Issue 16- February 2021

NATIONAL INSTITUTE FOR COMMUNICABLE DISEASES
Division of the National Health Laboratory Service

SCIENCE FOCUS

WE LOOK AT THE IMPORTANCE OF COVID-19 SURVEILLANCE DATA

The Science Focus acknowledges NICD members of staff who have published in peer-reviewed journals. This publication is a compilation of scientific publications where an NICD staff member is either the first or last author.

Editor's Note

The first Science Focus Issue for 2021 reflects research successes of the National Institute for Communicable Diseases (NICD) and highlights achievements for quarter three of the current fiscal year. This issue features a collection of noteworthy statistical findings, together with the number of peer-reviewed articles, top published authors and high-impact factor score articles.

The importance of COVID-19 surveillance and the tremendous weight it carries in guiding Government to make informed decisions takes center stage. Dr Kerrigan McCarthy reflects on how the philosophy of epidemiology has evolved to encourage a more holistic approach to the collection, analysis and interpretation of surveillance data.

Featured research abstracts for the quarter include, to name a few, *Antibody isotype switching as a mechanism to counter HIV neutralization escape,* by Prof Penny Moore and Cathrine Scheepers, Dr Jessica Coertse's *Lyssaviruses in Insectivorous Bats,* and Prof Janusz Paweska's *Shedding of Marburg Virus in Naturally Infected Egyptian Rousette Bats,* explaining that Marburg virus RNA was detected in the rectal swab samples from these bats.

By publishing important health papers and high-impact publications in the International Journal of Infectious Diseases, Journal of Clinical Virology and The Lancet Microbe, to name a few, it is clear that NICD researchers continue to make strides towards achieving scientific research excellence in informing policy. These research feats are commendable and the NICD celebrates the successes, and curiosity, of these brilliant minds.

Staff members are encouraged to continue to send their comments to the Communications Unit.

On behalf of the team,

Sinenhlanhla Jimoh
Senior Communications Manager

The Significance of Surveillance Data Interpretation

By Dr Kerrigan McCarthy

In the early days of the COVID-19 pandemic in South Africa, and soon after we reached the critical landmark of 100 laboratory-confirmed cases, the Minister of Health asked of me, as he no doubt asked many other disease specialists and epidemiologists, if a 'lockdown' was required. Whilst I had a lifetime of experience in public health response to communicable diseases, and I worked with the NICD – which is responsible for providing disease intelligence, and I knew theoretically that reduced social interaction would limit SARS-CoV-2 transmission, I remember being utterly unable to respond. A 'lockdown' would reduce cases of SARS-CoV-2, at least in the short term, by slowing the rate of increase, but the plethora of social and economic consequences of a 'lockdown' on individuals and communities could well worsen both individual and collective pandemic outcomes. I did not have the skills to understand how these composite and complex factors could interact and what the range of outcomes were.

As I reflect now on the Ministers question and my incapacity to respond, I realise that it begged a profound question about the nature of surveillance data, its interpretation and its role in the broader context of society. To answer this question, in the discussion that follows, I have delved into the 'philosophy of epidemiology.'

The official role of NICD is to serve as a resource of knowledge and expertise of communicable diseases to support the planning of policies and programmes that respond to communicable disease threats. Essentially we are required to provide and interpret surveillance data using the tools of epidemiology to report on the distribution of communicable diseases and establish their determinants or causal factors. All of our endeavours aim ultimately to determine cause of certain health events or outcomes (the 'determinants of disease') with a view to intervening to improve life for all. The NICD stands in a global context - public health institutes are created by governments across the world to generate public health insight and practical solutions for their countries health problems.

Whether we are aware of it or not, our thinking and approach to the health problems we face is determined by our philosophical context. At a philosophical level, all public health institutions are created with the hope that they will forge progress in society through the 'scientific method' which seeks to understand the world and improve it. Public health institutes operate within a context defined by a philosophy of epidemiology. Causal inference –how we prove an antecedent event or situation to be responsible for a subsequent event - is a key area in the philosophy of epidemiology.

The philosophy of epidemiology left to us by our immediate scientific forebears is this: theories of causality come and go as they are tested through observations and experiments, and those theories that best fit with reality remain. To support the process of discerning which theories fit best, scientists formulate questions based on creative conjecture, hypotheses and deductive logic, and measure outcomes of experiments in objective ways. Theories of causality which are so tested and 'hold up', are therefore 'truth', and reflect the nature of reality more closely.

However, epidemiologists of today have tended to reflect differently on causality. We do not simply measure risk ratios and assign causality if the ratio (and confidence interval) exceeds one. Nor, after we have 'established causality', do we simply say that if X seems to occur following Y, then X would not happen if Y were not present. In practice we unconsciously apply what Kuhn calls 'eco-epidemiology' – a post-modern philosophical approach to epidemiology. In our current thinking, social and biological contexts (also referred to as the 'social determinants of disease' by the WHO) are understood to shape both antecedent and subsequent events related to a health phenomenon. Causal 'association' is a function not only of biology but also of uncertainty and 'randomness' of antecedent and subsequent events. Furthermore, the epidemiologists recognise that formulation of questions, the design of experiments and interpretation of results by scientists is shaped by assumptions, and in turn by the values, beliefs and experiences of the questioner, and remain untestable – outside of the experiment and therefore not subject to interpretation. We have moved on from our scientific forebears to recognise this truth - that the 'goal of science' – namely to arrive at an ever more truthful account of the nature of reality - is in reality unachievable.

So where does this leave us, the NICD and the global community of epidemiologists, who are providers and interpreters of surveillance data? Firstly, I do believe that our philosophy of epidemiology has moved closer to an account of reality than the view our scientific forbears held. We now believe that reality is a product of innumerable contingent realities, and that no model of causation or scientific experiment can successfully predict the impact and outcome of public health interventions. The difficulties experienced by the modelling community in predicting the trajectory of the first wave of COVID-19 in South Africa illustrates this truth. Secondly, does this recognition render our surveillance endeavours worthless? I don't think so. Rather, it calls us to adopt a collective, multi-disciplinary approach to the collection, analysis and interpretation of surveillance data. We should no longer collect disease-specific data in isolation. We need to better understand the ecological, social, economic, psychological and cultural context of our surveillance data. We, as epidemiologists, need to work with geographers, anthropologists, environmentalists, economists, psychologists and sociologists so as to analyse and interpret our surveillance data. Where this is not possible we need at least to form collaborations and to make our data available to others for their consideration. At the least, epidemiologists need to form research questions first with a conceptual framework to outline theoretical causal pathways across multidisciplinary domains.

As I reflect on the philosophical context in which the NICD provides and surveillance data, I have a deeper understanding of why neither I, nor my colleagues I am sure – were able to provide a substantive answer to the Ministers question regarding 'lockdown'. The question was a profound one, not the least for its immense social importance at the time, but also as a challenge – to direct us, the NICD and globally - towards a more holistic approach to the collection, analysis and interpretation of surveillance data.

EXCEPTIONAL RESEARCH STATISTICS

NUMBER OF PEER-REVIEWED ARTICLES PRODUCED

QUARTER 1	53
QUARTER 2	44
QUARTER 3	51

TOP 3

MOST PUBLISHED AUTHORS IN Q3 OF 2020/2021

1

PROF LUCILLE BLUMBERG

2

PROF JOHN FREAN

3

DR MELINDA SUCHARD

3

DR. JENNY ROSSOUW

3

PROF CAROLINE TIEMESSEN

3

PROF JANUSZ PAWESKA

FEATURED RESEARCH ABSTRACTS FOR THE THIRD QUARTER OF 2020/2021

Ms Cathrine Scheepers

Prof Penny Moore

Antibody isotype switching as a mechanism to counter HIV neutralization escape

Scheepers *C, Bekker V, Anthony C, Richardson SI, Oosthuysen B, Moyo T, Kgagudi P, Kitchin D, Nonyane M, York T, Mielke D, Mabvakure B, Sheng Z, Lambson BE, Ismail A, Garrett NJ, Abdool Karim SS, Shapiro L, Williamson C, Morris L,* ***Moore PL***

Cell Reports
Impact Factor: 8.109

Neutralizing antibodies (nAbs) to highly variable viral pathogens show remarkable diversification duringinfection, resulting in an "arms race" between virus and host. Studies of nAb lineages have shown how somatic hypermutation (SHM) in immunoglobulin (Ig)-variable regions enables maturing antibodies to neutralize emerging viral escape variants. However, the Ig-constant region (which determines isotype) can also influence epitope recognition. Here, we use longitudinal deep sequencing of an HIV-directed nAb lineage,CAP88-CH06, and identify several co-circulating isotypes (IgG3, IgG1, IgA1, IgG2, and IgA2), some of which share identical variable regions. First, we show that IgG3 and IgA1 isotypes are better able to neutralize lon-gitudinal autologous viruses and epitope mutants than can IgG1. Second, detrimental class-switch recombination (CSR) events that resulted in reduced neutralization can be rescued by further CSR, which we term "switch redemption." Thus, CSR represents an additional immunological mechanism to counter viral escape from HIV-specific antibody responses. We declare no competing interests.

Dr. Jessica Coertse

Lyssaviruses in Insectivorous Bats, South Africa

Coertse J, *Grbler CS, Sabeta CT, Seamark ECJ, Kearney T, Paweska JT, Markotter W.*

Emerging Infectious Diseases
Impact Factor: 6.259

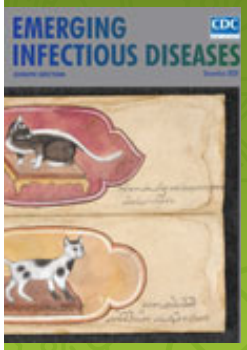
We detected 3 lyssaviruses in insectivorous bats sampled in South Africa during 2003–2018. We used phylogenetic analysis to identify Duvenhage lyssavirus and a potentially new lyssavirus, provisionally named Matlo bat lyssavirus, that is related to West Caucasian bat virus. These new detections highlight that much about lyssaviruses remains unknown.

Prof Janusz Paweska

Shedding of Marburg Virus in Naturally Infected Egyptian Rousette Bats, South Africa, 2017

Paweska JT, *Storm N, Markotter W, Di Paola N, Wiley MR, Palacios G, Jansen van Vuren P.*

Emerging Infectious Diseases
Impact Factor: 6.259



We detected Marburg virus RNA in rectal swab samples from Egyptian rousette bats in South Africa in 2017. This finding signifies that fecal contamination of natural bat habitats is a potential source of infection for humans. Identified genetic sequences are closely related to Ravn virus, implying wider distribution of Marburg virus in Africa.

Ms Faith Moyo

Dr Tendesayi Kufa

Achieving maternal viral load suppression for elimination of mother-to-child transmission of HIV in South Africa

Moyo F, *Mazanderani Haeri A; Murray T, Sherman GG,* ***Kufa T***

AIDS.2020
Impact Factor: 4.534

Objective: To describe changes in maternal viral control over time in South African women living with HIV (WLHIV) using surveillance data from the National Health Laboratory Service's Corporate Data Warehouse (NHLS CDW).

Design: A retrospective cohort analysis of maternal viral load during pregnancy and up to 15 months postpartum was performed amongst WLHIV (15–49 years) within the public health sector between 2016 and 2017.

Methods: HIV and pregnancy-related test data were used to create a synthetic cohort of pregnant WLHIV from the NHLS CDW. Syphilis-screening, in association with ward type and/or postpregnancy cervical screening and/or birth HIV test and/or positive β-hCG, was used as a proxy for pregnancy. The syphilis-screening date marked the first antenatal care visit (fANC). Fractional polynomial models described viral load evolution from fANC up to 15 months postdelivery. Piecewise linear regression models determined factors associated with viral load decline.

Findings: Among 178 319 pregnant WLHIV, 345 174 viral load tests were performed [median = 2 (IQR: 2–3) per woman]. At fANC, 85 545 (48%) women were antiretroviral therapy (ART) experienced; 88 877 (49.8%) were not and 3897 (2.2%) unknown. Proportions of viraemia (viral load ≥50 copies/ml) were 39 756 (53.6%) at first viral load performed during pregnancy, 14 780 (36.9%) at delivery and 24 328 (33.5%) postpartum. Maternal age at least 25 years, CD4+ cell count at least 500 cells/μl and viral load less than 50 copies/ml at baseline predicted sustained viral load suppression during follow-up.

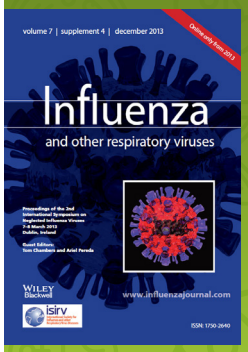
Conclusion: Despite high-ART coverage among pregnant women in South Africa, only 63% of WLHIV achieved viral load less than 50 copies/ml at delivery. Maternal viral load monitoring requires prioritization for maternal health and eMTCT.

Dr Jocelyn Moyes

Clinical characteristics, predictors, and performance of case definition Interim results from the WHO global respiratory syncytial virus surveillance pilot

Hirve S, Crawford N, Palekar R, Zhang W, Group WHORSV surveillance ***(J Moyes)***

Influenza Other Respi Viruses
Impact Factor: 3.288



Background: The lack of a uniform surveillance case definition poses a challenge to characterize the epidemiology, clinical features, and disease burden of the respiratory syncytial virus (RSV). Global standards for RSV surveillance will inform immunization policy when RSV vaccines become available.

Methods: The WHO RSV surveillance pilot leverages the capacities of the Global Influenza Surveillance and Response System (GISRS). Hospitalized and non-hospitalized medically attended patients of any age were tested for RSV using standardized molecular diagnostics throughout the year in fourteen countries. An extended severe acute respiratory infection (extended SARI) or an acute respiratory infection (ARI) case definition was used that did not require fever as a criterion.

Results: Amongst 21 221 patients tested for RSV between January 2017 and September 2018, 15 428 (73%) were hospital admissions. Amongst hospitalized RSV-positive patients, 50% were aged <6 months and 88% <2 years. The percentage of patients testing positive for RSV was 37% in children <6 months and 25% in those aged 6 months to 2 years. Patients with fever were less likely to be RSV positive compared to those without fever (OR 0.74; 95% CI: 0.63-0.86). For infants <6 months, 29% of RSV ARI cases did not have fever.

Conclusion: Requiring fever in a case definition for RSV lowers the sensitivity to detect cases in young children. Countries should consider ways to leverage the GISRS platform to implement RSV surveillance with an augmented case definition amongst the young pediatric population.

Dr Elvira Singh

The impact of the South African antiretroviral treatment programme on the age-standardised incidence rate of Kaposi sarcoma, 1999–2016: An interrupted time series analysis

Majaya E, Girdler-Brown B V, Muchengeti M, ***Singh E***

International Journal of Infectious Diseases
Impact Factor: 3.202

Objective: The objective of this study was to quantify the impact of the South African antiretroviral treatment programme on the age-standardised incidence rate of Kaposi sarcoma among black South African residents of all ages.

Methods: We performed an interrupted time series analysis using routinely collected, histologically confirmed surveillance data from the South African National Cancer Registry for the years 1999 to 2016. The analysis was performed using R statistical software. The total number of cases was 29,623 (12,475 females and 17,166 males). The background antiretroviral treatment coverage was less than 1% at the time that the antiretroviral programme was introduced and increased to over 50% in 2016.

Results: In 1999, the age-standardised rates were 1.48 and 2.82 cases per 100,000 per year for black females and males, respectively. These rates increased to 5.52 and 7.46 in 2008 before declining. The antiretroviral treatment programme was started in 2004. Five years after 2008 (nine years after the antiretroviral programme was introduced), the predicted standardised rates were 58.3% and 50.3% lower for females and males, respectively, than what they would have been without the treatment programme.

Conclusion: Introduction of the antiretroviral treatment programme was associated with a decrease of over 50% in the predicted age-standardised incidence rates of Kaposi sarcoma.

Dr Melinda Suchard

Plasma indoleamine 2, 3-dioxygenase activity, a sensitive screening tool for active pulmonary tuberculosis

Adu-Gyamfi CG, Snyman T, Makhathini L, Darboe F, Penn-Nicholson A, Fisher M, Savulescu D, Hoffmann CJ, Chaisson RE, Martinson NA, Scriba TJ, George JA, & ***Suchard MS***

International Journal of Infectious Diseases
Impact Factor: 3.202

Introduction: The World Health Organization has identified the need for a non-sputum-based test capable of detecting active tuberculosis (TB) as a priority. The plasma kynurenine-to-tryptophan (K/T) ratio, largely mediated by activity of the enzyme indoleamine 2,3-dioxygenase, may have potential as a suitable biomarker for active TB.

Method: We evaluated a commercial enzyme-linked immunosorbent assay (ELISA) in comparison to mass spectrometry for measuring the K/T ratio. We also used ELISA to determine the K/T ratio in plasma from patients with active TB compared to latently infected controls, with and without HIV.

Results: The two methods showed good agreement, with a mean bias of 0.01 (limit of agreement from –0.06 to 0.10). Using ELISA, it was found that HIV-infected patients with active TB disease had higher K/T ratios than those without TB (median, 0.101 [interquartile range (IQR), 0.091–0.140] versus 0.061 [IQR, 0.034–0.077], P < 0.0001). At a cutoff of 0.080, the K/T ratio produced a sensitivity of 90%, a specificity of 80%, a positive predictive value (PPV) of 82%, and a negative predictive value (NPV) of 90%. In a receiver operating characteristics analysis, the K/T ratio had an area under the curve of 0.93.

HIV-uninfected patients with active TB also had higher K/T ratios than those with latent TB infections (median, 0.064 [IQR, 0.040–0.088] versus 0.022 [IQR, 0.016–0.027], *P* < 0.0001). A cutoff of 0.040 gave a sensitivity of 85%, a specificity of 92%, a PPV of 91%, and an NPV of 84%.

Conclusion: The plasma K/T ratio is a sensitive biomarker for active TB. The K/T ratio can be measured from blood using ELISA. The K/T ratio should be evaluated as an initial test for TB.

Dr. Jenny Rossouw

First confirmed case of infant botulism in Africa, caused by a dual-toxin-producing *Clostridium botulinum* strain

Vosloo MN, Opperman CJ, Geyer, HDW, Setshedi GM, Allam M, Kwenda S, Ismail A, Khumalo ZTH, Brink AJ, Frean JA, ***Rossouw J.***

International Journal of Infectious Diseases
Impact Factor: 3.202

Botulism, a rare life-threatening toxemia, is probably underdiagnosed in all of its forms in Africa. This study reports the first laboratory-supported case of infant botulism on the African continent. A 10-week-old, previously well infant presented with progressive global weakness, feeding difficulty, and aspiration pneumonia. During a lengthy hospitalization, a rare bivalent *Clostridium botulinum* strain, producing subtype B3 and F8 toxins and with a new multilocus sequence type, was isolated from stool. The infant was successfully treated with a heptavalent botulinum antitoxin infusion and pyridostigmine. Despite the relative rarity of infant botulism, this case illustrates the importance of maintaining a high level of clinical suspicion when assessing hypotonic infants. The value of modern diagnostic modalities in identifying and characterizing this under-recognized condition is also demonstrated.

Dr Husna Ismail

Prof Olga Perovic

An outbreak of cutaneous abscesses caused by Panton-Valentine leukocidin-producing methicillin-susceptible *Staphylococcus aureus* among gold mine workers, South Africa

Ismail H, *Govender NP, Singh-Moodley A, van Schalkwyk E, Shuping L, Moema I, Feller G, Mogokotleng R, Strasheim W, Lowe M, Mpembe R, Naicker S, Maphanga TG, De Abreu C, Ismail F, Ismail N, Allam M, Ismail A, Singh T, Matuka O, Duba T****, Perovic O.***

BMC Infectious Diseases
Impact Factor: 3.143

Background: We aimed to describe an outbreak of cutaneous abscesses caused by Panton-Valentine leukocidin (PVL)-producing methicillin-susceptible *Staphylococcus aureus* (MSSA) among gold mine workers.

Methods: In February 2018, we retrospectively reviewed a random sample of 50 medical records from 243 cases and conducted face-to-face interviews using a structured questionnaire. Pus aspirates were sent to the National Institute for Communicable Diseases from prospectively-identified cases (November 2017–March 2018). Nasopharyngeal swabs were collected during a colonisation survey in February 2018. *Staphylococcus aureus* isolates were screened with a conventional PCR for *lukS*/F-PV. Pulsed-field gel electrophoresis (PFGE) was performed to determine the genetic relatedness among the isolates. A sample of isolates was selected for whole genome sequencing (WGS). We conducted an assessment on biological risks associated with mining activities.

Results: From January 2017 to February 2018, 10% (350/3582) of mine workers sought care for cutaneous abscesses. Forty-seven medical files were available for review, 96% were male ($n=45$) with a mean age of 43 years (SD = 7). About 52% (24/46) were involved in stoping and 28% (13/47) worked on a particular level. We cultured *S. aureus* from 79% (30/38) of cases with a submitted specimen and 14% (12/83) from colonisation swabs. All isolates were susceptible to cloxacillin. Seventy-one percent of *S. aureus* isolates (30/42) were PVL-PCR-positive. Six PFGE clusters were identified, 57% (21/37) were closely related. WGS analysis found nine different sequence types. PFGE and WGS analysis showed more than one cluster of *S. aureus* infections involving closely related isolates. Test reports for feed and product water of the mine showed that total plate counts were above the limits of 1000 cfu/ml, coliform counts > 10 cfu/100 ml and presence of faecal coliforms. Best practices were poorly implemented as some mine workers washed protective clothing with untreated water and hung them for drying at the underground surface.

Conclusions: PVL-producing MSSA caused an outbreak of cutaneous abscesses among underground workers at a gold mining company. To our knowledge, no other outbreaks of PVL-producing *S. aureus* involving skin and soft tissue infections have been reported in mining facilities in South Africa. We recommend that worker awareness of infection prevention and control practices be strengthened.

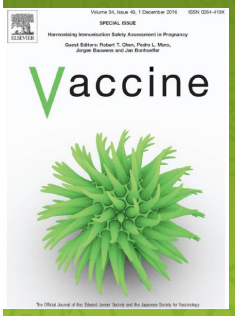
Prof Stefano Tempia

Prof Cheryl Cohen

Influenza economic burden among potential target risk groups for immunization in South Africa, 2013–2015

Tempia S, Moyes J, Cohen AL, Walaza S, McMorrow ML, Edoka I, Fraser H, Treurnicht FK, Hellferscee O, Wolter N, von Gottberg A, McAnerney JM, Dawood H, Variava E, ***Cohen C***

Vaccine
Impact Factor: 3.143



Background: Data on influenza economic burden in risk groups for severe influenza are important to guide targeted influenza immunization, especially in resource-limited settings. However, this information is limited in low- and middle-income countries.

Methods: We estimated the cost (from a health system and societal perspective) and years of life lost (YLL) for influenza-associated illness in South Africa during 2013–2015 among (i) children aged 6–59 months, (ii) individuals aged 5–64 years with HIV, pulmonary tuberculosis (PTB) and selected underlying medical conditions (UMC), separately, (iii) pregnant women and (iv) individuals aged ≥65 years, using publicly available data and data collected through laboratory-confirmed influenza surveillance and costing studies. All costs were expressed in 2015 prices using the South Africa all-items Consumer Price Index.

Results: During 2013–2015, the mean annual cost of influenza-associated illness among the selected risk groups accounted for 52.1% ($140.9/$270.5 million) of the total influenza-associated illness cost (for the entire population of South Africa), 45.2% ($52.2/$115.5 million) of non-medically attended illness costs, 43.3% ($46.7/$107.9 million) of medically-attended mild illness costs and 89.3% ($42.0/$47.1 million) of medically-attended severe illness costs. The YLL among the selected risk groups accounted for 86.0% (262,069 /304,867 years) of the total YLL due to influenza-associated death.

Conclusion: In South Africa, individuals in risk groups for severe influenza accounted for approximately half of the total influenza-associated illness cost but most of the cost of influenza-associated medically attended severe illness and YLL. This study provides the foundation for future studies on the cost-effectiveness of influenza immunization among risk groups.

Prof Olga Perovic

Diversity of SCC*mec* elements and *spa* types in South African *Staphylococcus aureus mec*A-positive blood culture isolates

Singh-Moodley A, Lowe M, Mogokotleng R, ***Perovic O***

BMC Infectious Diseases
Impact Factor: 3.143

Background: The prevalence of *Staphylococcus aureus* varies depending on the healthcare facility, region and country. To understand its genetic diversity, transmission, dissemination, epidemiology and evolution in a particular geographical location, it is important to understand the similarities and variations in the population being studied. This can be achieved by using various molecular characterisation techniques. This study aimed to provide detailed molecular characterisation of South African *mec*A-positive *S. aureus* blood culture isolates by describing the SCC*mec* types, *spa* types and to lesser extent, the sequence types obtained from two consecutive national surveillance studies.

Methods: *S. aureus* blood culture isolates from a national laboratory-based and enhanced surveillance programme were identified and antimicrobial susceptibility testing was performed using automated systems. A real-time PCR assay confirmed the presence of the methicillin-resistance determinant, *mec*A. Conventional PCR assays were used to identify the SCC*mec* type and *spa* type, which was subsequently analysed using the Ridom StaphType™ software. Multilocus sequence typing was performed on selected isolates using conventional methods. MRSA clones were defined by their sequence type (ST), SCC*mec* type and *spa* type.

Results: A detailed description of findings is reported in this manuscript. SCC*mec* type III predominated overall followed by type IV. A total of 71 different *spa* types and 24 novel *spa* types were observed. *Spa* type t037 was the most common and predominated throughout followed by t1257. Isolates were multidrug resistant; isolates belonging to all SCC*mec* types were resistant to most of the antibiotics with the exception of type I; isolates with *spa* type t045 showed resistance to all antibiotics except vancomycin. The most diverse SCCmec-spa type complex was composed of the SCC*mec* type IV element and 53 different *spa* types.

Conclusion: Although ST data was limited, thereby limiting the number of clones that could be identified, the circulating clones were relatively diverse.

Prof Janusz Paweska

Farm-Level Risk Factors of Increased Abortion and Mortality in Domestic Ruminants during the 2010 Rift Valley Fever Outbreak in Central South Africa

Paweska JT, *Rostal M.K,Cleaveland S, Cordel C, van Staden L, Matthews L, Anyamba A, Karesh BW, Haydon DT, Ross N.*

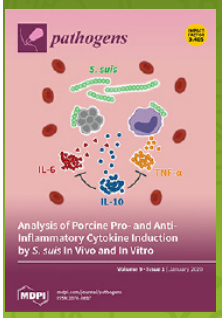
Pathogens
Impact Factor: 3.018

Background: Rift Valley fever (RVF) outbreaks in domestic ruminants have severe socio-economic impacts. Climate-based continental predictions providing early warnings to regions at risk for RVF outbreaks are not of a high enough resolution for ruminant owners to assess their individual risk.

Methods: We analyzed risk factors for RVF occurrence and severity at the farm level using the number of domestic ruminant deaths and abortions reported by farmers in central South Africa during the 2010 RVF outbreaks using a Bayesian multinomial hurdle framework.

Results: We found strong support that the proportion of days with precipitation, the number of water sources, and the proportion of goats in the herd were positively associated with increased severity of RVF (the numbers of deaths and abortions). We did not find an association between any risk factors and whether RVF was reported on farms.

Conclusions: At the farm level we identified risk factors of RVF severity; however, there was little support for risk factors of RVF occurrence. The identification of farm-level risk factors for Rift Valley fever virus (RVFV) occurrence would support and potentially improve current prediction methods and would provide animal owners with critical information needed in order to assess their herd's risk of RVFV infection.



Dr. Jenny Rossouw

Multi-Locus sequence analyses reveal a clonal *L. borgpetersenii* genotype in a heterogeneous invasive *Rattus* spp. Community across the City of Johannesburg, South Africa

Moselet M, Naidoo K, Bastos A, Retief L, Frean J, Telfer S, ***Rossouw J.***

Parasit Vectors
Impact Factor: 2.824

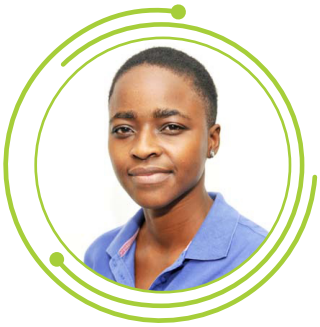
Background: *Rattus* spp. are frequently implicated as key reservoir hosts for leptospirosis, one of the most common, but neglected, bacterial zoonoses in the world. Although leptospirosis is predicted to be a significant public health threat in Africa, studies from the continent are limited.

Methods: *Rattus* spp. ($n = 171$) were sampled (January–May 2016) across the City of Johannesburg, South Africa's largest inland metropole. *Rattus* spp. genetic diversity was evaluated by full length (1140 bp) cyt b sequencing of 42 samples. For comparison, a further 12 *Rattus norvegicus* samples collected in Cape Town, South Africa's largest coastal metropole, were also genotyped. *Leptospira* infections were identified and genotyped using real-time PCR and multi-locus (*lfb1*, *secY* and *lipL41*) DNA sequencing.

Results: Five *R. norvegicus* haplotypes were identified across Johannesburg, four of which have not previously been detected in South Africa, and one in Cape Town. Across Johannesburg we identified a *Leptospira* spp. infection prevalence of 44% (75/171) and noted significant differences in the prevalence between administrative regions within the metropole. Multi-locus sequence analyses identified a clonal genotype consistent with *L. borgpetersenii* serogroup Javanica (serovar Ceylonica).

Discussion: The prevalence of infection identified in this study is amongst the highest detected in *Rattus* spp. in similar contexts across Africa. Despite the complex invasion history suggested by the heterogeneity in *R. norvegicus* haplotypes identified in Johannesburg, a single *L. borgpetersenii* genotype was identified in all infected rodents. The lack of *L. interrogans* in a rodent community dominated by *R. norvegicus* is notable, given the widely recognised host-pathogen association between these species and evidence for *L. interrogans* infection in *R. norvegicus* in Cape Town. It is likely that environmental conditions (cold, dry winters) in Johannesburg may limit the transmission of *L. interrogans*. Spatial heterogeneity in prevalence suggest that local factors, such as land use, influence disease risk in the metropole.

Conclusions: In South Africa, as in other African countries, leptospirosis is likely underdiagnosed. The high prevalence of infection in urban rodents in Johannesburg suggest that further work is urgently needed to understand the potential public health risk posed by this neglected zoonotic pathogen.



Dr Tsidiso Maphanga

Prof Nelesh Govender

Cross-reactivity of a *Histoplasma capsulatum* antigen enzyme immunoassay in urine specimens from persons with emergomycosis in South Africa

Maphanga TG, *Naicker SD, Gómez BL, Mhlanga M, Mpembe RS, Schwartz IS, Bamford C, Nel J,* ***Govender NP***

Medical Mycology
Impact Factor: 2.822

Histoplasma antigen detection in urine is a rapid diagnostic method for disseminated histoplasmosis, although cross-reactivity has been reported in specimens from patients with other thermally dimorphic fungal infections. We tested urine specimens, from persons with suspected invasive fungal infections, using a commercial monoclonal antibody *Histoplasma* enzyme immunoassay (EIA) at a South African national mycology reference laboratory from August 2014 through December 2018. Corresponding fungal culture and histopathology results were obtained from an electronic laboratory information system. In some cases, cultured fungal isolates were sent with the urine specimen for species-level identification by phenotypic and molecular methods. Cross-reactivity was confirmed using culture filtrates of several fungal pathogens. Of 212 referred cases, 41 (19%) were excluded since they had no recorded clinical history (n = 1), alternative diagnoses were confirmed (n = 2), or no fungal culture or histopathology results (n = 38). Eighty-seven of 212 (41%) had laboratory evidence of an invasive fungal disease, while 84 (40%) did not. Of the 87 cases, 37 (43%) were culture-confirmed mycoses: emergomycosis (n = 18), histoplasmosis (n = 8), sporotrichosis (n = 6), cryptococcosis (n = 2), talaromycosis (n = 1), and other fungi isolated (n = 2). The sensitivity and specificity of the EIA were calculated for two groups: culture-confirmed (n = 37) and histology-confirmed invasive fungal disease (n = 50). The sensitivity and specificity of the EIA for diagnosis of histoplasmosis compared to culture were 88% (7/8, 95%CI 47-100%) and 72% (21/29, 95%CI 53-87%), respectively, and for diagnosis of emergomycosis/histoplasmosis compared to histology was 83% (29/35, 95%CI 66-93%) and 93% (14/15, 95%CI 68-100%), respectively. Cross-reactions occurred in urine specimens of patients with *Emergomyces africanus* infection and in culture filtrates of *E. africanus, T. marneffei* and *Blastomyces* species. A commercial *Histoplasma* EIA had satisfactory accuracy for diagnosis of culture-confirmed histoplasmosis, but cross-reacted in urine specimens from patients with invasive disease caused by the closely-related pathogen, *E. africanus* and in culture filtrates of *E. africanus* and other related fungi.

Medical Mycology

Dr Sandrama Nadan

Prof Nicola Page

Circulation of classic and recombinant human astroviruses detected in South Africa: 2009 to 2014

Sandrama Nadan, *Maureen B Taylor,* ***Nicola A Page***

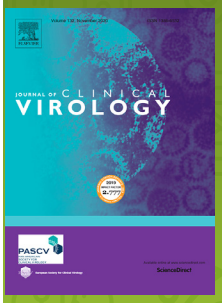
Journal of Clinical Virology
Impact Factor: 2.777

Background: Astroviruses (AstVs) are associated with diarrhoeal and extra-intestinal infections in human, animal and avian species. A prevalence of 7% was reported in selected regions in SA while AstVs detected from clinical stool specimens were almost identical phylogenetically to strains identified in environmental and water samples. This study investigated the molecular diversity of astroviruses circulating between 2009 and 2014 in South Africa (SA).

Methods: Astroviruses detected in stool specimens collected from hospitalised children were investigated retrospectively. Astroviruses were characterised using type-specific RT-PCR, partial nucleotide sequence analyses in ORF1 and ORF2 and whole genome sequencing. Different genotypes were compared with clinical features to investigate genotype-related associations. The Vesikari severity scale (VSS) was evaluated for scoring astrovirus diarrhoeal infections.

Results: Of 405 astroviruses detected, 49.9 % (202/405) were characterised into 32 genotypes comprising 66.3 % (134/202) putative-recombinants and 33.7 % (68/202) classic strains. No trends by year of collection, age or site were observed. Whole genome analysis in eight strains revealed that genotypes assigned by partial nucleotide sequence analyses to five astroviruses were incorrect. Bivariate analyses showed there were no significant associations between genotypes and clinical symptoms or severity of infection. A comparison of Vesikari parameters with astrovirus-positive proxy values demonstrated that Vesikari scores for duration of diarrhoea and admission temperatures would result in a milder infection rating in astrovirus-positive cases.

Conclusions: Diverse genotypes co-circulated with putative-recombinants predominating. Astrovirus classification was complicated by the lack of a consistent characterisation system and reliable reference database. The VSS should be used cautiously to rate astrovirus diarrhoea. While surveillance in communities and out-patient clinics must be continued, screening for human astroviruses in alternate hosts is needed to determine the reservoir species.

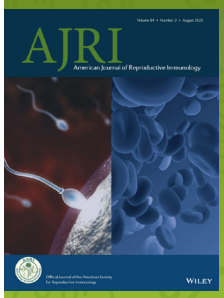


Dr Melinda Suchard

HLA antibody repertoire in infants suggests selectivity in transplacental crossing

Savulescu DM, Groome M, Malfeld SCK, Madhi S, Koen A, Jones S, Duxbury V, Scheuermaier K, de assis Rosa D and ***Suchard MS***

American Journal of Reproductive Immunology
Impact Factor: 2.739



Problem: Late in pregnancy, women produce and transfer high amounts of antibodies to the foetus. During gestation, women produce antibodies against human leukocyte antigens (HLA), including antibodies directed at foetal HLA. There is paucity of data on transplacental crossing, specificity and role of HLA antibodies in pregnancy and new-borns.

Method of study: Using highly sensitive Luminex technology, we measured prevalence of IgG HLA antibodies in 30 mother-infant pairs six weeks post-partum. Additionally, in six pregnant women, we measured HLA antibodies longitudinally and HLA-typed infant DNA to assess whether maternal HLA antibodies were directed at infant specificities.

Results: Overall, 68% of mothers and 44% of infants expressed HLA-I antibodies and 56% of mothers and 52% of infants expressed HLA-II antibodies. Infants shared up to 78% of antibodies with their mothers, suggesting that the remaining antibodies were self-made. Less than 25% of maternal HLA antibodies were detected in infants, possibly due to selection in transplacental crossing. We detected complement-fixing HLA antibodies in mothers and at low levels in infants. In a third of our pregnant subjects, we detected infant-directed HLA antibodies.

Conclusion: Our findings raise the possibility of selection in transplacental crossing of HLA antibodies. As HLA antibodies may act as autoantibodies in the neonate, the mechanism of a selective transfer may give important insights into immune tolerance. Findings also suggest that infants start producing their own HLA antibodies in the first weeks of life, which, together with maternally derived antibodies may impact the infant's immune reaction to HLA proteins.

Dr Shaheed Vally Omar

Dr Farzana Ismail

Systematic rifampicin resistance errors with Xpert® MTB/RIF Ultra: implications for regulation of genotypic assays

Omar SV, *Hillemann D, Pandey S, Merker M, Witt AK, Nadarajan D, Barilar I, Bainomugisa A, Kelly EC, Diel R, Vidanagama DS, Samarasinghe AIP, Cader MR, Gotsch U, Lavu E, Alabi A, Schon T, Coulter C, Niemann S, Maurer FP, Ismail NA, Kosher CU,* ***Ismail F***

The International Journal of Tuberculosis and Lung Disease
Impact Factor: 2.268

With nearly 12 million cartridges sold annually to the public sector alone, the Cepheid Xpert MTB/RIF (Xpert) is the most important commercial diagnostic assay for TB. It is increasingly being replaced with the Xpert MTB/RIF Ultra (Ultra) owing to the improved limit of detection of Ultra in samples with low bacterial loads. The underlying assumption for this transition has been that the sensitivity and specificity of Ultra to detect rifampicin resistance is either equivalent or potentially even superior to Xpert. In the attached study, three WHO Supranational Reference Laboratories reveal that this is not the case. Specifically, we show that systematic false resistant results are still possible, which should be addressed by adjusting the reporting language of the assay.

Dr Villyen Motaze

Dr Melinda Suchard

Rubella seroprevalence using residual samples from the South African measles surveillance program: a cross-sectional analytic study

Motaze NV, *Makhathini L, Smit SB, Adu-Gyamfi CG, Fortuin M, Wiysonge CS,* ***Suchard MS***

Human Vaccines & Immunotherapeutics
Impact Factor: 2.619

Introduction: South Africa is yet to introduce rubella-containing vaccines (RCV) into its routine immunization schedule. Selecting the target population when introducing RCV should take into account the ages of susceptible individuals in the population. We aimed to determine the seroprevalence of antibodies to rubella and characterize immunity gaps among individuals of all ages in South Africa.

Methods: We tested for rubella immunoglobulin G (IgG) antibodies with a commercial enzyme-linked immunosorbent assay. We used residual samples collected from 2016 through 2018 as part of the national measles surveillance program. We only tested samples that were negative for measles and rubella immunoglobulin M (IgM) and explored the association between rubella susceptibility (IgG negative) and predictor variables (year of sample collection, age, sex, and province of residence) using logistic regression analysis.

Results: We obtained results for 6057 records. Rubella susceptibility was highest among Individuals aged zero to 11 months (81.9%), followed by children 1 to 5 years old (71.5%), 6 to 10 y old (40.9%) and 11 to 15 y old (31.25) while the smallest proportion of susceptible individuals was among those 16 to 49 y old (19.9%). Females were less likely to be susceptible to rubella compared to males (OR = 0.79 (95%CI: 0.71–0.87), $P < .001$) in unadjusted analysis but this effect was not observed after adjusting for age and province. In multivariable logistic regression, age (OR = 6.24 (4.52–8.63), $P < .001$) and province of residence (OR = 0.97 (95%CI: 0.95–0.99), $P = .01$) were associated with rubella susceptibility.

Conclusion: In the absence of rubella vaccination in the Expanded Program on Immunization in South Africa, the bulk of individuals susceptible to rubella are children under 16 y old. About 20% of individuals 16 to 49 y old are susceptible to rubella. This susceptibility gap must be born in mind during RCV introduction.

Dr Anthony Smith

Dr Juno Thomas

Whole-genome sequencing to investigate two concurrent outbreaks of *Salmonella* Enteritidis in South Africa, 2018

Smith AM, *Tau NP, Ngomane HM, Sekwadi P, Ramalwa N, Moodley K, Govind C, Khan S, Archary M,* ***Thomas J.***

Journal of Medical Microbiology
Impact Factor: 2.156

Salmonella enterica serotype Enteritidis (*Salmonella* Enteritidis) is a major cause of foodborne disease outbreaks worldwide. In 2018, two concurrent outbreaks of *Salmonella* Enteritidis gastroenteritis in one district of South Africa were investigated. We describe the use of whole-genome sequencing (WGS) analysis of bacterial isolates to assist with the investigation of these outbreaks. Outbreak A affected children (*n*=27) attending a day-care centre, while outbreak B affected adults (*n*=16) who ate breakfast at the same restaurant. *Salmonella* Enteritidis was isolated from stool samples in both outbreaks (four children in outbreak A; 12 restaurant customers and three restaurant food-handlers in outbreak B). In outbreak B, *Salmonella* Enteritidis was isolated from three food retention samples (raw chicken egg, hollandaise sauce and rocket-herb). Available isolates from both outbreaks (*n*=13) were investigated using WGS analysis. Sequencing data for isolates were analysed at the EnteroBase web-based platform and included core-genome multi-locus sequence typing (cgMLST). Isolates with epidemiological links to the restaurant (*n*=10) and day-care centre (*n*=3), were shown by cgMLST to be highly genetically related, with no more than five allele differences when comparing one isolate against another. On food history, eggs and hollandaise sauce were the common food items consumed by ill restaurant customers. Unfortunately, *Salmonella* Enteritidis isolated from the egg and hollandaise sauce were not available for WGS analysis. Our investigation concluded that the two concurrent outbreaks were caused by a highly related strain of *Salmonella* Enteritidis, suggesting the possibility of a common contaminated food source, of which contaminated eggs are strongly implicated.

Prof Olga Perovic

Molecular diagnostics in South Africa and challenges in the establishment of a molecular laboratory in developing countries

Singh-Moodley A, Ismail H, ***Perovic O***

Journal of Infection in Developing Countries
Impact Factor: 0.703

The laboratory plays a significant role in public health surveillance, outbreak investigation and infection prevention and control strategies. Microbiology laboratories are moving towards incorporating molecular biology techniques for the surveillance and identification of pathogens causing infectious diseases as well as the genotypic characterisation of these organisms. These methods are accurate, rapid, reliable, and provide a wealth of information that are not available using conventional phenotypic methods. However, establishing such a laboratory can be challenging in developing countries due to poor infrastructure, the lack of funding and the required expertise. This manuscript discusses the essential issues that need to be addressed when establishing a molecular microbiology laboratory and the usefulness of molecular techniques in public health surveillance and outbreaks in developing countries. Molecular data on South African findings obtained from surveillance and outbreak studies are also presented in this manuscript.

Prof Basil Brooke

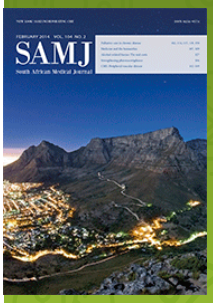
Prof Lucille Blumberg

Implementing malaria control in South Africa, Eswatini and southern Mozambique during the COVID-19 pandemic

Brooke BD, *Raman J, Frean J, Rundle J, Maartens F, Misiani E, Mabuza A, Barnes KI, Moonasar DP, Dlamini Q, Charles S,* ***Blumberg L***

South African Medical Journal
Impact Factor: n/a

The COVID-19 pandemic has strained healthcare delivery systems in a number of southern African countries. Despite this, it is imperative that malaria control and elimination activities continue, especially to reduce as far as possible the number and rate of hospitalisations caused by malaria. The implementation of enhanced malaria control/elimination activities in the context of COVID-19 requires measures to protect healthcare workers and the communities they serve. The aim of this review is therefore to present innovative ideas for the timely implementation of malaria control without increasing the risk of COVID-19 to healthcare workers and communities. Specific recommendations for parasite and vector surveillance, diagnosis, case management, mosquito vector control and community outreach and sensitisation are given.



Dr Howard Wayne

Prof Adrian Puren

Description of non-polio enteroviruses identified in two national surveillance programmes in South Africa

Howard W, *Savulescu D, Berrie L,* ***Puren AJ***

Southern African Journal of Infectious Diseases
Impact Factor: n/a

Background: Human enteroviruses (EV) consist of 106 serotypes and four species: EV-A, EV-B, EV-C and EV-D. Enteroviruses cause clinical symptoms varying from severe to mild. Knowledge of EV burden in South Africa is limited, and as non-polio EV are important causes of acute flaccid paralysis (AFP) and meningitis, information on the circulating serotypes is vital.

Methods: Between 2010 and 2012, a total of 832 stool and viral isolate specimens was obtained from two national surveillance programmes at the National Institute for Communicable Diseases: the Rotavirus Sentinel Surveillance Programme (RSSP) and the AFP surveillance programme. Real-time polymerase chain reaction and Sanger sequencing were performed to detect and serotype EV.

Results: Non-polio EV were detected in 446 specimens, of which 308 were sequenced. Stool specimens yielded a greater variety of serotypes than viral cultures. EV-B viruses were predominant (58.44%), whilst EV-C viruses were detected in 31% of the specimens tested. South African prevalence for these viruses was higher than other countries, such as France with less than 2%, and Spain and the United States with less than 10%. The most common serotype detected was Enterovirus 99 (EV-C, 8.63%), which has not been reported in other regions.

Conclusion: Direct sequencing from stool specimens yields a broader, more comprehensive description of EV infections compared to sequencing from viral cultures. Disease-associated serotypes were detected, but only in small numbers. This study provides a baseline for EV strain circulation; however, surveillance needs to be expanded to improve EV knowledge in South Africa.

Ms Unarine Makungo

Dr Kerrigan McCarthy

Epidemiological investigation of a typhoid fever outbreak in Sekhukhune District, Limpopo province, South Africa – 2017

Makungo UB, *Ramutshila TE, Mabotja MC, Thomas J, Lekalakala-Mokaba R, Smith AM, Ebonwu J, Williams SL, Khoza J, Ranoto Q, Muvhango N, Mosoma M, Phokane E, Ntshoe G, Calver K, Essel V, Ngobeni MF,* ***McCarthy K***

Southern African Journal of Infectious Diseases
Impact Factor: n/a

Background: Typhoid fever remains a public health concern in South Africa, where the risk of transmission is high because of poor access to safe water and sanitation. This study describes the investigation of typhoid fever outbreak in Limpopo province.

Methodology: Following notification of laboratory-confirmed cases, a descriptive study was conducted at Sekhukhune District, Limpopo province. A suspected case was defined as any person residing in Makhuduthamaga Municipality from November 2017 to January 2018, presenting with fever and gastrointestinal symptoms. Data were collected using case investigation forms. Whole-genome sequencing (WGS) was carried out on *Salmonella* Typhi isolates and polymerase chain reaction (PCR) test was done for *Salmonella* species from water samples. Location of cases and water sources were mapped using ArcGIS mapping tool.

Results: Amongst 122 cases, 54% ($n = 66$) were female and 6% ($n = 7$) laboratory-confirmed. The median age of the cases was 11 years (range 2–83 years), with 79% ($n = 102$) being children under the age of 14 years. *Salmonella* species were detected in 37% (10/27) of water samples and geographic information system (GIS) mapping showed clustering of cases in Tswaing-Kgwaripe and Vlakplaas villages. Six isolates were available for WGS analysis, with resulting data showing that five of the six isolates were genetically related. Phylogenetic analysis showed that the five isolates clustered together were genetically related showing <22 single nucleotide polymorphisms when compared to each other.

Conclusion: Molecular epidemiology of isolates suggests a common source outbreak, supported by the detection of *Salmonella* species from water sources. Consumption of water from contaminated open water sources, because of ongoing interruption of municipal water supply, was the likely cause of the outbreak. The investigation highlights the importance of consistent safe water supply and the ability of district surveillance systems to identify and contain outbreaks.

Dr Jaishree Raman

Maintaining focus on administering effective malaria treatment during the COVID-19 pandemic

Raman J, *Barnes KI, Baker L, Blaylock M, Blumberg L, Frean J, Misiani E, Ukpe IS*

Southern African Journal of Infectious Diseases
Impact Factor: n/a

As September marks the start of the malaria season in South Africa (SA), it is essential that healthcare professionals consider both COVID-19 and malaria when a patient who lives in or has recently travelled to a malaria area presents with acute febrile illness. Early diagnosis of malaria by either a rapid diagnostic test or microscopy enables prompt treatment with the effective antimalarial, artemether-lumefantrine, preventing progression to severe disease and death. Intravenous artesunate is the preferred treatment for severe malaria in both children and adults. Adding single low-dose primaquine to standard treatment is recommended in endemic areas to block onward transmission. Use of the highly effective artemisinin-based therapies should be limited to the treatment of confirmed malaria infections, as there is no clinical evidence that these antimalarials can prevent or treat COVID-19. Routine malaria case management services must be sustained, in spite of COVID-19, to treat malaria effectively and support SA's malaria elimination efforts.

Thank you for your contributions! Kindly continue to send your contributions to:

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