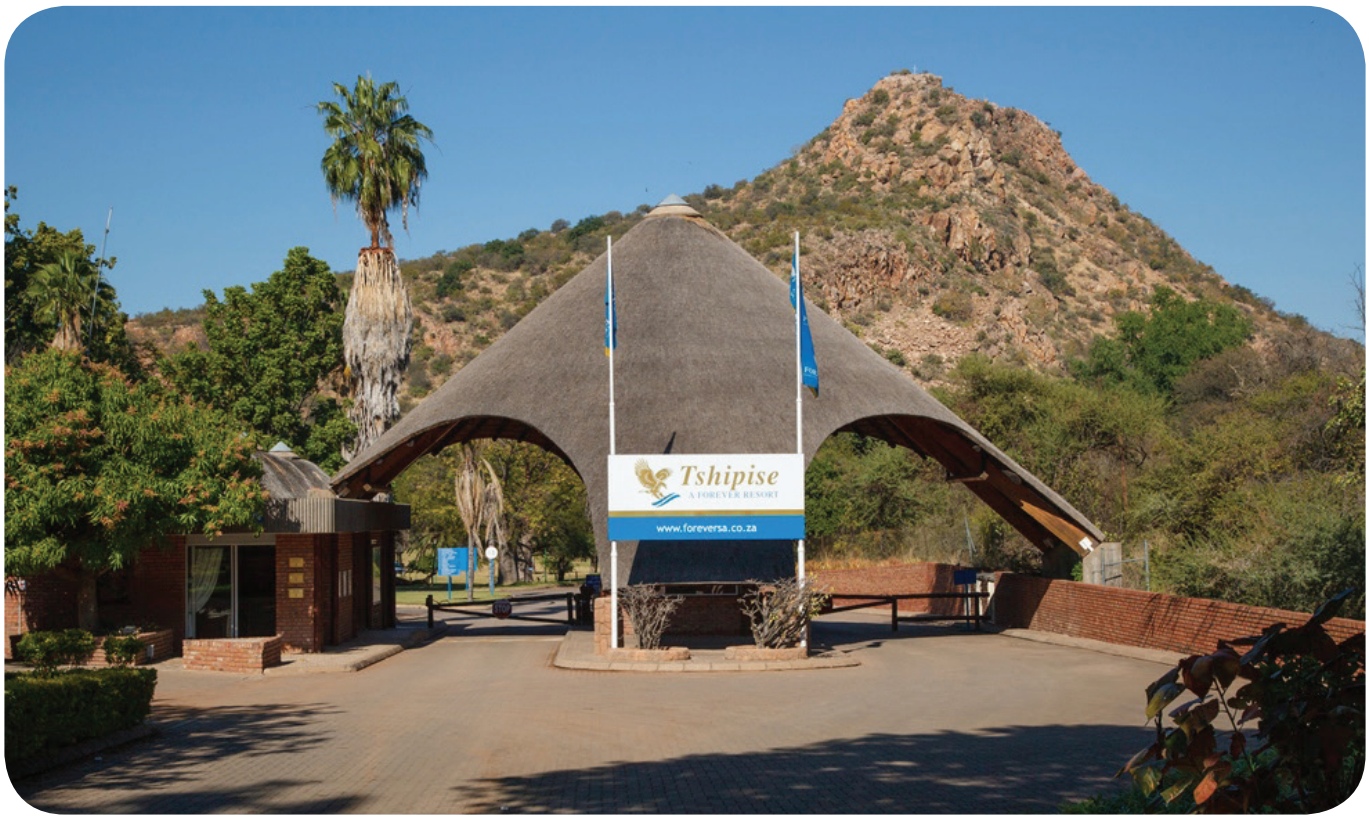
I am sure that you are all going to enjoy the relatively short programme and the presentations during the two days, especially the invited speakers, Prof Boris Krasnov from Ben-Gurion University, Prof Carol Gilchrist from the Department of Medicine, University of Virginia and Prof Helen Kutima from Jomo Kenyatta University of Agriculture and Technology.

On behalf of all participants I want to thank the local organizing committee from Venda University and ABEvents for all their hard work in organising the conference and accompanying events.

Enjoy the conference and the unique Limpopo Province scenery!

Sincerely

Susan Dippenaar



Programme

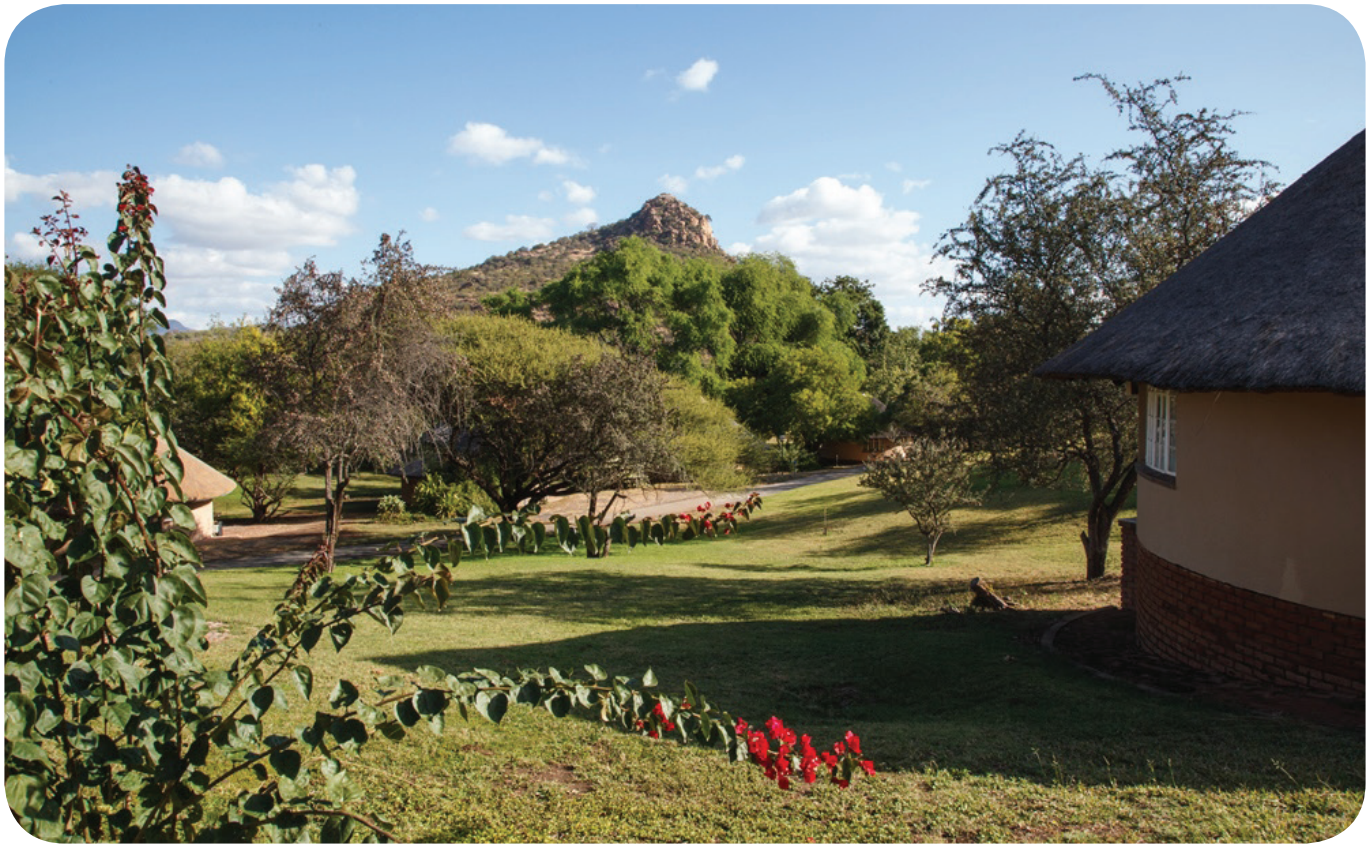
Sunday 16 September 2018		
16:00 – 19:00	**Arrivals and Registration**	
19:00	**Dinner in restaurant**	
Day 1: Monday 17 September 2018		
7:30	**Registration**	
Time	**Topic**	**Speaker**
Opening Session Chair: Matthee S		
8:00	Opening	**VC/DVC/Dean**
8:30	**Keynote address:** Sex-biased parasitism: patterns, mechanisms, consequences	**Krasnov BR**
Session1: Taxonomy and Parasite diversity Chair: Matthee S; Co-Chair: Nyangiwe N		
9:15	Barcoding of aquatic parasites: Bridging the divide between traditional and molecular taxonomy	**Dos Santos Q**
9:30	Parasite diversity of African penguins and the effect of host and environmental factors	**Espinase M**
9:45	Insights and research gaps in the systematics of amphistomes infecting ruminants and identity of intermediate host snails in eastern and southern Africa	**Mukaratirwa S**
10:00	Prevalence and diversity of avian haemosporidia in intra-African migratory land birds	**Chaisi ME**
10:15	Redescription of *Cichlidogyrus philander* (Monogenea, Ancyrocephalidae) using scanning electron microscopy (SEM) and molecular analysis	**Igeh P**
10:30	**TEA/Poster (30 min)**	
Session 2: Freshwater Biology Chair: Dippenaar S; Co-Chair: Traore A		
11:00	The expanding distribution of *Atractolytocestus huronesis*, an invasive alien parasite of the freshwater alien fish *Cyprinus carpio* in Limpopo Province, South Africa	**Chauke D**
11:15	The presence and abundance of *Lamproglena clariae* infecting the African sharptooth catfish (*Clarias gariepinus*) in comparison to water quality and metal concentration along the Vaal River, South Africa.	**Esterhuyze MMM**
11:30	First record of monogenean fish parasites in the Upper Lufira Basin (DR Congo): dactylogyrid and gyrodactylids infecting *Oreochromis macrochir* (Boulenger, 1912), and *Coptodon rendalli* (Boulenger, 1896), (Teleostei: Cichlidae).	**Kasembele GK**
11:45	Ecto- and endoparasites of the potential mariculture species, silver kob (*Argyrosomus inodorus*) (Actinopterygii: Sciaenidae), in Namibia	**Amakali A**
12:00	First molecular study and characterization of *Lamproglena monodi* Capart, 1944 (Copepod: Lernaeidae) infecting the Nile tilapia, *Oreochromis niloticus* L., 1758 in Kibos Fish Farm, Kenya	**Rindoria NM**

12:15	Morphological and molecular characterization of a new *Masenia* Chatterji, 1933 (Cephalogonimidae) from *Clarias gariepinus* in the Incomati River, Mozambique	**Dumbo J**
12:30	**LUNCH (60 min)**	
Session 3: Medical Parasitology Chair: Halajian A; Co-Chair: Oosthuizen M		
13:30	**Guest lecture:** Exposure factors associated with visceral leishmanosis (Kala-Azar) in Loima Sub-County of Turkana County, Kenya	**Kutima L**
14:00	Molecular detection and identification of *Cryptosporidium* species isolated from human and animal sources in Limpopo and Gauteng Provinces	**Samie A**
14:15	Modelling the influence of temperature and rainfall on malaria incidence in four endemic provinces of Zambia using semiparametric Poisson regression	**Shimaponda-Mataa NM**
14:30	Creating innovative diagnostic tests for African trypanosomosis, Dengue and Zika	**Grab D**
14:45	Autoprocessing and a second catalytic Cys-residue of metacaspase 5 of *Trypanosoma congolense:* two arrows in the quiver of a druggable virulence factor	**Eyssen L**
15:00	*Entamoeba* in South Africa: correlations with the host microbiome, parasite burden and first description of *E. bangladeshi* outside of Asia	**Ngobeni R**
15:15	**TEA/Poster (30 min)**	
Session 4: Macroparasites Chair: Chaisi M; Co-Chair: van der Mescht L		
15:45	Flea diversity of small mammals across South Africa: new records on host and geographic ranges	**Matthee S**
16:00	Seasonal distribution of *Culicoides* (Diptera: Ceratopogonidae) species in southern Africa	**Labuschagne K**
16:15	Diversity and seasonal dynamics of ticks in three agro-ecological zones in the Eastern Cape Province of South Africa	**Nyangiwe N**
16:30	Parasite performance as indicator of host resistance: laboratory studies with fleas and small mammals	**Khokhlova IS**
16:45	Dog ticks and their associated pathogens in the JB Marks Local Municipality, South Africa	**van Wyk CL**
17:00	Detection of pathogens harboured by *Stomoxys calcitrans*, a blood feeding ectoparasite of livestock in South African feedlots	**Makhahlela NB**
17:15	**POSTER SESSION WITH DRINKS (45 min)**	
18:00	**Free time**	

DAY 2: TUESDAY 18 September 2018

Time	Topic	Speaker
7:30	**Registration**	
Session 5: Molecular Parasitology Chair: Thekisoe O; Co-Chair: Shonai A		
8:00	**Guest lecture:** Whole genome sequencing of *Cryptosporidium* parasites: What can we learn?	**Gilchrist C**
8:30	Tick-borne disease dynamics in calves at the wildlife-livestock interface in the Mnisi Community area, Mpumalanga Province, South Africa	**Makgabo M**
8:45	16S rRNA-based bacterial community profiling of *Haemonchus contortus* infecting Dohne Merino sheep using next-generation sequencing	**Mafuna T**
9:00	Prevalence of *Ehrlichia ruminantium* in ticks collected from goats in Limpopo, South Africa	**Mathebula D**
9:15	Using a real-time PCR protocol to screen for avian haemosporidian parasites	**Wardjomto M**
9:30	Molecular characterization and distribution of rodent haemoparasites in the Lowveld regions of South Africa	**Moloro GT**
9:45	Development of a real time PCR assay to differentiate *Ehrlichia/ Anaplasma* spp. in dogs	**Nkosi NF**
10:00	Molecular characterization of haemoparasites infecting livestock at uMkhanyakude district	**Mofokeng L**
10:15	**TEA/Poster (30 min)**	
Session 6: Wildlife Parasitology Chair: Mukaratirwa S; Co-Chair: Collins N		
10:45	*Anaplasma* spp and *Hepatozoon* spp from free-ranging black-backed jackals (*Canis mesomelas*) in South Africa	**Penzhorn B**
11:00	Molecular and serological assessment of *Toxoplasma gondii* in definitive and intermediate hosts at the National Zoological Gardens of South Africa	**Mokgako N**
11:15	Parasites of the leopard (*Panthera pardus*) in South Africa, a review	**Rampedi KM**
11:30	Helminth parasites of *Crocodylus niloticus* (Reptilia: Crocodylidae) in the Kruger National Park, South Africa	**Junker K**
11:45	Species composition and the role of horse-flies in pathogen transmission in south-eastern KNP (Diptera: Tabanidae)	**Mazibuko X**
12:00	Parasitic helminths of rodents in South Africa: host range and spatial distribution	**Spickett A**
12:15	Diversity of the sporozoite antigen gene p67 in *Theileria parva* field isolates from cattle and buffalo in southern and eastern Africa	**Mukolwe DL**
12:30	**LUNCH (60 min)**	
Session 7: Therapeutic approaches to parasitic diseases Chair: van Zyl R; Co-Chair: Labuschagne K		
13:30	Oral administration of azithromycin ameliorates trypanosomosis in *Trypanosoma congolense* and *T. brucei brucei*-infected mice	**Molefe NI**

13:45	Ethno-veterinary medicine practices for the treatment of tick-borne diseases by rural farmers in the Eastern Cape Province, South Africa	**Mthi S**
14:00	The metal chelating properties of terpenes and effect on *Plasmodium* malaria and *Anopheles* vector	**van Zyl RL**
14:15	Investigation of the role of Hsp70 in Granzyme B-mediated in malaria therapy	**Ramatsui L**
14:30	Anthelmintic resistance in gastrointestinal nematodes of sheep in Limpopo Province, South Africa	**Mphahlele M**
14:45	Characterization of cytosol-localized Hsp70 isoforms from *Plasmodium falciparum* and their prospects as potential antimalarial drug targets	**Zininga T**
15:00	Antimicrobial activity of selected Venda medicinal plants against *Entamoeba moshkovskii*	**Maponya MM**
15:15	**TEA/Poster (30 min)**	
Session 8: Host-Parasite interactions Chair: Avenant-Oldewage A; Co-Chair: Junker K		
15:45	You are what you eat. Trace element and metal accumulation in a host–parasite relationship from the Vaal Dam, South Africa	**Gilbert B**
16:00	Investigating relationships among different facets of host specificity in haematophagous ectoparasitic arthropods	**van der Mescht L**
16:15	Establishment of the interacting partners of *Plasmodium falciparum* heat shock protein 70-x (PfHsp70-x)	**Monyai SF**
16:30 for 16:45	**AGM**	
17:45	**Free time**	
18:30	**Gala dinner**	



Keynote speaker

Boris Krasnov

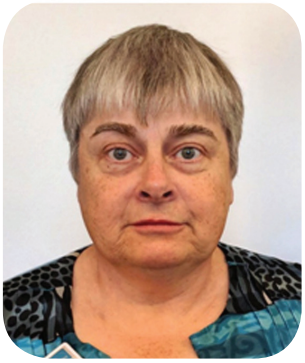
Boris Krasnov, Professor of Ecology in Ben-Gurion University of the Negev. Graduated from the Department of Zoology of Terrestrial Vertebrates of Lomonosov Moscow State University, Russia – M.Sc. in 1978, PhD in 1986. During first years of the scientific career, he mainly studied ecology and behavior of rodents at the Institute of Ecology and Evolution of the Russian Academy of Sciences. Following collapse of the former Soviet Union, in 1991 started to work at Ben-Gurion University of the Negev, Israel. After a few years of studying community ecology of ground dwelling desert animals (rodents, reptiles, insects), switched to study ecology of parasites in the mid 90ˢ and is doing this ever since. Krasnov's studies encompass various aspects of parasite ecology and evolution from physiology to biogeography and macroecology mainly on the model of haematophagous ectoparasites. In particular, he combines experimental work on physiological questions, field work on ecological questions, and comparative or meta-analyses on biogeographical or evolutionary questions. Krasnov is the author of 240 papers in peer-reviewed journals, 39 chapters in books, 3 authored monographs (one solo and two with co-authors) and co-editor of 5 collective volumes. He is also Section Editor of Parasitology Research, Subject Editor of Mammalia, Handling Editor of Israel Journal of Ecology and Evolution and member of editorial Board of European Journal of Ecology.

Guest speakers

Helen Kutima

Prof. Helen Lydiah Kutima has a BSc (Botany/Zoology) and MSc (Immuno-parasitology) from Jomo Kenyatta University of Agriculture and Technology (JKUAT), Kenya, and a PhD in Parasitology/Entomology from JKUAT, *icipe* and Ruhr Universität, Bochum, Germany. She has taught at JKUAT for 29 years and is currently an Associate Professor in the Department of Zoology and the Director of the Gender and Mentoring Centre. Her research and academic interest is in molecular parasitology and entomology, vector biology and tropical diseases of humans and animals. She has supervised many MSc and PhD students and published many papers in peer-reviewed journals. She is a member of many scientific bodies, among which are Entomological Society of Kenya, Microbiology Society of Kenya, African Academy of Sciences, African Women in Science and Engineering, World Association of the Advancement of Veterinary Parasitology, International Network of Women in Science and Engineering, African Regional Postgraduate Programme in Insect Science Scholars Association (ASA), American Society of Tropical Medicine and Hygiene (ASTMH), Kenya DAAD Scholars Association (KDSA), African Academy of Insect Scientists (AAIS), African Acarologists (AA) and American Association of Veterinary Parasitology (AAVP). She serves on the Editorial Boards of Journal of Vector-Borne Diseases (JVBD), Journal of Medical and Applied Science, Journal of Biological Science and Bioconservation. She is also a member of the Mentoring network of African Women in Academia.

Carol Gilchrist



Prof Carol A. Gilchrist received her BSc degree from University of Edinburgh, Scotland, UK, and PhD degree at the University of Western Ontario, Canada. She is currently an Associate Professor in the Division of Infectious Disease, Department and School of Medicine, University of Virginia., USA. Dr. Gilchrist's research has focused on using molecular methods to understand the biology and pathogenic phenotype of enteric parasites. She is currently part of a collaboration focused on obtaining whole genome sequencing of *Entamoeba histolytica* and *Cryptosporidium* protozoan pathogens to understand the genetic variability inherent in these human parasites.

Oral presentations abstracts

Sex-biased parasitism: Patterns, mechanisms, consequences

Krasnov BR
Blaustein Institutes for Desert Research, Ben-Gurion University of the Negev, Midreshet Ben-Gurion, Israel

In this talk, I consider patterns, mechanisms and consequences of sex biases in parasite infestation using small mammals (mainly) and their ectoparasites (mainly) as a model host-parasite association. I start with a description of sex biases in parasite infestation and discuss variation in these patterns among host and parasite taxa. Although, in most cases, parasite abundance, prevalence and species richness are often host male-biased, they may also be female-biased or host sex-independent. I show temporal and spatial variations in sex-biased parasitism and demonstrate that the extent of sex-bias can vary seasonally and be mediated by environmental conditions. In other words, that manifestation and strength of sex-biased parasitism may differ inter- and intraspecifically as well as being mediated by environmental conditions. Moreover, spatial variation in sex differences in parasite infestation can be affected by parasite, host and environmental factors with the set of factors affecting sex differences in infestation being different among parasite taxa. Then, I present main hypotheses aimed to explain mechanisms of sex-biased parasitism. One group of these hypotheses focuses on differences between male and female hosts in their probability to be attacked by parasites, while another group links gender-biased parasitism with differences in parasite performance in male versus female hosts. Further investigating the latter group of hypotheses and using data from laboratory experiments on fleas exploiting rodents, I will show that, in general, fleas perform better on male rodents. In particular, when feeding on male as compared to female hosts, fleas took more blood, digested it faster, produced more eggs and larger male offspring. Then, I consider consequences of sex-biased parasitism for parasite individuals, populations and communities. Regarding effect on parasite individuals, I show that parasites (in particular, fleas) appear to be able to distinguish between male and female hosts presumably by odour clues and select male hosts in Y-maze trials. I also show that male hosts drive population dynamics of parasites as well as their infracommunity structure. Finally, I talk about caveats in the analyses of sex-biased parasitism and demonstrate that straightforward analyses may sometimes lead to incorrect conclusions.

Barcoding of aquatic parasites: Bridging the divide between traditional and molecular taxonomy

Dos Santos Q, Avenant-Oldewage A
Department of Zoology, University of Johannesburg, P.O. Box 524, Auckland Park, Johannesburg 2006, South Africa

The taxonomic study of freshwater parasites in southern Africa has been a well-established and renowned field for many decades. Until recently, however, these studies were based almost entirely on the use of microscopy-based techniques and thus heavily reliant on morphology. Only a select number of species have enjoyed the addition of representative sequence data to their taxonomic information, be it through redescriptions or incorporation into the original descriptions. Thus, the need for standardised protocols with regard to the molecular study of both known and new parasite taxa is essential. During the molecular study of several parasitic species, it became clear that there are several hurdles when trying to incorporate barcoding principles into the taxonomic study of these organisms. In particular, the necessary incorporation of sequence data from publications and online data repositories such as GenBank proved to be a major factor in the effectiveness of barcoding approaches. As such, the possible obstacles encountered during molecular taxonomy were reviewed using lesser studied groups of parasitic organisms from South Africa, for which little or no molecular data is available. This was done by obtaining representative sequences for the species in question and assessing the level of ease with which their genetic identity could be inferred. Species from the Diplozoidae, Cichlidogyridae, Lernaeidae and Diplostominae were included. Five major factors which hinder the use of molecular information when studying parasite taxa were identified. These were: a) the use of different markers for different groups; b) publication of sequence data for unidentified species; c) publication of misidentified or incorrect sequence data; d) submission of sequences to data repositories without the publication of related study; and e) the generally limited numbers of taxa for which sequence data is available. Based on these findings, care needs to be taken when molecular approaches are incorporated into taxonomic studies. Hopefully, from the notes presented here, fully describing species, both morphologically and molecularly, will be easier in the future.

Parasite diversity of African penguins and the effect of host and environmental factors

Espinaze M, Hui C, Waller L, Dreyer F, Matthee S
Department of Conservation Ecology and Entomology, Stellenbosch University, South Africa

The African penguin is an endemic and endangered seabird of southern Africa. It breeds on island and mainland colonies distributed in Namibia and South Africa. This study explores the diversity and incidence of ecto- and endoparasites associated with African penguins and its natural habitat, as well as the effect of host and environmental factors on parasite diversity. Ecto-, haemo-, and helminth parasites were recorded from penguins (adults n=210 and chicks n=583) and their nests (n=628) across five localities (two mainland and three islands) along the Western Cape, South Africa, in 2016 and 2017. Mean nest density (total and active nests) and remote-sensed climatic conditions (temperature and precipitation) were obtained for each colony. Two flea (*Parapsyllus humboldti* and *Echidnophaga gallinacea*), one tick (*Ornithodoros capensis* s.s.) and one louse species (*Austrogonoides demersus*) were recorded from nests and penguins. *P. humboldti* was the most abundant and prevalent on penguins and in nests (69.10% and 57.80%, respectively). Haemoparasites (Piroplasmorida/ Haemospororida and Spirochaetales) and helminths (*Cardiocephaloides* spp., *Renicola* spp., *Contracaecum* spp. and *Cyathostoma* spp.) were also recorded. Piroplasmorida/Haemospororida (33.51%) and *Cardiocephaloides* spp. (56.17%) were the most prevalent from each group. Ecto- and haemoparasites abundance and prevalence increased in warmer seasons at Stony Point. Parasite prevalence and abundance was higher on mainland and in chicks compared to island colonies and adult penguins, respectively. A positive correlation was found between total nest density and Piroplasmorida/ Haemospororida, *Cardiocephaloides* spp. and in-nest tick abundance, and between active nest density and total ectoparasite abundance. Parasite diversity found in this study is similar to that previously reported for African penguins. Several factors are playing a role in shaping parasite infection patterns. Therefore, this information should be considered during management decisions at the colonies, in order to prevent further impact of parasites on the conservation of this endangered penguin species.

Insights and research gaps in the systematics of amphistomes infecting ruminants and identity of intermediate host snails in eastern and southern Africa

Mukaratirwa S, Pfukenyi DM
School of Life Sciences, Westville Campus, University of KwaZulu-Natal, Durban, South Africa

The main amphistome species infecting domestic and wild ruminants in east and southern Africa are reviewed including current advances and research gaps in the systematics and spectra of snail intermediate hosts. To date, 26 amphistome species belonging to nine genera from three families occur in domestic and wild ruminants. Over 70% of them belong to the genera *Calicophoron*, *Carmyerius* and *Cotylophoron*. Eighty-five percent of the amphistome species are shared between domestic and wild ruminant hosts; an important observation when considering the different options for their control. Seven snail species belonging to four genera from two families act as intermediate hosts of the identified amphistomes, with the genus *Bulinus* contributing 57% of the snail species. Some of the snails are intermediate hosts of amphistomes belonging to the same genus or to different genera; a phenomenon not yet fully elucidated as some snails are reported to be naturally infected with amphistome cercariae of unidentified species. Only nine (34.6%, 9/26) of the amphistome species have known snail hosts, while most (65.4%, 17/26) have unknown snail hosts. Future research in the use of molecular technology in elucidating the systematics of amphistomes as well as identification of snail intermediate hosts is discussed.

Prevalence and diversity of avian haemosporidia in intra-African migratory landbirds

Chaisi ME, Osinubi ST, Dalton D, Suleman E
National Zoological Gardens, South African National Biodiversity Institute, Pretoria, South Africa

Avian haemosporidian infections are widespread and can result in the decline or contribute to extinction of wild bird populations. We determined the prevalence and genetic variation of avian haemosporidia in 93 samples from intra-African migratory landbirds representing 22 species from South (n=76) and West Africa (n=17). The samples were analysed for the presence of haemosporidian DNA using real-time quantitative PCR (qPCR) and nested PCR assays targeting specific mitochondrial genes of these parasites. The cytochrome b (*cyt*b) gene was sequenced from all samples that tested positive by the nested PCR assay, and phylogenetic analysis was done to determine the relationships of the new sequences with previously published sequences from the MalAvi database. The overall prevalence of avian haemosporidia was 68.82% and 82.80% by qPCR and nested PCR, respectively. Eighteen (19.36%) birds had mixed infections of *Plasmodium/Haemoproteus and Leucocytozoon* spp. Infections by all haemosporidia were significantly higher ($p<0.05$) in samples from West Africa. Forty-five *cyt*b sequences obtained from 14 bird species grouped into three distinct clusters of *Haemoproteus* (35), *Leucocytozoon* (8) and *Plasmodium* (2) spp. These represent 8 published and 9 new MalAvi lineages. The most common lineage was *Haemoproteus* sp. (VIMWE1) which was identified from two bird species from West Africa and seven bird species from South Africa. This study indicates that haemosporidian infections in wild birds are common and genetically diverse. Future studies will determine the possible role of these birds in the spread and/or persistence of these infections across Africa.

Redescription of *Cichlidogyrus philander* (Monogenea, Ancyrocephalidae) using scanning electron microscopy (SEM) and molecular analysis

Igeh P, Dos Santos Q, Avenant-Oldewage A
Department of Zoology, University of Johannesburg, P.O. Box 524, Auckland Park, Johannesburg, 2006, South Africa

The taxonomy of monogeneans is based on the morphology of the sclerotized structures which is usually studied by light microscopy and different staining techniques; this enables morphological descriptions and morphometry. A recent trend for the study of these structures is the enzymatic digestion of the soft tissues of parasites, which is subsequently followed by scanning electron microscopy (SEM). This technique has led to the examination of structural details, which are not visible with light microscopy. In this study, the sclerites of *Cichlidogyrus philander* Douëllou, 1993 (Monogenea, Ancyrocephalidae), a minute parasite infecting *Pseudocrenilabrus philander* (Weber, 1897), were redescribed using SEM so as to reveal their ultrastructural details. *Pseudocrenilabrus philander* were collected from Padda Dam, Gauteng, South Africa. The parasites were removed from the gills in order to study their exterior and isolated hard parts, as well as to carry out molecular analysis. Based on SEM study, *C. philander* was found to have a large penis that ends in an almost 360° curve, with an opening running from the midpoint of the sharp lateral termination to its base, an accessory piece with a hook-like extremity that may appear forked terminally, and lacks a visible vagina. The ventral and dorsal bars have concave and convex surfaces with ribs on the concave surfaces, and the dorsal bar has fenestrations at the base of the auricles. The first pair of uncinuli displays a lateral wing on the left side of the base. On top of this wing, a ball-like structure with a small fenestration is visible. These features were not visible with light microscopy. Genetic characters derived from the 28S rDNA, the COI mitochondrial DNA and ITS1 rDNA regions distinguish *C. philander* from all other *Cichlidogyrus* species sequenced thus far.

The expanding distribution of *Atractolytocestus huronesis*, an invasive alien parasite of the freshwater alien fish *Cyprinus carpio* in Limpopo Province, South Africa

Chauke D, Luus-Powell WJ, Mokumo MF, Kunutu KD, Letlhage DS, Smit WJ, Halajian A
Department of Biodiversity, University of Limpopo, Polokwane, South Africa

Several alien freshwater fish species were introduced into South Africa for various reasons including aquaculture, biological control and the ornamental fish trade. An organism is regarded as an alien if it is introduced to an area outside its original natural range. The common carp, *Cyprinus carpio*, is an alien freshwater fish that originated from Europe and is widely distributed throughout South Africa. Through its introduction into aquatic systems globally this species has frequently co-introduced its parasites. The invasive tapeworm, *Atractolytocestus huronensis*, is one of the parasites frequently reported from *C. carpio* from different countries and has together with its host spread throughout the world. Although *A. huronensis* was originally described in North America, the parasite has previously been recorded in South Africa from Mpumalanga Province (Loskop Dam and Witbank Dam) and from Limpopo Province (Flag Boshielo Dam and Tzaneen Dam). From March 2017 to March 2018, 32 *C. carpio* were collected from different water bodies in Limpopo Province and examined for intestinal parasites. Fish were euthanized and the digestive tract removed and examined for parasites using a stereo-microscope. Cestodes found were relaxed, fixed, stained and mounted using standard techniques and identified using available keys. Cestodes found in the intestines were identified to be the alien parasite *Atractolytocestus huronensis*. No cestodes were recorded from carp collected from Tibani and Houtrivier Dams. However, this parasite was recorded from three specimens from Nkumpi Dam (prevalence, 37.5%), three specimens from Flag Boshielo Dam (prevalence, 75%), one specimen from Molepo Dam (prevalence, 100%) and six specimens from Albasini Dam (prevalence, 75%). The intensity ranged from 2 to 43. It is recommended that regulations and control measures for transport and introduction of alien fish should be followed to mitigate the spread of their parasites which may be a threat to native fish and thus local freshwater ecosystems.

The presence and abundance of *Lamproglena clariae* infecting the African sharptooth catfish (*Clarias gariepinus*) in comparison to water quality and metal concentration along the Vaal River, South Africa

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The Vaal River is important for South Africa by contributing to both the ecology and economy of the country. Pollution produced by anthropogenic activities or natural occurrences can have a negative effect on the ecology of this freshwater aquatic system. Various metals can bio-accumulate in aquatic systems and become a health risk. This study addressed the following aspects of metal pollution in the Vaal River: measuring water quality parameters and determining the concentration of metals in the water in comparison to the presence and abundance of the parasite *Lamproglena clariae* on *Clarias gariepinus*. During March 2017 a minimum of 10 and a maximum of 20 *C. gariepinus* specimens were collected with gill nets, electro-shocking or angling at each site. Specimens were collected from six sites along the Vaal River: Vaal River below Grootdraai Dam, Vaal Dam, Vaal River Barrage, Bloemhof Dam, Vaal-Harts Dam and Douglas Weir. All fish were inspected for *L. clariae* parasites. Water quality parameters (conductivity, oxygen saturation, dissolved oxygen, total dissolved solids, salinity, pH and temperature) were measured and recorded using a YSI 556 Multi-Probe meter. Water samples were analysed for metals using inductively coupled plasma mass spectrometry. Parasite prevalence, abundance and mean intensity were calculated for each site. Metal concentrations were found to increase and water quality was found to decrease downstream from the river's origin, with Vaal Barrage (Upper Vaal) and Douglas Weir (Lower Vaal) indicating poorer conditions in terms of electrical conductivity, total dissolved solids and salinity. *Lamproglena clariae* was found to be most prevalent upstream in the Vaal River below Grootdraai Dam (Upper Vaal). The second highest prevalence was observed in the Bloemhof Dam (Middle Vaal). Areas with a higher intensity of anthropogenic activities such as mining were identified as sites with the lowest parasite prevalence. Pollution caused by these activities has a significant effect on the presence and abundance of *L. clariae* on *C. gariepinus* in the Vaal River.

First record of monogenean fish parasites in the Upper Lufira Basin (DR Congo): Dactylogyrid and gyrodactylids infecting *Oreochromis macrochir* (Boulenger, 1912), and *Coptodon rendalli* (Boulenger, 1896), (Teleostei: Cichlidae)

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Parasites represent more than half of the total number of existing species on earth. They exhibit great species and phylogenetic diversity, and present a great variety of life cycles and body plans. Price (1980) stated that each free-living species harbours one or more parasites, indicating the existence of many more parasitic species than free-living ones. Parasites are hence an important component of biodiversity. They receive a lot of attention, not only because they can cause diseases, growth reduction or mortality, but also because they can be used as biological tools to investigate the ecology of their hosts, including diet, feeding, migration, and population structure. Additionally, they can be useful tools for historical biogeography, phylogeny or identification of their hosts (Williams *et al.*, 1992). Unfortunately, most studies undertaken on biodiversity concern larger organisms. All over the world, vast and species-rich communities, often dominated by smaller animals such as flatworms or various parasite taxa, remain understudied (Vanhove *et al.*, 2013). In view of the high biodiversity of potential host species in the tropics, it can be expected that parasitological surveys there can lead to the discovery of many parasite species, including species new to science (Whittington, 1998). We focus on monogenean fish parasites because of their diversity, wide distribution, high host-specificity and single-host lifecycle, rendering them interesting models for parasite biodiversity. We investigated the monogenean parasite fauna of cichlid fish in the Upper Lufira Basin (within the Congo Basin, DR Congo). These parasites had not formally been reported from this region. The work consisted of an inventory of the diversity of gill monogeneans, and an analysis of their infection parameters such prevalence, mean intensity and abundance, as defined by Bush *et al.* (1997).

Ecto- and endoparasites of the potential mariculture species, silver kob (*Argyrosomus inodorus*) (Actinopterygii: Sciaenidae) in Namibia

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Namibia is exploring aquaculture (both marine and freshwater) as a means of economic activity and food provision. The silver kob (*Argyrosomus inodorus*) has been identified as a potential candidate species for finfish culture. There is not much information available on the health of this species, especially parasite infestation that can pose a threat to fish cultivation and therefore to the success of mariculture. This study was designed to provide information on parasites that affect this fish. Fish were collected monthly for 11 months in 2017–2018 using conventional fishing gear (n=55) at Toscanini (20.8° S, 13.3° E), northern Namibia. Fish were examined for ecto- and endoparasites and the relationship between parasite prevalence/mean intensity and fish length, sex and season was analysed. Drawings and measurements were made using a camera lucida and calibrated eyepiece of an Olympus binocular microscope. Chi-square tests were used to determine differences in prevalence by season, sex and length. Different groups of parasites were found, including monogeneans (*Diplectanum* spp., *Calceostoma* sp., *Sciaenacotyle* sp.), digeneans (*Helicometrina* sp., *Stephanostomum* sp.), an unidentified cestode larva, nematode (*Anisakis* sp.), acanthocephalan (*Corynosoma* sp.) and copepods (species from Caligidae and Lernanthropidae). With the exception of digeneans, larger-sized fish (TL > 50 cm) were more subjected to parasitic infections than smaller-sized fish (TL ≤ 35 cm). Parasite prevalence and mean intensity levels were higher during the warmer months (October to March) than during colder months (April to September). Fish sex preference was not observed for parasite infections in silver kob.

First molecular study, review and redescription of *Lamproglena monodi* Capart, 1944 (Copepod: Lernaeidae) infecting the Nile tilapia, *Oreochromis niloticus* L., 1758 in Kibos Fish Farm, Kenya

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The crustacean copepod *Lamproglena monodi* was first described by Capart, 1944, from the gill filaments of *Haplochromis nobilis* (Boulenger), from Molindi River of same latitude with Lake Kibuga, *Haplochromis macrops* (Boulenger), from Rutshuru River, *Haplochromis eduardii* Regan and *H. serridens* Regan from Lake Edouard, *Haplochromis moffati* (Gastelnau), from Kafubu River and *Hemichromis fasciatus* Peters from Legide River, all in the Democratic Republic of Congo (DRC). Since then, four redescriptions of this parasitic copepod have been produced; three in Africa (Egypt) and one from South America (Brazil), all showing great discrepancies from the original description. The present study provides a further redescription of *L. monodi* using morphological features, with the aid of light microscopy (LM) and scanning electron microscopy (SEM), and the confirmation of the species by molecular analysis. This forms the first genetic study on *L. monodi*. The parasites were collected from the gills of *O. niloticus* (L., 1758) collected from Kibos Fish Farm, Kisumu County, Kenya. The prevalence, mean intensity and mean abundance were determined and recorded as 13.5%, 8.6 and 1.2 respectively. The 18S and 28S rDNA fragments were amplified, sequenced and compared to other Lernaeidae taxa. Both markers confirmed the distinctness of *L. monodi* from previously genetically characterized species, with distances of 1.36 – 2.80% observed for 18S and 17.10 – 20.32% for 28S. The morphogenetic data reported in these work can provide a useful point of reference for future studies.

Morphological and molecular characterization of a new *Masenia* Chatterji, 1933 (Cephalogonimidae) from *Clarias gariepinus* in the Incomati River, Mozambique

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During 2017, *Clarias gariepinus* were collected in the Incomati River, South Mozambique and examined for trematodes. Among others, an unidentified new species of the *Masenia* Chatterji, 1933 was found parasitizing the anterior section of the intestine of *C. gariepinus* (Burchell, 1822). The description of the morphology of the new form is based on histology, light microscopy (LM) and scanning electron microscopy (SEM). For LM, specimens were stained with acetocarmine and mounted in Canada balsam; for histology, parasites were embedded in resin, sectioned and stained with Hematoxylin and Eosin; for SEM study specimens were processed in gradually increasing concentrations of Hexamethyldisilazane. Ribosomal rDNA was extracted and fragments of the 18S and 28S regions amplified and sequenced. The new form is distinguished from other *Masenia* species by an ellipsoid body, a reniform seminal receptacle, a cirrus-sac ending anterior towards the ventral sucker, intestinal caeca extending into the hind body to the level of the posterior testis, an equatorial ovary, the vitelline follicles extending anteriorly to the ventral sucker and to the middle of the ovary and an hourglass-shaped excretory vesicle. It is further larger in overall size than other African *Masenia* species. Phylogenies from both maximum likelihood and parsimony methods based on 28S supported the placement of the new form in the Cephalogonimidae and its distinctness from species of *Cephalogonimus.* The 18S distinguished the new form from *M. bangweulensis* and thus represents a distinct haplotype.

Exposure factors associated with visceral leishmanosis (kala-azar) in Loima Sub-County of Turkana County, Kenya

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Visceral leishmanosis (Kala-azar) is a vector-borne disease caused by the obligate intra-macrophage protozoan parasites known as *Leishmania donovani* and *L. infantum* in the Old World and *L. chagasi* in the New World. It is classified as a neglected disease yet it is a public health problem: a debilitating disease causing an estimated 500,000 new cases each year. One tenth of these patients will die. The actual death toll from the disease may be higher than this estimate, considering the existence of unidentified foci. The new trend risks causing a public health crisis in Kenya and other African countries with weak economies, since there is no vaccine against the disease. In Kenya, kala-azar is common in arid and semi-arid regions of North Eastern and Rift Valley Provinces, especially the Loima Sub-County of Turkana County, West Pokot, Machakos, North Eastern, Marigat and Baringo East. The study was conducted between October 2015 and June 2016. The objective was to determine the exposure factors associated with kala-azar. Analytical cross- sectional research design was used to determine socio-demographic characteristics, socio-economic and cultural factors, health-seeking behaviour, vector dynamics, disease epidemiology, as well as local peoples' knowledge, perception and behaviour towards the existence of Kala-azar in the area. Cluster random sampling technique was used to identify study subjects in the purposively selected Loima Sub-county. A sample size of 341 respondents who were household heads or adult members and health facility workers were randomly sampled. The data collected were processed and analysed using Statistical Package for Social Sciences (SPSS), correlation and regression and multivariate analysis. The qualitative data were analysed thematically. Demographic data were summarised using percentages, means and standard deviations and are presented using tables and graphs. The chi-square test was used to compare the relationship between variables. The key exposure factors to the disease in the community include: Age, gender, educational level, socio-economic status, housing, presence of large number of termite mounds all over the area 60.1% (n=205), low ownership and usage of bed-nets (30%),inaccessibility to health services, varying health-seeking behaviour and lack of proper knowledge on transmission of disease. Also, human activities such as deforestation and hunting (52%; n=32), resting or sitting near termite mounds 70% (n=191) and dancing at night (Edong'a 64.8%; n=167), when the sand flies are active. There was a significant association between age (OR=0.7; 95% CI=0.4-1.1:; p=0.135) and exposure to kala-azar, gender (OR=0.6; 95% CI=0.4-0.9; p=0.012),education level (OR=1.2; 95% CI=0.1-1.4, p=0.0501), housing (OR=1.8; 95% CI=1.0-3.1;, p=0.029),presence of large amount of termite mounds (OR=0.6; 95% CI=0.2-2.0; p=0.0045) and resting or sitting near termite-mounds (OR=0.6; 95% CI=0.1-2.1; p=0.0043). The study concluded that kala-azar is prevalent in the area and though the community is aware of its existence, the residents have different beliefs about transmission. The study recommended the need for enhanced general health education and awareness on the transmission cycle of kala-azar. Community empowerment and participation should be emphasized as well as structural development plans that include sand fly management strategies and control methods that would ensure the removal of breeding and resting sites of the vectors within human habitation. In addition, integrated disease surveillance and response to be implemented to avert the disease situation should be initiated.

Molecular detection and identification of *Cryptosporidium* species isolated from human and animal sources in Limpopo and Gauteng Provinces

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Cryptosporidium infects humans as well as many different types of animals. However, very few studies have been conducted on the genetic diversity of these organisms in the region. Therefore, the aim of this study was to detect and identify the genetic diversity of *Cryptosporidium* species from humans and animals in Giyani, situated in the northern part of South Africa, and Pretoria, situated in the central part of the country. A total of 560 samples were collected from human and animals and were screened by microscopy using modified Ziehl-Neelsen staining technique. All the samples were tested by Enzyme-Linked Immunosorbent Assay (ELISA) using the *Cryptosporidium* II kits from Techlab, Virginia, USA. Positive samples from microscopy and ELISA were examined by conventional and real-time PCR protocols for the amplification of *Cryptosporidium* oocyst wall protein (COWP) region and 18S rRNA region. A number of samples from real-time PCR that gave clear bands on gel electrophoresis were sent for sequencing. The overall prevalence of *Cryptosporidium* as detected by ELISA method from the samples collected from humans was 41.2% and 56.5% among animals. Prevalence rates in cattle and goats were 55.8% (29/52) and 60.6% (20/33), respectively. Prevalence was higher from the rural area 73.0% (159/218) compared to the urban area 22.1% (80/362) (χ^2=145.1; p=0.0001). *C. hominis* and *C. muris* were the two species identified in humans, while *C. parvum* and *C. andersoni* were identified among animals. The present study identified *C. muris* from human samples in our area for the first time. However, *C. hominis* remains the dominant species that infects humans in our area. *Cryptosporidium* species was mostly found in samples from asymptomatic individuals. In animals, *C. parvum* was the most commonly isolated organism; *C. andersoni*, which was identified in our region for the first time as well, occurred in both goats and cattle. Populations in the affected areas need to be made aware of the infections so that care should be taken to avoid the spread of infection in water sources or in immunocompromised individuals.

Modelling the influence of temperature and rainfall on malaria incidence in four endemic provinces of Zambia using semiparametric Poisson regression

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Although malaria morbidity and mortality have been greatly reduced globally owing to great control efforts, the disease remains a major concern. In Zambia, all provinces are malaria endemic. Transmission intensities vary, however, mainly depending on environmental factors interacting with the vectors. Generally in Africa, possibly due to the varying perspectives and methods used, there is variation in the relative importance of malaria risk determinants. In Zambia, the role climatic factors play on malaria case rates has not been determined in combination of space and time using robust methods in modelling. This is critical considering the reversal in malaria reduction after the year 2010 and the variation by transmission zones. Using a geoadditive or structured additive semiparametric Poisson regression model, we determined the influence of climatic factors on malaria incidence in four endemic provinces of Zambia. We demonstrated a strong positive association between malaria incidence and precipitation as well as minimum temperature. The risk of malaria was 95% lower in Lusaka (ARR=0.05, 95% CI=0.04–0.06) and 68% lower in the Western Province (ARR=0.31, 95% CI=0.25–0.41) compared to Luapula Province. North-western Province was similar to Luapula Province. The effects of geographical region are clearly demonstrated by the unique behaviour and effects of minimum and maximum temperatures in the four provinces. Environmental factors such as landscape in urbanised places may also be playing a role.

Creating innovative diagnostic tests for African trypanosomosis, Dengue and Zika

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In rural Africa failure to diagnose infections by *Trypanosoma brucei gambiense* or *T. b. rhodesiense* in blood, lymph and/or CSF in the critical early stages of the disease is the single most important factor for fatalities due to failed treatment. Dengue is a major public health threat throughout the tropics and subtropics that can progress rapidly from benign febrile illness to dengue haemorrhage fever and/or dengue shock syndrome and death. Zika virus (ZK)is another flavivirus that has re-emerged as a public health threat throughout the tropics and subtropics. ZV causes microcephaly and serious birth defects in foetus, and has been associated with Guillen-Barre Syndrome in adults. Innovative point-of-care (POC) diagnostics of human African trypanosomosis (HAT), dengue and Zika with better analytical performance than current technologies would greatly improve disease surveillance and patient compliance at critical staging points so that appropriate drug regimens could be instituted earlier, important to limit morbidity and mortality from complicated disease outcomes. We previously created protein-DNA chimeras, termed 'LAMPoles, which coupled the specificity of antibodies with loop-mediated isothermal amplification (LAMP) to enable rapid measurement of protein analytes by DNA amplification. This technology dramatically lowered the limit-of-detection for anti-trypanosomal immune responses in mice and humans. Here, we present progress to expand the capability of our LAMPole platform to detect trypanosomes and virus in relevant body fluids utilizing a new class of affinity protein known as 'nanobodies' that can be tailored to bind virtually any molecular target. We summarize our progress towards development of sensitive and specific LAMPole-based platforms for host anti-pathogen antibodies and antigens in plasma/blood/CSF and ultimately the creation of non-invasive tests for these biomarkers. Overall, the combination of protein detection via LAMP using Nanobodies will facilitate implementation of highly sensitive and specific non-invasive diagnostics for HAT, dengue, zika and other relevant microbial pathogens that will facilitate treatment and surveillance at POC.

Autoprocessing and a second catalytic Cys-residue of metacaspase 5 of *Trypanosoma congolense:* two arrows in the quiver of a druggable virulence factor

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The metacaspases (MCAs) are attractive drug targets for the treatment of African trypanosomosis as they are not found in the metazoan kingdom and they have been implicated in cell cycle progression of kinetoplastid parasites and are considered virulence factors. We report the biochemical characterisation of MCA5 from the animal-infective *Trypanosoma congolense* that causes nagana in cattle in sub-Saharan Africa. Upon recombinant expression in *E. coli*, autoprocessing is evident, and MCA5 further autoprocesses when purified using nickel affinity chromatography, which we term nickel-induced autoprocessing. When the catalytic His and Cys residues were mutated (*Tco*MCA5H147AC202G), no nickel-induced over autoprocessing was observed and the peptidase was enzymatically active, suggesting the existence of a secondary catalytic Cys residue, Cys81. Immunoaffinity purification of native *Tco*MCA5 from the total parasite proteins was achieved using chicken anti-*Tco*MCA5 IgY antibodies. The full length native *Tco*MCA5 and the autoprocessed products of recombinant *Tco*MCA5H147AC202G were shown to possess gelatinolytic activity, the first report for that of a MCA. Both the native and recombinant enzyme were calcium independent, had a preference for Arg over Lys at the P1 site and were active over a pH range between 6.5 and 9. Partial inhibition (23%) of enzymatic activity was only achieved with leupeptin and antipain. Molecular modelling and docking were used to assess the binding of a library of ligands and commercial inhibitors to a homology model of *Tco*MCA5 towards the design of a novel trypanocide.

Entamoeba in South Africa: Correlations with the host microbiome, parasite burden and first description of *E. bangladeshi* outside of Asia

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High prevalence of *Prevotella copri* is a biological marker of a symptomatic *Entamoeba histolytica* infection. The goal of this work was to verify this Asian finding in a South African population and to examine the association of this biological marker with other related *Entamoeba* species common in our study population. A comprehensive assay was used which included probes to identify *Entamoeba bangladeshi,* first described in Bangladesh, which had not previously been described outside the Asian continent. A total of 484 stool samples were collected between November 2013 and June 2015 (rural=227) and (urban=257), from both diarrhoeal and non-diarrhoeal participants. DNA was extracted and a highly sensitive qPCR assay and amplicon sequencing of selected samples were used to detect and quantitate *Entamoeba* spp, *Prevotella copri* and Enterobacteriaceae. Approximately 27% (n=129) of the collected study samples were positive for *Entamoeba* species. *The prevalence of E. histolytica* was 6.4% (31/484)*, E. dispar* 8% (38/484), and *E. bangladeshi* 4.5% (22/484) (co-infections accounted for 2.3% (11/484) of the cases). Up to 10% (49/484) of samples were not initially identified at the species level by the qPCR assay. The amplicons of 34 of the 49 unassigned *Entamoeba* were purified and sequenced. Of these 10 were *E. histolytica* (adjusted prevalence 8.5%) and one *E. bangladeshi* (adjusted prevalence 4.75%) the remainder proved to be derived from *E. hartmanni* (2.6%), which was not discriminated against by the *Entamoeba* genus probe. *E. moshkovskii* was not identified in this population. A high parasite burden and expansion of the *P. copri* level was associated with diarrhoea due to *E. histolytica*. *E. bangladeshi,* first discovered in an urban cohort in Bangladesh, was identified in urban and rural South African settings. This is the first description of *E. bangladeshi* outside of Bangladesh. We were also able to observe changes in the host microbiome and the parasite burden associated with *E. histolytica* infections in South African diarrhoea cases versus infected asymptomatic controls but not with *E. bangladeshi* or the non-pathogenic *E. dispar*.

Flea diversity of small mammals across South Africa: New records on host and geographic ranges

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Small mammals host a large diversity of flea species. South Africa is no exception; however, most data are restricted to flea-host species lists. To date, few studies have included adequate host sample sizes per locality and as such the full flea species representation per host species, locality and biome is unknown. To address the paucity in data we studied the host range and distribution of flea species on rodents and insectivores across multiple biomes in South Africa. Habitat suitability for flea species considered as important vectors of disease in humans and domestic animals was modelled. Data comprised flea records from small mammals captured at 29 localities during 2009–2013 and published flea data. Climate□based predictor variables, widely used in arthropod vector distribution, were selected and habitat suitability modelled for 10 flea vector species. More than 2400 fleas representing 33 species and subspecies were collected from 1185 small mammals. Ten of each of the flea and rodent species are plague vectors and reservoirs, respectively. Multiple novel flea–host associations and locality records were noted. Three vector species were recorded from insectivores. Flea species varied in geographic distribution that ranged from broad, across□biome distributions to single biomes. Habitat suitability models identified regions of summer and all□year rainfall as representing suitable habitats for most vector species. The study yielded valuable data on host and geographic ranges of fleas, which can aid our understanding of vector and disease ecology.

Seasonal distribution of *Culicoides* (Diptera: Ceratopogonidae) species in Southern Africa

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Worldwide, *Culicoides* Latreille species are involved in disease transmission. In southern Africa viruses that cause African horse sickness, equine encephalosis and bluetongue are of major economic and veterinary importance. Cases of these diseases are reported annually throughout the region. Collection studies across the southern African region over the past 20 years have accumulated data for most of the region. Utilising these data, seasonal distribution of *Culicoides* species in general and the two main vectors species, *C. imicola* Kieffer and *C. bolitinos* Meiswinkel, were investigated. Data for 47 sites over 12 consecutive months were examined. In the analysis both average and maximum collection per month were used to quantify the total number of *Culicoides* and vector species collected. Mondrian matrixes were generated for the average and maximum numbers of total species, as well as for *C. imicola* and *C. bolitinos*. The Mondrian matrix shows that a better correlation exists between number of *Culicoides* per site and number of *C. imicola* rather than with number of *C. bolitinos* collected. Maps were generated to examine the distribution of vectors with rainfall. *Culicoides* species in general and especially *C. imicola* were more prevalent in summer and autumn with the highest number of *C. imicola* being collected in the summer rainfall areas. This study highlighted the importance of long-term collections in understanding the distribution of *Culicoides* species and role environmental factors play in their distribution.

Diversity and seasonal dynamics of ticks in three agro-ecological zones in the Eastern Cape Province of South Africa

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The objectives of this study were to determine the diversity, seasonal dynamics and abundance of ticks infesting cattle in three agro-ecological zones in the Eastern Cape Province. Ticks were collected from 10 cattle and from six drag samples during the 12-month study period. Adult ticks were removed from the right-hand side of each animal and placed in containers filled with 70% ethanol. Ten tick species were identified: *Rhipicephalus* (*Boophilus*) *decoloratus* (32.5%), *R. evertsi evertsi* (18.8%), *R. appendiculatus* (17.3%), *Amblyomma hebraeum* (16.3%), *R. simus* (7.7%), *Ixodes pilosus* (3.8%), *Hyalomma rufipes* (3.5%), *R. follis* (0.08%), *Haemaphysalis elliptica* (0.04%), and *H. silacea* (0.02%). The southern African yellow dog tick, *H. elliptica,* was only found on vegetation. The agro-ecological zones differ significantly in tick species and their distribution. The *A. hebraeum* and *R. evertsi evertsi* counts were higher in Kowie Thicket (KT) during summer season (2.05 ± 0.01 and 1.00 ± 0.09, respectively) compared to Bedford Dry Grassland (BDG) and Bhisho Thornveld (BT) veld types. In all vegetation types, *R. appendiculatus* had higher counts in KT in spring (0.91 ± 0.08), summer (0.78 ± 0.08) and winter (0.78 ± 0.08). *Rhipicephalus* (*Boophilus*) *decoloratus* was more frequent in the BT (1.78 ± 0.11) during the summer season. *Rhipicephalus* (*Boophilus*) *microplus* was not found in the present study, suggesting that this invasive tick is not yet established in the west-central region of the Eastern Cape Province. However, this study ascertained that agro-ecological differences and seasonal variations have influence on tick species distribution.

Parasite performance as indicator of host resistance: Laboratory studies with fleas and small mammals

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From ecological and evolutionary perspectives, host resistance is nothing other than loss of fitness in a parasite induced by a host. Consequently, ecoimmunological studies should focus not only on immunological responses of a host to parasites, but also on responses of parasites to host anti-parasitic defences. We present the results of several experimental case studies carried out on flea parasites of small mammals. We consider the effect of host resistance on feeding and reproductive performance of fleas. First, we focus on flea performance on individuals of the same host species that presumably differ in their resistance abilities (males versus females, young versus mature versus senile individuals, immune-naïve individuals versus individuals that acquired resistance against fleas). Second, we consider performance of individuals of the same flea species on several species of small mammals that presumably have different innate resistance against this flea species. Feeding performance of fleas is measured via bloodmeal size, rate of digestion and energy expenditure for digestion, while reproductive performance is evaluated via both quantity (egg and new imago production) and quality of offspring (development rate, survival under starvation and body size).

Dog ticks and their associated pathogens in the JB Marks Local Municipality, South Africa

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Ticks are blood-feeding pests of domestic and wild animals and are notorious for the transmission of pathogenic organisms to their mammalian hosts. Companion animals also suffer from tick infestations, which is problematic due to their close association with humans. The aim of this study was to identify tick species infesting urban dogs at Potchefstroom Animal Welfare Society (PAWS) and detect pathogens of veterinary importance by microscopy and PCR as well as to conduct 16S metabarcoding of their gut and salivary gland microbiota using Illumina next generation sequencing platform. Ticks species were identified as *Haemaphysalis elliptica* (39%) and *Rhipicephalus sanguineus* (61%) by both morphological and molecular analysis. Morphological detection of pathogens infecting *H. elliptica* indicated an infection rate of 29% (32/112) and 2% (2/112) with *Rickettsia* sp. and *Anaplasma phagocytophilum*, respectively. Whilst in *R. sanguineus* there was an infection rate of 14% (13/92) and 1% (1/92) with *Rickettsia* sp. and *Babesia* sp., respectively. Molecular analysis detected 2% (1/49) and 8% (4/49) of *A. phagocytophilum* and *Ehrlichia canis*, respectively in *H. elliptica* DNA. Whilst 2% (3/55) of *E. canis* was PCR positive in *R. sanguineus* DNA. Metagenomic analysis indicated the presence of veterinary, medically and ecological important bacterial genera, including *Mycobacterium*, *Bacillus*, *Clostridium*, *Ehrlichia*, *Rickettsia*, *Wolbachia*, *Helicobacter*, *Coxiella*, and *Borrelia*. Data obtained in this study showed that tick species infesting urban dogs in Potchefstroom, JB Marks Local Municipality, harbour various pathogenic and non-pathogenic microorganisms.

Detection of pathogens harboured by *Stomoxys calcitrans*, a blood-feeding ectoparasite of livestock in South African feedlots

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Stomoxys calcitrans, commonly known as stable flies, are hematophagous ectoparasites of economic importance worldwide due to their role in the transmittance of various pathogens including bacteria, viruses, and protozoa during blood feeding. The aim of this study was to characterize stable flies found in South African feedlots and investigate their gut microbiota, as well as to detect economically important bovine pathogens they are harboring. A total of 10195 stable flies were collected from 3 feedlots using Vavoua traps with n=9993 from Potchefstroom, North West Province (NW), n=175 from Sasolburg, Free State Province (FS) and n=27 from Polokwane, Limpopo Province (LP). The *S. calcitrans* flies were identified morphologically and further confirmed by PCR and phylogenetic analysis of *CO1* and *16S rRNA* genes. Analysis of gut microbiota by 16S metabarcoding using Illumina Mi-Seq Next Generation Sequencing (NGS) produced a total of 462 operational taxonomic units (OTUs). The most abundant genera in NW were *Wolbachia*, *Sphingomonas*, and *Flavobacterium,* whilst in FS *Sphigomonas, Agrobacterium,* and *Methyllobacterium* were most abundant. The NGS was not conducted on Polokwane samples due to poor DNA quality. Genera of medical, veterinary, and ecological importance found were *Anaplasma*, *Wolbachia*, *Rickettsia*, *Bacillus*, *Clostridium* and *Staphylococcus*. Nested PCR detected the presence of *Anaplasma marginale* DNA in pooled samples of *Stomoxys calcitrans* with 10% and 16% occurrence in FS and NW, respectively. No *Anaplasma marginale* was detected by PCR in LP. Although NGS revealed the presence of *Rickettsia* sp., none were detected by genusspecific probes. This study also showed the dominant high sensitivity of NGS as compared to conventional PCR.

Whole genome sequencing of *Cryptosporidium* parasites: What can we learn?

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Molecular tools, such as whole genome sequencing (WGS), are improving at a rapid pace and provide a robust and high-resolution method to identify, compare, and classify pathogenic organism. Whole genome sequencing of parasites allows us to identify region of genome under pressure from the host immune system and comparative genomics may allow us to identify pathways and proteins involved in disease. Cryptosporidiosis is one of the top 5 causes of diarrhoea in children □2 years of age from low-income countries and is one of the top 5 causes of diarrhoea in immune-compromised individuals. This parasite undergoes both sexual and asexual multiplication in the human host but very little is known about the degree and impact of parasite genetic diversity on disease. In a recent study we sequenced 32 *Cryptosporidium hominis* genomes to depth of ≥ 50X from oocysts directly isolated from faecal material collected between June 2014 and 2017 from disadvantaged children who were enrolled at birth at a Bangladesh study location. Each sequenced isolate represents not one individual parasite but the parasite population derived from a single infecting oocyst. We identified 36,780 single nucleotide polymorphisms in the Bangladesh *C. hominis* isolates; however, only 1,582 occurred with a frequency 20% (Common SNPs). No linkage was observed between common SPNs if they were separated by more than 300 bp in the genome, indicating that in Bangladesh parasites recombination was frequent. Several hypervariable regions encoding membrane or secreted proteins were identified. As expected the hypervariable gp60 gene was picked out – other regions spanned the Cops-1, cgd8_690 and secreted SKSR family protein and two members of the insulinase family. Both non-synonymous changes in then coded amino acids, internal deletions and the premature termination of some ORF occurred. Ongoing work is focused on determining if the identified genomic diversity plays a role in immune invasion and contributes to the high rates of reinfection that occur in this population.

Tick-borne disease dynamics in calves at the wildlife-livestock interface in the Mnisi Community area, Mpumalanga Province, South Africa

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Anaplasmosis, babesiosis, and heartwater are the three most important tick-borne diseases of cattle in South Africa and result in high mortalities. Endemic stability contributes to disease control, but little is known about the conditions required for maintenance of endemic stability. Through the Health and Demographic Surveillance System in Livestock in the study area of the Mnisi One Health Platform, Mpumalanga, a great deal of information is being collected about cattle in the area. Cattle have been identified for tick burden assessment, serological analyses and parasite identification. However, little is known about the timing of infection of cattle with various tick-borne haemoparasites. Therefore, this study aimed to investigate the time-course of infection in calves (n=10) and presence of haemoparasites in adult ticks over a 1-year period using pathogen-specific reverse line blot hybridization and quantitative polymerase chain reaction assays. Blood samples and adult ticks were collected monthly from calves in two areas of the Mnisi community: five located in a peri-urban area and five at the wildlife interface. A total of 119 blood samples and 818 adult ticks were collected. The results confirm the exposure of most calves to both non-pathogenic and pathogenic tick-borne haemoparasites from the age of 0–1 month, although some of the pathogens could not be detected in the calves until 6–7 months, and *A. marginale* was not detected at all in two calves at the wildlife interface. These calves were either infected at levels below the detection limit of our assays, or they were not infected at all. If the latter, it is possible that exposure to related non-pathogenic haemoparasites might help to establish and maintain endemic stability. Adult ticks were identified to species level and both non-pathogenic and pathogenic haemoparasites were identified in DNA extracted from ticks after digestion of their blood meal. A higher number of calves had detectable pathogenic haemoparasite infections throughout the year in the peri-urban area than calves at the wildlife-livestock interface. Cattle density and dipping methods differed at the two study sites and may therefore play a role in the time-course of infection.

16S rRNA-based bacterial community profiling of *Haemonchus contortus* infecting Dohne Merino sheep using next-generation sequencing

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Next-generation sequencing, or metabarcoding, has revolutionized the study of microbial communities in humans, animals and the environment. However, only few studies have applied these approaches to identify parasitic helminth microbial communities. Previous studies have shown that gastrointestinal nematodes (GIN), in particular *Haemonchus contortus* harbour bacterial communities associated with their different life cycle stages. In this study, we aimed to characterise bacterial populations of *H. contortus* and the *H. contortus* resistant and susceptible sheep using 16S sequencing. A total of 10 (6 resistant, 3 susceptible and 1 unclassified) Dohne Merino sheep were slaughtered and *H. contortus* and abomasum contents were sampled, of which 4/6 resistant sheep had *H. contortus*; 3/3 of the susceptible had *H. contortus* and unclassified sheep had no *H. contortus* present in the abomasum. In order to determine the bacterial community of *H. contortus* and sheep abomasum, 16S-based metabarcoding on Illumina Miseq Platform and Oxford Nanopore MinION sequencing platform were employed. Preliminary results from data generated by Oxford Nanopore MinION sequencing showed that the most abundant bacterial phylum detected in sheep abomasum was Firmicutes followed by Bacteroidetes and Proteobacteria which have also been reported to be abundant in *H. contortus* from literature. We present a comparative analysis of *H. contortus* and sheep abomasum microbiota.

Prevalence of *Ehrlichia ruminantium* in ticks collected from goats in Limpopo, South Africa

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Heartwater is a tick-borne disease that affects ruminants and wild animals in Africa south of the Sahara. It is caused by *Ehrlichia ruminantium* which is transmitted by the ticks of genus *Amblyomma* (*A. hebraeum* and *A. variegatum*). Heartwater, a great threat to livestock and economic development, is said to be endemic in the Vhembe district. However, very few studies have been conducted in the study region. Therefore, the present study sought to investigate the prevalence of *E. ruminantium* in the Northern region. A total of 106 *Amblyomma* ticks were collected in November 2017 from goats in three different villages. DNA was extracted, and a diagnostic conventional PCR was used for detection of PCS20 gene of *E. ruminantium*. One percent Agarose gel stained with ethidium bromide was used for visualization of the PCR products and the results were recorded. The overall prevalence of *E. ruminantium* in the ticks was 23% (24/106). The distribution of *E. ruminantium* in Nkomo, Tshaulu and Tshidzini was 20% (10/51), 38% (8/21) and 18% (6/34), respectively. The infection was found to be high in old animals (36%) compared to young animals (18%). Male *A. hebreum* were found to be most commonly infected with *E. ruminantium*. This study showed that *E. ruminantium* is prevalent in the study population, infecting both young and old animals, which may lead to a loss in livestock. Therefore, people should be educated about animal dipping to prevent the ticks from feeding on these animals; this practice could help to reduce/eradicate the disease. Further studies are needed to understand the circulating strains of *E. ruminantium* in the study population, since knowledge of these strains could help in vaccine development.

Using a real-time PCR protocol to screen for avian haemosporidian parasites

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Avian haemosporidian parasites of the genera *Plasmodium*, *Haemoproteus* and *Leucocytozoon* are widely distributed vector-transmitted blood parasites that can affect the fitness (survival and reproductive success) of birds. Although extensively studied in other parts of the world, there are gaps in our understanding of avian haemosporidian parasites in South Africa. Prevalence studies frequently use microscopy and/or the nested Polymerase Chain Reaction (PCR) protocols for detecting avian haemosporidian parasite infections. Recent studies have used newer PCR protocols to successfully identify the presence of haemosporidian parasites in birds in a single reaction, with a distinct advantage of reducing screening cost and time. Using genomic DNA extracted from 300 bird blood samples collected in the Kruger National Park, we conducted a real-time PCR (qPCR) protocol with primers R330F and R480RL designed from another study. Microscopic screening of these blood sampled showed a prevalence of 34%. Preliminary results (50 blood samples) revealed that the qPCR protocol successfully identified avian haemosporidian infections. Results of ongoing molecular analyses of the remaining 250 blood samples are presented. This study was the first to use a qPCR protocol to screen for avian haemosporidian parasites in South Africa. The study provides an opportunity to adopt new technologies with increased sensitivity and accuracy in avian parasitology studies.

Molecular characterization and distribution of rodent haemoparasites in the Lowveld regions of South Africa

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Haemoparasites are vector-borne parasites infecting many wildlife species, some of which can potentially have an impact on their population dynamics. They persist in the vascular system of animals. At a population level, haemoparasites affect their hosts by reducing fitness parameters such as body condition, reproductive success and survival. Haemoparasites are widespread in wild rodents. Rodents form an important part of South Africa's biodiversity and monitoring them is a relatively quick and inexpensive method of indicating ecosystem health and function. Several emerging zoonotic diseases may pose serious threats to public health We sampled rodents is several locations in the Lowveld region including sites in the Kruger National Park, and assessed the rodent blood for *Trypanosoma* spp, *Babesia* spp, *Theileria* spp*., Rickettsia* spp. etc. The represented rodent species were *Aethomys chrysophilus, Lemniscomys rosalia, Mastomys* spp and *Tatera leucogaster*. Blood smears were screened microscopically. Polymerase Chain Reaction and the Enzyme Linked Immunosorbent Assay were also used. Findings were used to characterise the prevalence of haemoparasite in rodents in this disease-endemic subtropical area in terms of species, locality, land-use and seasonal differences..

Development of a real-time PCR assay to differentiate *Ehrlichia/Anaplasma* spp. in dogs

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Ehrlichia/Anaplasma species are infectious pleomorphic Gram-negative bacteria which cause Ehrlichiosis/ Anaplasmosis in human, domestic and wild animal species. Ehrlichiosis and Anaplasmosis are potentially fatal tick-borne diseases. Although direct detection of the bacteria antigen in using ELISA has had success in diagnosis of disease, a challenge remains in dogs where co-infection of infectious agents is common. Cross-reactivity of the serology assays makes it difficult to make species-specific diagnoses. Thus a more sensitive and reliable molecular technique that can detect and identify pathogens at species level is needed to enhance disease diagnosis. Real-time PCR is highly sensitive but has limited multiplexing capabilities. In this study, we developed *Ehrlichia/ Anaplasma* spp. real-time PCR group-specific primers and species-specific TaqMan® MGB probes that can differentiate between *Ehrlichia canis and Anaplasma sp.* Dog targeting the 16S ribosomal RNA gene. We envisage developing these assays further by including other *Ehrlichia/Anaplasma* spp. and transferring the assays to a bead-based multiplex assays (Luminex xMAP) that is faster, reliable, specific and has a greater multiplexing capability than PCR. This technique allows testing of large number of analytes within a single biological sample with more efficiency, speed and dynamic range better than ELISA. This will be a great tool for early disease diagnosis, which will also aid in administering of correct treatment intervention.

Molecular characterization of haemoparasites infecting livestock in uMkhanyakude district

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The most important vector-borne diseases in sub-Saharan Africa include theileriosis, babesiosis, trypanosomosis, hepatozoonosis, toxoplasmosis and besnoitiosis. However, despite the considerable economic loss associated with these haemoparasitic diseases, information on their epidemiology in South Africa is inadequate. Therefore, this study was aimed at improving the current knowledge about occurrence and genetic diversity of haemoparasites in livestock from north-eastern KwaZulu-Natal. A total of 208 blood samples collected from apparently healthy animals in five different municipalities were tested using previously established PCR techniques for the detection of *Besnoitia besnoiti, Theileria* sp.*, Babesia* sp.*, Hepatozoon canis, Trypanosoma* sp. *Toxoplasma gondii*, species-specific genes encoding ITS1, 18S, B1, *RAP*-1 and *gp45* restriction fragment, respectively. Preliminary overall infection rates of *T. ovis, B. ovis, B. bigemina, B. bovis* and *Trypanosoma* sp. were 3 (6%), 3 (6%), 15 (13.76%), 11 (10.09%) and 12 (11%), respectively. Co-infection of two pathogens were detected in 4 (3.67%) for *B. bovis* and *B. bigemina*, 1 (2%) for *T. ovis* and *B. ovis*. PCR results were confirmed by sequencing amplicons of positive samples which matched with their respective parasites genes in the NCBI GenBank database. This is a preliminary data and the study is continuing and we hope it will play a role in understanding the occurrence of haemoparasites in KZN and ultimately contribute in formulation of control strategies.

Anaplasma spp and *Hepatozoon* spp from free-ranging black-backed jackals (*Canis mesomelas*) in South Africa

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Black-backed jackals, which are common and widespread in Southern Africa, have recently been shown to be natural hosts of *Babesia rossi*, the causative agent of virulent canine babesiosis in domestic dogs in sub-Saharan Africa. A large set (n=142) of blood specimens from free-ranging jackals from Mogale's Gate, on the border between Gauteng and North West Province, was available for screening for occurrence of other tick-borne protozoa and rickettsias. On reverse line blot, 82 (57.8%) specimens reacted with the *Ehrlichia/Anaplasma* genera-specific probe. Contrary to expectation, no specimens reacted with the *Ehrlichia canis* probe. Parasite 16S rRNA genes of specimens that tested positive on the RLB assay were subsequently amplified, cloned and the recombinants sequenced. Ten 16S rDNA sequences were obtained: 9 were identical (1 323 bp), the other sequence differing by 1 bp. BLASTn homology search results revealed no identical sequences in the public databases. The most closely related sequences, with approximately 99% identity were *Anaplasma* sp. South African Dog, various uncultured *Anaplasma* spp., as well as various *A. phagocytophilum* strains. This may pose a risk, however slight, to human health. Ninety-one specimens were screened for haemogregarines through PCR amplification using the 18S rRNA gene; 20 (21.9%) specimens reacted positively, of which 14 (15.4%) were confirmed positive for *Hepatozoon* genotypes from within *H. canis*. Two (2.2%) specimens were found positive for two different *Hepatozoon* genotypes.

Molecular and serological assessment of *Toxoplasma gondii* in definitive and intermediate hosts at the National Zoological Gardens of South Africa.

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Toxoplasmosis is a cosmopolitan zoonotic disease that affects approximately one third of the human population. The aetiological agent is *Toxoplasma gondii,* a coccidian microparasite of the family Sarcocystidae, phylum Apicomplexa, with a wide host range and several transmission methods .Infection is usually asymptomatic in healthy individuals but can result in debilitating disease in immunocompromised hosts. Horizontal transmission occurs by ingestion of oocysts from the environment, contaminated food or ingestion of tissue cysts in undercooked infected meat. Vertical transmission can occur from an infected female to its foetus during pregnancy. Felids play a pivotal role in the epidemiology of toxoplasmosis since they are the only definitive hosts of *T. gondii* and excrete infectious oocysts in their faeces. Previous cases of toxoplasmosis in lemurs at the Zoological Gardens of South Africa (NZG) resulted in death of these animals. This highlighted the need for a more thorough investigation of *T. gondii* infections at the zoo in order to implement effective prevention and control strategies. The aim of this study was to determine the occurrence of toxoplasmosis in selected definitive and intermediate hosts and feral cats at the NZG, and to characterise *T. gondii* strains. Blood and tissue samples (n= 152) from various zoo animals and suspected toxoplasmosis post-mortem cases were obtained from the NZG Biobank. The prevalence of infection was determined using nested PCR assays that target the 529bp repeat element (RE) and P30 gene of *T. gondii*, and a conventional PCR and real time PCR assay that target the B1 gene of the parasite. The sensitivities of these methods in detecting *T. gondii* infections will be compared. Strain genotyping of *T. gondii* was done in selected positive samples by Multi locus nested Poylmerase Chain Reaction Restriction Fragment Length Polymorphism (Mn-PCR RFLP), and high resolution melt (HRM) assays. Samples from intermediate and definitive hosts were analysed by an ELISA to determine previous exposure of these animals to *T. gondii*. Preliminary PCR results indicated that the prevalence of *T. gondii* infections in zoo animals and post-mortem cases is low. However, occurrence of *T. gondii* infections in zoo animals should be monitored to prevent disease outbreaks.

Parasites of the leopard (*Panthera pardus*) in South Africa: A review

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Leopards (Carnivora: Felidae) are the largest of the spotted felids found in Africa. Globally their population is declining (currently ‘vulnerable’ based on IUCN) due to anthropogenic factors such as habitat loss, decline in prey, poaching as well as conflict with farmers. Diseases may also contribute to this decline. However, limited information is available on leopard diseases and parasitic infections from South Africa. During 2016-2017 four leopards (three roadkill; one darted) were examined for parasites (only the darted one was examined for ectoparasites while all four for endoparasites). A tick species (*Haemaphysalis* sp.) was recovered, cestodes were found in the intestine of three leopards while two had nematodes in their stomach (*Cylicospirura* sp.) and small intestine (*Ancylostoma* sp.). The adult male leopard infected with *Cylicospirura* sp. (n=19) had gastric nodules that were collected for histopathology. Such pathology in the gastrointestinal tract can influence digestion and nutrient uptake. Furthermore, nodules have been re-ported to induce regurgitation by the host. A review on parasites of leopards reported in South Africa is included and the research gaps are identified.

Helminth parasites of *Crocodylus niloticus* (Reptilia: Crocodylidae) in the Kruger National Park, South Africa

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Sixteen Nile crocodiles were collected in the Kruger National Park, South Africa and vicinity during 2010 and 2011. The peritoneal and abdominal cavities, as well as the oesophagus were macroscopically examined for parasites. Aliquots were prepared from the ingesta of the stomach and small intestine. A total of 13 nematode taxa representing six families were recovered from the crocodiles. Nematodes infected all hosts, with species richness ranging from 1–7 (3.2). Heterocheilids were the dominant group, comprising five species, with *Dujardinascaris madagascariensis* being the most prevalent (75%), followed by *Ingwenascaris sprenti* (68.8%), which was also the second most numerous nematode. While less prevalent (31.3%), *Typhlophoros kwenae* was the most abundant species. *Micropleura huchzermeyeri* (Micropleuridae) was collected from the peritoneal and abdominal cavity and *Crocodylocapillaria* sp. (Capillariidae) occurred in the stomach of a single host. Three nematodes, *Camallanus* sp., *Spirocamallanus* sp. (both Camallanidae) and *Ascarophis* sp. (Cystidicolidae), are considered accidental infections, likely ingested with the hosts' prey. Similarly, metacestodes of *Cyclustera magna* (Gryporhynchidae), found in the stomach of a single crocodile, are typically parasites of fishes. The leech *Placobdelloides multistriata* (Hirudinea: Glossiphoniidae) was removed from the oesophagus of a single host and all crocodiles harboured a variety of digenean trematodes. Our findings of *D. dujardini*, *D. madagascariensis* and *M. agile* in Nile crocodiles in South Africa constitute new geographic records. *Typhlophoros kwenae*, *I. sprenti*, *M. huchzermeyeri* and *Crocodylocapillaria* sp. represent new host and geographic records, the former three having been described as new species from the crocodiles currently under discussion. No statistically significant differences were found in the prevalence and abundance of nematodes between male and female crocodiles.

Species composition and the role of horse-flies in pathogen transmission in south-eastern KNP (Diptera: Tabanidae)

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Tabanidae, commonly known as horse-flies, are a large family of important pollinators, generally believed to be among the most basal brachycerans (Diptera: Brachycera). In addition to their important ecosystem services, the females are haematophagous and capable vectors of pathogens. Despite their importance, the family has been severely neglected by science. The current study was aimed at determining the species composition of tabanids in south-eastern Kruger National Park. Using three different traps, we sampled tabanids in four habitats in south-eastern Kruger National Park (KNP) South Africa. Amongst the three types 14 traps were used, namely: Manitoba (6X), Ngu (4X) and H traps (4X). Manitoba trap captured an average of 1.7/trap/day flies, Ngu 0.7/trap/day and H trap 2.4/trap/day. A total of 247 flies were captured by the H trap, making it the most effective. Morphological analyses revealed five different genera, namely: *Haematopota, Tabanus, Philoliche, Chrysops and Atylotus* .Thirteen species were collected, of which the dominant species, *Tabanus minuscularius*, accounted for 55% (136/247) of the total flies sampled. Investigations to determine the role of tabanids in pathogen transmission in south-eastern Kruger National Park are on-going.

Parasitic helminths of rodents in South Africa: Host range and spatial distribution

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Descriptive information regarding host range and geographic distribution of parasitic helminths associated with naturally occurring rodents in South Africa is scarce. We commenced with a countrywide study to: 1) record the species and host range of helminths associated with these rodents, and 2) provide baseline data on the spatial distribution of helminths across the country. In total, 56 helminth taxa were recovered from at least 13 rodent species (n=1032) from 26 localities. The helminth taxa represented 25 genera, 15 nematodes, nine cestodes and one acanthocephalan. The most abundant and prevalent group were the monoxenous nematodes, while the incidence of heteroxenous nematodes and cestodes was generally lower. Single helminth species infections were common, although hosts with mixed helminth species infections were also recorded. *Rhabdomys dilectus*, collected from 10 localities, had the highest helminth diversity harbouring 26 helminth taxa, *Rhabdomys pumilio*, also from 10 localities, harboured 16 helminth taxa and *Gerbilliscus brantsii*, from a single locality, 11 helminth taxa. Six host species (46%) were infected with the nematode *Heligmonina boomkeri* with a mean abundance of 1.7±0.3 and seven host species (54%) with the cestode *Rodentolepis microstoma* with a mean abundance of 0.04±0.02. The study reported several novel helminth-host associations. A total of 11 helminth taxa were identified as previously unrecognised species. Monoxenous nematodes and some cestodes were recovered countrywide whereas heteroxenous nematodes were restricted to the eastern regions of South Africa, possibly due to the climate and vegetation being conducive to sustain large and diverse arthropod populations. The study highlighted the diversity of helminth species associated with naturally occurring rodent species and provides initial data on their spatial distribution in South Africa.

Diversity of the sporozoite antigen gene p67 in *Theileria parva* field isolates from cattle and buffalo in southern and eastern Africa

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Cattle theileriosis is a tick-borne disease caused by the haemoprotozoan parasite *Theileria parva* whose natural reservoir is the African buffalo. It is transmitted by the brown ear tick *Rhipicephalus appendiculatus*. East Coast fever (ECF) caused by the cattle-derived *T. parva* and Corridor disease (CD) caused by buffalo-derived *T. parva* are considered to be the most economically important tick-borne diseases of cattle in eastern Africa and South Africa, respectively. The gene encoding sporozoite surface antigen p67 of *T. parva* has been explored for the development of a recombinant vaccine. Vaccination of cattle using a subunit vaccine based on recombinant versions of p67 reduced severity of infection by approximately 70% in the laboratory and 30% under field tick challenge. Since heterogeneity of *T. parva* field isolates among other parasite dynamics contribute to differences in immunity, we assessed the diversity of p67 in buffalo- and cattle-derived *T. parva* isolates from South Africa, Mozambique, Kenya, Tanzania and Uganda. A 900bp fragment was amplified from DNA extracted from blood of *T. parva* positive samples, cloned and sequenced. Analysis of sequence data revealed four alleles from buffalo-derived isolates, and a single allele (allele 1) in the cattle-derived isolates from Kenya, Uganda and Tanzania. The South African cattle, CD isolates obtained from clinical cases had four alleles while carriers had one allele (allele 4). Analysis of two p67 immunodominant B cell epitopes in buffalo-derived isolates, including CD isolates from clinical cases in South Africa, revealed nine types of SNPs on epitope 1 (^{169}TKEEVPPADLSDQVP^{183}) and six on epitope 2 (^{209}LQPGKTS^{216}). However, both epitopes were conserved in the cattle-derived (ECF) isolates. For p67 subunit vaccine to confer protection against both buffalo- and cattle-derived *T. parva* infections, common conserved p67 B cell epitope(s) are significant. These findings therefore display the implications on the use of p67 subunit vaccine against buffalo-derived *T. parva*.

Oral administration of azithromycin ameliorates trypanosomosis in *Trypanosoma congolense* and *T. brucei brucei*-infected mice

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African trypanosomosis, a devastating disease of animals caused by parasites of the genus *Trypanosoma*, negatively affects the economic status of at least 36 African countries. Few available drugs for the treatment of trypanosomosis remain accessible in remote areas, are associated with severe toxicity and most importantly, resistance has widely developed against their usage. Therefore, safe, effective and easily administrable drugs are urgently needed. The objective of the current study was to determine efficacy of azithromycin (AZM), on *T. congolense* and *T. brucei brucei.* A 96-well luciferase assay was conducted in order to determine the trypanocidal effect of AZM on *T. congolense* and *T. b. brucei* as well as the cytotoxicity effect on the MDBK and NIH 3T3 cells. Additionally, TEM analysis was conducted to determine the morphological alteration of the AZM-treated trypanosomes. Mice were infected with *T. congolense* and *T. b. brucei* and orally treated with AZM for 7 and 28 days (referred to as the short and the long-term treatment). The *in vitro* IC_{50} values of AZM on *T. congolense* and *T. b. brucei* were 0.19 ± 0.17 and 3.69 ± 2.26 µg/mL, respectively, while the cytotoxicity effects values were > 25 µg/mL. A vacuole-like structure was observed in TEM images of AZM-treated *T. congolense*, while the presence of glycosomes and acidocalcisome-like structures were observed in *T. b. brucei*. AZM was more effective against *T. congolense*-infected mice than *T. b. brucei*, *in vivo*. Short-term treatment with AZM on *T. congolense* resulted in a relapse post–treatment, while long-term treatment resulted in trypanocidal activity at 300 and 400 mg/kg with observed survival rates of 80 and 100%, as compared to 70 and 70% survival rate observed in the long-term treatment of *T. b. brucei*-infected mice for the same AZM concentrations. In conclusion, AZM exhibited higher trypanocidal effects on *T. congolense*-infected mice as compared to *T. b. brucei*-infected mice.

Ethno-veterinary medicine practices for the treatment of tick-borne diseases by rural farmers in the Eastern Cape Province, South Africa

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South Africa is recognised as one of the most biodiverse countries in terms of fauna and flora in the Sub-Saharan region. More than 40% of farmers, who live in remote rural areas of the Eastern Cape Province, use medicinal plants for the treatment of different livestock ailments. The present study was conducted to assess the existing knowledge of ethno-veterinary medicine practices specifically for the treatment of tick-borne diseases in livestock. Semi-structured interviews were conducted among 48 respondents between November 2013 and February 2014. Nine plant species belonging to eight families were found to be used to treat three different tick-borne diseases (anaplasmosis, babesiosis and heartwater) in livestock. Most of the plant species used were from the families Xanthorrhoeaceae, Ebenaceae, Proteaceae, Malvaceae, Asteraceae, Vitaceae, Loganiaceae and Iridaceae. The most dominant life form of these plant families used were trees (55.6%), followed by herbs (33.3%) and shrubs (11.1%). The most frequent used plant part was the leaf (44.4%). Bark, root, stem, seed and bulb, contributed 11.1% respectively. The most common preparation methods were decoction (77.8%) and infusion (22.2%). Oral (100%) was the only used method of administration. The study showed that people in rural areas have preserved some knowledge of ethno-veterinary practices for the treatment of tick-borne diseases. However, the plants used for the treatment of these diseases need to be validated using standardized procedures in order to evaluate efficacy, safety (toxicity), quality (phytochemicals) and dosage standards.

The metal chelating properties of terpenes and effect on *Plasmodium* malaria and *Anopheles* vector

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A delicate balance is required to maintain an optimal metal homeostasis within the malaria parasite, where the parasite relies on its human host to provide key anti-oxidant enzymes, as well as iron and copper for critical enzyme function. As such, interruption in the homeostasis of either metal could kill the parasite. Several metal chelators have been evaluated as novel antimalarial agents, however with limited success. A potential class of metal chelators includes the lipophilic terpenes contained within the volatile, complex essential oils produced by plants. Aromatic plants are used as mosquito repellents and traditionally to treat patients with “fever” or “flu-like” symptoms. These oils have been reported to possess antimalarial and insecticidal activity, but there is limited data on their ability to interact with metals and as such have a potential inhibitory effect on the metal homeostasis of the malaria parasite or *Anopheles* vector. Eight structurally related terpenes including various isomers/enantiomers were evaluated for their metal chelating/reducing and free radical scavenging properties. The terpenes were evaluated for antimalarial activity, haemolytic effect and inhibition of haemozoin formation. The larvicidal activity against *Anopheles arabiensis* was compared to the mortality of *Artemia franciscana* nauplii. The cytotoxic effects were assessed on the human neuroblastoma cell line. The isomers/enantiomers displayed contrasting ability to either chelate copper, where (-)-α-pinene and (+)- and (-)-β-pinene potently chelated copper(I), whilst (-)-fenchone effectively chelated copper(II). None reduced either copper or iron, but (+)-β-pinene and (+)-α-pinene chelated ferrous ions. The terpenes had no free radical scavenging properties or able to prevent lipid peroxidation. Of the eight terpenes, (+)-α-pinene was the most potent against *P. falciparum*, by inhibiting haemozoin formation. The inhibitory effect by the terpenes was directed against the intra-erythrocytic parasite as indicated by minimal haemolysis. 1,8-Cineole and the pinenes killed the *Anopheles* larvae, whilst only (+)-β-pinene was toxic to the *Artemia* nauplii. An isomeric effect was observed with the α-pinenes inhibiting the neuroblastoma cells, compared to a minimal effect by the β-pinenes. These novel metal chelators warrant further investigation into possible use in metal overload pathologies or as antimalarial and insecticidal agents.

Investigation of the role of Hsp70 in Granzyme B-mediated in malaria therapy

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Plasmodium falciparum Hsp70-x is the only Hsp70 protein that is exported to the red blood cell (RBC) cytosol. Although PfHsp70-x is not essential for parasite survival, it is implicated in the development of malaria pathogenesis. This is largely on account of its association with P. falciparum erythrocyte membrane protein 1 (PfEMP1), an important virulent factor that is exposed to the exterior of the infected RBC (iRBC). However, the iRBC also contains Hsp70 (hHsp70) of human origin. Granzyme B (GrB) is a serine protease found in lytic granules of natural killer (NK) cells and cytotoxic T lymphocytes. GrB has been presented as a potent antimalarial. However, its mechanism of action remains poorly understood. In tumour cells, cell surface-bound Hsp70 is known to sensitise the cells to cytolytic attack that is mediated by NK cells. Cell surface bound Hsp70 is thought to recruit NK cells and GrB via its 14 amino acid sequence, TKDNNLLGRFELSG, known as the TKD motif. Both PfHsp70-x and hHsp70 contain a TKD motif. Thus, this study seeks to investigate the role of Hsp70 in facilitating the selective targeting of malaria parasite-infected RBCs by GrB. To this end, we expressed and purified recombinant human Hsp70 and PfHsp70-x and investigated the direct interaction of the two proteins with recombinant GrB using ELISA and SPR-based analyses. We further investigated the antimalarial activity of GrB. Our findings suggest that both PfHsp70-x and human Hsp70 directly interact with GrB. In addition, GrB exhibits potent antiplasmodial activity (IC50 of 0.5μM). Our findings suggest that GrB is taken up by Hsp70 (both of parasite and human origin) resident in the iRBC. Furthermore, GrB is a promising antimalarial agent. We intend to validate the presence of both human Hsp70 and PfHsp70-x on the surface of iRBC towards validating their role in uptake of GrB by iRBC.

Anthelmintic resistance in gastrointestinal nematodes of sheep in Limpopo Province, South Africa

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Anthelmintic resistance (AR) is a serious threat to animal health and has a major impact worldwide due to production and financial loss. The aim of this study was to determine occurrence of AR in nematodes of sheep in five districts of Limpopo Province, namely; Sekhukhune, Capricorn, Waterberg, Mopani and Vhembe using both *in vivo* and *in vitro* methods. A faecal egg count reduction test (FECRT) was used to determine AR against ivermectin (Ivomec®, Merial, 0.2 mg/kg), levamisole (Tramisol Ultra®, Coopers and Intervet®, 5 mg/kg) and albendazole (Valbazen®, Pfizer, 7.5 mg/kg). The egg hatch assay (EHA) was used to determine AR against thiabendazole (TBZ) and Micro Agar Larval Development Test (MALDT) was used for TBZ and Levamisosle (LEV). All three classes of drugs tested in all the districts had ≤ 95% FECR and ≤ 90% lower confidence interval (LCI). These results suggest occurrence of AR in all the districts, except in Sekhukhune where AR was suspected for LEV as 93% FECR and 99% LCI was recorded. The EHA results showed that TBZ had a minimal ovicidal effect (□1% hatchability) on nematode eggs at a discriminating dose (DD) of 0.1 µg/ml for all districts. The MALDT showed no AR against LEV as 100% larval development inhibition was recorded in all districts at 0.5 µg/ml DD. However, AR was detected against TBZ as development from L_1–L_3 was recorded in Sekhukhune, Capricorn and Waterberg districts at 6, 22 and 28%, respectively, at 0.02 µg/ml DD. Except for the similar results obtained for AR against the benzimidazole class (albendazole and TBZ), there was no correlation between FECRT and MALDT results in all the districts. However, a strong correlation existed between FECRT and EHA results as both tests confirmed the occurrence of AR in all the districts except for LEV in Sekhukhune. Further studies on risk factors associated with AR are therefore recommended in Limpopo Province.

Characterisation of cytosol-localised Hsp70 isoforms from *Plasmodium falciparum* and their prospects as potential antimalarial drug targets

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Heat shock proteins are conserved molecules whose main function is to facilitate protein folding. Heat shock protein 70 (Hsp70) is one of the most distinct families of heat shock proteins which is implicated in cytoprotection. The chaperone (protein folding) role of heat shock proteins is important in the survival of malaria parasites in their host. This is because the malaria parasites survive under physiologically distinct life-stages during its stint in the host. The variable physiological conditions under which it survives added to host defence responses requires that the parasite employs a robust protein-folding system to ensure proteostatic maintenance. Periodic fever associated with malaria adds further strain to the proteostatic stability of malaria parasites. One mechanism by which malaria parasites evade effects of cell stress is by upregulating the expression of certain heat shock proteins. The main agent of malaria, *Plasmodium falciparum*, expresses 6 Hsp70 proteins. Of these two (PfHsp70-1 and PfHsp70-z) are localised to the parasite cytosol. In the current study, we biochemically characterized the chaperone functions of the two proteins. In addition, we observed that the two proteins interact in a nucleotide-dependent fashion. Whereas the chaperone function of PfHsp70-1 is influenced by ATP, we established that the chaperone function of PfHsp70-z is nucleotide-independent. This suggests that the mechanism of action of their chaperone function is uniquely regulated. Structurally, PfHsp70-z belongs to the Hsp110 subfamily of the Hsp70 superfamily. For this reason, we speculate that PfHsp70-z could serve as a nucleotide exchange factor of PfHsp70-1 as Hsp110 proteins are thought to regulate nucleotide exchange function of their canonical Hsp70 counterparts. PfHsp70-z is the sole possible nucleotide exchange factor of PfHsp70-z based on parasite genomic information. This could present a bottle-neck for the design of possible inhibitors targeting it as its function is essential for parasite survival. To this end, we have identified inhibitors for both PfHsp70-z and PfHsp70-1 towards their development as possible antimalarial inhibitors.

Antimicrobial activity of selected Venda medicinal plants against *Entamoeba moshkovskii*

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Plants have been used as a medication for years all over the world for treatment of various diseases. There is a continuous and urgent need to develop new drugs against parasites such as *Entamoeba* spp which represent an important cause of morbidity and mortality throughout the world. The present study determined the activity of selected plants on *Entamoeba moshkovskii.* Nine plants, namely *Bridelia micrantha, Combretum molle, Mimusops obovata, Peltophorum africanum, Terminalia sericea, Ozoroa paniculosa, Ornithogalum ornithogaloides*, *Wrightia natalensis* and *Carpobrotuse edulis*, were tested for activity against *E. moshkovskii* cells using a microditulion procedure. Six plants, i.e. *M. obovata, P. africanum, O. paniculosa, O. ornithogaloides*, *W. natalensis* and *C. edulis*, were active against *E. moshkovskii* cells, with killing time less than 24hours. The methanol and acetone extracts were more active than ethyl acetate extracts with the IC90 of 2µg/ml. These plants could be used for the production of lead compounds that could help control *E. moshkovskii* in particular and *Entamoeba* spp in general. Further studies are warranted for the purification and identification of active compounds.

You are what you eat: Trace element and metal accumulation in a host–parasite relationship from the Vaal Dam, South Africa

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Parasitism is recognised as a common consumer strategy, although the interaction of parasites in communities and ecosystems is generally poorly understood. Studies have shown that helminths, such as acanthocephalans and tapeworms, are able to accumulate heavy metals to levels which are several times higher than concentrations in host tissues. This is achieved through the interruption of hepatic reabsorption of metals excreted from the liver with bile salts. This therefore suggests that heavy metal accumulation by endoparasites is in some way related to the acquisition of nutrients. As parasites are integral parts of food webs, analysis of the trophic interactions between parasites and hosts in combination with analysis of heavy metal content in each organism was assessed as a means of better understanding the fractionation of heavy metals between hosts and tapeworms. Largemouth yellowfish (*Labeobarbus kimberleyensis*) infected with the Asian tapeworm (*Schyzocotyle acheilognathi*) were collected from the Vaal Dam. Using stable isotope signatures for carbon ($^{13}C/^{12}C$) and nitrogen ($^{15}N/^{14}N$), the trophic relationship between host and parasite was assessed and compared to heavy metal accumulation. Analysis of stable isotopes signatures was performed for host muscle tissue and cestodes using an elemental analyser coupled with an isotope ratio-mass spectrometer (EA-IRMS) and trace element and heavy metal concentrations in hosts and parasites were analysed by inductively coupled plasma–mass spectrometry (ICP-MS). Comparison between isotope signatures and trace element and metal concentrations between host and cestode suggests that elements accumulated by parasites are sequestered from the host and are not simply taken up from the food available as gut content.

Investigating relationships among different facets of host specificity in haematophagous ectoparasitic arthropods

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Host specificity is a fundamental trait of a parasite species. Recently, multiple aspects of host specificity have been recognized, but the relationships between these facets are still poorly understood. Here, we studied pairwise relationships between basic, structural, phylogenetic and geographic host specificity in three taxa of haematophagous ectoparasites that differ in tightness of their association with the host. We asked which metrics of host specificity are correlated within each parasite taxon and whether the patterns of the association between different facets of host specificity are similar among parasite taxa. Basic, structural, phylogenetic and geographic specificity indices were calculated for 18 bat fly species recorded on 40 host species from 15 regions, 109 flea species recorded on 120 host species from 51 regions and 34 mite species recorded on 67 host species from 28 regions. Then, we tested for the correlation between any two measures of host specificity using model II regressions. We found that structural and basic specificity, as well as structural and geographic specificity, exhibited a positive association in all three taxa. However, basic and geographic specificity, as well as basic and phylogenetic specificity, were significantly positively associated in fleas but did not correlate in bat flies or mites. In addition, we found a significant negative association between structural and phylogenetic specificity in bat flies but no association in the remaining taxa. Moreover, geographic and phylogenetic specificity were not associated in any taxon. Our results suggest that different facets of host specificity were shaped differently by natural selection in different taxa.

Establishment of the interacting partners of *Plasmodium falciparum* heat shock protein 70-x (PfHsp70-x)

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Plasmodium falciparum Hsp70-x is a molecular chaperone (protein-folding facilitator) that is exported to the red blood cell at the erythrocyte stage of malaria parasite development in the human host. PfHsp70-x, though not essential is implicated in parasite virulence. For example, it is thought to associate with the virulent protein, *P. falciparum* erythrocyte membrane protein 1 (PFEMP1). Thus, PfHsp70-x is thought to modulate fold and function of some proteins that are implicated in the pathogenesis of *P. falciparum*. In addition, PfHsp70-x has been found to associate with proteins of human and parasite origin that are localised to the infected red blood cell. However, the global interactome of PfHsp70-x is yet to be determined. In the current study, we sought to elucidate the interacting partners of PfHsp70-x. In order to map out the interactome of PfHsp70-x, we conducted bioinformatics-based analyses and biochemical assays. Our study identified human Hsp70-Hsp90 organising protein (hHop) as a potential interactor of PfHsp70-x. In addition, we identified a distinct band representing a protein species of approximately 35, 70 and 170 kDa as a possible interactor of PfHsp70-x. We intend to conduct LC-MS analysis to further validate the possible interactors of PfHsp70-x. The possible interaction of PfHsp70-x with hHop suggests that PfHsp70-x could mediate partnership between PfHsp70-x and human Hsp90. However, this requires confirmation.

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Posters abstracts

Rhabdochona esseniae from *Labeobarbus aeneus* in the Vaal River with additional information on the morphology

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The Vaal-Orange smallmouth yellowfish, *Labeobarbus aeneus* (Burchell, 1822) is widely distributed in South Africa. Very limited information is available about the intestinal parasites of this species. Fish were collected with gill nets at Yellowfish Paradise near the town of Parys, on the Vaal River. After severing the spinal cord each fish was dissected and the mesenteric cavity and gastrointestinal tract were examined for nematodes. The nematodes were killed and fixed in warm glacial acetic acid (30°C) and preserved in 70% ethyl alcohol. The morphology of these parasites was studied with light and scanning electron microscopy; specimens for light microscopy were cleared in lactophenol, and for scanning electron microscopy (SEM), they were dehydrated in ethanol and dried in hexamethyldisilazane. Only *Rhabdochona esseniae* Mashego, 1990 previously reported from *Enteromius lineomaculatus*, *Enteromius trimaculatus* and *Labeobarbus marequensis*in the Limpopo and Olifants drainage systems was recovered. The morphology concurs with the species description and SEM clarified the structure of the V shaped deirids situated on the exterior and posterior to the buccal capsule. The anterior prostomal teeth in adults of *R. esseniae* is described for the first time. Light micrographs of the posterior end of male nematodes highlights key taxonomic features such as postanal papillae and spicules of unequal length; the short spicule bears a dorsal barb posteriorly. The posterior end of the female is characterised by a conical tail. The additional information enables placement of this species in the phylogenetic tree of the genus that is already available for the rest of the world.

The impact of IL4 serum concentration on the seroprevalence of schistosomiasis and chlamydia among HIV patients in Limpopo Province, South Africa

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After Malaria and intestinal helminthiasis, schistosomiasis is the most prevalent tropical disease in the world. The immune response to Schistosoma infection is characterized by increased production of Th type 2 cytokines such as interleukin-4 (IL-4), IL-5, IL-10 and IL-13.Studies have showed an overlap in the epidemiology of Sexually transmited infections and schistosomiasis in women living in schistosomiasis endemic areas, However, there are no studies conducted on the role of Il-4 serum concentration in schistosomiasis disease progression and severity. Therefore, the present study aims to determine the sero-prevalence of schistosomiasis and chlamydia as well as to investigate the role of IL-4 serum levels on the occurrence of the diseases' antibody response. Patients were recruited from different hospitals and clinics from three major Districts in Limpopo Province. About 500 blood samples were collected from HIV positive individuals and screened using ELISA for antibodies against Schistosoma and Chlamydia. IL-4 serum levels were measured from the samples using commercially available ELISA kits for human Il-4 from MABTECH. Out of the 500 samples tested, 69.5% were females. The age varied from 1 to 77 years with a mean of 35.4±12.8 years. The CD4 count from the studied participants varied from 5 to 4881 with a mean of 465.2 ± 415.1 cells/mm3. The overall prevalence of chlamydia IgG and IgM were 59.2% and 29.9% while that of Schistosoma IgG was 60.7% respectively. The highest prevalence for schistosomiasis IgG was found among participants who are 60 years and above while that of chlamydia was among younger participants aged between 26-45 years. Chlamydia infection was significantly associated with female gender (p=0.002). The infections were not associated with any of the socio-demographic characteristics. A strong association between chlamydia IgG and schistosomiasis IgG was observed (p=0.002). Higher IL-4 levels were associated with female gender (p=0.033).The present study demonstrated a high prevalence of schistosomiasis and chlamydia in Limpopo Province. Schistosomiasis is more common in males than in females while chlamydia was more common in females than males. The study did not show any significant association between schistosomiasis and IL-4.

Molecular characterisation of potential vaccine candidates from *Anaplasma marginale* strains in South Africa.

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Bovine anaplasmosis, caused by *Anaplasma marginale*, is a globally important tick-borne disease, with annual losses from cattle mortalities in South Africa estimated at R115 million. Despite the economic impact of the disease, few studies on prevalence and control exist in the country. An*Anaplasma centrale* blood vaccineis available, but it does not protect against all *A. marginale* field strains and may also transmit other blood-borne pathogens. Outer membrane protein (OMP) preparations are known to induce immune protection in nearly all animals tested, thus demonstrating the potential efficacy of a subunit vaccine. Five potential OMP vaccine candidates from North American *A. marginale* strains have been identified and are well-characterised in *A. marginale* strains from USA. However, their levels of conservation in other countries must be ascertained in order to inform their use in a vaccine with regional or global efficacy. This study aimed at evaluating the presence and genetic diversity of *A. marginale* in South Africa, and characterising five OMP vaccine candidates in *A. marginale*-positivesamples. *Anaplasma marginale* and *A. centrale* were detected using a duplex quantitative real-time PCR in, respectively, 57% and 17% of 517 bovine blood samples, with 15% being co-infected. High genetic diversity of South African strains was revealedby *msp1α* genotyping; 190 genotypes were obtained from 99 Msp1a amino acid repeat sequences. While 22% of the 99 repeat sequences were detected in other countries, only two South African genotypes were identified elsewhere in the world. OMPs Am854 and Am779 were highly conserved, with 99–100% amino acid identity; Omp7, Omp8 and Omp9 were conserved with 79–100% identity with US strains. As has been shown previously, Omp7–9 possess conserved N- and C- termini, along with a central hypervariable region. A previously identified, highly conserved T-cell epitope, FLLVDDAI/V, was found in the N-terminus of these three OMPs. Western analysis of recombinant OMPs indicated strong antigenic relationships between South African and US strains of *A. marginale*, suggesting that these five *A. marginale* OMPs are good candidates for use in a novel global vaccine cocktail, although further work on the best formulation and delivery methods will be necessary.

Seasonal occurrence of roundworms in goats reared under communal farming areas in the Eastern Cape, South Africa

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This study was conducted to establish the seasonal occurrence of roundworms in goats reared at the Wartburg community near Stutterheim. The study will greatly contribute to develop a health management guide for controlling roundworms infestations in goats. The randomly selected farmers each contribute 10 female goats (2- tooth) for a total of 30 experimental goats. Feacal samples were collected monthly for four years (January 2012 - December 2015) before the onset of the trial the experimental animals were dewormed with a broad spectrum remedy. The animals were only dosed when the roundworms egg per gram (e.p.g) counts exceeded the levels above 3000 e.p.g. The roundworms were significantly different $P<0.01$ in autumn than in spring for the study period. Winter roundworms were also significantly lower when compared with summer levels of roundworms. The results showed presence of roundworms in all selected animals, but with significant differences between and within seasons. The highest e.p.g were observed in summer (1038.09±638.38) and spring (851.13±734.80) and the lowest in autumn (336.11±489.98) and winter (68.91±97.43). High levels of roundworms infestations should be anticipated during the hot wet months of the year. The study confirms that seasons and management had an impact on the roundworms e.p.g levels of communal goats.

In silico screening of the *Theileria parva* proteome for identification of proteins responsible for transformation of infected host lymphocytes

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Theileria parasites can be grouped into schizont transforming and non-transforming species and the former are the leading cause for mortality and morbidity in livestock worldwide. Transforming *Theileria* species infecting cattle include *T. parva* and *T. annulata* and very little is known about their genetic basis of the virulence. The availability of the genome sequences of these parasites has provided an opportunity to study their biology and pathogenic processes. Thus, the aim of this study is to investigate the *T. parva* proteome for identification of host cell phenotype modulators (HCPMs). Possible HCPMs were predicted using a combination of bioinformatics tools targeting secreted, membrane and cytoplasmic proteins. Tools for signal peptide, subcellular localization, homology, protein-protein interactions and domain/motif prediction were among those employed, namely SIGNALP, CELLO2GO, KEGG, BLASTp, STRING, SMART and Pfam. Signal peptide and subcellular localization prediction analysis resulted in the identification of 1188 proteins from the proteome of 4035. Subsequently, 977 proteins with homologs or orthologs or paralogs, in non-transforming parasites were negatively selected. The remaining 211 proteins were further analysed for protein-protein interactions (PPI) and domain prediction. The PPI analysis revealed 20 proteins with 10 interacting partners in the *T. parva* proteome, with some of the interacting partners associated with up to 18 proteins. Among interactors were heat shock (HSP90) and proteinase protein families which are associated with oncogenic pathways playing a significant role in replication, activation of the NF-kB complex and regulation of apoptosis and metastasis. Further analysis of domains/motifs in proteins without homologs/orthologs/paralogs revealed three dominating groups, FAINT also known as DUF529 (n=118), PEST motif (typical of Tash protein) (n=37) and DUF1430 (n=9). Proteins possessing domain DUF1430 are reportedly involved in immunity and proteins with PEST motif, typical of Tash AT-hook proteins, are associated with pathogenesis while those with FAINT domain are predicted to play a role in host cell modification. Considering the predicted functions of these proteins in the parasite and the host, it will be worth it to investigate them further as possible host cell phenotype modulators.

Prevalence of cysticercosis in cattle and pigs slaughtered in Gauteng abattoirs

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Cysticercosis is a muscular infection caused by metacestodes of the zoonotic parasites, *Taenia saginata* and *Taenia solium* in cattle and pigs respectively. Humans are final host of these tapeworms whereas cattle and pigs are intermediate hosts. Meat inspection has been the only test used at abattoirs as a diagnostic tool for cysticercosis; however, serological and molecular assays have been developed and are more sensitive and specific. This study was conducted to determine prevalence of cysticercosis in cattle and pigs slaughtered around Gauteng Province using meat inspection, serology and molecular tools. Demographic information on slaughter animals was obtained and records (4-5 years) of visual diagnosis of cysticercosis in Gauteng province were reviewed. Blood and tissue samples (masseter, diaphragm, heart and tongue) were collected from 323 cattle and 106 pigs in 18 Gauteng abattoirs. Visual inspection was conducted to detect cysts. Commercially available B158C11A10/B60H8A4 Ag-ELISA (apDIA Cysticercosis) with manufacturer's instructions (ApDia n.v, 2004) was used to detect antigens of Taenia infections and a real-time polymerase chain reaction (qPCR) was used to detect *T. saginata* and *T. solium* DNA in tissue samples. Records revealed that the prevalence of bovine and porcine cysticercosis between the year 2013 and 2017 ranged from 0.0002 to 0.7% in cattle and 0.0002 to 0.0006% in pigs. No cysts were observed during meat inspection; sero-prevalence of cysticercosis in cattle and pigs was 2.1% (7/323) and 0.99% (1/106) respectively. Analyses of the data from PCR assays are nearing completion.

Use of a Chimeric Hsp70 to enhance the quality of recombinant *Plasmodium falciparum* S-adenosylmethionine decarboxylase protein produced in *Escherichia coli*

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S-adenosylmethionine decarboxylase (PfAdoMetDC) from *Plasmodium falciparum* is a prospective antimalarial drug target. The production of recombinant PfAdoMetDC for biochemical validation as a drug target is important. The production of PfAdoMetDC in *Escherichia coli* has been reported to result in unsatisfactory yields and poor quality product. The co-expression of recombinant proteins with molecular chaperones has been proposed as one way to improve the production of the former in *E. coli*. *E. coli* heat shock proteins DnaK, GroEL-GroES and DnaJ have previously been used to enhance production of some recombinant proteins. However, the outcomes were inconsistent. An Hsp70 chimeric protein, KPf, which is made up of the ATPase domain of *E. coli* DnaK and the substrate binding domain of P. falciparum Hsp70 (PfHsp70) has been previously shown to exhibit chaperone function when it was expressed in *E. coli* cells whose resident Hsp70 (DnaK) function was impaired. We proposed that because of its domain constitution, KPf would most likely be recognised by *E. coli* Hsp70 co-chaperones. Furthermore, because it possesses a substrate binding domain of plasmodial origin, KPf would be primed to recognise recombinant PfAdoMetDC expressed in *E. coli*. First, using site-directed mutagenesis, followed by complementation assays, we established that KPf with a mutation in the hydrophobic residue located in its substrate binding cavity was functionally compromised. We further co-expressed PfAdoMetDC with KPf, PfHsp70 and DnaK in *E. coli* cells either in the absence or presence of over-expressed GroEL-GroES chaperonin. The folded and functional status of the produced PfAdoMetDC was assessed using limited proteolysis and enzyme assays. PfAdoMetDC co-expressed with KPf and PfHsp70 exhibited improved activity compared to protein co-expressed with over-expressed DnaK. Our findings suggest that chimeric KPf may be an ideal Hsp70 co-expression partner for the production of recombinant plasmodial proteins in *E. coli*.

Monogeneans of *Hydrocynus vittatus* (Characiformes: Alestidae) from Schroda Dam, Limpopo Province, South Africa

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Characiformes constitute a large and most diverse component of the African ichthyofauna with the family Alestidae being the most speciose. However, the population of these fishes are declining due to pollution, overfishing, and constructions of dams and weirs. During health and parasitological investigations of 22 specimens of a commercially and economically important fish species,*Hydrocynus vittatus* Castelnau, 1861 commonly known as the tigerfish, 12946gill monogeneans were recorded. The fish were caught with fishing rods during two seasons(June and October) in 2017 from Schroda Dam (22°11’ S; 29°25’ E) located within the Mapungubwe National Park. Specimens of *Annulotrema* were mounted inglycerine ammonium-picrate (GAP)solution and identified based on the male copulatory organ (MCO) as well as the sclerotised structures of the haptor. The collected monogeneans were all from the genus *Annulotrema* Paperna and Thurston, 1969, with a high prevalence (72.7%), mean intensity(809.1) and mean abundance(588.5). The present study reports on three known species (*A. pikei*; *A. pikoides* and *A. pseudonili*) and proposed new species. The findings furthermore contribute to new geographical records for species of *Annulotrema* from Schroda Dam.

Parasite diversity of *Oreochromis mossambicus* and *Clarias gariepinus* from Tibani Dam, Limpopo Province

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This study was conducted to investigate the diversity and prevalence of parasites of Mozambique tilapia, *Oreochromis mossambicus* (Peters, 1852) and the Sharptooth catfish, *Clarias gariepinus* (Burchell, 1822) from Tibani Dam near Mokopane, Limpopo Province. A total of 59 fish were collected during autumn (n = 17) and winter (n = 25) of 2017 and summer (n = 17) of 2018. Microscopic analysis was performed and parasites were identified using available keys. A total of 1060 parasites were identified from *O. mossambicus* and 3120 from *C. gariepinus*. The following parasites were recorded from 35 specimens of *O. mossambicus* (prevalence indicated): Ciliophora, trichodinids (17.1%) from the skin; Monogenea, *Cichlidogyrus* spp. (42.9%) from the gills, *Enterogyrus* sp. (5.7%) from the stomach, *Gyrodactylus* sp. (14.3%) from the skin; Trematoda, *Clinostomum* sp. (20.0%) from the branchial cavity, *Diplostomum* sp. (8.6%) from the eye, *Euclinostomum* sp. (2.9%) from the branchial cavity; Cestoda, gryporhynchid larvae (48.6%) from the intestinal wall; Nematoda, *Contracaecum* sp. (14.3%) from the body cavity; Acanthocephala, *Acanthogyrus* sp. (25.7%) from the intestine; Branchiura, *Dolops ranarum* (5.7%) and *Argulusjaponicus* (5.7%) from the skin; and Copepoda, *Neoergasilusjaponicus* (22.9%) from the base of the fins. The following parasites were recorded from 24 specimens of *C. gariepinus*: trichodinids (8.3%) from the skin;Monogenea, *Quadriacanthus clariadis*(25.0%) from the gills; Trematoda, *Clinostomum* sp. (4.2%) from the body cavity, *Glossidium pendatum* (25.0%) from the intestine; cestode (4.2%) from the intestine; *Contracaecum* sp. (91.7%) from the body cavity, *Procamallanus* sp. (16.7%) from the intestine; *D. ranarum* (20.8%) and *A. japonicus* (25.0%) from the skin. *Oreochromis mossambicus* had a higher parasite diversity compared to *C. gariepinus*. This study contributes to new geographical records for parasites from both host species and a first record of *Gyrodactylus* sp. from the skin of *O. mossambicus* from Limpopo Province.

Risk factors associated with occurrence of anthelmintic resistance in sheep of resource poor farmers in Limpopo province, South Africa

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Anthelmintic treatment is the most common way of controlling nematode infections in ruminants even though several countries have reported anthelmintic resistance (AR) resulting in limitation for sustainable small ruminant production. The aim of this study was to evaluate the knowledge of resource poor sheep farmers in Limpopo province on the use of anthelmintics. A questionnaire regarding helminth control practices was administered tosmall ruminant farmers (n=77) in five districts of Limpopo province, South Africa, namely Capricorn, Sekhukhune, Waterberg, Vhembe and Mopani. Farmers were interviewed using a structured questionnaire with open-ended questions such as questions on clinical signs linked to parasitism, time and reason for anthelmintic treatment, anthelmintic products used and dose determination. None of the farmers weighed their sheep before dosing them instead visual appraisal of individual weight was the most common means of estimating the anthelmintic dose. About 57% of the respondents could identify the clinical symptoms of nematode infection. Farmers that owned sheep for ≥10 years were better informed ($p<0.05$) than their inexperienced counterparts in identifying symptoms of nematode infection and knowledge of infection mode. However, no significant difference (p□0.05) was found to exist between the three groups of farmers with regard to the usage of anthelmintics. Although 67.5% of farmers mentioned that they never dose their sheep, 32.5% use anthelmintics at varying times in a year and this percentage falls within the farmers that never weigh their animals before dosing.Relying solely on the visual appraisal of an animal to determine its weight leads to incorrect weight estimation and under/over dosing which could be the primary cause of AR development in Limpopo province.In order to prevent or reduce the emergence of AR, correct use of anthelmintics and on-farm training on gastrointestinal nematodes infecting small stock must be provided and focus on aspects such the importance of correct dosage, when to alternate anthelmintic classes, treatment frequency and new treatment strategies, such as targeted drenching in combination with faecal egg counts.

Infection biology for *Chonopeltis australis* in the Vaal River System

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Chonopeltis australis Boxshall, 1976 is an ectoparasite of freshwater cyprinid fish that is endemic to Africa. Previous records have listed Boskop Dam on the Mooi River, Wuras Dam on the Vet River and Maselspoort Dam on the Modder River as localities. These rivers are all tributaries of the Orange-Vaal River System. The current study considers infection biology of *C. australis* at two sites on the main stream of the Vaal River, comparing it to published infection biology data from Boskop Dam reported by Avenant-Oldewage and Knight (2008) in order to understand the life history strategy of this species.

During this study, fish collections were conducted seasonally in Standerton, the Vaal Dam, and Visgat using gill nets of varying mesh size. Nets were checked every two hours for fish. On shore, fish were identified, measured and the external parasite load recorded. Only adult *C. australis* were recorded. Data for the Boskop Dam showed that prevalence of *C. australis* was at its highest in autumn, and decreased towards spring. A similar trend is seen at the two river sites where results show that the prevalence peaks at the start of summer but, decreases at the height of summer and increases again in autumn and start of winter. This suggests that egg hatching occurs in spring, organisms mature in summer and deposit eggs that hatch in autumn to supplement the population, thereafternew eggs overwinter until spring.The data for Boskop Dam showed a higher prevalence of *C. australis* on *Labeo umbratus*, where more of this species was caught (151 *Labeo umbratus* to 18 *Labeo capensis*), while both river sites show a higher prevalence on *Labeo capensis*, mirroring the species composition of the hosts (193 *Labeo capensis* to 33 *Labeo umbratus*). This difference could be due to the habitat preference of the host. Additionally, *C. australis* shows aggregation however, the extent differs between the two river sites. In Standerton, the variance/mean ratio is 2.17 indicating a moderate aggregation whereas in Visgat, the variance/mean ratio is 6.14 indicating increased aggregation. Aggregation supports sexual reproduction and would benefit*C. australis*which are weak swimmers.

Monogenean parasites of the genus *Enterogyrus* Paperna, 1963 in cichlid hosts of South Africa: how little is known and how much is hidden

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Monogenean parasites of the genus *Enterogyrus* parasitise the stomach of cichlids. To date, there are 12 known species and only two, *Enterogyrus coronatus*Pariselle, Lambert & Euzet, 1991 and *Enterogyrus cichlidarum* Paperna, 1963, have been recorded in South Africa.From August 2014 till September 2016, cichlids Mozambique tilapia, *Oreochromis mossambicus* and Banded tilapia, *Tilapia sparrmanii* were sampled at three localities in three provinces of South Africa. Nwanedi-Luphephe Dam, Limpopo Province (L), Mooi River, North-West Province (NW) and Modderrivier, Northern Cape Province (NC). Based on the morphometry on the hard parts five *Enterogyrus* species were identified. *Enterogyrus coronatus* was found in *T. sparrmanii* in L and NC. These findings represent a new host record for this parasite. *Enterogyrus cichlidarum* was identified from *T. sparrmanii* in NC and also representing new host record. *Enterogyrus malmbergi* was indentified in South Africa for the first time from L and NC in *O. mossambicus* and *T. sparrmanii*, respectively, and both hosts are new host records. Two new species. *Enterogyrus* sp. 1 and *Enterogyrus* sp. 2 found from *T. sparrmanii* and *O. mossambicus* in L and NW, respectively, are currently being described as a part of another study.The present study indicates that a small scale study can give an insight into the diversity of these parasites in South Africa. A more intensive study in future will probably result in additional new host records and new species from South Africa.

Interleukin 10 (IL-10) Production, and Seroprevalence of *Entamoeba histolytica* infection among HIV-infected patients in South Africa

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Infections by the parasite *E. histolytica* is increasing in HIV infected individuals. Interleukin (IL-10) plays an important role in maintaining the mucosal barrier. Therefore, the sero-prevalence of E. histolytica was investigated in relation to IL-10 serum concentration among HIV- infected patients was examined. A total of 647 blood samples were collected from HIV-infected patients. The *Entamoeba histolytica* antigen (Galnac lectin) and serum antibodies were assessed using specific ELISAs (TechLab, Virginia, USA). IL10 blood levels were determined using a commercial ELISA test and the results were analyzed using parametric and non-parametric statistical tests. The Gal/galnac lectin was detected in only 0.5% (3/ 647) individualsand the serum antibodies against *E. histolytica* were detected in 65.2% (422/647) of the samples. A significant increase in IL-10 levels was found in 68.1% of patients who were positive for *E. histolytica* antibodies compared to patients who were seronegative. There is a high level of exposure to *E. histolytica* among HIV patients in South Africa although the prevalence of amoebic liver abscess might be low. This study revealed that elevated levels of IL-10 is associated with reduced risk of amebiasis.

Molecular characterization of *Entamoeba histolytica* tRNA genes

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Entamoeba histolytica is a eukaryotic protozoan parasite responsible for the disease called amoebiasis and it generally occurs through ingestion of cysts from food or water contaminated by feces. The specific objectives of this study was to determine the prevalence of *E. histolytica* infection from stool samples using microscopy and Enzyme-Linked Immunosorbent Assay (ELISA), to identify different genotypes of *E. histolytica* based on the tRNA genes that circulate in the population in a rural area and in an urban population, and also to identify any potential association that may exist between parasite genotype and the presence of diarrhea in the patients.

A total of 774 stool samples were tested by microscopy, TechLab enzyme-linked immunosorbent assays (ELISAs) and multiplex PCR. For genotype analysis, six different loci (NK, RR, AL, DA and **S^{TGA}-**D) of the tRNA genes were amplified by PCR. The genotyping as well as the demographic data were analyzed using the Statistical Package for Social Sciences (SPSS for WINDOWS version 21.0) program. The overall prevalence of *Entamoeba* species by microscopy was16.7%. The analysis by the TechLab ELISA based antigen detection kit specific only for *E. histolytica* infection showed that the prevalence of *E. histolytica* among diarrheal samples from Pretoria was10.5% while in Giyani it was 5.4%. Males were more infected (12%) than females (8.4%). The genetic profiling of *E. histolytica* indicated that some strains were specific to each of the two locations while some were common. Profile Number 1 of the NK locus was more prevalent in diarrhea samples and was from Pretoria. Similarly, some profiles were more associated with diarrhea compared to others further indicating that the outcome of the infection by this parasite might be associated with the genotype. The results also indicated the possibility of strains clustering by region.The results obtained in this study confirm that tRNA genes might have a role in the presentation of amoebiasis (symptomatic and asymptomatic infections) depending on the genetic profile of the infecting strain. This genotyping system could also be used to identify the origin of the infection once it has been standardized.

Genetic polymorphism at the Interleukin 6 gene promoter 174G/C and its correlation with the sero-prevalence of *E. histolytica* among HIV positive patients in Northern South Africa

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Entamoeba histolytica is causative agent of amebiasis, which causes 100000 death annually. It has been suggested that the Host genetic polymorphism has been identified as a potential role player in the pathogenesis of several diseases. It was shown that cocultured of human and epithelial cell lines with trophozoite is shown to increase expression of cytokines including IL-6. In the present study we determine the impact of IL-6 promoter genetic polymorphism on the sero-prevalence of *Entamoeba histolytica*. Patients with HIV/AIDS attending different clinics in Limpopo province were recruited. Serum was used to detect Anti-*E. histolytica* antibodies using ELISA, the DNA was extracted from buffy coat using GeneElute blood Genomic DNA kit from Sigma Aldrich according to manufacturer's instruction. RFLP-PCR protocol targeting IL-6 174G/C was used for host genotyping. A total of 139 blood sample were obtained from patients and examined. Out of these 69.8% were females. The overall seroprevalence of *Entamoeba histolytica* in the study population was 77%. The prevalence was higher in females (78.4%) compared to males (73.8%) patients (p=0.346). The age group within which the highest prevalence was found was between 21-45 years. Of all samples that were genotyped for IL-6 only one was found to be GC genotype and the rest were GG genotype. IL-6 concentrations were above 300pg/ml in most of patients. The present study shows a high seroprevalence of amebiasis in the study population. IL-6 genotypes and IL-6 expression were not associated with amebiasis. However there is association between IL-6 levels and gender, CD4 count, origin and age. Additional studies are required using a larger population size in order to confirm these findings.

Prevalence of diarrhoea causing parasites in 10 river water sources used by rural communities in the Vhembe District, South Africa.

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Waterborne diseases contribute more than 75% of the outbreaks of water associated infectious diseases globally. The protozoan parasites such as *Giardia* and *Cryptosporidium*, are among the etiological agents linked with waterborne diseases. Reports in Africa have shown that almost five times as many people in rural areas lack potable water sources relative to those living in urban areas. The lack of portable water sources forces people to supplement their water needs by using raw unsafe surface waters from rivers, dams, wells and streams which are vulnerable to faecal contamination. The objectiveof this study was to determine the prevalence of diarrhoea causing parasites in 10 river water sources used by rural communities in the Vhembe District.Water was sampled once during the winter months (June-August) and once during the summer months (October-December) over a 12 months period. The USEPA Method 1623.1 was employed for the detection of *Giardia* and *Cryptosporidium* cysts and oocysts respectively. *Giardia* and *Cryptosporidium* were only detected in 20% (2 cysts/10L and 62 cysts/10L) and 10% (2 oocysts/10L) of winter samples respectively. Only *Giardia* was detected in 10% (1 cyst/10L) of summer samples. The maximum number of cysts detected was 62 from a sewage contaminated river sample. Generally, most of the rivers were within the South African target water quality range for cysts (*Giardia*) or oocysts (*Cryptosporidium*) in water for domestic use which is 0/10L. The results clearly indicate that raw surface water could be a potential health hazard if consumed without any treatment because doses greater than 1 may pose a risk of parasite infection.

Characterization of the chaperone activity of a plasmodial Hsp40 type II, PFE0055c

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The causative agent for the most dangerous form of malaria is *Plasmodium falciparum*, and its survival and development in red blood cells is maintained by exporting proteins required for remodeling of the host erythrocyte. Heat shock proteins play a wide range of roles in the cell and they are required to assist the parasite as it moves from a cold blooded insect vector to a warm blooded mammalian host. The expression of heat shock proteins increases during these heat shock conditions, and this is considered to play a role in differentiation of these vector-borne parasites. Heat shock protein 70 (Hsp70) is an important molecular chaperone that is involved in protein homeostasis, Hsp40 acts as a co-chaperone and stimulates its intrinsically weak ATPase activity. Hsp40s are classified into four types and are characterized by the presence of a highly conserved J-domain at the N-terminus and the histidine proline-aspartic acid (HPD) motif within the J-domain. Hsp40 Type I, II, and III contain the highly conserved J-domain; type IV contains a corrupted HPD motif. Hsp40 acts as a co-chaperone of Hsp70 and targets the substrates to Hsp70 for folding and interaction stability, interaction of Hsp40 with Hsp70 is regulated by the HPD motif. However, Hsp40 also demonstrates independent chaperone activity, functioning as a holdase. *P. falciparum* comprises 49 Hsp40s of which 19 contains the *Plasmodium* export element (PEXEL). Three of these exported Hsp40s are type II proteins (PFE0055c, PFA0660w and PFB0090c) and are exported to the red blood cell. PFE0055c is homologous to human Hsp40, DnaJB4, which interacts with human Hsp70. In infected erythrocytes, PFE0055c has been shown to co-localize with exported PfHsp70-x and PFA0660w. This study aims to biochemically characterize the exported form of PFE0055c to further enhance our understanding of parasite biology. Bioinformatics was used to characterize the structure/function features of PFE0055c.The gene coding for PFE0055c was cloned into expression vector pQE30. The expression, purification and solubility of the PFE0055c will be investigated using *Escherichia coli* XL1-Blue cells. The ability of PFE0055c to function as a holdase will be determined using aggregation suppression assays.The data generated from the characterization of PFE0055c may lead to understanding of its role in malaria towards identifying a successful biomarker.

Notes