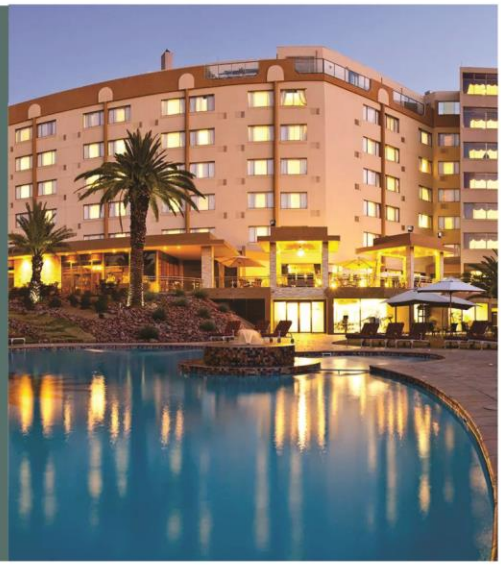

48th Annual Conference of the Parasitological Society of Southern Africa

15-17 September 2019
Hotel Safari, Windhoek, Namibia

BOOK OF ABSTRACTS

PARASITOLOGICAL SOCIETY OF SOUTHERN AFRICA
PARASITOLOGIESE VERENIGING VAN SUIDELIKE AFRIKA
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PROGRAMME

Sunday 15 September 2019		
16h00	Registration Opens	
18h00	Welcome Reception - Foyer of the Safari Court Conference Centre	
Day 1 - Monday 16 September 2019		
07h30	Registration Opens	
Plenary 1 ***Chair: S. Matthee***		
Time	**Title**	
08h00	Welcome & Opening *Prof. J.K.E. Mfune, University of Namibia and Prof. K. Matengu, University of Namibia*	
08h30	Keynote: Drivers of aquatic parasite diversity: the complex interaction of hosts, parasites and ecosystems *Prof. W. Luus-Powell, University of Limpopo*	
Session 1 - Freshwater fish parasites ***Chair: A. Kolo***		
Time	**Title**	**Speaker**
09h00	Determination of metallothioneins in *Lamproglena clariae* infecting *Clarias gariepinus* from Vaal River System, South Africa	J. Ndaba
09h15	Identification of an unidentified *Spinitectus* Fourment, 1883 collected from *Clarias gariepinus* (Burchell, 1822), in the Vaal River system, South Africa	L. Austin
09h30	The effects of water hardness on the rate of parasitic infestations on fishes collected from the Supersands Dam, South Africa	E.B.M. Moema
09h45	Trojan horses in South Africa: introduction of invasive fish and the distribution of their cestode parasites	Q.M. Dos Santos
10h00	Trophic position is a lot like real estate, it's all about location, location, location, as shown by stable isotopes in a fish host – parasite system	B.M. Gilbert
10h15	Mid-morning Refreshment Break & Posters - Omatako 2 (Safari Court Conference Centre)	
Session 2 - Freshwater and marine fish parasites ***Chair: Q.M. dos Santos***		
10h45	The infection statistics of *Trypanosoma* sp. in the African sharptooth catfish (*Clarias gariepinus*) collected along the Vaal River, South Africa, in correlation to water quality	M.M. Pretorius
11h00	Diversity of parasites from different fish species at Loskop dam	N.N. Shakwane
11h15	Relationship between host body size and sex, and mean abundance of three gill monogeneans (*Calceostoma* spp., *Sciaenacotyle* sp. and *Diplectanum* spp.) of silver kob (*Argyrosomus inodorus*) (actinopterygii; sciaenidae), Northern Namibia	A.M. Amakali
11h30	Conservation of ancient relationships – elasmobranchs and parasites	B.C. Schaeffner
11h45	Notes on some lernaeopodids (siphonostomatoida: Copepoda) collected from elasmobranchs off South Africa	S.M. Dippenaar
12h00	Mediterranean Gobiids: small hosts and their Gyrodactylid parasites	I. Přikrylová
12h15	*Prosorhynchoides* sp. (Trematoda: Bucephalidae): an exceptional trematode in monkfish, *Lophius vormerinus* sampled off Cape Town, South Africa	P.H. King
12h30	Lunch - Akasia Restaurant (Safari Court Hotel)	

Session 3 - Ectoparasites ***Chair: N. Nyangiwe and S. Matthee***		
Time	**Title**	**Speaker**
13h30	The effect of biotic and abiotic factors on the distribution of ixodid ticks on cattle in the Eastern Cape Province, South Africa	N. Nyangiwe
13h45	Genetic Diversity of *Amblyomma hebraeum* and *Rickettsia* spp. in the Eastern Cape, South Africa	A. Pillay
14h00	Ecological preferences and seasonal dynamics of ticks (Acari: Ixodidae) on and off bovine hosts in the Eastern Cape Province, South Africa	M. Yawa
14h15	Phylogenetic distance between hosts and parasite performance: is the close relatedness to the principal host better?	B.R. Krasnov
14h30	The energetic costs of tick infestation in four-toed sengis (*Petrodromus tetradactylus*)	H. Lutermann
14h45	The abundance, composition and barcoding of Tabanidae within Kruger National Park and their role in the transmission of pathogens	A. Smit
15h00	Species diversity of small mammals and the prevalence and intensity of infestation of the associated fleas (Siphonaptera) across an altitudinal gradient along the Ugab River, Namibia	A.I. Frans
15h15	Mid-afternoon Refreshment Break & Posters - Omatako 2 (Safari Court Conference Centre)	
Session 4 - Micro- and macroparasites ***Chairs: M. Chaisi and S. Matthee***		
15h45	A survey of captive penguins for avian haemosporidiosis	M. Chaisi
16h00	Diversity of avian Haemosporidian parasites in the lowveld region of South Africa	M.B. Wardjomto
16h15	Association of gametocyte profile and socio-demographic factors with malaria burden at Chipulukusu Clinic in Ndola, Zambia	N.M. Shimaponda-Mataa
16h30	Genetic diversity of *Entamoeba* spp based on the analysis of the 18SrRNA gene	A. Samie
16h45	Characterization of the bacterial blood microbiome of dogs and cattle in the Mnisi community, Mpumalanga, South Africa	A.O. Kolo
17h00	Geographic variation in the body size of ectoparasites on rodents	S. Matthee
17h30	**PARSA Annual General Meeting**	
18h30	Free Time	
19h00	Dinner - Welwitschia Restaurant, Hotel Safari	

Day 2 - Tuesday 17 September 2019	
07h30	Registration Opens
Plenary 2 ***Chair: A. Spickett***	
Time	**Title**
08h00	Announcements/housekeeping *Prof. J.K.E. Mfune, University of Namibia*
08h15	Keynote: My research experiences with selected zoonotic parasites endemic in southern Africa *Prof. S. Mukaratirwa, University of KwaZulu-Natal*

Session 5 - Microparasites ***Chair: J.K.E. Mfune***		
Time	**Title**	**Speaker**
08h45	Assessment of resistance of indigenous Nguni cattle to African animal trypanosomosis primary – challenge	A.A. Latif
09h00	In vitro and in vivo evaluation trypanocidal efficacy of curcumin and curcumin nanoparticle on animal trypanosomes	N.I. Molefe
09h15	Identification and genotyping of predicted host cell phenotype modulators in *Theileria parva*	N. Komani
09h30	Can we build a genetic trap for drug resistant malaria parasites?	G.I. McFadden
09h45	Prevalence of rodent haemoprotozoan infections in the Lowveld region of South Africa	G.T. Moloro

10h00	Investigating *Rickettsia africae* infection in *Amblyomma hebraeum* ticks in Mnisi, Bushbuckridge Municipality, South Africa	E. Mazhetese
Time	**Title**	**Speaker**
10h15	Mid-morning Refreshment Break & Posters - Omatako 2 (Safari Court Conference Centre)	
Session 6 - Diagnostics, tools and taxonomy ***Chair: T. Sibanda***		
10h45	First molecular detection and identification of *Abbreviata kazachstanica* isolated from roadkill European glass lizard, *Pseudopus apodus* in northern Iran	A. Vafae Eslahi
11h00	Next generation sequencing Technologies in public health laboratories: a case study of a national outbreak of listeriosis caused by ready-to-eat processed meat products, South Africa, 2017-2018	Z.T.H. Khumalo
11h15	Novel arenavirus isolates from Namaqua rock mice, Namibia, Southern Africa	J.M.P. Musuuo
11h30	Validation of multiplex *Ehrlichia ranis*, *Babesia rossi* and *Babesia vogeli* real time PCR assay	N.F. Nkosi
11h45	Phenotypic and genotypic screening for acaride resistance in Ixodid ticks from Southern Zambia	S. Chitanga
12h00	Molecular detection and genetic characterization of *Rickettsia* species in ticks collected from southern Zambia	S. Chitanga
12h15	Forensic entomology research and application in Southern Africa: a systematic review	D. Tembe
12h30	Lunch - Akasia Restaurant (Safari Court Hotel)	
Session 7 - Helminthology ***Chair: A. Halajian***		
13h30	Gastrointestinal helminth assemblages of the common warthog, *Phacochoerus africanus*, in KwaZulu-Natal Province, South Africa	K. Junker
13h45	Farmers' perceptions on the incidence of gastro-intestinal parasites in sheep in the Eastern Cape, South Africa	M.S. Jansen
14h00	Host-parasite-vector interactions and elucidation of an Amphibian Filarial Nematode life cycle	E.C. Netherlands
14h15	Larval Stages of Trematodes associated with snails from Lake Victoria, Kenya	J.O. Outa
14h30	Species and site contributions to beta diversity of gastro-intestinal nematodes and cestodes in *Rhabdomys dilectus* and *Rhabdomys pumilio*	A. Spickett
14h45	Population genetic structure of *Haemonchus contortus* in Limpopo Province, South Africa: a preliminary study	M. Mphahlele
15h00	Mid-afternoon Refreshment Break & Posters - Omatako 2 (Safari Court Conference Centre)	
Session 8 - Fish parasites ***Chair: R. Block***		
15h30	Exploring the diversity of *Diplostomum* (Digenea: Diplostomoidea) in freshwater fishes in South Africa	C. Hoogendoorn
15h45	An update on the taxonomy of freshwater gastropods and their complete trematode fauna in Lake Kariba, Zimbabwe	K.C. Muzarabani
16h00	Parasite Fauna of Nile Perch *Lates niloticus*, Nile Tilapia Oreochromis *niloticus* and *Haplochromis* spp. from Lake Victoria Kenya	J.O. Outa
16h15	New geographical distribution record, scanning electron (SEM) and phylogenetic analyses of *Glossidium pedatum* and *Orientocreadium batrachoides*	J.C. Dumbo
16h30	A fluke discovery – digeneans of *Clinus superciliosus* from the coast of South Africa	A. Vermaak
16h45	Scanning electron microscopy and molecular analysis of male *Lamproglena monodi* (Copepoda: Lernaeidae) infesting *Oreochromis niloticus* (Pisces: Cichlidae) in Kenya	N.M. Rindoria
17h00	Free Time	
19h00	Gala Dinner & Prize Giving - Omatako 4 (Safari Court Conference Centre)	

LIST OF ABSTRACTS IN ALPHABETICAL ORDER

KEYNOTE PRESENTATIONS

Monday, 16 September 2019

Drivers of aquatic parasite diversity: the complex interaction of hosts, parasites and ecosystems

W.J. Luus-Powell

DST-NRF SARChI Chair (Ecosystem Health), Department of Biodiversity, Private Bag X1106, University of Limpopo, Sovenga, 0727 - wilmien.powell@ul.ac.za

Species introductions and habitat loss are leading factors of biodiversity change. Fishes are one of the most commonly introduced aquatic taxa globally. These introductions and translocations into non-native regions have an effect on native biota with consequent potential economical and ecological risks. One of these risks includes the co-introduction of parasites and the potential of host switching to native species. Parasites account for nearly a quarter of IUCN's list of invasive species. Together with the introduction of alien parasite species is the constant risk of habitat change due to anthropogenic pollution as well as overexploitation and climate change. From 2000 to 2019, freshwater fish parasites and water parameters were recorded from ecosystems with different levels of environmental pollution from Mpumalanga and Limpopo provinces, South Africa. Parasite species richness and diversity indices were used to analyse impacted ecosystems. Results indicated that heteroxenous and monoxenous species should be considered separately as they are affected differently by pollution. In addition, several alien invasive fish parasites have been recorded including monogeneans (*Alcolpenteron* sp., *Clavuculus bursatus*, *Dactylogyrus extensus*, *Gyrodactylus* sp., *Onchocleidus furcatus*, *Onchocleidus principalis*, *Pseudodactylogyrus anguillae*), nematodes (*Camallanus cotti*, *Anguillicola crassus*), cestodes (*Atractolytocestus huronensis*, *Schyzocotyle* (*Bothriocephalus*) *acheilognathi*), a branchiuran (*Argulus japonicus*) and copepods (*Lernaea cyprincaea*, *Neoergasilus japonicus*). Host switching have been reported and some parasites previously known as specialists are now considered generalists. Native fish species have no co-evolution history with introduced alien parasites thus affecting host resistance and tolerance which may lead to greater pathogenic effect in native hosts with a risk to loss in biodiversity. This report includes new host records, new species and distribution records as well as the complexity of the use of fish parasites as a biomonitoring tool of ecosystem health.

Biography: Prof. Luus-Powell is a Professor in the Department of Biodiversity and Chair-holder of a DST-NRF SARChI Chair in Ecosystem Health (University of Limpopo). She received her BSc degree in Zoology from UJ (then RAU) in 1992, BSc Hons in 1994 and MSc in Parasitology from the same university in 1997. She obtained her PhD in 2004 at UL on metazoan parasites of mormyrid fishes. Her research has focused on aquatic ecosystem health including biomarkers of fish health; metal levels in fish and the human health risk associated with consuming contaminated fish; water and sediment quality; stable isotopes in aquatic food webs; parasite diversity and the effect of parasites on their host; new parasite species descriptions and biology of selected parasites. She has been teaching on undergraduate and Hons (Parasitology) level for more than 24 years. Prof Luus-Powell has supervised

more than 50 postgraduate students (Hons to PhD level) with more than 75 scientific papers and technical reports published. She received funding from the NRF, CSIR, WRC, VLIR-OUS and DST-NRF SARChI for various research projects. She is a NRF rated scientist and serves as reviewer for several journals and external examiner for national and international institutions. She was also an Associate Editor for African Journal of Aquatic Science from 2016 – 2018. She received the Vice-Chancellor's Research Award for 'Best Established Researcher' in the School of Molecular and Life Sciences in 2013 and 'Best Overall Researcher' of UL in 2016.

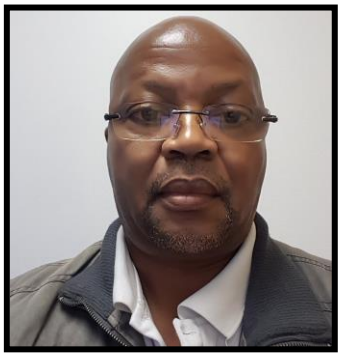
Tuesday, 17 September 2019

My research experiences with selected zoonotic parasites endemic in southern Africa

S. Mukaratirwa

School of Life Sciences, University of Kwazulu-Natal, Westville Campus, Durban, South Africa - Mukaratirwa@ukzn.ac.za

The impact of Parasitic Zoonoses (PZs) in sub-Saharan Africa can never be underestimated as they affect poor and marginalized communities that lack access to health services. Consequence of their footprint is that they have created a cycle of ill-health and poverty due to high morbidity and mortality to both animals and humans in low resource communities in Africa. What has aggravated the situation in sub-Sharan Africa is the lack of readily available context oriented information of these diseases in the public domain, the lack of a coordinated approach in addressing the challenges posed by these diseases and the absence of serious commitment from both decision makers and funding bodies to combat these PZs. In the last two decades I have focused my research interest in a variety of tropical parasitic diseases of economic and public health importance and in the process developed passion on "Neglected Parasitic Zoonoses" affecting the resource-poor communities in Africa and capacity building in the field of veterinary and medical parasitology. The diseases include malaria, schistosomiasis, *Taenia solium* cysticercosis, fasciolosis and trichinellosis and the application of a "One Health" approach in the prevention and control of these diseases. My presentation will focus on the highlights of my experiences and research findings of a selected few of these diseases.



Biography: Prof. Samson Mukaratirwa is holder of DVM, MVSc (Veterinary Parasitology) and a PhD (Parasitology) qualifications. He attained the rank of Full Professor in 2003 at the University of Zimbabwe and currently a Professor of Parasitology at the School of Life, College of Agriculture, Engineering and Science, University of KwaZulu-Natal, a post he has held since 2007. He was the Head of School of Biological and Conservation Sciences, University of KwaZulu-Natal (2009-2011) before becoming the inaugural Dean of School of Life Sciences, a merger with the School of Biochemistry, Genetics and Microbiology to form the current School of Life Sciences during the University reconfiguration to a College system. Before moving to the University of KwaZulu-Natal, he had occupied the post of Dean of Faculty of Veterinary Science, University of Zimbabwe for 8 years (2000-2007). He is a prolific researcher in the field of veterinary and medical parasitology and has published more than 150 articles in peer-

reviewed journals and has successfully supervised 17 PhD and more than 25 MSc students in parasitology.

Oral Presentations

Session 1 – Freshwater fish parasites

Determination of metallothioneins in *Lamproglena clariae* infecting *Clarias gariepinus* from Vaal River System, South Africa

J. Ndaba[1], B.M. Gilbert[1] & A. Avenant-Oldewage[1]

[1]*Department of Zoology, University of Johannesburg, P.O. Box 524 Auckland Park, Johannesburg, 2006, South Africa - JabulaniNdaba33@gmail.com; bericg@uj.ac.za; aoldewage@uj.ac.za*

The response of parasites toward changes in the chemical composition of aquatic ecosystems have been observed at population and individual levels. Therefore, this has led to the suggestion that these organisms may be suitable sentinels for monitoring the quality of the environment. Historically this has been assessed in two ways, firstly by examining changes in parasite population structure in response to pollutant exposure and secondly as accumulation indicators based on the ability of certain taxa to accumulate pollutants in their tissues. Despite this, little is known regarding the effects of pollution on parasites at a physiological level. Exposure to heavy metals and trace elements has been found to result in the production of metalloproteins such as metallothioneins (MTs) in free-living organisms as a regulatory mechanism. Metallothioneins are low molecular, cysteine rich proteins that are induced by the presence of heavy metals and trace elements in the environment and function to reduce metal toxicity. In parasites, however, metals become sequestered in eggshells and sclerotized structures as a possible means of reducing metal toxicity. Knowledge on the regulation of metals in parasites from a biochemical and/ or physiological perspective along with identification of biomarkers such as MTs and other metalloproteins in parasites is lacking. The aim of this study was to identify the expression of MTs in the host, *Clarias gariepinus* and ectoparasite, *Lamproglena clariae. Clarias gariepinus* were collected at two sites in the Vaal River, namely the Vaal Dam and below the Vaal River Barrage using gill nets. The gills were examined for the presence of *L. clariae*, which were collected with host muscle and liver tissue for assessment of metallothioneins in the host. Metallothioneins were identified using 10% sodium-dodecyl sulphate polyacrylamide gel electrophoresis (SDS-PAGE) and spectrophotometric assay. The identified MTs had a size range of 9–16 kDa for both *C. gariepinus* and *L. clariae*, and the concentrations expressed in host and parasite tissues were variable. This study is the first to report on the identification of MTs in an ectoparasite.

Identification of an unidentified *Spinitectus* Fourment, 1883 collected from *Clarias gariepinus* (Burchell, 1822), in the Vaal River system, South Africa

L. Austin[1], A. Avenant-Oldewage[1] & Q.M. Dos Santos[1]

[1]*Department of Zoology, University of Johannesburg, P.O. Box 524, Auckland Park, Johannesburg 2006, South Africa - lucinaustin@gmail.com; oldewage@uj.ac.za; quintonds@uj.ac.za*

Spinitectus spp. are enteric nematodes of the parasitic family Rhabdochonidae, characterized by annular spines decreasing in size towards the posterior. Eleven species have been recorded from Africa, with two from southern Africa. *Spinitectus petterae* Boomker, 1993 (Crocodile River) from *Clarias gariepinus* (Burchell, 1822) and *Spinitectus polli* Campana-Rouget, 1961 (Sabie River) from *Synodontis zambezensis* (Peters, 1852). Both species have only been recorded from the Kruger National Park (Limpopo River Catchment). During standard parasitological investigations in the Vaal River system (Vaal Dam, below the Donkerpoort Dam, and below the Vaal River Barrage), *Spinitectus* specimens were collected from *C. gariepinus.* As the documented distribution of *P. petterae* and *P. polli* does not include the Vaal River system, the present study aimed to identify the species collected. This was done using light microscopy, scanning electron microscopy and DNA analysis to compare the morphology, morphometrics and molecular distinctness of the specimens collected to other African species. Light microscopy specimens were cleared and mounted in anhydrous glycerol, while for scanning electron microscopy specimens were dehydrated using hexamethyldisilazane (HMDS) and spicules isolated to study the external morphology. Important structures measured and observed were the number, size and length of spines, and length ratios of specific structures, which were compared with all African species. Taxonomic structures used for comparison were the number and location of cephalic and caudal papillae, the shape of the pseudolabia, and the configuration of the annular rings. For DNA analysis, small ribosomal subunit (18S) and large ribosomal subunit (28S) rDNA were used to determine the identity and distinctness of the specimens collected. The spines of the specimens studied were similar to those of *S. polli* and *S. petterae.* Both the length ratios of the vulva to anterior and anus, and the length ratios for the spicules were similar to that of *S. petterae.* The 18S showed the specimens were genetically distinct from *S. petterae* and, while these species were closely associated, bootstrap support for this association was low. Thus, there was limited correlation between the genetic and morphological results of the specimens collected in the Vaal River and other *Spinitectus* species from Africa.

The effects of water hardness on the rate of parasitic infestations on fishes collected from the Supersands Dam, South Africa

Z.M. Tseka[1] & E.B.E. Moema[1]

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Fish are aquatic animals and the surrounding water has an impact on their lives in general. Water quality parameters such as pH, turbidity and water hardness affect the aquatic organisms and the symbiotic relationships amongst them. The effect of water hardness, which by definition is the concentration of dissolved minerals, particularly calcium and magnesium in water, on the life of aquatic organisms has not been extensively studied. The present work was undertaken to evaluate the effects of different levels of water hardness on the survival of ecto-parasites on fish species collected from the Supersands Dam. Juvenile fish from three genera, *Barbus, Tilapia* and *Pseudocrenilabrus* were collected from the dam

and were examined for presence of ecto-parasites before being exposed to water media of different hardness levels (i.e. soft, moderately hard, and very hard water). The fish species were then exposed for five days, after which the number of surviving ecto-parasites were compared to the initial number of parasites and recorded. The experiment was done in triplicate. After the five-day observation period, the average number of ecto-parasites, mostly cilliates and monogeneas per fish was recorded from the different media as follows: 23.5 in dam water, 4.5 in tap water, 0.6 in soft water, 1.4 hard water and 3 in very hard water. The results show that an increase in water hardness leads to a decrease in the number of ecto-parasites on fish. These results may in the future help combat challenges of ecto-parasitism in the aquaculture industry.

Trojan horses in South Africa: introduction of invasive fish and the distribution of their cestode parasites

Q.M. Dos Santos[1] & A. Avenant-Oldewage[1]

[1]Department of Zoology, University of Johannesburg, P.O. Box 524, Auckland Park, Johannesburg, 2006, South Africa - gmdossantos@live.co.za; aoldewage@uj.ac.za

The cestode *Atractolytocestus huronensis* Anthony, 1958 (Cestoda: Caryophyllidea) is a well-documented parasite of the globally occurring *Cyprinus carpio* Linnaeus, 1758. This parasite was first described from common carp in North America, but has since been recorded regularly alongside the distribution of its host. Carp has been known to occur in the freshwater systems of southern Africa for more than a century, with the first documented introduction in 1859. Recently, *A. huronensis* has been collected from two river systems in South Africa. The Olifants River runs through Mpumalanga and Limpopo provinces, with *A. huronensis* recorded from Loskop, Witbank, Flag Boshielo and Tzaneen Dams. The Vaal River flows from Limpopo, through Gauteng, Freestate and North-West, entering the Orange River in the Northern Cape, with *A. huronensis* only recently collected in the middle reaches of the river. In 2018, an unidentified cestode was collected from *C. carpio* for the first time in the upper reaches of the Vaal River (below Grootdraai Dam). Using both light and scanning electron microscopy, in addition to genetic characterisation, the cestodes were identified as *A. huronensis*. In 2019, carp infected with this cestode were collected at two additional sites along the Vaal River system, in the Vaal Dam and below the Vaal River Barrage. The cestodes displayed high prevalence (between 60 and 100%), with some individuals infected with as many as 64 cestodes. The fact that these cestodes are suddenly so prevalent in a river system where they had only recently been recorded for the first time, even though their host species has populated and been studied in this system for many decades, is worrisome. This may indicate that the parasite was recently introduced into the Vaal River system, probably in the upper reaches of the system, by means of infected common carp. However, the occurrence and possible introduction of this cestode in the upper reaches of the Vaal River could suggest that carp in the entire catchment, including the Orange River and its tributaries, may be infected. Further investigation is needed to accurately determine the origin of this recent introduction of invasive cestodes in the Vaal River.

Trophic position is a lot like real estate, it's all about location, location, location, as shown by stable isotopes in a fish host – parasite system

B.M. Gilbert[1], M. Nachev[2], M.A. Jochmann[3,4], T.C. Schmidt[3,4], D. Köster[4], B. Sures[1,2] & A. Avenant-Oldewage[1]

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Differences in the abundance of naturally occurring stable isotopes of carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) have been used as a tool in ecological studies for analysing food web architecture and trophic interactions between organisms. Ratios of stable isotopes represent a unique fingerprint of the nutrients assimilated by organisms in ecosystems. Ecological studies of food webs have mostly ignored parasites despite parasitism being recognised as a common consumer strategy. Most investigations of the trophic interactions between hosts and parasites have identified variable enrichment of different parasite taxa, which can be related to their feeding strategies. Comparison of stable isotope ratios for carbon ($\delta^{13}C$, $^{13}C/^{12}C$) and nitrogen ($\delta^{15}N$, $^{15}N/^{14}N$) of hosts and parasites were performed to investigate the trophic interaction between the host fish, *Clarias gariepinus*, and four co-occurring parasites: *Lamproglena clariae* (Copepoda), *Tetracampus ciliothaeca* (Cestoda), *Proteocaphalis glanduligerus* (Cestoda) and L3 *Contracaecum* sp. (Nematoda). Hosts and parasites were collected from six sites along the Vaal River. Analysis of stable isotope signatures was performed for host muscle tissue and parasites using an elemental analyser coupled with an isotope ratio-mass spectrometer (EA-IRMS). Based on enrichment by ^{15}N isotope, *L. clariae* were enriched relative to the host in a manner similar to that observed for predators and other blood – feeding ectoparasites. Endoparasite taxa were depleted in this isotope, which has similarly been found in other host – parasite systems for similar taxa. For ^{13}C isotope, hosts and parasites shared similar enrichment factors indicating that hosts and parasites assimilate nutrients from similar sources. Spatial comparisons showed that isotopic enrichment was variable between the different collection sites. This suggests that food items utilised in the diet of *C. gariepinus* are variable and likely determined by the availability of prey organisms at each of the different sites. Such spatial variation in stable isotope signatures in parasites further reflects baseline isotope gradients across the host distribution.

Session 2 - Freshwater and marine fish parasites

The infection statistics of *Trypanosoma* sp. in the African sharptooth catfish (*Clarias gariepinus*) collected along the Vaal River, South Africa, in correlation to water quality

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The Vaal River contributes to both the ecology and economy of South Africa. Anthropogenic activities produce organic and inorganic pollution. which may have a negative effect on the ecology of this freshwater system. A previous study in the Vaal River has indicated that the occurrence of *Trypanosoma* sp. infecting *Clarias gariepinus* are variable and probably related to the presence of the leech vector. Another study reported that leech numbers increase when organic pollution increases. The current study recorded aspects of pollution in the Vaal River by measuring water quality parameters in comparison to the prevalence of *Trypanosoma* sp. in *C. gariepinus*. During October 2018, a minimum of 10 and a maximum of 20 *C. gariepinus* specimens were collected with gill nets, electro-shocking or angling at six sites along the Vaal River including: Vaal River below Grootdraai Dam, Vaal Dam, Vaal River Barrage, Bloemhof Dam, Vaal-Harts Dam and Douglas Weir. Blood smears were made from each fish to determine *Trypanosoma* sp. infection. 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. Parasite prevalence as well as erythrocyte and leukocyte counts and their ratio from each fish was determined for each site. *Trypanosoma* sp. had the highest prevalence (70%), in the Vaal Dam (Upper Vaal) a site considered to have good water quality, and the lowest prevalence at Grootdraai (Upper Vaal) with 20% and Douglas Weir (Lower Vaal) with 0%. The occurrence of leech infections on *C. gariepinus* was low and only three specimens were found on catfish at Vaal Dam, below the Vaal Barrage and below Vaal-Harts Weir respectively. It was not possible to conclusively indicate whether infections of both leech and *Trypanosoma* sp. in *C. gariepinus* are related to the impact of water quality on both parasite species or only on the leeches. Further investigation is required to establish the relationship within this host – parasite system with water quality as an impacting factor.

Diversity of parasites from different fish species at Loskop dam

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Fish populations are negatively affected due to habitat degradation through pollution and land expansion as well as diseases and parasites. A parasitological study was conducted to investigate the parasite diversity of four fish species, *Labeo rosae, Schilbe intermedius, Oreochromis mossambicus* and *Clarias gariepinus* from a dam in the upper Olifants River, a river known to be contaminated from various sources. The fish were caught with fishing rods and gill nets during two seasons (summer and winter) in 2018 from Loskop Dam (25˚ 26' 57. 05" S 29˚ 19' 44. 36" E) located about 32 km south of the town of Groblersdal in Mpumalanga Province. Standard methods were used for parasite fixation and preservation. A total of 731 ectoparasites and 4778 endoparasites were recovered from all fish species. The highest number of monogeneans was recorded from the gills of *S. intermedius,* i.e. *Schilbetrema* spp. with a prevalence of 76.2%, mean intensity of 35.4 and mean abundance of 27.0. The highest number of endoparasites was recorded from *C. gariepinus* from the genus *Contracaecum* with a 100% prevalence, and mean intensity and mean abundance of 177. This nematode larva was also recorded from *S. intermedius* with lower intensity levels. Two different copepods were recovered, *Lamproglena clariae* and *Neoergasilus japonicus* (an alien invasive parasite). One branchiuran, *Argulus japonicus,* was found. In addition, the following monogeneans were recovered from different fish species: *Cichlidogyrus* sp. from *O. mossambicus*, and *Quadriacanthus* sp., *Macrogyrodactylus* sp. and *Gyrodactylus* sp. from *C. gariepinus*. Other endoparasites recorded included *Paracamallanus* sp., *Procamallanus* sp., *Tetracampus* sp., *Proteocephalus* sp., an unidentified cestode larva and adult cestode from *C. gariepinus*. Encysted digenean larvae and *Clinostomum* sp. was recorded from *O. mossambicus*. None of the parasites caused excessive pathology but future monitoring of fish health at Loskop Dam is recommended.

Relationship between host body size and sex, and mean abundance of three gill monogeneans (*Calceostoma* spp., *Sciaenacotyle* sp. and *Diplectanum* spp.) of silver kob (*Argyrosomus inodorus*) (actinopterygii; sciaenidae), Northern Namibia

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This is the first study to assess the influence of silver kob (*Argyrosomus inodorus*) size and sex on the mean abundance of three of its gill monogenean parasites (*Calceostoma* spp., *Sciaenacotyle* sp. and *Diplectanum* spp.) in northern Namibia. Silver kob were collected monthly (2017–2018 for 11 months) using conventional fishing gear (n = 55) in Toscanini, Mile 108 and Henties Bay, northern Namibia (21°- 24°S). Morphometrical characters of fish (e.g. Total Length (TL) and sex) were recorded and the gills were checked thoroughly and *Calceostoma* spp., *Sciaenacotyle* sp. and *Diplectanum* spp. were collected and counted. Chi-square tests were used to determine differences in parasite mean abundance by three length groups and by sex. *Calceostoma* spp. showed a significant decrease in mean abundance with increasing host length (X^2 = 28.22, p <0.01). *Sciaenacotyle* sp. showed no significant difference in mean abundance by host length group (X^2 = 1, p 0.30). *Diplectanum* spp. showed a significant increase in mean abundances with increasing host length (X^2 = 41.1, p <0.001). Reasons for these differences between the three gill monogeneans are discussed, with suggestions for further studies. No fish sex preference was observed for

parasite mean abundance in silver kob. As these monogeneans can be pathogenic for the host, knowing about their epidemiological characters may help for better prevention and control in mariculture practices.

Conservation of ancient relationships – elasmobranchs and parasites

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Southern Africa is an exceptionally biodiverse region for cartilaginous fishes. Many species are threatened, and conservation efforts are restricted to certain iconic species. To preserve natural resources, marine protected areas are established. However, information on the biology and ecology of elasmobranchs across management zones is sparse. In addition, current conservation schemes only apply for species at higher trophic levels, while parasitic organisms, forming the majority of biodiversity worldwide, are ignored as eligible conservation targets. Elasmobranchs and their internal parasites display one of the most 'ancient' host-parasite interrelationships on this planet, whose co-evolution is strongly interconnected, dating back at least 270 million years. Incorporation of parasites, together with their threatened hosts, becomes essential for future conservation efforts, since co-extinction events (i.e. parasite species facing extinction), which happen entirely unrecognized, may lead to cascading negative impacts within ecosystems. This project aims to assess the ecosystem health and anthropogenic impacts using cartilaginous fishes and their endoparasites as model organisms to establish and support the preservation of marine life and natural resources, and the conservation of threatened elasmobranch species and their associated parasites to maintain ancient host-parasite interactions and interrelationships. Testing the condition of the marine environment between Betty's Bay and Gaansbai, covering approximately 200km^2 of South African coastline, we focus on the most dominant and least threatened species of elasmobranchs and their internal parasites as biological indicator organisms. An assessment of the biodiversity of internal parasites of selected elasmobranch species endemic to the Western Cape province will further provide useful information on the hosts' biology and ancient relationships with their specific parasites.

Notes on some lernaeopodids (Siphonostomatoida: Copepoda) collected from elasmobranchs off South Africa

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Lernaeopodidae is one of the 40 families of the Siphonostomatoida. The family currently has 48 accepted genera and has marine and freshwater members. The females are uniquely adapted to attach to their hosts with the maxillae, mostly through a bulla – an anchor-like attachment organ. This way of attaching to the host has caused significant changes to their morphology when compared to other siphonostomatoids as the maxillae and maxillipeds switched position on the cephalothorax and the maxillae are modified. Males of the lernaeopodids are dwarf males with an ephemeral existence. Copepods were collected from elasmobranchs caught in the nets of the KwaZulu-Natal Sharks Board, caught as bycatch

during demersal surveys of DAFF and ORI and from fish caught by commercial fishermen off Gansbaai. They were fixed and preserved in 70% ethanol and studied using stereo- and light microscopy. Representatives of five genera of lernaeopodids were collected from 16 different host species. Host infection is quantified by prevalence, mean intensity and mean abundance values. Some of the lernaeopodids seem to be very host specific while others are generalists. Many of the reports represent new geographical and or new host records.

Mediterranean Gobiids: small hosts and their Gyrodactylid parasites

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In the Mediterranean, Gobiids represent one of the most diverse group of fish, with almost 70 species known in the area. From all of these, only six species have been noted to host representatives of the genus *Gyrodactylus*. Monogenan parasites of the genus parasitize predominantly bony fishes, and worldwide nearly 500 species have been described so far. Seven of 16 *Gyrodactylus* spp. known from marine gobiids have been recorded in the Meditteranean and Adriatic Sea. During three sampling trips in the northern Adriatic Sea, in June 2013, May and September 2014, a total of 858 individuals of 30 gobiid species belonging to 16 genera were collected. Collection was done by scuba diving collectors from various deeps and habitats off Croatia in the vicinity of Selce town. Fishes were caugth by small nets being temporally immobilized by solution of quinaldin and examined for the presence of parasites. Morphometric and molecular identification, using morphometrical evaluation of the hard parts of attachment organ and ITS rDNA gene respectively, confirmed the presence of five *Gyrodactylus* species parasitizing six host species. These were as follow: *Buenia affinis, Gobius niger, Gobius roulei, Odontobuenia balearica, Pomatoschistus marmoratus* and *Pseudaphya ferreri.* Only one of five identified *Gyrodactylus* species, *Gyrodactylus arcuatoides*, is currently known and remaining four species represent new species to the science. Three hosts, *B. afinis, G. roulei* and *P. ferreri*, are reported to host these parasites for the first time. *Gyrodactylus* species identified as a part of the present study represent morphologically and genetically very diverse individuals what makes them interesting objects to study co-evolution with their diverse hosts. Mediterranean gobiids are very small hosts and their parasites fauna is still poorly known. Present study brings a slight insight on new gyrodactylid parasites of the unique fish samples. The collection of such variety of fishes using a specific technique represents valuable contribution to the understanding of host-parasites interaction in marine environment.

Prosorhynchoides sp. (Trematoda: Bucephalidae): an exceptional trematode in monkfish, *Lophius vormerinus* sampled off Cape Town, South Africa

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The Bucephalidae Poche, 1907 is a cosmopolitan family found in marine, brackish and freshwater fishes that differs from all other digeneans by the configuration of their digestive system, the configuration of their terminal genitalia and by the presence of an anterior rhynchus for attachment. Monkfish were collected from the west and south coasts of South Africa during demersal research cruises of the Department of Agriculture, Forestry and Fisheries. Specimens for light microscopy were fixed in 70% EtOH and stained in Van Cleave's haematoxylin and fixed in 2.5% gluteraldehyde for scanning electron microscopy. A *Prosorhynchoides* sp. was sampled from the intestine of the monkfish, *Lophius vormerinus* south of Cape Town, South Africa. These parasites are small, measuring 1162-1586 (1336) µm long and 412-548 (471) µm wide. The anterior sucker is called a rhynchus and it is not connected to the pharynx. The acetabulum is absent. The mouth opens in the middle of the body, leading to a pharynx that continues into a sac-like intestine that does not reach to the posterior end of the body. The cirrus-sac is extremely long, measuring 390-509 (458) long x 70-119 (99) µm wide and occupies the posterior third of the body. The genital atrium opens at the posterior end of the body. The vitellaria form two clusters with 10-16 follicles posterior to the rhynchus. *Prosorhynchoides gracilescens* is a common intestinal parasite of *Lophius piscatorius* in European marine waters. This is the first record of a *Prosorhynchoides* sp. in *L. vormerinus* in the southern Atlantic and Indian Oceans but requires a species description in order to verify the status of this small unique trematode.

Session 3 - Ectoparasites

The effect of biotic and abiotic factors on the distribution of ixodid ticks on cattle in the Eastern Cape Province, South Africa

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The objectives of the study were to determine the effect of vegetation type on the number of species and the species composition of ticks on cattle and on the vegetation in four

vegetation types (Albany Coastal Belt, Amathole Montane Grassland, Bhisho Thornveld and Great Fish Thicket) in the south-west region of the Eastern Cape Province. A study was carried out between October 2012 and February 2013 in 20 communal cattle-farming areas. A total of 20,212 ticks belonging to 12 species were collected from adult cattle, calves and on the vegetation at the 20 localities. *Rhipicephalus microplus* (35.7%) was the most abundant tick species, followed by *R. appendiculatus* (24.8%) and *Amblyomma hebraeum* (17.4%). The overall tick abundance and species richness on cattle and on the vegetation was not significantly affected (P>0.05) by vegetation type. Rather, there was a strong within-vegetation type effect with activities related to individual villages facilitating this pattern. Significant differences (P<0.05) were observed in species composition between the vegetation types with Grassland having a more distinct composition compared to the other vegetation types. The species that contributed most to the dissimilarity was *R. microplus* and *A. hebraeum* with A. hebraeum being absent from cattle in Grassland and *R. microplus* being more abundant on cattle in Grassland compared to the other three vegetation types. These findings highlight the importance of ecological studies regarding tick occurrence and species composition.

Genetic Diversity of *Amblyomma hebraeum* and *Rickettsia* spp. in the Eastern Cape, South Africa

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Ticks are among the most important vectors of disease, responsible for the transmission of a wide range of pathogens that are of veterinary importance and public health concern. In South Africa, *Amblyomma hebraeum* is the principal vector of *Rickettsia africae* and *Ehrlichia ruminantium,* responsible for the transmission of African tick bite fever and Heartwater respectively. The detection of emerging zoonotic agents from cattle in a rural community in South Africa highlights the potential risk of human and cattle infection.Clinical detection of rickettsial infections is difficult due to the symptoms resembling other diseases (Malaria) and cross-reactivity not allowing the discrimination of *Rickettsia spp*. Due to this, molecular techniques are the currently used method that enable the identification and the delimitation between *Rickettsia* spp. Therefore, the aim of this project was to determine the genetic diversity of *Rickettsia* spp. in *A. hebraeum* using phylogenetic analyses. *Rickettsia* spp. were detected by screening ticks collected from cattle, pastures and blood samples over a period of a year in the Eastern Cape province, South Africa. A multi-locus sequence typing (MLST) approach was used by amplifying the *gltA, ompA, ompB, sca4, 17kDa* and *16Sr*RNA genes of *A. hebraeum* ticks to characterise rickettsial pathogens. Results thus far revealed that 54% of *A. hebraeum* adult ticks were positive for *Rickettsia* spp. From the blood samples, 32% of the samples were positive for *Rickettsia* spp. Sequencing of the *gltA* and *ompA* gene confirmed the infection of *A. hebraeum* ticks and cattle blood samples with *Rickettsia africae*. This multigenic approach will allow for the differentiation of the spotted fever group *Rickettsia* that includes a great number of closely related species. This information will further aid in the epidemiology of tick-borne zoonotic diseases both locally and internationally by raising awareness about tick-borne diseases in the veterinary, medical and tourism sectors that are most affected.

Ecological preferences and seasonal dynamics of ticks (Acari: Ixodidae) on and off bovine hosts in the Eastern Cape Province, South Africa.

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**Deceased*

The objective of this study was to determine the seasonal abundance and species composition of free-living and engorged ticks of cattle in three agro-ecological zones. The occurrence of ticks on Bonsmara and Nguni cattle was studied for the period of 12 months from April 2016 till March 2017. Tick collections were performed from a total of 1080 with 360 in each locality and 10 were randomly selected during tick sampling on monthly interval. Ticks were collected on monthly interval. From the cattle, a half body tick collection was performed and visible ticks were collected by means of fine-forceps after the animals had been restrained. Body regions that were examined included the ear, head, neck, chest, abdomen, flank, front and hind leg and feet, tail, and peri-anal region. At each site, six replicate drags of 100 m, approximately 50 m apart, were performed. All ticks collected per animal and from vegetation were stored in labelled sample tubes containing 70% ethanol, subsequently counted and identified to genus and species level using a standard stereomicroscope. A total of 31425 ticks belonging to ten species were collected during the study. *Rhipicephalus* (*Boophilus*) *decoloratus*, was the most observed tick species (32.50%), all other ticks like *R. evertsi evertsi* (18.84%), *R. appendiculatus* (17.26%), *A. hebraeum* (16.27%), *R. simus* (7.69%) were also commonly collected. *Ixodes pilosus* (3.84%), *H. rufipes* (3.46%), *R. follis* (0.08%) together with *Haemaphysalis silacea* (0.02%) were sporadic infestation. *Haemaphysalis elliptica* (0.04%) was only found on the vegetation. No *H. rufipes* was collected in the thicket vegetation. Agro-ecological zones differ significantly ($P<0.05$) in ticks' abundance and distribution. Engorged ticks from cattle were significantly higher during the summer season across different farms. Free-living ticks were widely distributed across different seasons but significantly lower during the winter season. It was concluded that agro-ecological differences and seasonal variations had significantly influenced tick species distribution and abundance.

Phylogenetic distance between hosts and parasite performance: is the close relatedness to the principal host better?

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Parasites achieve maximum abundance on their principal host, and lower abundances on their auxiliary hosts. The aim of our study was to test whether phylogenetic relatedness between the principal and auxiliary host species determines what abundance a parasite can achieve on its auxiliary hosts, as relatedness should reflect similarities among host species in ecological, physiological and/or immunological characters. We tested this hypothesis with fleas parasitic on small mammals. Field data demonstrated consistent support for this relationship. Then, we tested for this hypotheses in experiments and found that energy expenditure of blood digestion increased and reproductive output of fleas decreased with an increase of phylogenetic relatedness between a given host and the principal host of this flea species but only when auxiliary hosts belonged to the same family as the principal host. However, when fleas exploited a host that was very distant from the principal host their performance appeared to be unexpectedly high. Furthermore, when a flea characteristic for rodents was forced to exploit hosts from different order (Chiroptera) but co-occurring with the principal host of this flea, flea feeding performance (blood meal size and duration of digestion) was either as high as that on the principal host or much lower in dependence of a bat species. Finally, we measured flea performance on hosts that were both distant from to the principal host (gerbillids versus heteromyids) and inhabit other geographic reasons (Palearctic vs Nearctic) modelling thus host invasions. Flea performance was either high or low on different invasive hosts being thus context dependent. We conclude that among-host variation in parasite performance may result from interplay of several factors including co-occurrence between hosts, susceptibility of a host to parasite attacks, species-specific level of immunocompetence and the level of host specificity of a parasite. High performance in an unusual but co-occurring host may be one of the reasons for host switching to unrelated lineage. From the conservation perspective, we conclude that the response of a resident parasite to an invasive host is unpredictable.

The energetic costs of tick infestation in four-toed sengis (*Petrodromus tetradactylus*)

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Parasites may induce energetically costly host defences and thus, they should increase the energy demand of their hosts. However, attempts to quantify these costs have yielded contradictory results such that negative, positive and neutral effects have been reported. In the current study, we aimed to evaluate the energetic costs associated with tick infestation of four-toed sengis (*Petrodromus tetradactylus*) from Bonamanzi Game Park, KwaZulu-Natal, South Africa. We measured resting metabolic rate (RMR) of nine sengis over a 24h period after capture and four weeks later and calculated the daily energy expenditure (DEE) for these time points. In addition, we determined the basal metabolic rate (BMR) after capture and during each consecutive week until all ticks had detached. We retrieved a total of 4374 immature ticks (i.e. larvae and nymph) of which 4222 (96.5%) were identified as *Rhipicephalus muehlensi* while the remaining belonged to four other species (*Rhipicephalus appendiculatus*, *Rhipicephalus maculatus*, *Haemaphysalis elliptica* and *Haemaphysalis silacea*). The abundance of *R. muehlensi* decreased significantly from week to week and all ticks had detached from their hosts from all but one sengi four weeks after capture. The

BMR peaked during week 3 when tick burdens were low but reached its nadir in week 4, however, the latter did not differ significantly from the initial BMR measurement. Conversely, RMR was significantly higher in infested compared to uninfested sengis. RMR exhibited a clear diurnal cycle in infested animals only while significant differences in BMR between infested and uninfested animals were not apparent during daytime. As a result, the DEE was significantly larger in infested compared to uninfested sengis. Our findings suggest that the assessment of energetic costs of parasitism can be affected by the type as well as the time the measurement is taken. This may at least partially account for the contradictory results reported in other studies of the energetic costs of parasitism.

The abundance, composition and barcoding of Tabanidae within Kruger National Park and their role in the transmission of pathogens

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Tabanidae (Diptera) are a diverse hematophagous fly family. They are a nuisance to humans and animals alike with their persistent and painful biting behavior. Tabanids are known to transmit diseases to humans including loiasis and tularemia; and over 35 livestock pathogens both mechanically and biologically. Very little modern taxonomical work has been done on tabanids within South Africa despite their environmental importance as pollinators and disease vectors. This study aimed at comparing the traditional alpha-taxonomic approach to species delimitation with molecular methods using two protein coding gene regions, mitochondrial cytochrome oxidase I (COI) and nuclear alanyl-tRNA-synthetase (AATS). Furthermore, the study aimed to elucidate the role of tabanids in the transmission of selected pathogens. Tabanids were captured in two major locations within the Kruger National Park. The flies where morphologically identified then homogenized for DNA extractions, which was then sequenced and phylogenetically analysed. A random subset of samples was screened for *Besnoitia besnoiti, Trypanosoma theileri*, *Anaplasma marginale*, *Anaplasma centrale* and *Babesia bigemina*. In total, 854 flies belonging to 14 species under five genera in three subfamilies were captured. The phylogenetic analysis indicated sufficient correspondence to that of the morphological identification. However, several discrepancies were apparent. The genera *Haematopota*, *Philoliche* and *Chrysops* were well supported across all analyses and clustered into monophyletic groups. Tabaninae, however, formed an unsupported monophyletic group with an unresolved *Tabanus* cluster. It is apparent that the classification of Tabanidae should be placed under scrutiny. A larger sample size, especially with regards to the *Tabanus* genus will aid in clarifying their relationships. Furthermore, in-depth research should also be conducted in other regions of South Africa; not only on tabanid ecology and composition but also on their role as pathogen vectors.

Species diversity of small mammals and the prevalence and intensity of infestation of the associated fleas (Siphonaptera) across an altitudinal gradient along the Ugab River, Namibia

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Studies have shown that changing altitude is strongly associated with changes in environmental factors such as climate and vegetation. We hypothesized that altitude should be associated with variation in species diversity of small mammals and fleas that they harbor. The study investigated patterns in species diversity of small mammals and the prevalence and intensity of infestation of associated fleas across an altitudinal gradient along the Ugab River, Namibia in January (hot wet season) and May (cold dry season) in 2018. Three sites were selected namely: Outjo (1300m asl), Vingerklip (1000m asl) and Brandberg (400m asl). The species trapped at the 3 sites included: *Micaelamys namaquensis*, *Mastomys natalensis*, *Aethomys chrysophilus*, *Crocidura hirta*, *Saccostomus campestris*, *Gerbilliscus leucogaster*, *Thallomys nigricauda*, *Thallomys paedulcus* and *Elephantulus intufi*. The t-test revealed that there was no significant difference in the species diversity of small mammals at the 3 sites. The overall median number of fleas per host did not differ significantly amongst the 3 sites in the hot wet season (p=0.6065). The prevalence of infestation of fleas was generally higher in Outjo (high altitude site) and lower in Vingerklip (medium altitude). The overall prevalence of infestation of fleas did not differ significantly in the hot wet season amongst the 3 sites (p=0.6051). Prevalence of infestation was high in female hosts at Brandberg and no male hosts were recorded. Male hosts had the highest prevalence of infestation of fleas at Outjo and Vingerklip in the hot wet season. Male hosts had the highest prevalence of infestation of fleas at Outjo and Vingerklip in the hot wet season. Overall species diversity of small mammals was not significantly different among the 3 sites (Outjo (1300m asl), Vingerklip (1000m asl) and Brandberg (400m asl) in the hot wet season and dry cold season (p=9535 for hot wet season and p=9682 for dry cold season). Therefore, the study revealed that species diversity of small mammals is not affected by altitude. Prevalence of infestation of fleas did not differ significantly with altitude during the wet hot season.

Session 4 – Micro- and macroparasites

A survey of captive penguins for avian haemosporidiosis

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The African penguin (*Spheniscus demersus*) is endemic to coastal areas of southern Africa. The species has experienced rapid population declines over the past century as a result of overexploitation for food, habitat modification of nesting sites, oil spillages, invasion by alien species and diseases, and competition for food resources with commercial fishing. It is therefore considered as endangered by the IUCN Red List and a protected species under the National Environment Management: Biodiversity Act (No. 10 of 2004). Furthermore, penguins have evolved without exposure to avian haemosporidia infection (malaria, haemoproteosis and leucozytozoonosis) in their natural environments and are therefore highly susceptible to infection. In such cases, infection can cause severe disease and mortality, especially in captivity where animal densities are high. Multiple mortalities due to avian malaria in a wide range of avian species submitted for necropsy examination to the

National Zoological Gardens (NZG) from aviaries both within and outside of the NZG highlighted the need to investigate this disease further. Blood samples and smears were collected from 46 live penguins at the NZG and analysed by microscopy, PCR-based methods and sequencing of the cytochrome b (*cytb*) gene to determine the prevalence, diversity and phylogenetic relationships of *Plasmodium*, *Leucozytozoon* and *Haemoproteus* spp. in these birds. Infection was correlated with sex, and body condition. Preliminary results of postmortem samples from three penguins that recently died at the NZG indicate that infection by novel haemosporidian lineages (*cytb* haplotypes) may be associated with the death of the penguins. Additional results of the study will be presented and recommendations for better management of the disease will be discussed.

Diversity of avian Haemosporidian parasites in the lowveld region of South Africa

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Avian Haemosporidian parasites of the genera *Plasmodium, Haemoproteus* and *Leucocytozoon*, are widely distributed vector-borne blood parasites that affect most birds and can have serious negative effect on their avian hosts. A large number of these parasites has been described worldwide, but studies in Southern Africa and South Africa in Particular, are lacking and if existent, often make use of the traditional microscopy technique. In the present study, we characterise parasites from the three genera in 1037 birds belonging to 46 species trapped in the Lowveld region of South Africa between 2015 and 2017. We screened for avian haemosporidian parasites using the Nested PCR, targeting the parasites' mitochondrial cytochrome b gene (*cyt-b*). Preliminary results reveal the presence of all three genera in the Park with varying prevalence and diversity. The presence of previously undescribed parasite haplotypes was also observed. We present phylogenetic analyses of all haplotypes as well as preliminary results on the prevalence these parasites. We discuss these findings in the context of host-parasite relationships and draw attention to the need for disease prevalence maps, necessary for biodiversity conservation.

Association of gametocyte profile and socio-demographic factors with malaria burden at Chipulukusu Clinic in Ndola, Zambia

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Malaria in Zambia has increased in some provinces. Gametocytaemia contributes to the variation in spread of malaria. Socio-demographic factors of infected individuals may play a role. Chipulukusu compound in Ndola, Zambia, a city within the medium malaria transmission zone has been known for high malaria burden compared to other compounds. Chipulukusu is also known for high population density but transmission has not been investigated in this light and using sensitive methods. Sixty-seven (67) malaria-infected individuals that tested malaria positive both by RDT and microscopy at Chipulukusu Clinic, Ndola, Zambia were enrolled into the study. Data were collected from June to October 2018 using structured questionnaires and laboratory analyses employing microscopy and Reverse Transcriptase PCR. Using a logistic regression approach, we related malaria burden to the gametocyte profile (Pfs25) and socio-demographic factors of the participants. Gametocytaemia (Pfs25) by PCR was higher (73.1%) than by microscopy (3%). There was no significant difference between parasitaemia positivity by microscopy and gametocytaemia (Pfs25) by PCR among all the socio-demographic variables considered. Income and family size had a significant effect on malaria microscopy positivity. A unit increase in family size had a 19.7% chance of developing gametocytaemia [AOR 1.197 (95% CI: 1.013. 1.414)]. Whether houses were sprayed or not (OR 5.495% CI: 1.593-18.594 and used ITN or not, malaria positivity was still high. Gametocytaemia and hence transmission potential may be underestimated by microscopy. Parasite burden mirrors gametocyte burden and may thus be used as a proxy for estimating transmission. Family size and income are risk factors for malaria in Chipulukusu area.

Genetic diversity of *Entamoeba* spp based on the analysis of the 18SrRNA gene

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Entamoeba histolytica is a significant cause of dysentery and liver abscess disease worldwide. Due to the existence of different clinical outcomes caused by the parasite, one would want to understand the profile of the virulent gene. The present study aimed to determine the genetic diversity of *E. histolytica* through analysis of the nucleotide sequence of the gene. A total of 300 unidentified stool samples were collected and observed under a light microscope for the detection of *Entamoeba* cysts and trophozoites. Genomic DNA was extracted from stool samples using ZR Fecal DNA Zymo MiniPrep extraction kit and was used in a real-time PCR protocol for the detection of *E. histolytica* using specific primers. Another set of specific primers were used in a Conventional PCR to detect the 18SrRNA gene of *Entamoeba* spp. from the positive *E. histolytica* samples. PCR amplicons were sent to Inqaba for sequencing and the results were analysed using mega-5 software. Of the 300 stool samples analysed for *Entamoeba*, 42(14%) were positive by microscopy. The overall prevalence of *Entamoeba* species was 34% when the molecular methods were used. When the samples are subjected to Real-Time PCR for specific detection of *E. histolytica*, 27(9%) samples revealed positive results. However, this study also demonstrated that the qPCR and cPCR showed little correlation because they were unable to detect the same species in an individual. The sequencing of the amplicon obtained from conventional PCR showed that the

species found were *Entamoeba coli* 18 (12%), 6 (4%) were *Entamoeba hartmanni* and 3 (2%) were *Entamoeba dispar*. Amplicon obtained from Real-time PCR showed that the species found were *Entamoeba dispar* 10 (6.7%) and *Entamoeba nuttali* 3 (2.0%). When taken together the most common *Entamoeba* spp. was *E. hartmanni.* The results demonstrate that amebiasis is a major health concern for our region. The causative agent of amoebiasis is currently attributed to two distinct species (*E. dispar* and *E. histolytica*). The findings from the present study showed little correlation between qPCR and cPCR. The differences could be due to the fact that the method used for sequencing was a standard method.

Characterization of the bacterial blood microbiome of dogs and cattle in the Mnisi community, Mpumalanga, South Africa

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All over the world, dogs are nurtured and kept as domestic pets. In sub-Saharan Africa, the majority of dogs are owned by households yet wander about freely. In the Mnisi community, livestock farming is by far the most significant agricultural activity and cattle form the vast majority of domestic livestock reared. Recent research conducted in the area detected tick-borne pathogens of zoonotic and economic importance from domestic dogs and cattle. The aim of this study was the surveillance of the bacterial populations in the blood of dogs and cattle in order to understand the role these domestic animals play as reservoirs of zoonotic and other tick-borne pathogens in the community. The bacterial blood microbiome of 10 dogs and 9 cattle was characterized using circular consensus sequencing on the Pacific Biosciences platform. The average number of sequence reads obtained per sample was 3,034 sequences for dogs and 3,839 sequences for cattle, sufficient to satisfy rarefaction curves that all operational taxonomic units (OTUs) were captured. Approximately 24% of the sequences obtained from the blood of the dogs corresponded to *Ehrlichia canis*, 19.3% to *Anaplasma platys*, 14.8% to *Anaplasma* sp. ZAM dog strain, and 21.4% to *Achromobacter xylosoxidans*. Species with low representation included *Mycoplasma haemocanis* (5%) and *A. phagocytophilum* (0.6%), while 1.6% of the total sequences obtained from canine blood corresponded to other *Anaplasma* spp. From cattle blood, 54% of bacterial sequences obtained corresponded to *A. marginale*, 22.2% to *Anaplasma* sp. Mymensingh, 10.5% to *Anaplasma* spp., and 5.4% to *Anaplasma* sp. Dedessa. Species with low representation included: *Anaplasma* sp. Hadesa (2.7%), *A. centrale* (1.4%), *Bartonella* spp. (0.5%), *A. platys* (0.2%), *Anaplasma* sp. Saso (0.2%) and *A. phagocytophilum* (0.01%). Sequences of *Borrelia* sp., *Brucella* sp., *Bartonella bovis* and the novel pathogen *Ehrlichia minasensis* were also detected in cattle. This study reveals that domestic dogs and cattle in the Mnisi community serve as reservoirs of zoonotic and other economically important *Anaplasma* spp. This study also serves as the first report of the detection of recently described *Anaplasma* species and *E. minasensis* in cattle and the pathogen *M. haemocanis* in dogs in South Africa.

Geographic variation in the body size of ectoparasites on rodents

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Fleas and mites are temporary parasites that spend a variable, but significant, part of their lifecycle in the nest of their host. Microclimatic conditions in the nest can impact the development and body size of nest ectoparasites. Little is known with regard to the effect of climatic conditions on the body size of rodent fleas and mites in natural conditions at a regional scale. Using the flea *Chiastopsylla rossi* and mite *Laelaps giganteus* as models, the aims of the study were to establish 1) if there is a relationship between latitude and parasite body size and 2) whether similarity in parasite body size follows a distance decay pattern or is better explained by extrinsic biotic (host body size) and abiotic (temperature and precipitation) factors. Both species were recorded on the four-striped mouse, *Rhabdomys pumilio*, along the western side of South Africa. Adult rodents were trapped during spring-summer at 13 localities, starting in the south (-34°) and ending in the north (-29°). Each locality was characterized using seven abiotic variables. A total of 86 fleas (42 female and 44 male) and 423 female mites were removed and measured. The effect of latitude on flea and mite body size was tested for each species and sex (fleas) separately while controlling for the effect of host body size. If latitude had an effect, we proceeded to test for the effect of particular climate variables on flea and mite body size. The study found that female flea and mite body size (but not male flea) increased significantly with an increase in latitude (north to south). None of the climate variables affected the body size of female *C. rossi*. In contrast, body size of female *L. giganteus* was significantly affected by PC1, which correlated negatively with several precipitation variables and PC2, which correlated positively with several temperature variables. The effect of latitude on the body size of the two ectoparasites supports previous studies that recorded a similar latitudinal effect on parasitic mites on rodents and other insects.

Session 5 – Microparasites

Assessment of resistance of indigenous Nguni cattle to African animal trypanosomosis primary challenge

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Trypanotolerant cattle are able to survive and remain productive under active trypanosome transmission in endemic areas due to their ability to control parasitaemia and development of severe anaemia. Trypanotolerance (innate resistance) is a genetic trait, heritable and with individual variations within the breed. Currently, there is no information reported on the level of tolerance or susceptibility to trypanosome infections in the indigenous Nguni cattle, though their natural resistance to tick infestation has been demonstrated. Thus, this study was conducted to determine the susceptibility of Nguni and Friesian cattle to Savannah-type *Trypanosoma congolense* strains. Twenty-four Nguni cattle and six Friesians susceptible to animal trypanosomosis were challenged with two *T. congolense* strains previously characterized in Balb/c mice as of high virulence (HV) and the other of low virulence (LV). Additionally, a group of six Nguni cattle were used as uninfected control. Cattle were monitored daily for temperature, blood packed cell volume (PCV) and parasitaemia (scores 1-4). Infected cattle with a PCV ≤19 % for three consecutive days were considered anaemic and required treatment. The Nguni and Friesian cattle infected with the HV and LV did not show elevated body temperature for more than one day. Infected Friesian bovines with either the HV or LV isolates developed parasitaemia (score 3) by day 6, which progressed to severe anaemia thereafter and all animals in the two groups received treatment. All Nguni animals injected with the LV isolate did not require treatment while 5/12 (41.7%) animals injected with HV had to be treated. Moreover, the animals in the untreated group attained higher frequencies of very low to low parasitaemia (scores 1 and 2) compared to those in the treated group. Overall, 19/24 (79.2%) of Nguni animals did not require treatment compared to 100% of Friesians which received treatment. The present study showed that Nguni cattle manifested a high degree of trypanotolerance to infection comparable to the well characterized trypanotolerant cattle in West Africa and can be used in an integrated disease control employing strategic treatment with trypanocidal drugs and insecticides/acaricides treatment.

In vitro **and** ***in vivo*** **evaluation trypanocidal efficacy of curcumin and curcumin nanoparticle on animal trypanosomes**

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African animal trypanosomosis (AAT) is a devastating disease of animals caused by *Trypanosoma congolense, T. brucei brucei, T. evansi*, and *T. vivax*. The disease is endemic in 36 African countries, negatively affecting the economy of these countries by reducing animal production. In the absence of vaccines, various trypanocidal drugs are used for the treatment of nagana. However, few available drugs were discovered decades ago and are either inaccessible to farmers in remote areas or are associated with severe toxicity. Most importantly resistance has widely developed against their usage. Therefore, safe, effective and easily administrable drugs are urgently in need. This study aimed to demonstrate the effect of CUR and CUR-nanoparticle *in vitro* on *T. congolense, T. b. brucei and T. evansi*, and the cytotoxicity effects on the MDBK and NIH 3T3 cells. Additionally, CUR and CUR nanoparticle trypanocidal efficacy were demonstrated *in vivo* against *T. congolense*-infected mice. CUR-nanoparticles were synthesized using the antisolvent precipitation with a syringe pump (ASPS), evaporative precipitation of nanosuspension (EPN) and wet-milling (WM). All the CUR-nanoparticles were 2-fold more effective on the *T. congolense* as compared to free CUR *in vitro,* nonetheless, the in *vivo* tests were conducted only on the ASPS produced nanoparticles. The *in vitro* efficacy of CUR on the trypanosomes was 1.36 ± 0.31; 2.82 ± 1.22 and 2.37 ± 3.07 µg/mL, while the ASPS prepared CUR-nanoparticle efficacy was 0.58 ± 0.50; 10.43 ± 9.43 and 4.86 ± 0.13 µg/mL on *T. congolense*, *T. b. brucei* and *T. evansi*, respectively. Both CUR and CUR-nanoparticles showed moderate efficacy orally, as compared to the intraperitoneal treatment. The efficacy of CUR and CUR-nanoparticle *in vivo* were evidently affected by the solvent, the presence of food and the treatment period. None of the CUR treated mice were cured from the infection, however, the survival rate of the orally treated mice was significantly prolonged as compared intraperitoneally treated mice. CUR-nanoparticles, resulted in a significant suppression of parasitaemia in treated mice although relapse was observed in mice. In conclusion, CUR and CUR-nanoparticles possess moderate efficacy orally on the trypanosomes as compared to the intraperitoneal treatment.

Identification and genotyping of predicted host cell phenotype modulators in *Theileria parva*

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Theileria parva is one of the two *Theileria* species known to cause reversible transformation of their host cells. This protozoan parasite belongs to the Apicomplexan phylum, and causes cattle theileriosis in eastern, central, and southern Africa. The cattle-derived *T. parva* isolate is responsible for the form of theileriosis known as East Coast fever while the buffalo-derived causes Corridor disease. The schizont is the pathogenic stage of the parasite, responsible for the transformation of infected lymphocytes, yet little is known about virulent proteins responsible for host cell transformation in this stage. Thus, the aim of this study was to predict *T. parva* host cell phenotype modulators (HCPMs) using *in silico* methods.

Determination of their gene and protein expression and conservation was analysed using a combination of bioinformatics tools targeting secreted, membrane and cytoplasmic proteins. One hundred and forty-five proteins with predicted functions associated with host cell transformation were identified after excluding proteins with homologs, orthologs or paralogs in non-transforming parasites and the bovine host. The expression of genes encoding the predicted HCPMs was determined using RNA sequence data, previously obtained from the analysis of the transcriptome of the schizont stage of two *T. parva* stocks, representing the cattle-derived and buffalo-derived parasites. The transcriptome analysis revealed expression of 47 genes encoding possible HCPMs, in both *T. parva* stocks, with average RPKM values ranging from just above 10 to >3000. Therefore, the top three genes with the highest expression in both isolates were selected for genetic diversity studies, comparing gene, and protein sequences from cattle and buffalo-derived parasites. Genetic diversity analysis is ongoing, which will give us insight on the evolution, diversification, and migration of this parasite in the African continent based on the allelic diversity pattern among strains.

Can we build a genetic trap for drug resistant malaria parasites?

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Malaria parasites experience an abrupt and radical change in environment when taken up by a blood feeding mosquito. The mosquito host presents a new fitness landscape with very different selection pressures to those previously imposed in the blood of the vertebrate, particularly if that vertebrate was a human using antimalarials. We are seeking to exploit this abrupt change in fitness landscape—especially the switch from drug pressure in humans to absence of drug pressure in mosquitoes—as a genetic trap to minimise the spread of drug resistance. The basic premise of the trap is that mutations conferring drug resistance are no longer advantageous when the parasite transfers to a mosquito host and can even be detrimental to transmission via mosquitoes. We showed that the genetic trap works for the safe and effective drug atovaquone (Goodman et al. *Science* 352:349-353 2016). Common atovaquone resistance mutations cannot transmit via mosquitoes because the mutants, which are no longer under drug pressure, cannot respire sufficiently well in mosquitoes to proliferate. Resistance is trapped in the patient in which it arises. Our recent work has focused on identifying other drugs for which resistance can be trapped. We generated resistance to approved antimalarials azithromycin and clindamycin, which target the malaria parasite apicoplast, in human and rodent malaria parasites and tested the ability of the resistance mutants to transmit via mosquitoes. Both azithromycin and clindamycin resistant parasites develop poorly in mosquitoes and fail to infect new vertebrate hosts suggesting that resistance to these drugs will also not spread readily. We believe that these safe, cheap antimalarials will make ideal partner drugs for emerging, fast acting antimalarials allowing combination therapies that are better able to constrain resistance. The genetic trap drugs could protect the efficacy of the primary compound and also alleviate the spectre of combinatorial use selecting for multi-drug resistance, as has happened previously with sulfadoxine/pyrimethamine (SP) and artemisin combination therapies (ACTs).

Prevalence of rodent haemoprotozoan infections in the Lowveld region of South Africa

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Haemoprotozoan infections are vector borne diseases found in several wildlife species, some of which can affect their ecology and survival. Most of these pathogens have a wildlife reservoir and are potentially zoonotic diseases. Understanding the prevalence and drivers of these infections may contribute towards our understanding of several understudied and even unknown wildlife disease dynamics in subtropical regions. This study was undertaken to understand the effects of season and different habitats on the prevalence of rodent haemoprotozoan infections. A total of 129 rodents from seven species were sampled from four locations in and around the Kruger national park during the dry and wet seasons. Rodents were caught inside and outside long-term herbivore exclosure experimental plots. Blood was collected and screened for *Trypanosoma* spp, *Babesia* spp and *Theileria* spp. Some ectoparasites were collected from the sampled rodents and screened microscopically. All blood smears have been screened microscopically and Initial findings suggest that there is mostly *Theileria* and *Babesia* infections from these rodents. This study will unravel the identity of the currently circulating rodent parasites in the Lowveld region of country which is also home to our largest national park and conservation area. Understanding how haemoprotozoan infections persist in hosts living in varied environments will help us understand the role of the environment in disease prevalence.

Investigating *Rickettsia Africae* infection in *Amblyomma hebraeum* ticks in Mnisi, Bushbuckridge Municipality, South Africa.

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Rickettsia africae is a gram-negative bacterium which causes African Tick Bite Fever (ATBF) in humans. African TBF is a febrile disease mainly affecting travellers to Southern Africa. This bacterium is known to be transmitted by *Amblyomma hebraeum* and *Amblyomma variegatum* ticks. In Southern Africa the principal vector is *A. hebraeum*. This project was performed in a rural community in Mpumalanga province and aimed at addressing knowledge gaps in *R. africae* infection in *A. hebraeum* ticks. Infection rates in adult ticks, eggs and larvae as well as transovarial transmission efficiency of *R. africae* from the tick to its offspring was determined. To accomplish this, adult *A. hebraeum* ticks were collected from cattle and larvae were collected by dragging at the targeted dip tanks. Engorged female *A. hebraeum* ticks were also collected and they were put in a humidifier to oviposit. DNA was extracted from the engorged ticks and the egg masses as well as from the adult ticks and the larvae. After DNA quantification, a *Rickettsia* qPCR was performed to screen all samples

for the target gene, *gltA*. Samples positive for *gltA* were subjected to conventional PCR targeting the *ompA* gene, which is specific for the Spotted Fever Group to which *R. africae* belongs. From the sampled adult ticks, engorged females and egg masses 95.75%, 87.74% and 91.67% were positive for the *gltA* gene respectively. Results from the *ompA* gene screening revealed that 14.28% of adult ticks, 26.56% engorged females and 19.81% egg masses and were positive. The samples positive for amplicons of *ompA* gene will be sequenced to determine the rickettsia species present.

Session 6 – Diagnostics, tools and taxonomy

First molecular detection and identification of *Abbreviata kazachstanica* isolated from roadkill European glass lizard, *Pseudopus apodus* in northern Iran

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Roadkills are frequently found on roads and they are considered as biodiversity loss. However, they are valuable for ecological research as well as parasitological studies. Iran fauna has 125 species of lizards assigned to 36 genera in 8 families including *Pseudopus apodus* (Anguidae) that is the only surviving member of an ancient genus of western Palearctic anguid lizards distributed from southern Europe to central Asia. Guilan Province in northern Iran is a suitable habitat for *P. apodus*, since southern parts of this region are surrounded by Alborz mountains which provides a Caspian climatological situation similar to Mediterranean-like climate. The first and only report regarding parasitic infection of *P. apodus* in Iran has been conducted by Halajian *et al*. (2013). During April 2016 to September 2018, 6 roadkill carcasses (4 male and 2 female) of *P. apodus* were collected and studied for presence of parasites. Internal organs such as esophagus, stomach, small and large intestine as well as lungs were separately examined for helminths. Collected helminths were fixed and preserved in 70% ethanol and later cleared by lactophenol solution. After drawing, the measurements were made using a light microscope and finally the helminths were identified based on keys given by Anderson *et al*. (2009). One of the lizards harbored *Entomelas entomelas* (Rhabdiasidae) in the lungs while three harbored *Abbreviata kazakhstanica* (Physalopteridae) in their stomachs. Physalopteridae nematodes infect a large number of vertebrates, including mammals, birds, reptiles, and amphibians. The genus *Abbreviata*, which currently contains 47 species, has been bisected by Travassos, 1920 from the genus *Physalopteris* and it has a worldwide distribution. Additionally, molecular work was done on *A. kazakhstanica* using COX1 gene and the sequences were deposited in the GenBank. As the available parasitological records are limited in Iranian reptiles, more precise and comprehensive studies with the aim of identifying the parasite fauna and particularly the helminthes diversity are required.

Next-generation sequencing technologies in public health laboratories: a case study of a national uutbreak of listeriosis caused by ready-to-eat processed meat products, South Africa, 2017-2018

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Advances in Next Generation Sequencing (NGS) technologies have transformed public health surveillance and outbreak investigations by providing more accurate pathogen identification and typing, antimicrobial resistance profiling, monitoring vaccines and detect emerging infectious diseases, all with high-throughput capacity, decreasing complexity and cost, turn-around times and efficiency. The Sequencing Core Facility was established in 2016 to provide NGS and Bioinformatics solutions to the National Institute for Communicable Diseases (NICD) centres as well as serve researchers from academic and public institutions in South Africa. In South Africa, between 2017 and 2018, a large and multi-province outbreak of listeriosis, a serious foodborne disease caused by Listeria monocytogenes, was traced to ready-to-eat (RTE) processed meat products using epidemiological investigations and whole-genome sequencing (WGS) analyses. In addition to standard laboratory techniques, WGS was performed for additional subtyping of the outbreak isolates and to support investigation into the source of the outbreak. As of 19 June 2018, 1056 cases leading to 214 deaths were reported to the NICD, South Africa. Multilocus sequence typing of 628 clinical isolates using WGS determined that 571/628 (91%) belonged to the L. monocytogenes sequence type 6 (ST6) and the remainder (9%) to 17 other sequence types. Furthermore, WGS-based core genome multilocus sequence typing (cgMLST) analysis using 1748 core genes showed that all ST6 (the outbreak sequence type) clinical isolates belonged to the cgMLST type CT4148. Isolates of the same cgMLST type were found in RTE processed meat products (including a widely consumed product called "polony") and in the processing environment of the manufacturer (0-4 allelic differences), strongly suggesting that the polony and the other RTE products made in this facility is the source of the outbreak. Popular RTE processed meat products from a single food production facility caused this listeriosis outbreak, which is to date the world's largest. The high-throughput sequencing in conjunction with epidemiological investigation was instrumental in tracing the source of contaminated food, in return preventing further illnesses and possible deaths.

Novel arenavirus isolates from Namaqua rock mice, Namibia, Southern Africa

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Small mammals host diverse communities of both ectoparasites (e.g. fleas and ticks) and endoparasites (e.g. bacteria and viruses). Some parasites are vectors of diseases that infect both humans and wildlife. The main objective of the study was to discover and molecularly characterize novel Hantaviruses and Arenaviruses in small mammals in selected areas of Namibia. Small mammals were trapped from six different regions and screened for both Hantavirus and Arenavirus. The regions are, namely, Hardap, Khomas, Kunene, Okavango,

Omaheke, and Otjozondjupa. Nucleotide sequence analysis of PCR of the partial large segments of Arenavirus were amplified from RNA extracted from the lungs of small mammal hosts. Hantaviruses were not prevalent in any of the small mammals trapped. However, two new Arenaviruses, not previously recorded in Namibia were both isolated from the Namaqua rock mouse, *Micaelamys namaquensis*, trapped in Okahandja and Mariental. The Arenavirus isolated from rodents trapped in Okahandja and Mariental respectively, is closely related to the Merino Walk virus isolated from the Bush vlei rat, *Myotomys unisulcatus,* captured in South Africa and Luna virus discovered in Zambia in the host, the Natal multimammate mouse, *Mastomys natalensis*.

Validation of multiplex *Ehrlichia canis*, *Babesia rossi* and *Babesia vogeli* real time PCR assay

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Canine babesiosis and canine ehrlichiosis are tick-borne diseases caused by *Ehrlichia canis*, *Babesia rossi* and *Babesia vogeli* and transmitted by *Rhipicephalus sanguineus* ticks*. B. rossi* and *B. vogeli* present different clinical signs, *B. rossi* can cause biliary also known as "tickbite fever". Infection with *B. vogeli* is usually mild, while as *B. rossi* is virulent and ehrlichiosis normally presents with non-specific clinical signs. Coinfection with these pathogens is common and there is a need to differentiate the infections so that animals can be treated accordingly. Real-time PCR is fast, specific and sensitive and multiplexing of the assay to detect multiple pathogens from the same sample drastically reduce the costs of testing. In this study we multiplexed existing in-house *E. canis*, *B. rossi* and *B. vogeli* real-time PCR assays using TaqMan® MGB probes. The probes were labelled with different dyes namely FAM (6-carboxyfluorescein), VIC and NED respectively. Validation of this assay was performed with diagnostic samples submitted to Department of Veterinary Tropical Diseases laboratories and results compared to the reverse line blot hybridization assay.

Phenotypic and genotypic screening for acaricide resistance in Ixodid ticks from Southern Zambia

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Ticks are obligate haematophagus arthropods associated with maintenance and transmission of various pathogens to humans and animals. In animals, ticks cause direct or indirect health effects, leading to reduced productivity, morbidity and/or mortality amounting to billions of dollars annually. In order to curb the threat posed by ticks on their livestock, farmers often have to implement various tick control strategies, the most common of which is use of acaricides. Unfortunately, due to rampant misuse, there have been increased reports of acaricide resistance in tick populations. In Zambia, resistance towards organophosphate-

based compounds has previously been reported in some regions of the country. The purpose of this study was thus to screen for resistance in the most important tick species (*Rhipicephalus* and *Amblyomma* species) against commonly available acaricides in the country using both bioassay and molecular methods. The Larval Packet Test (LPT) assay was used to detect phenotypic resistance in Rhipicephalus ticks against commonly available drugs covering the organophosphates, pyrethroid and amitraz classes. Presence of previously reported resistance – associated genetic mutations in the Carboxylesterase, Octopamine and Voltage-gated sodium channel was screened using PCR-RFLP and sequencing to detect resistance against organophosphates, armitraz and pyrethroids, respectively. Probit analysis of LPT assay results at doses of 0.0002%, 0.002%, 0.02%, 0.2%, 0.4%, 1% & 2% Cypermethrin (Pyrethroid) showed an LD50 of 0.006% and LD99 of 0.55%, with the manufacturer's recommended dose of 0.003% causing 34.5% mortality. On molecular analysis, none of the previously reported resistance – associated genetic mutations were detected in all the ticks used in our study. Preliminary indications from LPT are that there is acaricide resistance in the ticks. Based on the findings from our molecular analysis, it seems that the ticks in our study area have developed acaricide resistance using as yet to be identified mechanisms. The implications of these findings are discussed.

Molecular detection and genetic characterization of *Rickettsiae* species in ticks collected from southern Zambia

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Tick-borne rickettsioses caused by spotted fever group (SFG) *Rickettsiae* are systemic febrile illnesses currently considered as emerging or re-emerging diseases across the globe. They are usually associated with ixodid ticks which serve as vectors, reservoirs, and amplifying hosts, with transmission to vertebrate hosts occuring through salivary secretions during the tick feeding process. Screening of ticks for SFG *Rickettsiae* using specific and sensitive molecular techniques is used as a measure of risk for emergence of disease in a given locality. This study sought to determine the prevalence of *Rickettsiae* pathogens in ticks. Determination of *Rickettsiae* prevalence in ticks was done by amplification of the *omp*A gene and confirmation by amplification of the *omp*B gene. Genetic characterization of positive samples was done by sequence analysis of the *omp*B gene. The Basic Local Alignment Search tool (BLAST) was used to search for nucleotide sequence similarity between those determined in this study and reference strains in GenBank. A phylogenetic tree was constructed using a Maximum Likelihood method with bootstrap analysis (1000 replicates) to determine the evolutionary relatedness of Zambian rickettsia species. A total of 136 adult ticks were collected from Chirundu and Namwala Districts of Southern Zambia. The adult ticks were subsequently identified morphologically into *Amblyomma spp.* (15/136), *Hyalomma spp.* (31/136), *Rhipicephalus spp.* (90/136). The overall prevalence of *Rickettsiae* in ticks was 8.8% (12/136). By tick species, the prevalence of infection in *Amplyomma*, *Hyalomma* and *Rhipicephalus* was 26.7% (4/15), 9.7% (3/31) and 5.6% (5/90) respectively. Through BLAST sequence analysis based on the ompB gene, all the Rickettsia in our study were identified as *Rickettsia africae* (98.8% - 99.5% similarity). Further characterization using phylogenetic analysis grouped the *Rickettsia africae* in our study with those from Kenya. We report the presence of *Rickettsia africae* in the ticks collected from southern

Zambia. Considering the potential exposure of humans in this area to ticks through agricultural practices, the finding of Rickettsia in the ticks indicates that the human population is at risk of infection. There is thus need to raise awareness about this pathogen in the community and also amongst health professionals.

Forensic entomology research and application in Southern Africa: a systematic review

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The use of forensic entomology is well established in the northern hemisphere whereas it is still emerging in the southern hemisphere, where most of the current research is not explicitly undertaken in the context of forensics. This review aims to provide an update on the current status of forensic entomology research and its application in relation to estimation of post-mortem interval (PMI) in various criminal investigation such as, murder cases, human neglect and poaching of wildlife in southern Africa, among other issues. A literature search was conducted on Google Scholar, PubMed and Sci-hub databases using the Boolean operators in combination with the key words. Viewed studies focused on arthropod diversity during different stages of carcass decomposition, effect of seasons on the abundance and diversity of carrion feeding arthropods species during carcass decomposition and diurnal and nocturnal oviposition of forensically important insect species during carcass decomposition. It was observed that arthropod species establishing on a decomposing carcass are potentially useful in the estimation of PMI and determining clues in case of criminal investigations. To date, there is scarcity of research of forensic entomology hence its application in southern Africa. Therefore, future studies on application of forensic entomology in various criminal investigations such as murder cases, human neglect, and wildlife poaching in southern Africa is still needed.

Session 7 – Helminthology

Gastrointestinal helminth assemblages of the common warthog, *Phacochoerus africanus*, in KwaZulu-Natal Province, South Africa

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Little work has been done on the helminth parasites of one of the most iconic suids on the African continent, *Phacochoerus africanus*, with most papers focusing on taxonomy, and only a few studies describing the composition of its helminth assemblages. In the present study, thirty common warthogs from the Pongola Game Reserve, South Africa were examined for helminth parasites. Their gastrointestinal helminth assemblages comprised the digenean trematode *Gastrodiscus aegyptiacus*, representatives of the cestode genus *Moniezia* and seven species of nematodes. A metacestode of *Taenia hydatigena* was found in the mesenteries of a single warthog. No helminths were detected in the heart, lungs or liver of the hosts examined. *Probstmayria vivipara*, *Murshidia hamata* and *M. pugnicaudata* had the highest prevalence and were the most numerous of all helminth species collected, followed by *Physocephalus sexalatus*. No effect of host sex or age on the incidence of *Moniezia* sp. was found. The abundance of *Murshidia* spp. did not differ between males and females but was higher in adult than in juvenile hosts. Conversely, host age did not affect burdens of *Trichostrongylus thomasi*, but males harboured more worms than females. Furthermore, helminth assemblages in male warthogs were found to be more species rich than those in females, although this finding was not highly significant. Helminth communities in the three genera of wild suids in sub-Saharan Africa are largely unique, but more helminth species are shared between *P. africanus* and *Hylochoerus meinertzhageni*, than between these hosts and *Potamochorus larvatus*, possibly as a result of the closer relatedness of the former two species.

Farmers' perceptions on the incidence of gastro-intestinal parasites in sheep in the Eastern Cape, South Africa

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Gastrointestinal parasites (GIPs) are known to affect small ruminant productivity worldwide especially in tropics and sub-tropics. Globally the most common nematode species known to affect small ruminants are *Haemonchus contortus*, *Trichostrongylus colubriformis*, *Teladorsagia circumcincta.* The aim of the study was to investigate the farmers' perceptions on the incidence of gastrointestinal parasites in small ruminants in the Eastern Cape of South Africa. A questionnaire survey was carried out in three agro-ecological zones, humid (Wartburg), semi-humid (Allen waters) and arid region (Cradock commonages) from September to November 2018. Of the 108 farmers interviewed during the survey, 68% were males and 32% were females. The highest percentage (85%) of livestock is owned by older age > 55 years and younger age <36 years owned 15% across all vegetation types. Farmers reported that 83% of their animals are affected by wire worm during hot-wet months,

followed by hot dry months (14%) and least during cold months (2%). Farmers perceived that 85% of young animals were susceptible to parasitic infection mostly by wireworm than mature sheep (15%) across all vegetation types. Very few farmers (8%) had knowledge of the GIP lifecycle, the rest of respondents (92%) do not know about the GIPs infection and their biology across all vegetation types. It was also perceived in all vegetation types that the increase in the occurrence of worms over the years is greatly associated with resistance of the strain (wireworm) to deworming remedies (67%) and followed by changes on the weather patterns (33%). Farming experience were significantly ($p < 0.05$) affected by gender and age. Significantly higher helminths were reported in humid zones than other agro-ecological zones ($p<0.05$). From the study, it is evident that farmers perceive that young animals are more susceptible to gastrointestinal parasites. Therefore, knowledge concerning gastrointestinal helminth biology and epidemiological infection patterns caused by nematode species is essential in the development of appropriate control strategies. Farmers must adhere to remedy instruction and alter deworming remedies to avoid the build-up of resistance.

Host-parasite-vector interactions and elucidation of an Amphibian Filarial Nematode life cycle

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Filarial nematodes are important microscopic, thread-like roundworms known as agents of medical and veterinary disease. That includes lymphatic, subcutaneous and dirofilariasis, e.g. elephantiasis, river blindness, and hearthworm, respectively. These predominantly tissue-dwelling nematodes are characterised by a broad-host-range including amphibians, reptiles, mammals, and birds, infecting up to 170 million people worldwide. They have an evolved lifecycle, comprising a vertebrate host in which adult worms release microfilaria into the bloodstream. Microfilariae are then ingested with the blood meal of a haematophagous arthropod. Following complete development, the parasite can be transmitted to a new host. In the present study we were able to elucidate this cycle in the Guttural toad host, *Sclerophrys gutturalis* and the mosquito vectors, *Uranotaenia* (*Pseudoficalbia*) *mashonaensis* and *U.* (*Pseudoficalbia*) *montana*. Mosquitos enticed to feed on infected toads were progressively dissected, and nematodes extracted and fixed according to the stages of their development. Light and scanning electron microscopy were used to study morphology, and PCR amplification to molecularly characterise and link each of these developmental stages. Additionally, we report on the unique host-seeking behaviour of *U. mashonaensis* and *U. montana* who respond to the calls of their toad hosts. This unique host-vector evolution may be the reason that only 12% of the male (6/50) *S. gutturalis* were infected with microfilaria as compared to 0% of the females (0/21). This study is the first to elucidate the life history of an African amphibian filarial nematode and to provide information on the complex interaction between host, parasite and vector.

Larval Stages of Trematodes associated with snails from Lake Victoria, Kenya

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Lake Victoria is the second largest freshwater lake in the world and is home to numerous flora and fauna. Most studies on snails in Eastern Africa have always focused on specific pulmonates that are widely known as intermediate hosts for trematode parasites of livestock and man. This study aimed to investigate the parasite fauna of a variety of prosobranch and pulmonate snails of Lake Victoria, Kenya. Out of the 1,106 snail specimens inspected, 65 % belong to prosobranch species (*Bellamya unicolor*, *Melanoides tuberculata* and *Pila ovata*), which tend to receive less attention in parasitological studies in Africa. Of all the snails studied, 13 % had infections with larval stages of digenean trematodes. The highest trematode prevalence (P) was recorded in *M. tuberculata* (Thiaridae) (66 %), followed by *Radix natalensis* (Lymnaeidae) (18 %), *P. ovata* (Ampullariidae) (15 %), *Bulinus ugandae* (Planorbidae) (13 %), *B. unicolor* (Viviparidae) (8.9 %) and *Biomphalaria* spp. (Planorbidae) (3.9 %). Morphological study of the trematode larvae revealed 15 different cercarial types: echinostome, fucocercariae, gymnocephalous, megalurous, monostome, parapleurolophocercous and 9 xiphidiocercarial types. Armatae xiphidiocercariae with long sword shaped stylet (50 – 67 µm) which did not match existing literature records was recovered from *Pila ovata* (P = 12 %). This study also gives the second report of *Thapariella* sp. (Thapariellidae) in Africa. *Thapariella* sp. metacercariae were recovered from *B. unicolor* (P = 7.6 %) and *P. ovata* (P = 4.6 %)). Parapleurolophocercous cercariae of: *Haplorchis pumilio* (Heterophyidae) which encyst in fish, and potentially infect domestic animals and humans, were recovered from the thiarid snails *M. tuberculata* (P = 57 %). *Fasciola gigantica* rediae (Fasciolidae), parasite of livestock was recovered from *R. natalensis* (P = 3.6 %). Further studies are on course including detailed morphological descriptions and molecular analyses of the specimen.

Species and site contributions to beta diversity of gastro-intestinal nematodes and cestodes in *Rhabdomys dilectus* and *Rhabdomys pumilio*

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A fundamental aim of parasite ecology is to understand the mechanisms behind spatial variation in diversity and structure of parasite assemblages. We examined the Species Contributions to Beta Diversity (SCBD) and Local Contributions to Beta Diversity (LCBD) of

parasitic gastrointestinal helminths (nematodes and cestodes) from phylogenetically similar South African rodents, *Rhabdomys dilectus* and *Rhabdomys pumilio*, from 20 localities across South Africa. Despite the two *Rhabdomys* spp. being morphological similar, they differ substantially in body size, habitats and sociality. We asked whether the variation in life history traits and infection parameters are associated with SCBD of helminths and whether variation in environmental factors, host population density and species richness of host communities are associated with LCBD of component assemblages of helminths. We also took into account spatial factors to test whether LCBD of helminth assemblages demonstrate geographic structure. The contribution of helminth species parasitic in both hosts to beta diversity significantly increased with characteristic prevalence of these species, whereas mean abundance, type of life cycle and location in the host's gut had no effect on SCBD. The LCBD of helminth assemblages showed a significant positive correlation with environmental factors in both host species: helminth assemblages of *R. pumilio* from the Karoo-type biomes contributed significantly more to total beta diversity than those of *R. pumilio* trapped in the Fynbos. Our results suggest that predictors of variation in SCBD and LCBD may substantially differ between parasites of different life history and/or parasite communities at different hierarchical scales.

Population genetic structure of *Haemonchus contortus* in Limpopo Province, South Africa: a preliminary study

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Haemonchus contortus is a parasite of high biotic potential, together with high infection rates in small ruminants and a direct lifecycle, which results in a large effective population with marked genetic variability. Genetic information such as genetic diversity portrayed on the phylogenetic tree is essential for a better understanding of ecology, epidemiology and evolution of *Haemonchus contortus*. The aim of this study was to assess the genetic diversity amongst *H. contortus* populations in 5 districts of Limpopo province of South Africa. DNA was extracted from the *Haemonchus* L_3 stage larvae infecting sheep in Capricorn, Mopani, Sekhukhune, Vhembe and Waterberg districts. Thereafter the 18S rRNA gene was amplified by PCR and sequenced. Phylogenetic analysis of the 18S rRNA gene revealed that there is no significant genetic diversity amongst the *H. contortus* populations in Limpopo. All analysed *H. contortus* isolates appeared to be homogenetic. Furthermore, the South African *H. contortus* Limpopo isolates clustered in the same group with Asian isolates. The observed homogenecity can be attributed to the fact that the *H. contortus* and their host (sheep) in Limpopo are exposed to the same environmental conditions (weather, rainfall, humidity etc.) resulting in similar selective pressure for the isolates. Data obtained from this study can possibly be used to analyse the virulence of the *H. contortus* isolates as well as their response to anthelmintic treatment by continually studying the ecological changes and changes in the genetic make-up of the parasite. This is an ongoing study where other conserved genes of *H. contortus* populations in Limpopo districts will be analysed.

Session 8 – Fish parasites

Exploring the diversity of *Diplostomum* (Digenea: Diplostomoidea) in freshwater fishes in South Africa

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The genus *Diplostomum* is a large group of widely distributed trematodes utilising a three-host life cycle with freshwater snails and fishes as intermediate and piscivorous birds as definitive hosts. The metacercarial larval stages parasitising fish are important pathogens. Application of molecular methods based on multiple markers rapidly increased our knowledge of the species diversity within *Diplostomum* in the last decade. Out of 80 nominal species within *Diplostomum* known worldwide, 10 species, 11 species-level genetic lineages and 27 unidentified species from North America, Europe, Asia and Africa have been molecularly characterised. Nine species of *Diplostomum* have been reported from fishes in Africa, with molecular confirmation available only for a *Diplostomum* sp. from Nigeria. In South Africa, only two species have been morphologically characterised. Thus, our knowledge on the diversity of *Diplostomum* as well as their effect on the fish hosts is limited and studies dedicated to this group of parasites are of high importance. Our study aimed to explore the diversity of *Diplostomum* in freshwater fishes in South Africa and the effect of the infection with *Diplostomum* on fish behaviour. A total of 141 fishes of 16 species from 9 families were examined in the Phongolo, Riet and Mooi Rivers during expeditions to Ndumo Game Reserve, Mokala National Park, and Boskop Dam in 2016–2019. Metacercariae were recovered from the eye lenses of 38 fishes of five species: *Anguilla bengalensis labiata*, *Oreochromis mossambicus*, *Pseudocrenilabrus philander*, *Synodontis zambezensis* and *Tilapia sparrmanii*. Twenty-two *S. zambezensis* were also used in behaviour experiments. Morphological and molecular analyses based on three genetic markers (*cox*1, 28S rDNA and ITS1-5.8S-ITS2), revealed the presence of three species. One species was conspecific with *Diplostomum* sp. reported in Nigeria. The remaining two species were closely related to *Diplostomum* spp. reported in Asia. No significant effect of infection with metacercariae on the behaviour of *S. zambezensis* was recorded. Our study is the first dedicated assessment of *Diplostomum* in fishes in South Africa, the first to provide detailed morphological descriptions with molecular data for South African species and the first to study the effect on host behaviour.

An update on the taxonomy of freshwater gastropods and their complete trematode fauna in Lake Kariba, Zimbabwe

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Studies carried out on gastropod-trematode associations in Lake Kariba have largely focused on *Schistosoma hematobium* and *Schistosoma mansonii*, and their intermediate gastropods-host *Bulinus globosus* and *Biomphalaria pfeifferi*, respectively. The most recently documented study was carried out in 2003. Effective strategies to prevent health, veterinary and economic losses; caused by trematodes, largely depend on the existence of accurate current information on freshwater gastropod-trematode associations. This current study serves as an update on the gastropod- and trematode fauna in Lake Kariba. Sampling was carried out monthly between May-2017 and April-2018. Sixteen sites along the littoral zone of Lake Kariba were sampled for snails using standardised methods. All snails were identified morphologically using malacological keys, and snail shedding experiments were carried out monthly to determine trematode species present in Lake Kariba, and the prevalence of trematode infection in the snails. Molecular barcoding was carried out on the snails after shedding experiments, to determine their species as well as pre-patent infections by trematodes. Morphological and molecular results for gastropod identification were compared and molecular barcoding results were used to conclude the identity of obtained species. It was concluded that there are at least 9 gastropod species present in Lake Kariba, including six previously undocumented species; namely, *Bulinus truncatus, Bulinus forskalii, Gyraulus* sp., *Pseudosuccinea columella, Radix* sp. and *Succinea* sp. Six trematode families were detected; namely, Notocotylidae, Psilostomatidae, Paramphistomoidae, Fasciolidae, Diplostomatidae and Leucochloridiidae,in five of the gastropod species, namely *P. columella, Radix* sp., *Succinea* sp. and *B. forskalii* that were each infected by at least one trematode species, with *B. truncatus* being infected by at least three species. It was concluded that there has been a shift in potential trematode intermediate host composition in Kariba. These results are a good foundation for further research into the exact species of trematodes in Kariba and the Zambezi basin at large, which would be essential in the development of effective strategies to prevent public and veterinary health, aquaculture and economic losses in Kariba. The study will also shed light on our understanding of invasion biology through studying such host-parasite systems.

Parasite Fauna of Nile Perch *Lates niloticus*, Nile Tilapia *Oreochromis niloticus* and *Haplochromis* spp. from Lake Victoria, Kenya

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Lake Victoria is the largest inland water body in Africa and supports numerous flora and fauna. The fish community has been influenced over the years by the introduction of new species, especially Nile perch *Lates niloticus* and Nile tilapia *Oreochromis niloticus*. For this study, 103 specimens of *L. niloticus* (Latidae), 165 specimens of *O. niloticus* (Cichlidae) and 144 specimens of *Haplochromis* spp. (Cichlidae) were examined. This is the first comprehensive description of the parasite community of these fish taxa from the lake. A total of 21 parasite taxa were recovered from the cichlids and only six from *L. niloticus*. Myxosporea was the most dominant taxon with *Henneguya* sp. cysts (Prevalence (P) = 79

%, Mean intensity (MI) = 26) from *L. niloticus* and *Myxobolus* sp. cysts from *O. niloticus* (P = 44 %, MI = 6). For the haplochromines, digenean *Neascus* sp. was the most dominant taxon (P = 37 %, MI = 13). From *O. niloticus*, 5 monogenean species: *Cichlidogyrus sclerosus*, *C. halli*, *C. tilapiae*, *Scutogyrus* sp. and *Gyrodactylus cichlidarum* were recorded, while from *Haplochromis* spp., only one monogenean: *C. haplochromii*, was recorded. Other taxa that were recorded from the cichlids include: digeneans *Clinostomum* sp., *Euculinostomum* sp., and *Tylodelphys* sp.; acanthocephalan *Acanthogyrus* (*Acanthosentis*) *tilapiae*; cestode *Amirthalingamia macracantha*; two nematode species; bivalve glochidia; leeches and crustaceans *Lamproglena monodi*, *Ergasilus* sp. and *Argulus* sp. Fish size was an important factor for infection levels in *L. niloticus*. The P and MI for *Henneguya* sp. was higher in smaller fish, while the Monogenea *Diplectenum lacustris* was prevalent in bigger fish. In addition, nematodes *Cucullanus* sp. and unidentified species were found only in bigger fish, though at very low prevalence and intensities. Further studies are on course including detailed morphological and molecular analysis for species description and identification of the parasite specimen.

New geographical distribution record, scanning electron (SEM) and phylogenetic analyses of *Glossidium pedatum* and *Orientocreadium batrachoides*

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Examination of the intestine of *Clarias gariepinus* (Burchell, 1822) from the Incomati Basin, southern Mozambique revealed the occurrence of two digeneans, *Glossidium pedatum* Looss, 1899 and *Orientocreadium batrachoides* Tubangui, 1931. Based on SEM, only *G. pedatum* revealed an undescribed feature, two terminal lobes covered with spines. Currently, for both species only morphological characterization is available, and for the first time, molecular data are provided and a systematic position of *Glossidium* within the major taxa Plagiorchioidea is analysed. Sequences were analysed through Bayesian inference and Maximum likelihood estimation using 18S and 28S rRNA genes. The obtained phylogenies revealed that the family to which *Glossidium* should be assigned is not identifiable, although it is closely related to members of the family Haematoloechidae. This finding confirms the current position based on morphology, as *insertae sedis*. The Orientocreadiidae is a monotypic family. The genus *Orientocreadium* contains 28 species; one of these, *O. pseudobagri,* has been molecularly characterized. Based on the topology, this species formed a sister clade with *O. batrachoides*, which in turn is closely related to members of the family Leptophallidae. However, phylogenetic analysis supported the placement of both species within the major clade Plagiorchioidea, confirming the position currently conceived in systematic classification based on morphology.

A fluke discovery – digeneans of *Clinus superciliosus* from the coast of South Africa

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The Super klipfish, *Clinus superciliosus* (Linnaeus, 1758) (Perciformes: Clinidae), is a common residential intertidal fish species that is distributed from the Kei River mouth in South Africa to the Skeleton Coast in Namibia. As it is a top predator in the intertidal zone, it is invaluable for the functioning of the vibrant intertidal ecosystem, since this fish species has been reported as a host for several species of digenean trematodes. Bray (1987) reported that the Super klipfish collected from Port Elizabeth are hosts for two digenean species, *Helicometra fasciata* (Rudolphi, 1819) Odhner, 1902 and *Coitocaecum capense* Bray, 1987, the latter being a newfound species that he then described. However, literature suggests that our knowledge on this parasitic group from South Africa, especially those found in marine habitats, is fragmentary. Thus, the aim of our study was to further explore the trematode parasitic fauna of this fish species. A total of 29 specimens of *C. superciliosus* were collected from Tsitsikamma National Park (n = 7), Jeffreys Bay (n = 14) and Hermanus (n = 8) on the South Coast of South Africa in 2017−2018. Morphological and molecular analyses based on 28S rDNA, ITS2 and *cox*1 sequence data confirmed the presence of five digenean species from the families Opecoelidae Ozaki, 1925 (adults of *Coitocaecum* spp. and *Helicometra* sp. from the intestine) and Strigeidae Rialliet, 1919 (metacercariae of *Cardiocephaloides* spp. from the eyes). The overall parasite prevalence was high (P = 66 %) and the highest diversity of digenean trematodes (n = 5) was recorded in *C. superciliosus* from Hermanus. Not only does our study provide the first report of *C. superciliosus* as a second intermediate host for digeneans, but it is also the first study to molecularly characterise marine digenean trematodes from South Africa, thereby contributing valuable additions to the trematode molecular database. From a modest study such as this, it has become evident that the general speculation regarding the under-description of marine digeneans from fishes in South Africa might in fact be realistic. This further supports the notion that additional investigative studies on the diversity of this enigmatic parasitic group should be conducted.

Scanning electron microscopy and molecular analysis of male *Lamproglena monodi* (Copepoda: Lernaeidae) infesting *Oreochromis niloticus* (Pisces: Cichlidae) in Kenya

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The crustacean copepod *Lamproglena monodi* was first described by Capart in 1944, from Lake Mweru in the Democratic Republic of Congo from the gill filaments of cichlid fishes. He mentioned the presence of a male but did not provide morphological details. Fryer in 1961 reported a single male, associated with a female on the gills of *Haplochromis* sp. and provided a line drawing from this specimen. He also mentioned that the rami of the

swimming legs are two segmented and that the maxillipedes have five terminal spines of which two are very small. He collected *L. monodi* from Lake Victoria and Victoria Nile. In the present study, non-parasitic *L. monodi* males were collected from the gills of *Oreochromis niloticus* from Kibos Fish Farm, Kisumu County, Kenya. The study provides a detailed characterization of the adult male of *L. monodi* using morphological features, with an aid of scanning electron microscopy (SEM) and the confirmation of the species by DNA analysis. The small subunit (18S rDNA) fragments and larger subunit (28S rDNA) 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 providing the first genetic information on *L. monodi*. This study provides additional morphological characteristics which include: a single pair of uniramous maxillae which terminates in a sickle shaped claw, two small setae arising from the base of the terminal claws of maxilliped, two segmented antennule, tri-segmented antenna with setae and claws and serrations on the mandibles,he biramous first four pairs of swimming legs with a double segmentation on the ramus of each leg, spine, and setae, a pair of atrophied unsegmented fifth legs, a pair of testes, And rows of papilla and setae on the terminal end of either side of the genital segments. In conclusion, the additional morphological details and DNA typing leads to a better understanding of the elusive male of *L. monodi* and extend the geographical range of the species to Kenya.

POSTER PRESENTATIONS

Investigating the relationship between novel farming practices and zoonotic disease transmission at the wildlife/livestock interface: A case study at a commercial farm and conservancy in Beaufort West, South Africa

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As agricultural activities in South Africa continue to intensify, wildlife farming continues to gain popularity among farmers within the sector. Although the adoption of this strategy has improved economic growth and development, it has elevated the risk of zoonotic disease transmission between wildlife and livestock. While a wealth of information exists for emerging and reemerging zoonotic diseases, the dynamics of disease transmission between livestock and wildlife remains poorly understood. This is concerning as environmental conflicts between domestic populations and wildlife are capable of compromising both agricultural productivity and human health. This study was aimed at elucidating bacterial community composition of wildlife and livestock populations at a commercial farm and conservancy in Beaufort West, South Africa using 16S rRNA Next Generation Sequencing (NGS). Our results showed no significant differences in bacterial diversity between wildlife and livestock populations. There was however a tendency for livestock bacterial assemblages to dominate the interface between the two populations. Our analysis also led to the detection of five pathogenic bacterial strains: *Eschericia coli*, *Egerthella lenta*, *Clostridium gasigenes*, *Lactobacillus acidophilus* and *Exiguobacterium sibiricum*, all of which carry a human health risk and ability to compromise economic activities at the farm.

Assessment and genotyping of a novel vaccine candidate for *Theileria parva* infections

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Theileriosis is a lymphoproliferative tick-borne disease of cattle and other wild ruminants. It is caused by infection with a protozoan, *Theileria parva,* which is vectored by *Rhipicephalus sp.* ticks. This disease is prevalent in cattle throughout most parts of Central, East and southern Africa. There are various control and treatment methods in place for *T. parva* infections; however, they all have drawbacks and limitations. The available vaccine, Muguga cocktail does not confer protection against all field strains, particularly buffalo-derived *T. parva* infections. Attempts to develop a subunit vaccine using different candidate antigens have been promising but these have shown limited efficacy due to antigenic and genetic

diversity of *T. parva* strains in the field. Thus, there is a need to search for additional vaccine candidates. Our laboratory identified potential vaccine candidates using a genome-wide *in silico* approach. Secreted antigens expressed in the pathogenic schizont stage of the parasite were the main targets. Ideally, a vaccine candidate that is genetically conserved in both cattle- and buffalo-derived *T. parva* isolates is preferred to provide broad spectrum immunity against the different types of *T. parva* strains. Thus, the aim of this study was to assess and genotype one of the possible antigens identified as a novel vaccine candidate. The identified candidates were ranked based on their level of expression in the schizont stage of the parasite. RNA sequence data produced from the transcriptome analysis of two *T. parva* stocks representing cattle- and buffalo-derived parasites, using next-generation sequencing, was employed. The candidate with the highest expression levels in the schizont stage was selected for genetic diversity analysis, in cattle- and buffalo-derived *T. parva* isolates. Thus, specific primers were designed and optimised for PCR amplification and sequencing of the gene encoding the target 'antigen'. The selected candidate seems to be conserved across both buffalo and cattle samples as well as between geographical areas. These findings indicate that this hypothetical protein could be a good vaccine candidate.

The development of a colorimetric nanoparticle assay for diagnosis of African trypanosomiasis

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African Trypanosomiasis is a vector-borne disease that is caused by protozoan parasites of the genus *Trypanosoma* infecting both humans and animals and is a major health and economic problem in Africa. The aim of this study was to develop a colorimetric nanoparticle assay for diagnosis of African trypanosomiasis caused by subgenus *Trypanozoon* species, including, *T. brucei* subspecies, *T. evansi* and *T. equiperdum*. Gold nanoparticles were synthesized, coated with 11-Mercaptoundecanoic acid and fused with the nucleic acid probe of RIME gene of *Trypanozoon* species. The particles stability was tested by undergoing a salt stress test, PBS (pH)- and BSA test. The results revealed that the particles were stable whereby they constantly produced same absorbance of 520 nm. The reaction of nanoparticle fused with RIME probe with DNA amplicons of *Trypanozoon* species gave a positive reaction whereby there was a colour change from dark purple to ruby red whilst reaction with negative controls remained dark purple. The development of a colorimetric diagnostic assay for the detection of *Trypanozoon* species was successful. Future studies will be conducted to determine the detection limit and then validate the assay by screening field derived samples for the presence of trypanosome infections.

Morphological and molecular characterization of *Eimeria* species among chickens in KwaZulu-Natal Province, South Africa

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Coccidiosis is a renowned intestinal disease of chicken and other livestock, which affects chicken's productivity and welfare. The economic impact of this disease has been a major constraint in the poultry industry. Effective diagnosis of these parasite species is imperative for efficient anticoccidial control. Despite the prevalent of this parasite species in different regions, there is paucity of information on these species in South Africa. The objective of this study was to use both morphological and molecular diagnostic techniques to determine the prevalence of *Eimeria* species in broiler chicken in KwaZulu-Natal, South Africa, from January to October 2018. Using standard saturated salt fecal floatation method, *Eimeria* infection was detected in 10 out of 12 farms (10/12; 83.3%) sampled which include 19 out of 41 pens (19/41; 46.3%). Molecular diagnosis based on species-specific primer targeting the internal transcribed spacer (ITS)-1 region confirmed the presence of four different species of *Eimeria* in all positive pens detailed as follow: *Eimeria tenella* (13/19; 42.1%); *E. maxima* (9/19; 47.4%) and *E. mitis* (8/19; 42.1%) while *E. acervulina* (5/19; 26.3%) had the lowest prevalence. Mixed species infection was also detected in 7 out of the 12 farms (7/12; 58.3%) and 11 out of 19 positive pens (11/19; 57.9%). This study shows that coccidiosis is widespread in chicken and an efficient control strategy is therefore important to curtail this disease in KwaZulu-Natal province.

Preliminary study of parasites of three ornamental fish species in Limpopo Province, South Africa

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Keeping ornamental fish as pet is a popular global hobby. In South Africa, the majority of ornamental fish sold locally are imported from mostly Asian countries including Singapore, Malaysia, Japan and Taiwan. Knowledge on diseases/parasites is important as ornamental fish can transfer diseases/parasites to indigenous species. Thus, if management strategies

are not efficient, alien parasites can be transferred to native aquatic ecosystems. However, limited information is available on parasitic infections of ornamental fishes imported to South Africa. This study was designed to assess the parasites of three of the popular ornamental fish in Limpopo Province i.e. goldfish, *Carassius auratus* (Linnaeus, 1758), guppy, *Poecelia reticulata* Peters, 1859 and X-ray fish, *Pristella maxillaris* (Ulrey, 1894). Twenty specimens (10 from pet shop and 10 from a local farm) of each fish species were purchased. Fishes were euthanised and examined for ecto- and endo-parasites using a Stereo-microscope (Leica EZ4). A crustacean parasite *Argulus* sp. was found attached to the fins and skin of goldfish (Yellow comet). Monogenean parasites were found attached on the gills of goldfish (Red comet). Moreover, several cysts containing echinostomatid larvae (digenean parasite) were found on the gills of several individuals of goldfish (Red fantail). Monogeneans were mounted on the slides using glycerine ammonium picrate (GAP) and were identified as *Dactylogyrus* spp. The also identified the parasites to species level and determined if any of them are alien parasites. In conclusion, our preliminary investigation shows that ornamental fish harbour parasites. Quarantine measures should be considered (for those species imported into South Africa) as an important key to minimise threats to biodiversity.

What can monogenean parasites of *Clarias ngamensis* Castelnau, 1861 (Teleostei: Clariidae) tell us about aquatic ecosystem health in the Upper Lufira basin?

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Some parasites are considered as useful biological tags through their use as sentinels of environmental health, among other aspects, as bioindicators of pollution. In the Democratic Republic of Congo (DRC), metal ores are mined without considering environmental standards. In the Upper Lufira basin, situated in the Katanga Copper belt, southern DRC, untreated effluents from mining plants and the hydrometallurgical complex of Shituru, containing mainly Cu, Co, Zn, Cd, and Pb are continuously discharged into aquatic systems. This may lead to biodiversity loss. Between 2015 and 2017, we carried out a study aimed at assessing the biological effects of mining pollution using monogenean gill parasites of the Blunt-toothed African catfish, *Clarias ngamensis* Castelnau, 1861. Objectives included: (i) analyzing metallic trace elements in water and sediment; (ii) analyzing monogenean parasite communities in unpolluted (Lufira River) and polluted (Lake Tshangalele) areas, and (iii) comparing infection parameters of parasites between the unpolluted and polluted areas. Results showed that concentrations of Cu in water and sediment of the Lufira River and Lake Tshangalele were 8 µg/l *vs.* 10 µg/l and 41.9 µg/g *vs.* 495.5 µg/g, respectively. The concentration of Cu in Lake Tshangalele is higher than the probable effect concentrations mentioned in the Sediment Quality Guidelines for metals in freshwater ecosystems (149 µg/g) above which harmful effects are likely to be observed. For the Lufira River and Lake Tshangalele, the species richness was 7 *vs* 5 parasites; 1.69 *vs* 1.47 as Shannon's indices and 0.87 *vs* 0.91 as indices of equitability. In both sites, prevalence was 100% (all

monogenean species together), whereas the mean intensity was higher in the Lufira River (31.28 ± 28.95 parasites per infected fish) than in the Lake Tshangalele (3.23 ± 2.89 parasites per infected fish). In conclusion, it appears that some metallic pollutants can affect aquatic ecosystems by decreasing parasite diversity and load. Whether this is beneficial for the host or not depends on the effects of pollution on the fish.

Genome-wide analysis of *Theileria Parva* proteases

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Theileria parva is a protozoan parasite that infects cattle and African buffalo, causing East Coast fever, Corridor disease and Zimbabwean theileriosis in cattle while buffalo do not show clinical symptoms. *Theileria parva* infections can incur major economic losses in cattle populations due to the high mortality rate and thus it is important to control and treat these infections. The current drug treatment available, Buparvaquone, has several shortfalls. The most concerning is the fact that this drug does not eliminate the infection thus creating a carrier status within recovered cattle herds, which can serve as a source of future infection. Therefore, there is a great need for a curative anti-parasitic drug treatment for *T. parva* infections. Proteases are reported to play a major role in the pathogenesis of related apicomplexan parasites and are considered to be virulence factors for some parasitic protozoans; hence they are attractive targets for drug therapy. The aim of this study was to identify possible drug targets for the treatment of theileriosis by identifying and analysing the proteases of *T. parva in silico*. Using a combination of bioinformatics tools, a genome-wide search was conducted and resulted in the identification of 128 proteases within the genome of *T. parva*. Subsequently, a sequence homology analysis through reverse BLAST was performed for negative selection of *T. parva* proteases with orthologs in the host proteome. Proteases involved in pathogenesis were identified by subcellular localization prediction analysis using SignalP 5.0 and Phobius for prediction of secreted proteases and HMMTOP and TMHMM for membrane proteases, assuming that virulent proteases will be secreted or found on the membrane. The function of the selected proteases was predicted by performing pathway analysis using KEGG. These results gave insight to which proteases are the best candidates for drug targets. Finally, inhibitors for selected proteases were identified by analysing protein-ligand interactions and will be evaluated in future studies as potential candidates for the development of chemotherapeutic drugs for *T. parva* infections.

Parasite diversity of different fish species from Hardap dam, Namibia

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Very few parasitological studies have been done on freshwater fish in Namibia. A survey was done during June 2016 at Hardap Dam (24°29′58″S, 17°51′31″E). The dam forms part of the Fish River System and has a surface area of 25 km2 with a 40 m high dam wall

forming the largest reservoir in Namibia. This dam is of strategic importance because of the size and location in Hardap Nature Reserve. The Namibia Fish River System joins the Orange River in South Africa and thus any changes in this system may affect aquatic ecosystem health of South Africa. Ten fish species occurs in Hardap Dam of which seven were examined for parasites: *Enteromius paludinosus* (n=92), *Cyprinus carpio* (n=1), *Labeo capensis* (n=11), *Labeo umbratus* (n=1), *Labeobarbus kimberlyensis* (n=9), *Oreochromis mossambicus* (n=6) and *Clarias gariepinus* (n=5). Standard methods were used to fix and preserve parasites. Numerous ecto- and endoparasites were recorded from these fish species including trichodinids, monogeneans, digenean larvae, adult and larval cestodes, adult and larval nematodes and copepods. High prevalence and mean intensity levels were recorded for *Lernaea* sp. from some of the fish species. Focal inflammation and hemorrhage occurred at the attachment site of these imbedded copepods, resulting in ulcerated lesions. These lesions may lead to secondary infections and have detrimental effects on the health of the host. Several lernaeids, commonly known as anchor worms, have been described with *Lernaea cyprinacea* being invasive and reported worldwide. A high prevalence was recorded for Ligula sp. from the body cavity of *E. paludinosus* with atrophy of gonads observed for several specimens. The gonadal suppression caused by this plerocercoid larva is of concern. *Contracaecum* larvae were recorded for some fish species with high intensity levels reported from *C. gariepinus*. The study includes new host and distribution records.

Gene expression profiling of small intestine of village chickens from *Ascaridia galli* infected environment

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Ascaridia galli is one of the most common nematodes affecting chickens. This study characterized *A. galli* parasites collected from South African village chickens of Limpopo (18) and KwaZulu-Natal (22) provinces using the 510bp sequences of the mitochondrial DNA (cytochrome C oxidase subunit 1 gene). PCR amplicons were directly sequenced using ABI3500 XL genetic analyzer and sequence data was assembled and edited using GAP4 of the Staden package (Version 1.6.0). Furthermore, this study investigated differential gene expression profiles of the *A. galli* infected small intestines of village chickens using RNA-seq strategy. Total RNA was isolated from the small intestine of infected and non-infected village chickens and sequenced using Illumina HiSeq2500 to generate between 3,908,924 and 3,994,946 reads. An average of 83.50% quality-controlled reads mapped to the reference chicken genome (*gallus.galgal4.74*). The results show that there was no genetic differentiation between *A. galli* from Limpopo and KZN provinces. Fourteen polymorphic positions were observed, which defined 6 haplotypes. Haplotype diversity of the two populations was moderate for both provinces and ranged from 0.749 for Limpopo province to 0.758 for KZN province. The utility of cytochrome C oxidase subunit 1 gene as a potential genetic marker for studying *A. galli* in village chicken populations is presented. Between any two-way comparisons of the intestines, 277 and 190 transcripts were significantly expressed in Limpopo and KZN chickens, respectively. Gene ontology analysis of the differentially expressed genes revealed an enrichment of genes proposed to function in immune response, defense response, inflammatory response and cell signalling genes. T cell

receptor signalling pathways and arachidonic acid metabolism pathways were among the most significantly impacted pathways. Overall, this study suggests expression patterns and functional annotation of genes involved in response to *A. galli* infection in chickens. Such information could aid in better understanding host-parasite interactions in the development of effective control strategies.

Molecular detection of tick-borne haemoparasites in cattle and buffalo samples from Manicaland province, Zimbabwe

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In sub-Saharan Africa, the most important tick-borne diseases (TBDs) affecting cattle are babesiosis caused by *Babesia bovis* and *B. bigemina*, theileriosis caused by *Theileria parva*, anaplasmosis caused by *Anaplasma marginale* and heartwater caused by *Ehrlichia ruminantium*. These TBDs cause major constraints to livestock production in developing countries such as Zimbabwe, and the lack of epidemiological information results in inadequate control measures. The present study used molecular tools to investigate haemoparasites occurring in cattle and buffalo, in the Manicaland province, Zimbabwe. DNA was extracted from 87 (80 cattle and 7 buffalo) whole blood samples and subjected to Reverse-line blot hybridization (RLB) and quantitative real-time polymerase chain reaction (qPCR) analyses. The RLB analysis revealed TBD infections in 58 samples (67%); 48 (55%) of which hybridized to the genus-specific probes only. The species detected by the genus-probes will be confirmed by 16S and 18S rRNA gene sequencing. Nonetheless, TBDs detected by RLB included three *Theileria* (*T. mutans*, *T. velifera*, and *Theileria* sp. sable), two *Anaplasma* (*A. marginale, A. centrale*) and two *Babesia* (*B. bigemina, B. bovis*) species. The most commonly occurring infections detected by qPCR assays in cattle, were *A. marginale* (n=22, 70.97 %) and *B. bigemina* (n=7, 22.58 %); followed by *A. centrale* (n=6, 19.35 %) and *B. bovis* (n=2, 6.45 %). While in buffalo (n=6), the most commonly occurring TBD was *A. marginale* (n=6, 85.71 %), followed by *A. centrale* (n=1, 14.28 %). Notably, all the buffalo samples tested negative for *B. bigemina* and *B. bovis*. Moreover, none of the samples (n=87) tested positive for either *T. parva* or *E. ruminantium* by any of the assays used. Our results did not follow the common trend for the prevalence of TBDs in Zimbambwe. According to recent reports, cattle theileriosis is the major killer of cattle, followed by babesiosis, heartwater, then anaplasmosis. Thus, our data suggests that the trend of occurrence of TBDs may vary between provinces depending on the vector-parasite-host-environment dynamics for each province. Finally, this study confirms that buffalo in this area are carriers of TBDs that pose risk to the cattle population.

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

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The present study emanates from a comprehensive parasitological investigation of a fish host from the Characiformes. This order constitutes a large and diverse component of the African ichthyofauna with the family Alestidae contributing about 95% of the known species within this order. The targeted fish species for this study was a commercially and economically important fish species, *Hydrocynus vittatus* Castelnau, 1861 commonly known as tigerfish. Despite being globally popular, little has been reported on any ecological aspects of tigerfish, particularly on associations with helminth parasites in South Africa. Additionally, some population of alestid hosts are declining due to commercial exploitations, pollution and overfishing. Fish were caught using fishing rods and gill nets during winter (n = 11; mean total length 360.2) and spring (n = 11; mean total length 375.9 mm) seasons from Schroda Dam, Limpopo Province. Monogeneans were collected in both seasons (June 2017; n = 197; mean intensity 39.4; prevalence 45.5%; mean abundance 17.9 and October 2017; n = 12946; mean intensity 809.1; prevalence 72.7%; mean abundance 588.5) and mounted in glycerine ammonium-picrate (GAP) solution and identified based on the male copulatory organ (MCO) and sclerotised structures of the haptor. The study presents seven new and four known species of *Annulotrema paperna* and Thurston, 1969 all collected from the gill lamellae. The four known species (*A. pikei*, *A. pikoides*, *A. nili ruahae* and *A. pseudonili*) represents new geographical records in South Africa.

Diversity and abundance of rodents in the lowveld region of South Africa

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Small mammals are hosts to a myriad of diseases linked to major global environmental problems with important public health, economic, and political consequences. High diversity within the host community is correlated with low disease risk or prevalence, and in turn protects ecosystems against infectious diseases. As part of a larger project investigating the prevalence of vector-borne blood parasites in rodents within the Kruger National Park, we examined the diversity of rodents sampled using Sherman traps. A total of 200 rodents were captured at 5 sites within the Park. Twelve rodent species were recorded: Bushveld Gerbil (46,73%), Highveld Gerbil (25,63%), Vlei rat (12,06%), Hairy-footed Gerbil (3,02%), Southern multimammate mouse (3,02%), Namaqua Rock Mouse (2,51%), Red Veld Mouse (2,51%), Marsh rat (1,51%), African Mole Rat (1,01%), Pygmy Mouse (1,01%), Acacia rat (0,5%) and Spiny Mouse (0,5%). Analyses showed statistically significant variations in the abundance of rodents between sites during wet and dry seasons.

Monogenea of the genus *Protogyrodactylus* Johnstin & Tiegs, 1922 from *Terapon jarbua* (Forsskal, 1775) from Kosi Bay, South Africa

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Monogenean parasites of the genus *Protogyrodactylus* parasitise hosts living in marine and brackish environments. The first four species of this genus were described from terapontids in Australia by Johnston and Tiegs in 1922. Recently, in 2018, five new species were described from two gerreid hosts from Moreton Bay, Queensland, Australia by Kritsky, bringing the number of valid species of this genus to 33. In South Africa, only one record of a single species exist namely *Protogyrodactylus amacleithrium* decribed from the Juarbua terapon, *Terapon jarbua*. by Price and McClellan from various localities in KwazZulu-Natal in 1969. During August 2015, five specimens of *Terapon jarbua* with a mean total length of 116.4 +- 7.4 mm were collected from the canal between Lake Mpungwini and Nhlange, Kosi Bay, KwaZulu-Natal, South Africa. Gills were screened for the presence of monogeneans and found specimens were cleared on microscope slides in glycerin amonium picrate. Morphometric analysis of the hard parts of attachement haptor was perform in order to identify the species. All five fish were infected by monogenean parasite. The differences in the shape and size of the haptoral hard parts suggested the presence of four different species of *Protogyrodactylus*. All identified species have a pair of ventral anchors with a large inner root and very short outer root. The pair of slender dorsal anchors are usually smaller than the ventral anchors, with both roots well developed of which size differ, as well as the opening between them. One of the species was identified as *P. amacleithirum* and the remaining three represent new species. As shown by our present results, just a small study can bring interesting insigths on limited knowledge on the parasite fauna of the marine fish of South Africa and specifically monogeneans. Additional sampling of the studied host would be needed in order to collect specimens to complete species description and to obtain samples for molecular characterisation.

Conservation of ancient relationships – elasmobranchs and parasites

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Southern Africa is an exceptionally biodiverse region for cartilaginous fishes. Many species are threatened, and conservation efforts are restricted to certain iconic species. To preserve natural resources, marine protected areas are established. However, information on the biology and ecology of elasmobranchs across management zones is sparse. In addition, current conservation schemes only apply for species at higher trophic levels, while parasitic organisms, forming the majority of biodiversity worldwide, are ignored as eligible conservation targets. Elasmobranchs and their internal parasites display one of the most

'ancient' host-parasite interrelationships on this planet, whose co-evolution is strongly interconnected, dating back at least 270 million years. Incorporation of parasites, together with their threatened hosts, becomes essential for future conservation efforts, since co-extinction events (i.e. parasite species facing extinction), which happen entirely unrecognized, may lead to cascading negative impacts within ecosystems. This project aims to assess the ecosystem health and anthropogenic impacts using cartilaginous fishes and their endoparasites as model organisms to establish and support the preservation of marine life and natural resources, and the conservation of threatened elasmobranch species and their associated parasites to maintain ancient host-parasite interactions and inter-relationships. Testing the condition of the marine environment between Betty's Bay and Gaansbai, covering approximately 200km^2 of South African coastline, we focus on the most dominant and least threatened species of elasmobranchs and their internal parasites as biological indicator organisms. An assessment of the biodiversity of internal parasites of selected elasmobranch species endemic to the Western Cape province will further provide useful information on the hosts' biology and ancient relationships with their specific parasites.

Single-step synthesised benzyltriazole derivatives and their in vitro efficacy on animal trypanosomosis causative agents

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Animal African trypanosomosis is a disease of economic importance resulting in decreased production of animal products. The currently available drugs for the treatment and prevention of the disease are fraught with negative impacts, which include high costs, variable efficacy at different stages of the infection on various subspecies of *T. brucei*, and most importantly, drug resistant strains have developed widely. Therefore, new, effective and cost-effective agents are in urgent need. The current study aimed to determine the efficacy of synthesized benzyltriazole (BZT) derivatives on *Trypanosoma congolense*, *T. brucei brucei* and *T. evansi*. The BZT derivatives were easily synthesized in a single-step resulting in high yields (60 - 90%) by employing Click chemistry. Overall, *T. b. brucei* and *T. evansi* parasites were more susceptible to the BZT compounds than *T. congolense*. Furthermore, there was observed efficacy variability between *T. b. brucei* and *T. evansi*. The efficacy of the compounds was found to be structure-specific. The most anti-trypanosomal active BZT derivative was FJS-402 compound which featured a CF3 on the phenyl ring and n-hexyl chain on the triazole moiety. It had IC50 values of 2.316, 6.49 and 1.49 µg/mL on *T. congolense*, *T. b. brucei* and *T. evansi*, respectively, and stands as hit for further investigation in the search for new trypanocidal agents. Furthermore, 12 out 15 BZT derivatives synthesized in this study had cLogP values within targeted range of 1 – 5 which indicates their hydrophilicity/lipophilicity balance for crossing biological membranes through passive diffusion. In conclusion, BZT derivatives synthesized in this study possess

trypanocidal activity against *T. congolense*, *T. b. brucei* and *T. evansi*. Further studies are necessary to determine the efficacy of the derivatives *in vivo* on experimentally infected mice.

Comparison of four methods for effective DNA extraction on arthropods

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Molecular methods have become common place in several different biological fields. Even though these methods have greatly become more cost-effective, it remains a financial burden in labs in developing countries. Generally, molecular studies require nucleic acid extractions with a high quality and quantity yield from the available specimens. Currently, three main categories of DNA extraction methods are known, including: conventional extractions, solid-phase nucleic acid extractions and all-in-one biomolecules extractions. Automated DNA extraction methods, also known as kits, have made an appearance and are believed to be the golden standard for DNA extractions. In this study, four DNA extraction methods were compared, to determine an effective, time and cost-efficient method that can be used on arthropods. We compared the Chelex 100 resin extraction, Qiagen DNeasy tissue kit, phenol chloroform isoamyl extraction (PCI) and phenol chloroform isoamyl sodium acetate extraction (PCI-SA). The DNA extracted was quantified (ng/uL) and qualified (260/280 ratio) using two separate nanodrop apparatus (Thermo Scientific™ NanoDrop™ 1000 spectrophotometer) of which the average values were calculated. The extractions were compared based on quantity and quality of the DNA, time and cost per extraction and the final total volume obtained. These methods were conducted on three groups of arthropods: horseflies (Tabanidae), mosquitoes (Culicidae) and ticks (Ixodidae). Five individuals per group where selected. One leg per individual per extraction method was used to allow for comparison purposes. The Qiagen DNeasy tissue kit produced the highest final total volume of the extracted sample. However, it is the most expensive and the quality and quantity of DNA obtained was low. The quickest and easiest of the methods were the Chelex 100 resin extraction, with no overnight incubation of samples. It produced the highest quantity of DNA, with a varying quality. Both PCI and PCI-SA extractions were significantly cheaper than both the Chelex 100 resin and the Qiagen kit, but both were time consuming and produced chemical waste that required specialised disposal. The PCI produced the least amount of final total volume, whereas PCI-SA quality of DNA was highly comparable between the nanodrops used.

Distribution of Branchiuran fish parasites in the Limpopo river system, South Africa

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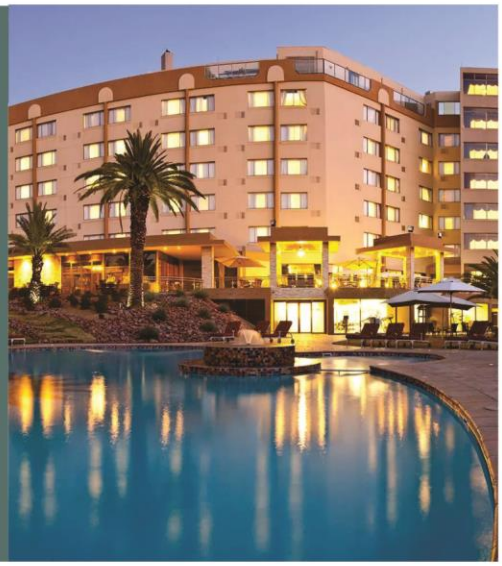
Fish parasitological surveys conducted from 2000 to 2019 at different localities in the Limpopo River System revealed the occurrence of three branchiuran species from three

genera. These include endemic species *Chonopeltis inermis*, *Dolops ranarum* and the alien parasite *Argulus japonicus*. Branchiurans were recorded from the skin, fins, mouth cavity and branchial chamber of various freshwater fish species. Standard methods were followed for parasite fixation and preservation. *Argulus japonicus* and *D. ranarum* were recorded from *Cyprinus carpio* (alien), *Clarias gariepinus* (native), *Oreochromis mossambicus* (native) as well as *Schilbe intermedius* (native) and appear to be generalist parasites. In addition, *D. ranarum* was also recorded from *Chetia flaviventris* (native) and *Micropterus salmoides* (alien). *Chonopeltis inermis* was only recorded from *C. gariepinus* and is a specialist parasite recorded from clariid hosts. Mean intensity levels were low for all branchiuran records, but pathology was noted for *D. ranarum* from some fish species. This may lead to secondary infections and affect fish health. This study includes several new hosts and geographical records contributing to our understanding of the biodiversity of African branchiurans.

LIST OF SPEAKERS IN ALPHABETICAL ORDER



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