



Stellenbosch University Animal TB Research Group

2019 ANIMAL TB RESEARCH GROUP

IN THIS ISSUE

Welcome to the 2019 Animal TB Research Group Newsletter!

The Animal TB Research Group has had a busy 2019. The group continues to grow with the addition of two new Masters' and two BSc Honours' students. We are proud of our three 2019 graduates (2 BSc Hons and 1 PhD).

We are also growing our networks and collaborations with the veterinary, medical, and wildlife communities in South Africa and abroad. Our research findings are shared through scientific publications, conference presentations, and interactions with stakeholders. In addition, our group continues to actively participate in community engagement events to increase TB awareness and inspire young learners to pursue careers in science.

We are excited to share some of this news with you!

Meet the Team

The Animal TB Research group has continued to grow, with 13 members in 2019!

Pictured above, (left to right first row): Dr. Leanie Kleynhans (Senior scientist), Kat Smith (MSc student), Josephine Chileshe (PhD student), Rachiel Gumbo (Honours student), Tina Meiring (MSc student), Prof. Michele Miller (professor); (left to right second row): Sam Goldswain (MSc student), Dr. Tanya Kerr (post-doctoral fellow), Dr. Wynand Goosen (post-doctoral fellow). Missing: Netanya Bernitz (PhD student), Candice De Waal (MSc student), Pam Ncube (MSc student), Sune Mostert (Honours student).



CONTENTS

Updates on projects:	
BovineTB testing in buffalo	2
Update on elephant TB	3
Rhinoceros TB assays	3-4
Wild dog genomics	4
Understanding disease in buffalo	6
New projects:	
Focus on One Health	5
Cheetah and leopard TB tests	6
Focus on:	
Pam Ncube, MSc	4
Dr. Leanie Kleynhans	4
Dr. Tanya Kerr	5
Dr. Wynand Goosen	5
Netanya Bernitz, PhD Students	5 8
Communication & Collaborations	
	7
Publications	9-10
Acknowledgements	11



Bovine Tuberculosis Testing Program for Buffaloes 2019

As our buffalo TB diagnostic research progresses, we have worked annually to test herds of buffaloes in the Hluhluwe iMfolozi Park, applying new techniques to improve the detection of *M. bovis* infected animals. The test and cull program appears to be reducing the percentage of infected animals detected each year.

Importantly, we have begun to investigate how the tests perform in uninfected herds. Working in collaboration with buffalo owners and wildlife veterinarians, we have tested over 500 buffalo in 2019. These results have provided better evaluation of sensitivity and specificity of the blood-based tests which still appear to have suboptimal performance for an effective disease program. This work has also allowed us to collect samples to expand our research into how the buffalo's immune system responds to infection and develops disease which will improve our understanding and management of the disease.

The buffalo testing programme is a highlight of the Animal TB Group, every year, and we thank Dr. Dave Cooper, JP van Heerden, Alicia and Warren McCall, Debbie Cooke, Dr. Andy Fraser, Dr. Emma Rambert, Dr. Douw Grobler, Dr. Angela Bruins, and others for supporting our work.



Buffalo field teams in 2019

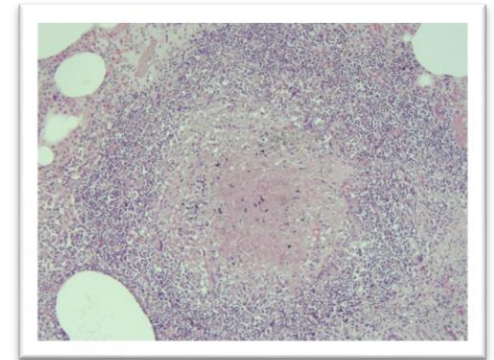
Left: Wynand Goosen, Leanie Kleynhans, Sam Goldswain, Michele Miller, Kat Smith, Adeleine Goosen

Above: Tanya Kerr, Jos Chileshe

Characterizing the immune cell populations involved in *M. bovis* granuloma formation

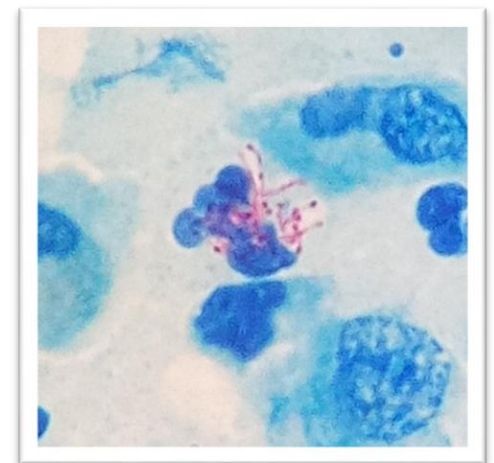
Granulomas are the hallmark lesion of tuberculosis, and have been well-described in humans and cattle, but not in wildlife. Since histopathological lesions of bTB have not been well described in buffalo, the stages of infection and disease progression are not really understood in this species. Understanding the bTB immune response in buffaloes is important for advancing knowledge of the immunobiology and pathogenesis of TB. In this project, MSc student Sam Goldswain will use immunohistochemistry, a tissue staining technique that involves selecting antibodies that can identify different cell types, to characterise

the immune cell types present and their distribution in granulomas from *M. bovis*-infected buffalo. This study will improve our understanding of bTB pathogenesis and investigate the connection between our diagnostic test results and disease in buffalo.



*Above: Microscopic image of a granuloma in the lung of a *M. bovis*-infected buffalo; S. Goldswain.*

*Below: *M. bovis* bacilli in smear from a tissue lesion in a buffalo.*



Above: Sam, Wynand and Kat preparing samples in the field.

UPDATE ON ELEPHANT TUBERCULOSIS

Since the initial case of *M. tuberculosis* as the cause of death in a bull elephant in Kruger National Park in 2016 (Miller et al. 2019), a surveillance project has been initiated. Using the validated serological assay for TB in elephants, the Chembio VetTB DPP, banked serum samples were tested by post-doctoral fellow Dr. Tanya Kerr and approximately 6% of elephants in KNP had detectable antibodies, suggesting previous exposure/infection (Kerr et al., 2019). Research by MSc student Candice de Waal has also detected upregulation of several cytokine genes that will be developed for a blood-based test of immune sensitization to TB in elephants. In addition, opportunistically immobilized elephants are currently being screened using endoscopic collection of respiratory samples from the lungs and trunk to identify mycobacterium. Tissue samples are also collected for testing. A new rapid test for direct detection of the bacteria's DNA (used in human TB clinics worldwide) has recently been tested with elephant and rhinoceros samples and shown to be promising. The Cepheid GeneXpert MTB/RIF Ultra assay can be performed in the field using a swab or respiratory sample with results in less than 2 hours. In collaboration with the Cepheid (the company that produces the GeneXpert), this test is currently being tested in the field in Kruger National Park.

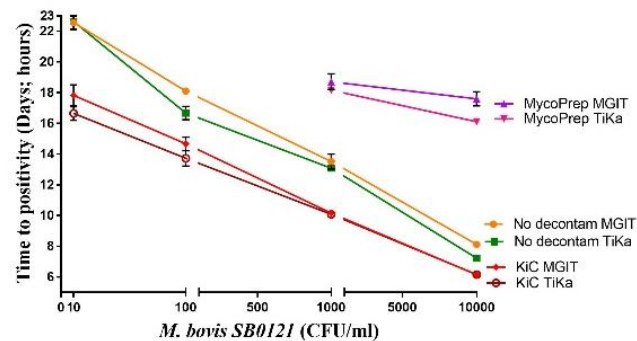


Above: Prof. Gerhard Walzl, assisted by Dr. Leanie Kleynhans and Guy Hausler, collects respiratory samples from an immobilized elephant using a flexible endoscope. Below: Drs. Leanie Kleynhans, Tanya Kerr, Wynand Goosen work with SANParks vet tech Tebogo Manamela to process samples in Veterinary Wildlife Services laboratory.



Direct Detection of Mycobacteria using Novel Methods in Elephants and Rhinoceros

Since rejoining the Animal TB Research group in 2018, post-doctoral fellow Dr. Wynand Goosen has been focusing on rapid methods of detecting TB using bacterial DNA detection and enhanced culture of elephant and rhinoceros samples. Through collaborations with Cepheid and TiKa Diagnostics, he is evaluating the human GeneXpert MTB/RIF Ultra assay (Ultra) and the new TiKa-MGIT culture test, respectively. He has shown that Ultra is able to detect TB bacilli as low as 2 CFU/ml and the new TiKa-MGIT system to enhance mycobacterial culture of samples with low levels of bacteria.



Culture time to positivity of sterile phosphate buffer spiked with different concentrations of *M. bovis* SB0121 (10000 – 10 CFU/ml) prior to culturing using the conventional MGIT system versus the new TiKa-MGIT system.

Validation of Interferon-gamma Release Assay for Rhinoceros

PhD student Jos Chileshe has been able to develop and validate a blood-based test to screen rhinoceros for TB.



Her work was published in 2019 in the scientific journals *Veterinary Immunology and Immunopathology* and *Journal of Veterinary Diagnostic Investigation*. This test is currently being used to test rhinos for translocation in KNP.

Alternative Biomarkers for Rhinoceros

Since no single test has been found to be perfect for diagnosing TB in either humans or animals, our research continues to search for additional biomarkers that can be used to increase confidence in the interferon-gamma assay results. This includes screening rhinoceros RNA from stimulated blood cells in a multiplex assay to identify potential cytokines for further investigation. Jos will be including this work in her PhD thesis which she will be submitting in 2020.

FOCUS ON PAM NCUBE, MSC STUDENT

Pam Ncube is a MSc student that joined the Mycobacteriomics and Animal TB Research groups in 2019. Her research question is focused on answering the question of whether *M. bovis* can cause latent infections in animals. She is using an in vitro model to "stress" bacteria isolated from various wildlife species to determine if they have the potential to become "persisters" (bacteria that are viable but don't replicate). This novel approach has been developed by Prof. Sampson and her group to study latency in human TB. Pam will investigate whether different stains of *M. bovis* are more likely to result in latent infections.

Dr. Leanie Kleyhans, Senior Scientist

Dr. Kleyhans joined the Animal TB group in 2017, to assist with the growing number of students and projects. She received her PhD in Molecular Biology and completed her post-doctoral training with the Immunology group at SUN. Her research has been focused on immune-endocrine interactions in the context of TB. She continues her research as well as supervising post-graduate students in both the Immunology and Animal TB groups. Her expertise and management skills make her ideal for helping steer the progress and future of Animal TB research.

Investigating genome variation in a South African wild dog population: Towards understanding their susceptibility to *Mycobacterium bovis* infection

African wild dogs (*Lycaon pictus*) are key role-players in the ecosystem and biodiversity in Africa, however, they are now considered critically endangered. MSc student Tina Meiring has undertaken research to investigate genetic diversity in this species. Using whole genome



sequencing of DNA extracted from banked whole blood of 22 wild dogs in Kruger National Park, her initial findings suggest that these wild dogs have low levels of genetic diversity. Since the samples were from wild dogs selected from different packs, it was expected that they would be unrelated. However,

based on our relatedness estimations, only 13% were completely unrelated. Based on bioinformatic calculations, the inbreeding occurred many years ago. Tina is currently sequencing 50 additional wild dogs from KNP and will try to identify regions in the wild dog genome that may impact susceptibility to infection. With her recent upgrade to a PhD student, she will be developing tools to identify genetic factors conferring adaptive advantages in this species.

Tina pictured with immobilized hyena in KNP.

RECOGNITION

Congratulations to Dr. Tanya Kerr for being recognized as one of the top 20 post-doctoral fellows at Stellenbosch University in 2019!



We are also proud of post-doctoral fellow Dr. Wynand Goosen (top left) who was awarded a spot in the top 20 young emerging scientists in South Africa selected by the National Science and Technology Forum and his inauguration into the National Geographic Society as an explorer.



Focus on One Health

The "One Health" initiative aims to merge animal and human health with the environment, providing a more holistic perspective to conservation, research, and public health activities. To address global concerns such as food security and emerging diseases, a multidisciplinary approach is required. Zoonotic diseases, such as tuberculosis, rabies and tick-borne pathogens remain a major concern for domestic animal/livestock, wildlife and public health in South Africa. Furthermore, controlling these diseases present a complex socio-economic challenge in resource-limited countries. The Animal TB Research group is evaluating logistics required to embark on a one health project investigating the transmission of TB and other important zoonotic pathogens between wildlife, livestock/domestic animals, and humans, especially in communities bordering wildlife reserves. The initial aim of this project is to determine the prevalence of diseases in various species to evaluate the relative importance they have in human and animal health. By facilitating co-learning opportunities with stakeholders and multidisciplinary experts, a greater understanding can be gained regarding appropriate interventions and prevention strategies.

CONGRAULATIONS TO NETANYA BERNITZ!

Netanya successfully completed her PhD, entitled "Novel approaches to the diagnosis of *Mycobacterium bovis* infection in African buffaloes (*Syncerus caffer*)". Her research investigated new assays and algorithms to improve detection of bovine TB in buffaloes. Netanya has graduated in December and relocated to the United Kingdom. We wish her success in the future!



Understanding How *M. bovis* Disease in Buffaloes Impacts TB Test Interpretation



Tuberculin skin test in a buffalo - swelling, erythema and slight necrosis is observed at 72 hours after infection of bovine PPD.

Bovine tuberculosis is a chronic disease in buffaloes with signs often only showing up months to years after infection. Therefore, tests to detect infection are based on measuring the host's immune response. Since techniques used in other species, such as chest X-rays,

are not feasible in buffaloes, other indications are needed to differentiate infection from disease. This is important because animals with disease are more likely to shed bacteria and thus infect other individuals.

Research to be published in the *Journal of Wildlife Diseases* (Bernitz et al., 2020) suggests that background levels of cytokines interferon-gamma and IP-10 are higher in buffaloes with TB lesions. The combination of increased antigen-specific IFN-gamma and IP-10 along with unstimulated cytokine concentrations may be useful in detecting animals with disease.

Kat Smith, who joined as an MSc student in 2019, is continuing to evaluate additional cytokines that may be able to distinguish between early infection and disease in buffaloes. These blood-based assays will facilitate interpretation of other TB tests such as the tuberculin skin test, which commonly causes confusion due to cross-reactions with environmental bacteria.

NEW PROJECTS

Cheetah and Leopard TB Diagnostic Test Development

Bovine tuberculosis in lions has been recognized in a number of wildlife parks and reserves where *M. bovis* is present, especially in prey species. However, the presence of this pathogen in leopards and cheetahs has only sporadically been reported in South Africa. In early 2019, a private wildlife veterinarian contacted the Animal TB group regarding a suspected case of TB in a cheetah. Although we had previously developed a cytokine (CXCL9) gene expression assay as a diagnostic test for lions (Olivier et al., 2017), it was unknown if it could be used in other wild felids. Using the QuantiFERON TB Plus system to incubate whole blood from the cheetah, and using the qPCR assay with lion CXCL9 primers, a significant upregulation was detected, suggesting that the cheetah was infected. This was confirmed by mycobacterial culture of *M. bovis* from postmortem tissue samples. The case report is currently under review for publication which includes more details.

LEOPARD SAMPLE COLLECTION



The immobilized leopard will have blood collected and an intradermal tuberculin test performed to screen for TB. Positive animals are euthanized and tissues collected for mycobacterial culture and PCR. These data are important for improving our understanding of this disease in wild felids. Digital radiography capabilities have recently been acquired and will be used to evaluate the lungs, bones and other organs for TB lesions.

Since movement restrictions can be implemented for populations with controlled diseases such as bovine TB, this can significantly impact conservation programs. The cheetah metapopulation actively translocates animals between fragmented reserves to preserve genetic diversity. Working with the Endangered Wildlife Trust and reserves in Kwa-Zulu Natal, cheetah samples have been collected in order to develop tests that can be used to minimize risks of moving infected cheetah. Dr. Tanya Kerr and Honours student Rachiel Gumbo, as well as U.S. veterinary student, Elin Crockett, are currently working on this project. Preliminary data suggest that cytokine gene expression assays can identify *M. bovis*-infected leopards and cheetahs.

SCIENCE COMMUNICATION, INSPIRATION AND COLLABORATION



Animal TB Research team members actively participate in interactive events to promote TB awareness in stakeholders (above: Dr. Goosen talking with wildlife industry stakeholders), inspire interest in science (above right: Dr. Kerr answers questions from young learners), and provide insights to collaborators (below right: Cepheid senior staff work with Animal TB scientists and South African National Parks veterinary team to screen immobilized rhinoceros).

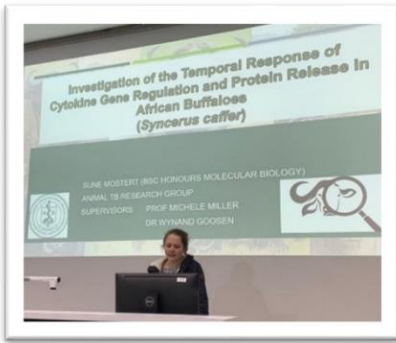


Communication with the scientific community, stakeholders, and the public are a crucial part of making research findings relevant and translation. The Animal TB research group regularly shares “in progress” research results with their collaborators to elicit feedback and strengthen relationships to improve our work.

Increasing awareness among stakeholders is key to active participation in research. Attending meetings and discussing new findings and questions can provide important insight and create new partnerships for research, as demonstrated by the collaboration with Endangered Wildlife Trust and private reserves to obtain cheetah and leopard samples. Cooperation can accelerate projects and reduce costs as well as provide important resources to the wildlife community.

Collaborations with Industry

Research funding to purchase state-of-the-art equipment and reagents is often difficult to acquire, especially for wildlife. The Animal TB Research group has been fortunate to develop a number of partnerships with industry to provide in-kind donations or lease of equipment that facilitates research projects, such as the evaluation of Cepheid’s GeneXpert platform for animal samples. Opportunities to get first-hand knowledge of the challenges experienced in wildlife research by collaborators often provide a common ground to discuss strategies to overcome these obstacles. We are grateful to all our partners, especially Cepheid, Chembio Diagnostics, TiKa, MabTech, Qiagen, Thermofischer Scientific and others.



HONOURS STUDENTS 2019

Congratulations to our Honours students, Sune Mostert and Rachiel Gumbo, who graduated in December! We are proud of their accomplishments this year in both their exams and projects.

Sune's project investigated how cytokine gene expression and cytokine protein production in buffaloes changes over different incubation times. This work provides important information to optimize the TB diagnostic tests in this species.

Rachiel's project determined whether the lion CXCL9 gene expression assay could detect antigen-specific upregulation in stimulated cheetah and leopard whole blood from *M. bovis*-infected animals. This research provided the foundation to launch a larger study of diagnostic test development for cheetah and leopards which will be the basis for Rachiel's MSc project starting in 2020.

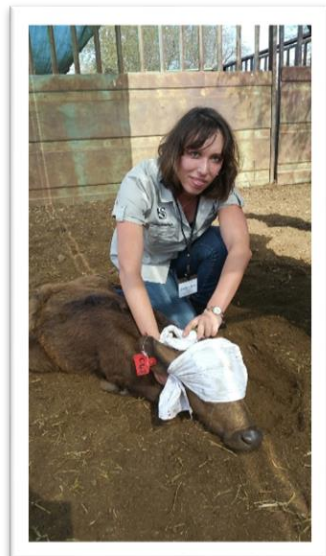
*Top: Sune Mostert
Below: Rachiel Gumbo*

Continuing Students

Candice de Waal, Kat Smith, Sam Goldswain, and Pam Ncube will be continuing their MSc studies with the group in 2020.

Tina Meiring will be upgrading to a PhD project, continuing her work on wild dog genomics.

Jos Chileshe will be finalizing her rhino studies and working on her PhD which will be completed in 2020.



Rachiel Gumbo, MSc student, returned to her studies in 2019 to complete her BSc Hons with the Animal TB Group. She will continue her research on leopard and cheetah TB diagnostics for her MSc.

Charlene Clarke, PhD student, is returning to our group where she completed her BSc Hons and MSc. She will be evaluating systemic and local immune responses during *M. bovis* infection in buffaloes.

Left: Charlene Clarke with buffalo calf.

WELCOME NEW STUDENTS IN 2020

The Animal TB Research group will have 2 new PhD and 2 new MSc students joining in 2020.

Rebecca Dwyer, MSc student, completed her BSc Hons in Port Elizabeth and has been working at a commercial laboratory during 2019. She will be continuing to investigate rhinoceros immune responses for her MSc project.

Debbie Cooke, PhD student, is a senior Animal Health Technician at the State Veterinary Epidemiology unit in Pietermaritzburg, KZN. She has worked with the Animal TB group on buffalo TB projects and completed her MSc in 2017 working on *Mycobacterium avium* in poultry. Her PhD project will investigate the diagnosis and epidemiology of TB in goats.

Publications by Animal TB Research Group 2019

1. Gibson, K.M., Nguyen, B.N., Neumann, L.M., **Miller, M.**, Buss, P., Daniels, S., Ahn, M.J., Crandall, K.A. and Pukazhenthi, B., 2019. Gut microbiome differences between wild and captive black rhinoceros—implications for rhino health. *Scientific reports*, 9(1), p.7570.
2. Edwards, K.L., **Miller, M.A.**, Carlstead, K. and Brown, J.L., 2019. Relationships between housing and management factors and clinical health events in elephants in North American zoos. *PloS One*, 14(6), p.e0217774.
3. Byrne, A.W., Allen, A., O'Brien, D.J. and **Miller, M.A.**, 2019. Bovine tuberculosis—international perspectives on epidemiology and management. *Frontiers in Veterinary Science*, 6, p.202.
4. **Miller, M.A.**, Buss, P.E., **Roos, E.O.**, **Hausler, G.**, Dippenaar, A., Mitchell, E., Van Schalkwyk, O.L., Robbe-Austerman, S., Waters, W., Sikar-Gang, A. and Lyashchenko, K.P., 2019. Fatal tuberculosis in a free-ranging African elephant and one health implications of human pathogens in wildlife. *Frontiers in Veterinary Science*, 6, p.18.
5. **Bernitz, N.**, **Kerr, T.J.**, **Goosen, W.J.**, **Clarke, C.**, **Higgitt, R.**, **Roos, E.O.**, Cooper, D.V., **Warren, R.M.**, **van Helden, P.D.**, **Parsons, S.D.** and **Miller, M.A.**, 2019. Parallel measurement of IFN- γ and IP-10 in QuantiFERON[®]-TB Gold (QFT) plasma improves the detection of *Mycobacterium bovis* infection in African buffaloes (*Syncerus caffer*). *Preventive Veterinary Medicine*, 169, p.104700.
6. **Chileshe, J.**, **Roos, E.O.**, **Goosen, W.J.**, Buss, P., **Hausler, G.**, Rossouw, L., Manemela, T., **van Helden, P.**, **Warren, R.**, **Parsons, S.D.** and **Miller, M.**, 2019. An interferon-gamma release assay for the diagnosis of the *Mycobacterium bovis* infection in white rhinoceros (*Ceratotherium simum*). *Veterinary Immunology and Immunopathology*, 217, p.109931.
7. **Kerr, T.J.**, **de Waal, C.R.**, Buss, P.E., Hofmeyr, J., Lyashchenko, K.P. and **Miller, M.A.**, 2019. Seroprevalence of *Mycobacterium tuberculosis* complex in free-ranging African elephants (*Loxodonta africana*) in Kruger National Park, South Africa. *Journal of Wildlife Diseases*, 55(4), pp.923-927.
8. **Chileshe, J.**, **Goosen, W.J.**, Buss, P.E., **van Helden, P.D.**, **Warren, R.**, **Parsons, S.D.** and **Miller, M.A.**, 2019. A commercial ELISA for detection of interferon-gamma in white rhinoceros. *Journal of Veterinary Diagnostic Investigation*, p.1040638719843955.
9. **Miller, M.A.**, Buss, P., **Sylvester, T.T.**, Lyashchenko, K.P., deKlerk-Lorist, L.M., Bengis, R., Hofmeyr, M., Hofmeyr, J., Mathebula, N., **Hausler, G.** and **van Helden, P.**, 2019. *Mycobacterium bovis* in free-ranging lions (*Panthera leo*)—evaluation of serological and tuberculin skin tests for detection of infection and disease. *Journal of Zoo and Wildlife Medicine*, 50(1), pp.7-15.
10. Pohlin, F., Buss, P., **Miller, M.**, Steenkamp, G., Glead, R., Poore, L., Boesch, J. and Zeiler, G., 2019. Etorphine-ketamine constant rate infusion for maintenance of anaesthesia in a compromised white rhinoceros (*Ceratotherium simum*). *Case Reports in Veterinary Medicine*, 2019.
11. Rosen, L.E., Olea-Popelka, F., Deem, S.L., Iszaz, R., Schmitt, D. and **Miller, M.**, 2019. Survey of antituberculosis drug administration and adverse effects in elephants in North America. *Journal of Zoo and Wildlife Medicine*, 50(1), pp.23-32.
12. **Bernitz, N.**, **Kerr, T.J.**, **Goosen, W.J.**, **Higgitt, R.L.**, **de Waal, C.**, **Clarke, C.**, Cooper, D.V., **Warren, R.M.**, **van Helden, P.D.**, **Parsons, S.D.C.** and **Miller, M.A.**, 2019. Impact of *Mycobacterium bovis*-induced pathology on interpretation of QuantiFERON[®]-TB Gold assay results in African buffaloes (*Syncerus caffer*). *Veterinary Immunology and Immunopathology*, <https://doi.org/10.1016/j.vetimm.2019.109923>
13. **Viljoen, I.M.**, **Sylvester, T.T.**, **Parsons, S.D.C.**, Millar, R.P., **van Helden, P.D.** and **Miller, M.A.**, 2019. Performance of the tuberculin skin test in *Mycobacterium bovis*-exposed and -unexposed African lions (*Panthera leo*). *Journal of Wildlife Diseases*, 55(3), pp. 537-543.
14. **Roos, E.O.**, **Scott, L.A.**, **Ndou, S.**, Olea-Popelka, F., Buss, P.E., de Klerk-Lorist, L.-M., **Warren, R.M.**, **van Helden, P.D.**, **Sylvester, T.T.**, **Miller, M.A.** and **Parsons, S.D.C.**, 2019. Cytokine gene expression assay as a diagnostic tool for detection of *Mycobacterium bovis* infection in warthogs (*Phacochoerus africanus*). *Scientific Reports* 9:16525. doi:10.1038/s41598-019-53045-0
15. Neiffer, D., Buss, P., Hewlett, J., **Hausler, G.**, Rossouw, L., Manemela, T., Grenus, B., Thulson, E., Olea-Popelka, F. and **Miller, M.**, 2019. Evaluation of physiological parameters and effectiveness of an immobilization protocol

- using etorphine, azaperone, and butorphanol in free-ranging warthogs (*Phacochoerus africanus*). *Frontiers in Veterinary Science*, doi: 10.3389/fvets.2019.00402
16. **Miller, M.A.**, Gortazar, C., Roos, E.O., Rivalde, M.A., Johnathan-Lee, A., Sridhara, A.A. and Lyashchenko, K.P., 2019. Serological reactivity to MPB83 and CFP10/ESAT-6 antigens in three suid hosts of *Mycobacterium bovis* infection. *Veterinary microbiology*, 235, pp.285-288.
 17. Love, D.M., Garner, M.M., Lyashchenko, K.P., Sikar-Gang, A., Bradway, D.S., Robbe-Austerman, S., **Miller, M.**, and Ramer, J. Tuberculosis caused by *Mycobacterium orygis* in a greater one-horned rhinoceros (*Rhinoceros unicornis*): first report in the western hemisphere. *Journal of Zoo and Wildlife Medicine*, IN PRESS.
 18. **Bernitz, N., Kerr, T.J., de Waal, C., Cooper, D.V., Warren, R.M., van Helden, P.D., Parsons, S.D.C., and Miller, M.A.** Test characteristics of assays to detect *Mycobacterium bovis* infection in high-prevalence African buffalo (*Syncerus caffer*) herds. *Journal of Wildlife Diseases*, IN PRESS.
 19. **Warren, R.M.**, Ismail, N., Chegou, N., Walzl, G., Kinnear, C., **Goosen, W., Miller, M.**, Diacon, A. and **van Helden, P.D.** Tuberculosis in South Africa over the past 50 years: from bench to bedside. *South African Medical Journal*, IN PRESS.
 20. Hewlett, J., Buss, P., Olea-Popelka, F., Koeppel, K., Neiffer, D., **Hausler, G.**, Rossouw, L., Manamela, T., Stout, E. and **Miller, M.** Evaluation of a partially reversible immobilization protocol using medetomidine, butorphanol, zolazepam-tiletamine, and ketamine in free-ranging warthogs (*Phacochoerus africanus*) in Kruger National Park, South Africa. *Journal of Zoo and Wildlife Medicine*, IN PRESS.
 21. Hooijberg, E.H., Cray, C., Steenkamp, G., Buss, P., Goddard, A. and **Miller, M.** Assessment of the acute phase response in healthy and injured southern white rhinoceros (*Ceratotherium simum simum*). *Frontiers in Veterinary Science*, IN PRESS.
 22. **Kerr, T.J., Gumbo, R., Goosen, W.J., Rogers, P., Last, R.D. and Miller, M.A.** Novel detection techniques for *Mycobacterium bovis* infection in a cheetah (*Acinonyx jubatus*). *Emerging Infectious Diseases*, IN PRESS.
 23. Krajewska, M., Didkowska, A., Sridhara, A., Elahi, R., Johnathan-Lee, A., Radulski, L., Lipiec, M., Anusz, K., Lyashchenko, K., **Miller, M.** and Waters, R. Transboundary tuberculosis: importation of alpacas infected with *Mycobacterium bovis* from the United Kingdom to Poland and potential for serodiagnostic assays detecting tuberculin skin test false-negative animals. *Transboundary and Emerging Diseases*, In revision.
 24. **Goosen, W.J., Kerr, T.J., Kleyhans, L., Warren, R.M., van Helden, P.D., Persing, D., Parsons, S.D.C., Buss, P. and Miller, M.A.** The GeneXpert MTB/RIF Ultra assay detects *Mycobacterium tuberculosis* complex DNA in white rhinoceros (*Ceratotherium simum*) and African elephants (*Loxodonta africana*). In review.
 25. Achkar, J.M., Adams, L.B., Dupnik, K.M., Hall-Stoodley, L., Kanipe, C., Kraigsley, A., Kurtz, S., **Miller, M.A.**, Salvador, L., Larsen, M. and Robinson, R. The many hosts of mycobacteria: a conference report. In review.
 26. Radcliffe, R.W., Jago, M., Morkel, P.V., Parry, S.A., Morkel, E., du Preez, P., Beytell, P., du Preez, J., **Miller, M.A.**, Felipe, J. and Gleed, R.D. The pulmonary and metabolic effects of suspension by the feet compared with lateral recumbency in black rhinoceroses (*Diceros bicornis*) recently captured by aerial darting. In review.
 27. **Goosen, W.J., Kerr, T.J., Kleyhans, L., Buss, P., Warren, R.M., van Helden, P.D., Schroder, B., Parsons, S.D.C. and Miller, M.A.** The VetMAX™ *M. tuberculosis* complex PCR Kit detects MTBC DNA in antemortem and postmortem samples from white rhinoceros (*Ceratotherium simum*), African elephants (*Loxodonta africana*) and African buffaloes (*Syncerus caffer*). In review.

Book Chapter

Buss, P. and Miller, M., 2019. Update on Field Anesthesia Protocols for Free-Ranging African Lions. In *Fowler's Zoo and Wild Animal Medicine Current Therapy, Volume 9* (pp. 536-538). WB Saunders.

Stellenbosch University Animal TB Research Group

P.O. Box 241
Cape Town 8000
South Africa

Follow Us!



For Information:

http://www.sun.ac.za/english/faculty/healthsciences/Molecular_Biology_Human_Genetics/animaltb/Pages/default.aspx

Or contact:

Prof. Michele Miller miller@sun.ac.za

Thank you to all our collaborators and supporting partners!



UNIVERSITEIT VAN PRETORIA
UNIVERSITY OF PRETORIA
YUNIBESITHI YA PRETORIA

