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FOREWORD

The West African Ebola epidemic began in December 2013, peaked during the period September – December 2014, and is now gradually declining. To date, the vast majority of cases have occurred in Sierra Leone and Liberia although the highest case fatality rate comes from Guinea. Fortunately, no Ebola cases have been confirmed in South Africa since the outbreak was announced. An update of the epidemic is given in this issue, which also includes three surveillance reports.

The 2015 winter influenza season in South Africa will once again be carefully monitored by several influenza surveillance programmes coordinated at the NICD. Last year, these programmes showed that the season in South Africa was predominated by influenza A(H3N2), followed by influenza B and influenza A(H1N1)pdm09. This is also the first report that combines viral pathogens with additional testing for bacterial pathogens as well as some of the atypical causes of pneumonia.

Sentinel site surveillance of transmitted HIV drug resistance (TDR) is ongoing and the results from specimens collected as part of the 2012 national antenatal sentinel HIV & Herpes Simplex Type-2 prevalence survey in South Africa are reported here. These data provide the first national TDR estimate for South Africa and indicate that levels of TDR are low to moderate.

Antimicrobial resistance surveillance is also conducted at the NICD, and aims to determine the extent of resistance amongst the most important disease causing pathogens in South Africa. Data presented in this issue show the extent of antimicrobial resistance by pathogen for 2014.

All contributors are thanked for their inputs and I trust you will find these reports useful and interesting.

Basil Brooke, Editor

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EBOLA VIRUS DISEASE OUTBREAK IN WEST AFRICA: ONE YEAR DOWN THE LINE AND THE NICD RESPONSE

Jacqueline Weyer[1], Petrus Jansen van Vuren[1], Genevie Ntshoe[2], Chikwe Ihekweazu[2], Veerle Msimang[1], Lucille Blumberg[2] & Janusz T. Paweska[1]

[1]Centre for Emerging and Zoonotic Diseases, NICD
[2]Division of Public Health Surveillance and Response, NICD

Background

Ebola virus disease (EVD) is caused by zoonotic RNA viruses belonging to the family Filoviridae. Five diverse species of Ebola virus have been described to date but only the *Zaire* and *Sudan ebolaviruses* have been associated with sizeable and highly fatal human outbreaks.[1] Natural outbreaks caused by *Zaire ebolavirus* (ZEBOV) have been described mostly from equatorial Africa, but the current outbreak in West Africa suggests that the geographic distribution of the virus may be more widespread than anticipated.[2] The Ebola viruses in Africa are thought to be maintained in specific species of arboreal bats, but exact mechanisms of spill-over into other susceptible animal and human populations are not fully understood.[3-5] Various outbreaks have been traced back to index patients that had contact with bushmeat, for example chimpanzees during the Gabon outbreak in 1996, and bats during the 2014 Democratic Republic of Congo outbreak.[6-7] The likely index case of the ongoing West Africa outbreak was a two-year-old patient that died in December 2013, who supposedly had contact with bats while playing in a hollow tree near his home.[8-9] Once the virus has spilled over to humans, outbreaks are perpetuated by direct contact with infected bodily fluids, secreta and excreta. This mode of transmission predisposes healthcare workers, relatives and friends caring for the sick.[10-12] Burial ceremonies have been recognized as a major risk factor for transmission.[10-12]

A more comprehensive overview of EVD was provided in the previous NICD Communicable Disease Surveillance Bulletin (2014, Volume 12 number 4, pages 109-116). Here we give an update on the outbreak developments in West Africa and summarize the response launched by the South African National Institute for Communicable Diseases (NICD) to this event.

EVD in West Africa

On 29 March 2015, one year since the outbreak was first reported, a cumulative total of 25 178 EVD cases (laboratory-confirmed, probable and suspected), including 10 445 deaths with an overall case fatality rate (CFR) of 41% has been reported by the World Health Organization (WHO) in the three countries currently affected with intense transmission, i.e. Guinea, Liberia and Sierra Leone.[13]

The outbreak started in Guinea's southern district, Guéckédou in December 2013, with subsequent spread to other districts (including the capital Conakry), as well as neighbouring countries Liberia and Sierra Leone. A marked increase in the number of EVD cases was noted from July 2014 onward with a peak occurring in September/October 2014 in Liberia and November/ December in Sierra Leone, before gradually declining to lower levels in 2015. Rates of disease incidence have differed by country, with Sierra Leone and Liberia being the most affected. As of 29 March 2015, the total number of EVD cases reported per country was 3 492 EVD cases in Guinea, despite the fact that the epidemic started there, and totals of 9 712 and 11 974 EVD cases in Liberia and Sierra Leone, respectively.[13] Over the past 21 days, one new confirmed case was reported from Liberia on 20 March (who later died on 27 March),

while Guinea and Sierra Leone continued reporting relatively many new cases (197 and 113 respectively) and deaths over the same period (8–29 March).[13]

Sustained transmission of infection occurred at community level for a prolonged time in numerous regions and towns across all three affected countries. However, the numbers of cases differ within regions with certain districts accounting for a higher proportion of total cases than others, while other areas remain unaffected. In Guinea, transmission remains centred in the west of the country, in and around the capital Conakry and nearby prefectures (Boffa, Coyah, Dubreka and Forecariah). Two additional prefectures, Fria in the west and Siguiri, to the north on the border with Mali, reported cases for the first time in over 50 days. In Sierra Leone confirmed cases are still being reported from the northern and western districts as well as in and around the capital Freetown (Bombali, Kambia, Port Loko, and Western Rural) in the week leading to 29 March 2015 (figure 1).

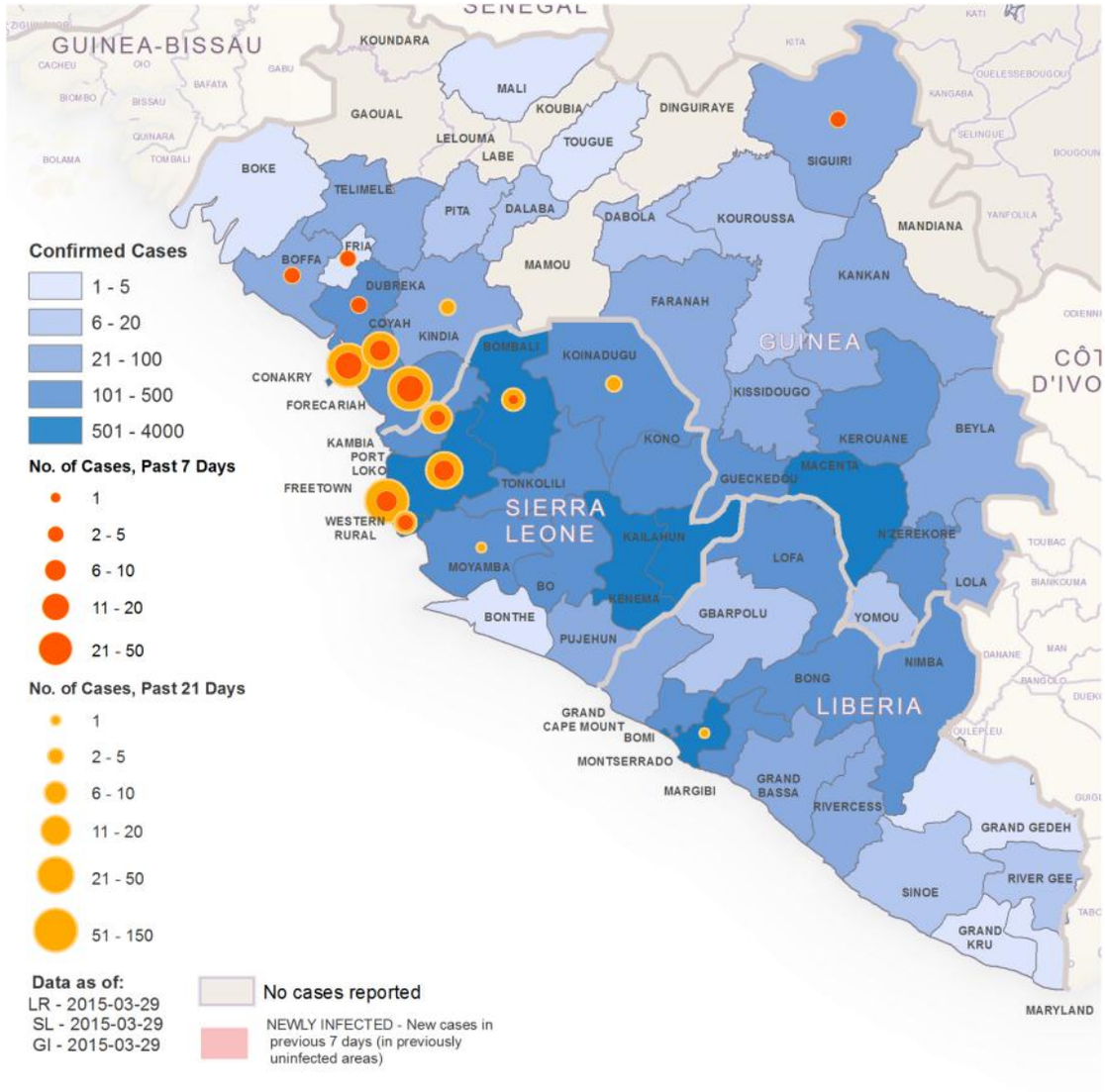

Figure 1: Geographical distribution of Ebola virus disease (EVD) cases in Guinea, Liberia and Sierra Leone as at 29 March 2015. Source: World Health Organization: Ebola situation report of 1 April 2015 (http://apps.who.int/ebola/current-situation/ebola-situation-report-1-april-2015-0).

It is also noted that case fatality rates (CFR) vary from country to country and per location within each country: the highest overall CFR is recorded for Guinea (66%; 2314/3492), followed by Liberia with a CFR of 45% (4332/9712) and the lowest death rate (32%; 3799/11974) amongst all cases is documented for Sierra Leone.

Healthcare workers (HCWs) have frequently been infected while treating patients with suspected or confirmed EVD and their case numbers and deaths continue to be reported up until present. To date, 861 cases amongst HCWs have been reported, with 495 deaths (CFR: 57%); the highest HCWs CFR is being reported from Sierra Leone (73%).[13]

Where information on age and gender is available, people aged 15-44 are three times more likely to be affected than children (<15 years), and those 45 and over, three to five times more likely. Rates of disease incidence are similar between males and females within all three affected countries (figure 2.)[13]

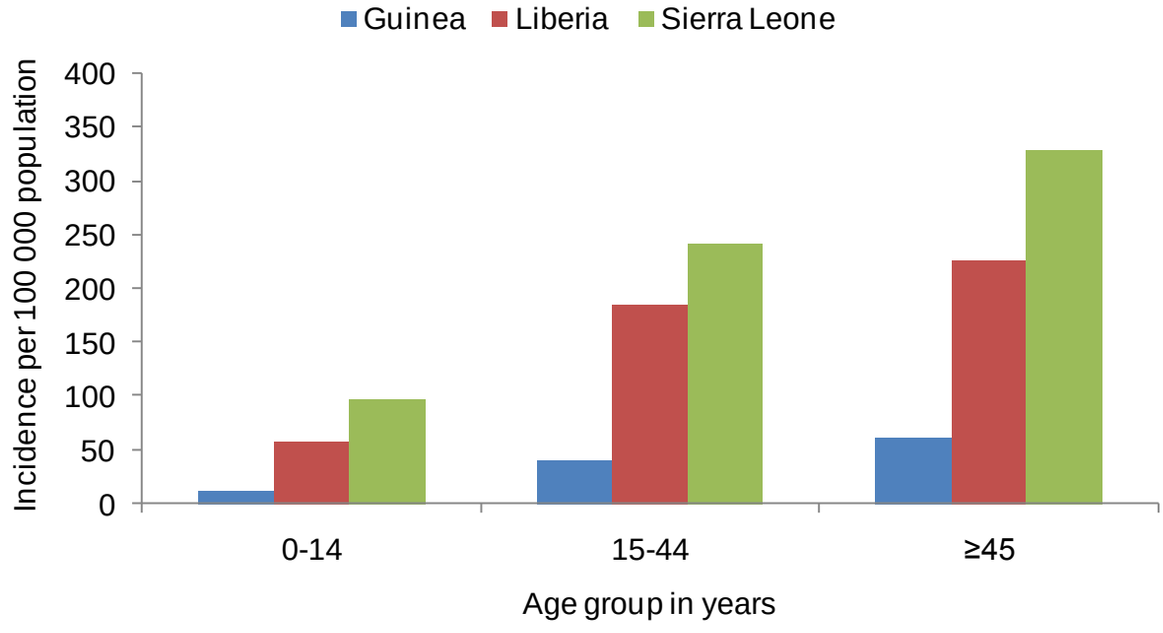

Figure 2: Age-specific incidence of Ebola virus disease cases in Guinea, Liberia and Sierra Leone as at 29 March 2015 (created from data in WHO Ebola situation report of 1 April 2015).

Several interventions to contain spread have been instituted with the primary goal of breaking the chains of transmission, and include: early identification of cases and contacts, isolating and treating patients, infection prevention and control, laboratory diagnosis, contact tracing and safe burials. However, unsafe burials and unknown chains of transmission (those who were not known to be contacts of a previous case) continue to be reported, especially in Guinea.

EVD status in South Africa, SADC region and other countries

Even though the outbreak has been largely confined to Guinea, Liberia and Sierra Leone, there is a concern about importing EVD cases into other countries.

No EVD cases have been confirmed in South Africa since the West African outbreak was announced. Close monitoring and testing is being carried out on returned

travellers from the affected countries that present with features suggestive of EVD. No EVD cases have been documented from other SADC countries to date.

Thus far there are six countries i.e. Nigeria (20 cases, 8 deaths), Mali (8 cases, 6 deaths), Senegal (1 case), United States of America (4 cases, 1 death), Spain (1 case) and the United Kingdom (1 case) that have reported localised transmission or imported a case or cases from Guinea/Liberia/Sierra Leone. Nonetheless, the EVD outbreaks in these countries have been declared over. The latest imported EVD case-patient, a healthcare worker who returned from volunteering at an Ebola treatment centre in Sierra Leone, was confirmed EVD positive on 29 December 2014 in Scotland in the United Kingdom, but has recovered and gained EVD-negative status on 24 January 2015.[14]

NICD response to the EVD outbreak

EVD testing of returned travellers to South Africa or SADC, and other national activities

The Centre for Emerging and Zoonotic Diseases (CEZD) of the NICD operates the only maximum biosafety laboratory (biosafety level 4) in Africa. With more than 30 years history in the laboratory diagnosis and investigation of viral haemorrhagic fevers (VHFs) including EVD, the NICD represents a critical national and international resource during the current EVD outbreak in West Africa. The CEZD is recognized by the WHO as one of nine laboratories worldwide for expertise in diagnosis of filoviruses, and has a status of World Health Organization Collaborating Centre for Reference and Research of VHFs and Arboviruses. From February 2014 to date, samples from a total of 37 suspected cases were subjected to testing at the CEZD at the NICD in Johannesburg. Eight of these cases were referred from other African countries including Zimbabwe (n=2), Namibia (n=4), Angola and Ethiopia (not SADC). The remainder were patients presenting to South African health-care facilities, who required follow-up for suspected EVD. All of these cases tested negative for EVD. The differential diagnosis of these cases revealed infectious and non-infectious causes of disease. A total of ten cases was diagnosed with malaria, but laboratory testing also supported trypanosomiasis (n=1), dengue (n=1) and parvovirus infection (n=1). Two cases involving Nigerian patients were related to complications of sickle-cell anaemia, a common hereditary condition in the Nigerian population. One patient was diagnosed with severe complications of autoimmune disease, whilst another died from a possible drug reaction.

In addition to follow up of suspected cases, the NICD also contributed to drafting of guidelines, EVD risk communication to the general public and other stakeholders, and various training activities to support responses to EVD in other countries. With the outbreak of Ebola virus disease in West Africa, the NICD has also initiated an Emergency Operations Centre (EOC) in South Africa to prepare and respond better to high-risk outbreaks such as Ebola in the country, in line with its mandate by the National Department of Health.

Ebola molecular diagnostic facility in Sierra Leone

With the momentum of the EVD outbreak in Liberia and Sierra Leone building after June 2014, and in response to international calls for assistance, the NICD of South Africa has been actively involved in the “hot zone”. In mid-August 2014, the NICD deployed its field Ebola Molecular Diagnostic Facility (EMDF) to Freetown, Sierra Leone. Under Professor Janusz Paweska’s leadership, volunteers from the NICD and from the University of Pretoria were deployed under the management of the Global Outbreak Response Network of the World Health Organization. The EMDF established by CEZD-NICD in Freetown-Lakka, Sierra Leone, is a significant collaborative effort of the Sierra

Leone Ministry of Health and Sanitation, the South African Department of Health and the World Health Organization, in response to the public health crisis caused by the unprecedented outbreak of EVD. The full operationalisation of the EMDF in August 2014 has been critically important in supporting the response to the EVD outbreak in Sierra Leone.

Clockwise from top left: Dr Jacqueline Weyer setting up a polymerase chain reaction (PCR) for testing a suspected EVD case in South Africa; the team from the Ethiopian Public Health Institute (including Director Ahmed Kebede, left front) received training in EVD diagnosis at CEZD in October 2014; Alan Kemp and Professor Janusz Paweska working in the only biosafety level 4, maximum containment facility in Africa, which is housed at the NICD.

Up to March 2015 the NICD had deployed eight teams to operate the EMDF in Freetown-Lakka, each comprising of 2-5 members, rotating every 4-6 weeks. Since the second week of the NICD EMDF operation, the NICD staff undertook intensive training of Sierra Leonean scientists and technical personnel in facility operational logistics, biosafety and Ebola diagnostic procedures. The national staff became fully integrated into all aspects of functions and operation of the EMDF.

The Western Urban Area of Sierra Leone, where the NICD EMDF operates, remained a hotspot of the EVD epidemic for months. Consequently, this facility played a major role in providing Ebola diagnostic services for Sierra Leone, especially during the first months of the epidemic, when it was the only Ebola diagnostic facility in Freetown. As of 31 March 2015, the NICD EMDF has tested 7 261 clinical specimens from suspected EVD cases, of which 32.67% were positive by RT-PCR. The

NICD contributes to the following aspects of EVD outbreak response in Sierra Leone:

- Provides assistance to the Sierra Leonean national laboratory counterparts, the WHO Country Office, and international response teams in the diagnosis of EVD.
- Provides adequate bio-hazard protection of staff working in the EMDF.
- Contributes to the improvement of the national Ebola diagnostic capacity.
- Provides technical assistance to the Government of Sierra Leone in controlling the EVD outbreak.

Clockwise from top left: Professor Janusz Paweska and the first team deployed to Sierra Leone; Dr Petrus Jansen van Vuren setting up a PCR run at the Lakka Laboratory; Günther Meier assists Cardia Fourie in donning of personal protective equipment before entering the containment area; staff performing inactivation of specimens from suspected EVD cases. Note the containment infrastructure associated with this procedure.[15]

Epidemiology support to the West African outbreak

In addition to laboratory expertise, two medical epidemiologists were deployed to Liberia for epidemiology and surveillance of EVD cases. Dr Ngormbu Ballah, a Liberian medical doctor who is completing a Field Epidemiology Training Programme (FETP) in SA, returned to his country during July-August 2014. Dr Chikwe Ihekweazu was deployed in

Montserrado County (including the capital city of Liberia, Monrovia) during January-February 2015. Both experts worked with the World Health Organization team while in Liberia.

Dr Ihekweazu fed back on the challenges of contact tracing during the response. Contact tracing is the process of finding everyone who has come in direct contact with a sick Ebola patient. Contacts are monitored for signs of illness for 21 days from the last day they came in contact with the Ebola patient. If the contact develops a fever or other Ebola symptoms, they are immediately isolated, tested, provided care, and the cycle starts again – all of the new patient's contacts are found and watched for 21 days. In January 2015, 22 cases in the St. Paul Bridge cluster in Liberia led to about five hundred contacts who were followed every day for 21 days by the team. Contacts were urged to stay at home for 21 days. Ensuring the comfort of contacts during the 21-day follow-up period was essential in achieving the isolation objectives. A major challenge was ensuring that all contacts received food and other essential commodities while at home. A complicating factor at this stage of the outbreak was the increasing number of suspected cases that turned out to be negative, but all of whom required isolation until testing was completed. During this period, the full public health response, including contact tracing, had to be instituted. The large number of contacts that required monitoring made paper-based questionnaires burdensome and impracticable. Making use of personal mobile phones for collection of data may be a more efficient solution for future contact tracing and outbreak control. Finally, while carrying out contact tracing and monitoring, a compassionate approach is required. It is important to remember that these "contacts" are indeed people that most likely suffered loss due to Ebola, and it would be unfair to expect them to behave in a completely rational way in such circumstances. Coordination of the numerous resources, in particular human capacity and the technical skills from different organisations with same mission and expertise, was a major task in itself.

Dr. Chikwe Ihekweazu visiting a treatment centre (left); at the Ministry of Health in Liberia (right).

Concluding remarks

The ongoing and unprecedented Ebola outbreak in West Africa has resulted in one of the largest public health responses to an outbreak to date. These efforts far exceeded the borders of the countries reporting EVD cases, and have not only seen governments around the world putting measures in place to prevent introduction of EVD into their countries, but also to be prepared to handle EVD cases if they should occur. As numbers of EVD cases are declining in the three most-affected

countries, the experts are calling the “end game” for the EVD outbreak. Nonetheless, efforts are required in the coming weeks and months to ensure that the final case of EVD during this outbreak can be concluded.

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References

1. Kuhn JH, Boa Y, Bavari S, *et al*., Virus nomenclature below the species level: a standardized nomenclature for natural variants of viruses assigned to the family Filoviridae. *Arch Virol* 2013; 158: 301-11.
2. Centres for Disease Control and Prevention. Outbreaks chronology: Ebola virus disease. http://www.cdc.gov/vhf/ebola/outbreaks/history/chronology.html. (Accessed: 23 March 2015)
3. Pourrut X, Kumulungui B, Wittmann T, *et al*. The natural history of Ebola virus in Africa. *Microbes and Infection* 2005; 7: 1005-1014.
4. Groseth A, Feldmann H, Strong JE. The ecology of Ebola viruses. *Trends in Microbiology* 2007; 15(9):408-416.
5. Olival KJ, Hayman DT. Filoviruses in bats: current knowledge and future directions. *Viruses* 2014; 6: 1759 – 1788.
6. World Health Organization. Outbreak(s) of Ebola haemorrhagic fever, Congo and Gabon, October 2001-July 2002. *Weekly Epidemiological Report* 2003; 78(26): 223-225. PMID: 15571171.
7. World Health Organization. WHO declares end of Ebola outbreak in the Democratic Republic of Congo. http://www.who.int/mediacentre/news/statements/2014/drc-ends-ebola/en/. (Accessed: 17 December 2014)
8. Baize S, Pannetier D, Oestereich L, *et al*. Emergence of Zaire Ebola virus disease in Guinea – preliminary report. *NEJM* 2014; 371: 1418-1428.
9. Saéz M, Weiss S, Nowak K, *et al*., Investigating the zoonotic origin of the West African Ebola epidemic. *EMBO Mol Med.* 2014; 7(1):17-23.
10. Drosten C. What transmission trees reveal about Ebola. *The Lancet* 2015; 15:258-259.
11. Shears P, O’Dempsey TJD. Ebola virus disease in Africa: epidemiology and nosocomial transmission. *J Hosp Infect* 2015; https://dx.doi.org/10.106/j.jhin.2015.01.002.
12. Faye O, Boële P-Y, Heleze E, *et al*. Chains of transmission and control of Ebola virus disease in Conakry,

Guinea, in 2014: and observational study. *The Lancet* 2015; 15: 320-326.

13. World Health Organization. Ebola situation report – 1 April 2015. http://apps.who.int/ebola/current-situation/ebola-situation-report-1-april-2015-0; (Accessed: 6 April 2015)
14. World Health Organization. Ebola situation report – 31 December 2014 and 28 January 2015. http://apps.who.int/ebola/en/status-outbreak/situation-reports/ebola-situation-report-31-december-2014; http://apps.who.int/ebola/en/ebola-situation-report/situation-reports/ebola-situation-report-28-january-2015 (Accessed; 6 April 2015)
15. Paweska JT. Third Report on the operation of the Ebola Mobile Laboratory of the National Institute for Communicable Diseases in Sierra Leone, Freetown. National Institute for Communicable Diseases, Sandringham, South Africa. http://www.nicd.ac.za/assets/files/NICD%20EML%20SL%203rd%20J%20Paweska%20report%20Dec%202014%20final%20pdf.pdf (Accessed: 30 March 2015)

RESPIRATORY PATHOGENS FROM INFLUENZA-LIKE ILLNESS AND PNEUMONIA SURVEILLANCE PROGRAMMES, SOUTH AFRICA 2014

Cheryl Cohen[1], Andries Dreyer[2], John Frean[3], Florette Treurnicht[1], Samantha Iyaloo[3], Nazir Ismail[2], Jo Mcanerney[1], Jocelyn Moyes[1], Bhavany Poonsamy[3], Anne von Gottberg[1], Sibongile Walaza[1], Nicole Wolter[1]

[1]Centre for Respiratory Diseases and Meningitis, NICD
[2]Centre for Tuberculosis, NICD
[3]Centre for Opportunistic, Tropical and Hospital Infections, NICD

Introduction and methods

The National Institute for Communicable Diseases (NICD) has been conducting active, prospective, hospital-based sentinel surveillance for severe acute respiratory illness (SARI) since February 2009. Surveillance was initially conducted in three of South Africa's provinces: Gauteng - Chris Hani-Baragwanath Hospital (CHBH) (this site stopped enrolling patients in December 2013 and was replaced by the Helen Joseph and Rahima Moosa hospital Complex (RMMCH/HJ) in July 2014); KwaZulu-Natal - Edendale Hospital (EDH); and Mpumalanga - Matikwana and Mapulaneng Hospitals (Matikwana/Mapulaneng). This programme has been described previously.[1] Patients were enrolled based on standardised clinical case definitions (table 1). The programme initially focused on the detection of influenza, but included testing for other respiratory viruses and *Streptococcus pneumoniae*. In June 2010, Klerksdorp-Tshepong Hospital Complex (KTHC), North West Province, was included as a new site. At the same time, the case definition was expanded at KTHC and Edendale Hospital (enhanced surveillance sites) to include cases with severe respiratory illness irrespective of symptom duration, and patients with a clinician admission diagnosis of suspected tuberculosis (TB). This expanded case definition was termed severe respiratory illness (SRI). In 2012, the surveillance was further expanded at the two enhanced surveillance sites (Edendale and KTHC) to include expanded testing of specimens (naso- and oropharyngeal swabs and aspirates) for additional pathogens and collection of additional specimens (induced sputum and oral washes) from patients with SRI (table 2). Also in 2012, the NICD initiated a programme of systematic influenza-like illness (ILI) surveillance at public health clinics. Two primary health care clinics serviced by the two enhanced SRI surveillance sites (Edendale and KTHC) commenced systematically enrolling patients with ILI. Dedicated staff screened and enrolled patients from Monday to Friday each week.

The Viral Watch sentinel surveillance programme, which started in 1984, was specifically designed to monitor influenza activity and has been fully described

previously.[2] Participation in the programme is voluntary, and it is mainly composed of general practitioners who are requested to submit specimens from patients with influenza-like illness. During 2014, 171 practitioners registered across South Africa submitted specimens throughout the year. Clinical case definitions for the surveillance programmes are described in table 1 and types of specimens collected and pathogens tested for are described in table 2.

Table 1: Case definitions by age group and surveillance site/programme for the clinical syndromes included in the influenza-like illness and pneumonia surveillance programmes, South Africa, 2014.

Case definition	Criteria	Surveillance site/programme
Influenza-like illness (ILI)	**Patients of all ages** Acute fever of ≥38° Celsius and/or self-reported fever within the last 10 days AND Cough The absence of other diagnoses	Viral watch programme Public health clinic ILI surveillance at Jouberton (Klerksdorp) and Edendale Gateway clinics
Severe acute respiratory illness (SARI) Patient presenting within 10 days of the onset of illness	**2 days-<3 months** Any child hospitalised with diagnosis of suspected sepsis or physician diagnosed lower respiratory tract infection (LRTI) irrespective of signs and symptoms.	EDH, KTHC Complex, Matikwana/ Mapulaneng, RMMCH/HJ
	3 months-<5 years Any child ≥3 months to <5 years hospitalised with physician-diagnosed LRTI including bronchiolitis, pneumonia, bronchitis and pleural effusion.	
	≥5 years Any person hospitalised with an acute respiratory infection with fever (≥38ºC) or history of fever AND cough.	
Severe chronic respiratory illness (SCRI)	Any child or adult meeting the above case definitions presenting with symptom duration >10 days or Any patient with a clinical diagnosis of suspected pulmonary TB AND not meeting any of the above criteria	RMMCH/HJ, EDH, KTHC
Severe Respiratory illness (SRI)	Anyone who meet either SARI or SCRI definitions at RMMHC/HJ, EDH and KTHC hospitals	RMMCH/HJ, KTHC, EDH,

EDH = Edendale Hospital, KTHC = Klerksdorp-Tshepong Hospital Complex, RMMHC/HJ = Helen Joseph and Rahima Moosa Hospital Complex

Table 2: Pathogens tested for by clinical syndrome, surveillance site, type of specimen collected and test conducted - influenza-like illness and pneumonia surveillance, South Africa, 2014.

Pathogen	Programme (syndrome)	Surveillance site	Specimen collected	Test conducted
Influenza and respiratory syncytial virus	Viral watch (ILI)	All Viral watch sites in 9 provinces	Nasopharyngeal (NP) and oropharyngeal (OP) flocked swabs	Multiplex Real-time reverse transcription polymerase chain reaction (RT-PCR)
	Systematic ILI surveillance (ILI)	Edendale Gateway Clinic and Jouberton clinic Klerksdorp	NP and OP flocked swabs > 5 years Nasopharyngeal aspirates (NPA) in children ≤5 years of age	
	Pneumonia surveillance (SARI and SCRI)	EDH, KTHC, Matikwana/Mapulaneng	NP and OP flocked swabs > 5 years. NPA ≤5 years Induced sputum (IS), all ages.	
Human metapneumovirus, Parainfluenza viruses, 1, 2 and 3	Systematic ILI surveillance (ILI)	Edendale Gateway Clinic and Jouberton clinic Klerksdorp	NP and OP flocked swabs > 5 years NPA ≤5 years	Multiplex RT- PCR
	Pneumonia surveillance (SARI and SRI)	EDH, KTHC, Matikwana/Mapulaneng RMMCH/HJ	NP and OP flocked swabs > 5 years NPA ≤5 years	
Bordetella pertussis, Mycoplasma pneumoniae, Legionella spp, Chlamydophila pneumonia	ILI	Edendale Gateway Clinic and Jouberton clinic Klerksdorp	NP and OP flocked swabs > 5 years NPA ≤5 years	Real Time -PCR
	Pneumonia surveillance (SRI)	EDH, KTHC	NP and OP flocked swabs > 5 years NPA ≤5 years Induced sputum	
Streptococcus pneumoniae	SARI, SCRI	EDH, KTHC, Matikwana/Mapulaneng, RMMHC/HJ	Whole blood	PCR for *Lyt A*
Tuberculosis	SRI	EDH, KTHC	Induced or expectorated sputum	GeneXpert and culture
Pneumocystis jirovecii	Pneumonia surveillance (SRI)	EDH, KTHC	Oral washes, Nasopharygeal swabs/ aspirates Induced or expectorated sputum	Real time-PCR

ILI = influenza like illness, SRI = severe respiratory illness, SARI = severe acute respiratory illness, SCRI = severe chronic respiratory illness, EDH = Edendale Hospital, KTHC = Klerksdorp-Tshepong Hospital Complex, RMMHC/HJ = Helen Joseph and Rahima Moosa hospital Complex

The primary objective of the pneumonia and systematic ILI surveillance programmes is to describe the burden and aetiology of outpatient ILI and inpatient severe respiratory illness in HIV-infected and HIV-uninfected children and adults in selected sites in South Africa. This report presents the findings from these surveillance programmes for 2014 for the following pathogens: influenza, respiratory syncytial virus (RSV), human metapneumovirus (hPMV), parainfluenza viruses, 1, 2 and 3 (PIV1-3), *Streptococcus pneumoniae, Bordetella pertussis* and atypical bacterial causes of pneumonia (*Legionella species, Chlamydia pneumoniae* and *Mycoplasma pneumoniae*), tuberculosis and *Pneumocystis jirovecii* (PCP). Data from this

surveillance programme for 2013 were reported in the March 2014 and November 2014 editions of the Communicable Diseases Surveillance Bulletin. [3, 4]

Sample collection and processing

Upper respiratory tract specimens (oropharyngeal - OP, nasopharyngeal - NP and nasopharyngeal aspirates - NPA) were collected in viral transport medium. Whole blood specimens were collected in EDTA containing tubes. Oral washes and sputum were collected in universal containers. Following collection, upper respiratory and blood samples were kept at 4°C at the local laboratory, and were transported to the NICD on ice within 72 hours post-collection. Sputum samples were stored separately at -20^{0}C at the local laboratory before being transported to the NICD on dry ice on a weekly basis. One sputum sample was tested at the surveillance site laboratory for *M. tuberculosis* using GeneXpert and a second sample was tested at the NICD for *M. tuberculosis* by culture, as well as for PCP and bacterial pathogens by PCR (table 2).

Detection of viral pathogens

Respiratory specimens were tested by multiplex real-time reverse-transcription PCR assay for 10 respiratory viruses (influenza A and B viruses, parainfluenza virus 1, 2 and 3; respiratory syncytial virus; enterovirus; human metapneumovirus; adenovirus and rhinovirus). Influenza positive specimens were subtyped using the US Centers for Disease Control and Prevention (CDC) real-time reverse-transcription PCR protocol for characterisation of influenza virus.[5] *Streptococcus pneumoniae* was identified by quantitative real-time PCR detecting the lytA gene from whole blood specimens.

Detection of bacterial pathogens other than tuberculosis

Induced sputum and nasopharyngeal samples were tested for *M. pneumoniae*, *C. pneumoniae, Legionella* spp. and *B. pertussis.* DNA was extracted from the clinical specimens and tested for bacterial pathogens by RT-PCR. A specimen was considered positive for *M. pneumoniae* if the *MP181* target was detected (Ct<45), *C. pneumoniae* if the *CP-Arg* target was detected (Ct<45) and *Legionella* spp. if the Pan-Leg target was detected (Ct<45).[6] A positive result for pertussis was obtained when a specimen was positive for *IS481* and/ or *ptxS*1 genes.[7] Blood specimens were tested using quantitative real-time PCR for the presence of pneumococcal DNA (*lyt*A gene). For *lytA* testing, specimens with a *lytA* Ct-value <40 were considered positive.[8]

Detection of tuberculosis

Tuberculosis testing at the site laboratory was based on the GeneXpert System (Cepheid, Sunnyvale, CA) using the cartridge-based Xpert MTB/RIF (Xpert) assay.[9] Tuberculosis microscopy for acid-fast bacilli was conducted for some patients. All induced sputum specimens were also tested for *M. tuberculosis* by smear for acid-fast bacilli and culture in liquid media using BD Bactec MGIT 960 at the NICD. Positive cultures were identified as *M. tuberculosis* complex using Ziehl-Neelsen staining and antigen testing. Genotypic resistance to isoniazid and rifampicin was tested using the Hain MTBDRplus v2 assay.[10]

Detection of Pneumocystis jirovecii (PCP)

Pneumocystis jirovecii was tested for on one or more of the following specimens from each patient- oral wash, naso/oropharyngeal sample and induced sputum. DNA was extracted from the clinical specimens using an automated DNA extraction system. Fungal load was determined using a quantitative real-time PCR targeting the region coding for the mitochondrial large subunit rRNA for *P. jirovecii*.[11] All specimens with copy numbers >0 copies/µl were included as positive. These include both cases of infection and colonisation with *P. jirovecii*.

Determination of HIV status

HIV status data was obtained from two data sources. Firstly, for some patients HIV testing was requested by admitting physicians as part of clinical management. This included HIV enzyme-linked immunosorbent assay (ELISA) testing with confirmation by ELISA on a second specimen for patients ≥18 months of age, and qualitative HIV PCR testing for confirmation of HIV-infection status in children <18 months of age. Secondly, for consenting patients, linked anonymous HIV PCR testing for children <18 months of age or ELISA for patients ≥18 months of age was performed using a dried blood spot or whole blood specimen.

Data management

Data management was centralised at the NICD where laboratory, clinical and demographic data from enrolled patients were recorded on a Microsoft Access database.

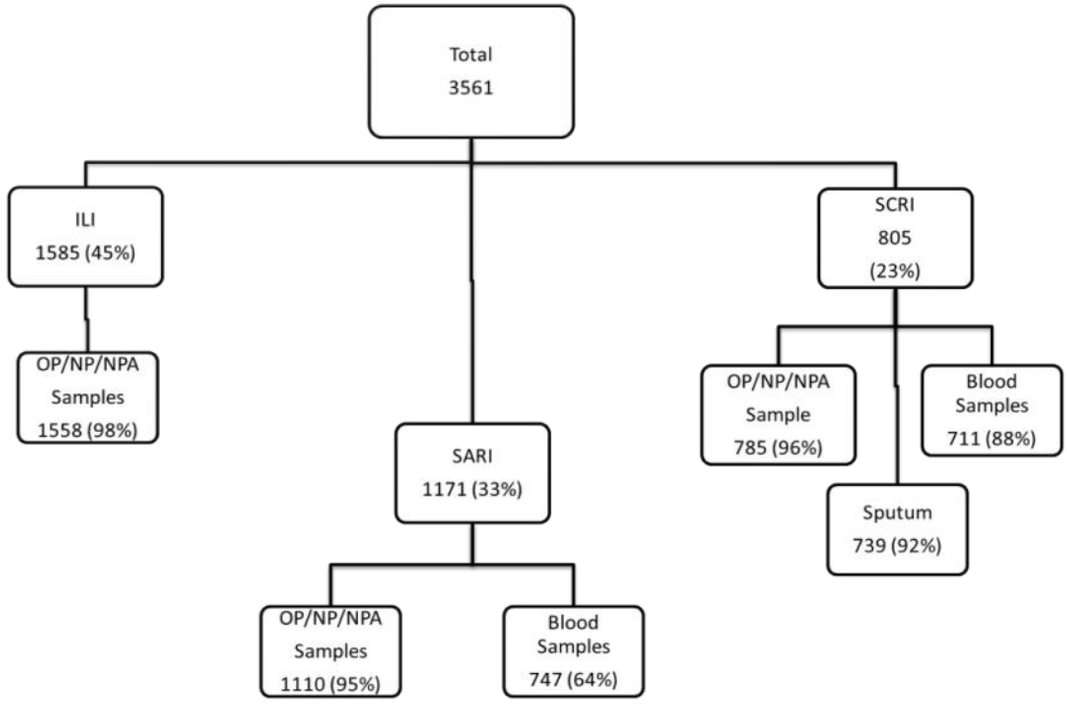
Ethical considerations

The protocol was approved by the Research Ethics Committees of the University of the Witwatersrand and University of KwaZulu-Natal.

Results

Of the 3693 patients enrolled into the surveillance programme, 3561 had full data on case definition available, 1585 (45%) fitted the case definition of ILI, 1171 (33%) were diagnosed with SARI and 805 (23%) were diagnosed with SCRI. The demographic characteristics of patients enrolled into the programme are described in table 3. The number of samples tested for each of these groups of patients depended on the samples available and suitable for testing. The number of samples tested for each case definition and pathogen are detailed in figure 1.

Figure 1: Numbers of samples collected by case definition in the influenza like illness and pneumonia surveillance programmes, South Africa, 2014.



ILI = influenza like illness, SARI = severe acute respiratory illness, SCRI = severe chronic respiratory illness, OP=Oropharyngeal, NP= Nasopharyngeal, NPA= nasopharyngeal aspirate

The HIV prevalence varied by case definition and age group; the overall prevalence was lowest in the SARI group (147/638, 23% vs 387/1424, 27% in the ILI group), likely driven by the high numbers of young children in the SARI group. HIV prevalence was highest in the SCRI group (522/723, 72%) and was highest in the 25-44 age group across all case definitions (ILI 236/403, 59%; SARI 51/58, 88%; SCRI 313/337, 93%) (figure 2).

Table 3: Demographic and clinical characteristics of patients with an upper respiratory sample available for testing and enrolled into the systematic influenza-like illness and pneumonia surveillance programmes, South Africa, 2014.

Characteristic	**Influenza-like illness n/N (%) N=1585**	**Severe acute respiratory illness n/N (%) N=1171**	**Severe chronic respiratory illness n/N (%) N=805**
Age group years			
0-4	525/1584 (33)	827/1165 (70)	48/800(6)
5-14	231/1584 (14)	39/1165 (3)	14/800 (2)
15-24	187/1584 (11)	28/1165 (2)	53/800 (7)
25-44	424/1584 (27)	147/1165 (13)	375/800 (47)
45-64	180/1584 (11)	91/1165 (8)	243/800 (30)
≥ 65	37/1584 (2)	33/1165 (30)	67/800 (8)
Female gender	1019/1582 (64)	607/1168 (48)	390/803 (49)
Site			
Edendale Gateway clinic	1025/1585 (67)	N/A	N/A
Jouberton clinic	560/1585 (33)	N/A	N/A
EDH	N/A	324/1171 (28)	204/805 (55)
KTHC	N/A	248/1171 (21)	601/805 (75)
Mapulaneng/Matikwana hospitals	N/A	246/1171 (23)	N/A
RMMCH/HJ	N/A	353/1171 (30)	N/A
In hospital case-fatality ratio	N/A	35/1095 (3)	95/792 (12)

EDH= Edendale Hospital, KTHC= Klerksdorp-Tshepong hospital complex, RMMCH/HJ= Rahima Moosa Mother and Child Hospital/Helen Joseph Hospital

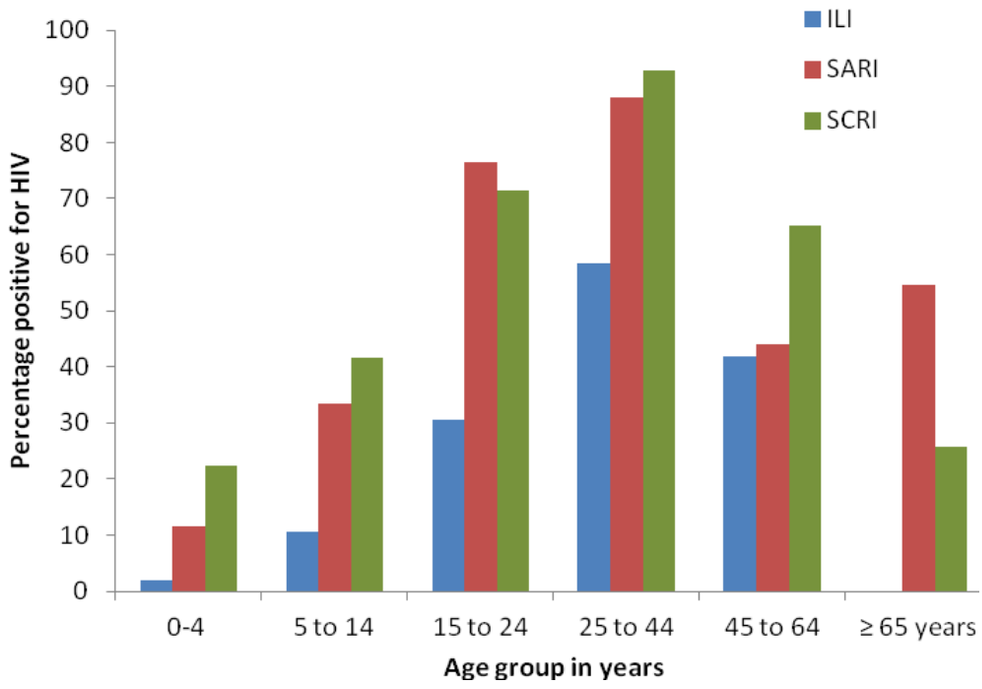


Figure 2: HIV prevalence by age group for case definitions of influenza-like illness (ILI), severe acute respiratory illness (SARI) and severe chronic respiratory illness (SCRI), among patients enrolled in pneumonia and systematic ILI surveillance, South Africa, 2014.

Pneumonia surveillance programme results

Viral pathogens (SARI and SCRI)

Of individuals tested for influenza 4% (74/1895) were positive. The season was dominated by influenza A (H3N3) (46/74, 62%) followed by influenza B (25/74, 34%) and influenza A (H1N1)pdm09 (3/74, 4%). The season started in week 30 and continued through week 33. The peak detection rate was 26% in week 33 (figure 3).

The detection rate for RSV was 11% (205/1689). The RSV season preceeded the influenza season, started in week 4 and continued through week 19. The peak detection rate of 52% was in week 10 (figure 4). Parainfluenza viruses 1-3 were detected in 5% (98/1894) of samples and hMPV in 3% (56/1894) of samples. In the same group of patients *S. pneumoniae* was detected in 15% (212/1458) of blood samples (figure 5). Across these pathogens the highest number of cases were in the <5 year age group (influenza 35/73 (48%), RSV 184/205 (90%), PIV1-3 76/97 (78%), hMPV 42/56 (75%) and *S. pneumoniae* 72/211 (34%). The case fatality ratio was highest for patients testing positive for *S. pneumoniae* 18/206 (9%), table 4.

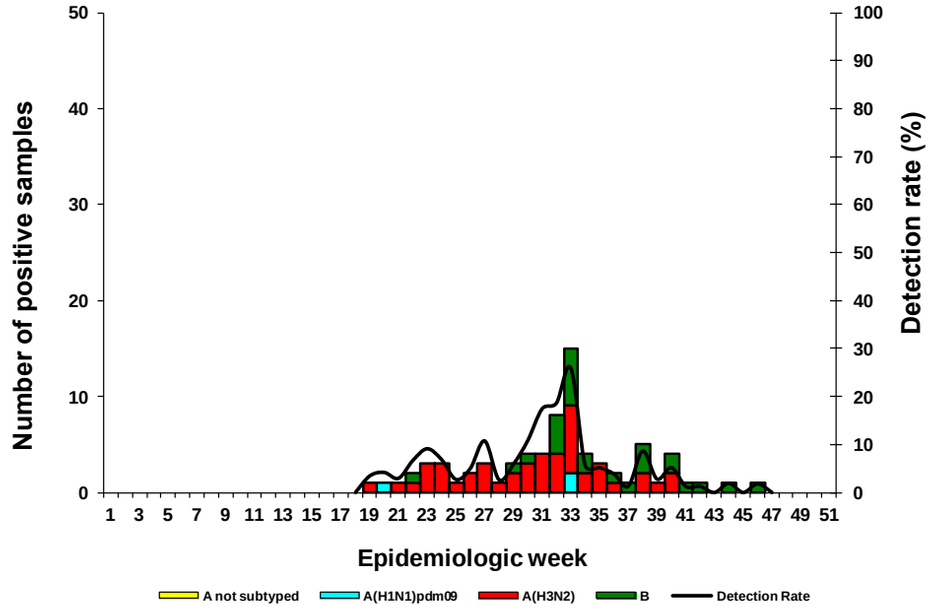


Figure 3: Numbers of samples positive for influenza and influenza detection rate, by subtype and week, in patients enrolled into the pneumonia surveillance fitting the case definition of severe acute respiratory illness (SARI) or severe chronic respiratory illness (SCRI) in South Africa, 2014.

Table 4: Characteristics of patients who tested positive for viral pathogens and *Streptococcus pneumoniae* amongst those patients with severe acute respiratory illness (SARI) or severe chronic respiratory illness (SCRI).

	Influenza n/N(%)	RSV n/N(%)	PIV 1-3 n/N(%)	hMPV n/N(%)	*S. pneumoniae* n/N(%)
Age group, years					
0-4	35/73 (48)	184/205(90)	76/97 (78)	42/56 (75)	72/211 (34)
5-14	2/73 (3)	2/205 (1)	3/97 (3)	0	4/211 (2)
15-24	5/73 (7)	3/205 (1)	4/97 (4)	0	10/211 (5)
25-44	12/73 (16)	7/205 (3)	8/97 (8)	7/56 (13)	71/211 (34)
45-64	13/73 (18)	4/205 (2)	5/97 (5)	5/56 (9)	43/211 (20)
≥ 65	6/73 (8)	5/205 (2)	1/97 (1)	2/56 (4)	11/211 (5)
Female gender	44/73 (59)	88/205 (43)	49/98(5)	31/56 (55)	94/211 (45)
Site					
EDH	14/74(19)	89/205 (43)	33/98 (34)	18/56 (32)	66/212 (31)
KTHC	34/74 (46)	58/205 (28)	25/98 (26)	16/56 (29)	96/212 (45)
Matikwana/ Mapulneng	17/74 (23)	57/205 (28)	10/98(10)	4/56 (7)	38/212 (18)
RMMCH/HJ	9/74 (12)	1/205 (1)	30/98 (31)	18/56 (32)	12/212 (6)
In hospital case fatality ratio	1/73(1)	4/205 (2)	5/98 (5)	0	18/206 (9)

RSV= Respiratory syncytial virus, PIV= parainfluenza virus, hMPV= human metapneumovirus

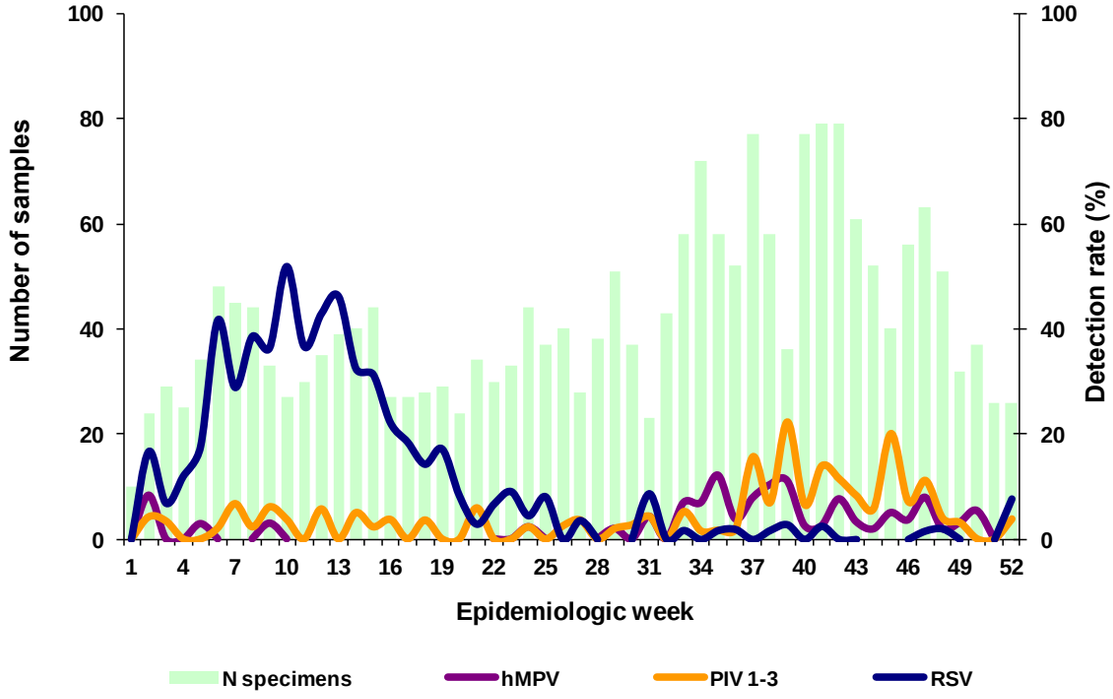


Figure 4: Numbers of samples collected and detection rates for respiratory syncytial virus (RSV), parainfluenza virus 1 -3 (PIV1-3) and human metapneumovirus (hMPV) in patients fitting the case definition for severe acute respiratory illness (SARI) or severe chronic respiratory virus (SCRI), South Africa, 2014.

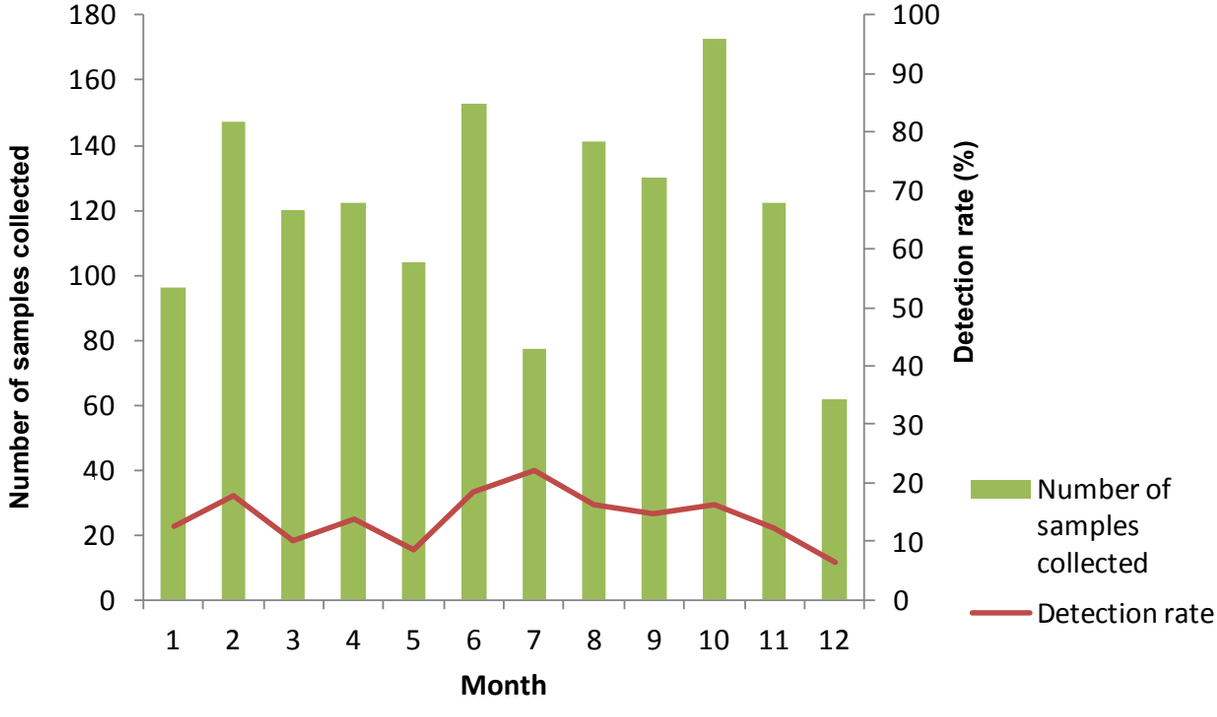


Figure 5: Numbers of blood samples collected and detection rate of *Streptococcus pneumoniae* from patients with severe acute respiratory illness (SARI) and severe chronic respiratory illness (SCRI) by month, South Africa, 2014.

Bacterial pathogens (SARI and SCRI)

Of the 1667 respiratory samples tested for *B. pertussis*, 46 tested positive (3%), 14/1662 (1%) for *M. pneumoniae*, 3/1662 (0.2%) for *C. pneumoniae* and 1/1661 (0.06%) for *Legionella spp*. The highest number of positive samples for *B. pertussis* and *M. pneumoniae* were in the <5 year and 25 to 44 year age groups (table 5). Similarly, the highest number of cases for *C. pneumonia* was in the <5 year age group. There was no clear seasonality for any of the bacterial pathogens (figure 6).

Table 5: Numbers of samples collected, detection rate and characteristics of patients fitting the severe acute respiratory illness (SARI) and the severe chronic respiratory illness (SCRI) case definition who were tested for *Streptococcus pneumoniae, Bordetella pertussis, Mycoplasma pneumoniae, Chlamydia pneumoniae and Legionella spp,* pneumonia surveillance, South Africa, 2014.

	B. pertussis **n/N (%)**	***M. pneumoniae*** **n/N (%)**	***C. pneumoniae*** **n/N (%)**	***Legionella spp*** **n/N (%)**
Age group, years				
0-4	13/45 (29)	5/14 (36)	2/3 (67)	0
5-14	2/45 (4)	1/14 (7)	0	0
15-24	2/45 (4)	2/14 (14)	0	1/1 (100)
25-44	13/45 (29)	5.14 (36)	1/3 (33)	0
45-64	11/45 (24)	1/14 (7)	0	0
≥ 65	4/45 (9)	0	0	0
Female gender	23/46 (50)	5/14 (36)	0/3	0/1
Site				
EDH	14/46 (30)	5/14 (36)	1/3 (33)	0/1
KTHC	30/46 (65)	7/14 950)	2/3 (67)	1/1 (100)
RMMCH/HJ	2/46 (4)	2/14 (14)	0/3	0/1

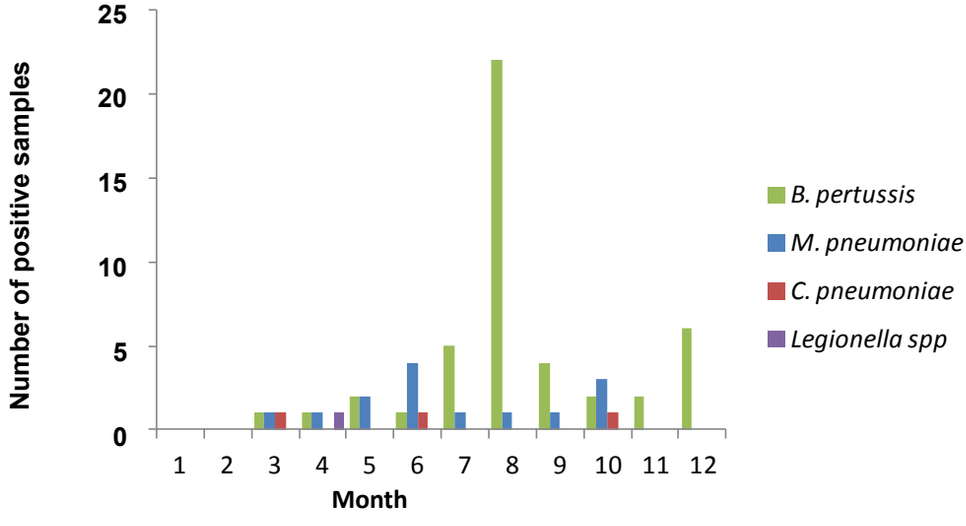


Figure 6: Numbers of positive samples of *Bordetella pertussis*, *Mycoplasma pneumoniae*, *Legionella spp* and *Chlamydia pneumonia* among patients with severe acute respiratory illness (SARI) and severe chronic respiratory illness (SCRI) by month, South Africa, 2014.

Tuberculosis and Pneumocystis jirovecii (PCP)

Of the 945 samples tested for TB, 195 (24%) were positive. Tuberculosis and PCP had no obvious seasonality (figures 7 and 8). The majority of samples that tested positive for tuberculosis were collected at the KTHC site (138/195, 71%) and were in the 25 to 44 year age group (113/223, 51%) (table 6).

Of the 266 samples that tested positive for PCP, 108 (41%) were from nasophyngeal samples, 34 (13%) were oral washes and 124 (48%) were from sputum. Half the patients with positive samples were in the age group 25 to 44 years (133/266, 50%) and the majority was from KTHC 176/266 (66%).

Table 6: Detection rate and characteristics of patients fitting the case definition of severe respiratory illness enrolled into pneumonia surveillance and testing positive for tuberculosis and *Pneumocystis jirovecii*, South Africa, 2014.

	Tuberculosis n/N(%)	***Pneumocystis jirovecii* n/N(%)**
Age group, years		
0-4	11/195 (6)	61/266 (23)
5-14	2/195 (1)	2/266 (1)
15-24	24/195 (12)	10/266 (4)
25-44	113/195 (58)	133/266 (50)
45-64	44/195 (23)	49/266 (18)
≥ 65	4/195 (2)	11/266 (4)
Female gender	111/195 (57)	145/266 (60)
Site		
EDH	57/195 (29)	90 (34)
KTHC	138/195 (71)	176 (66)

EDH= Edendale Hospital, KTHC= Klerksdorp-Tshepong hospital complex

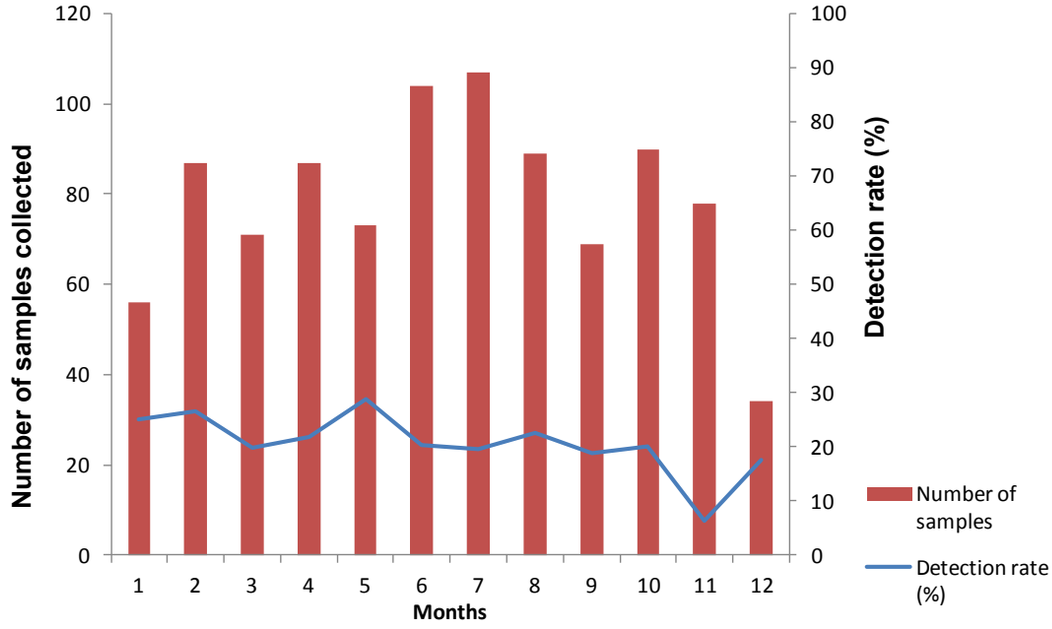


Figure 7: Numbers of samples collected for tuberculosis testing and TB detection rate for patients fitting the severe acute respiratory illness (SRI) case definition, pneumonia surveillance, South Africa, 2014.

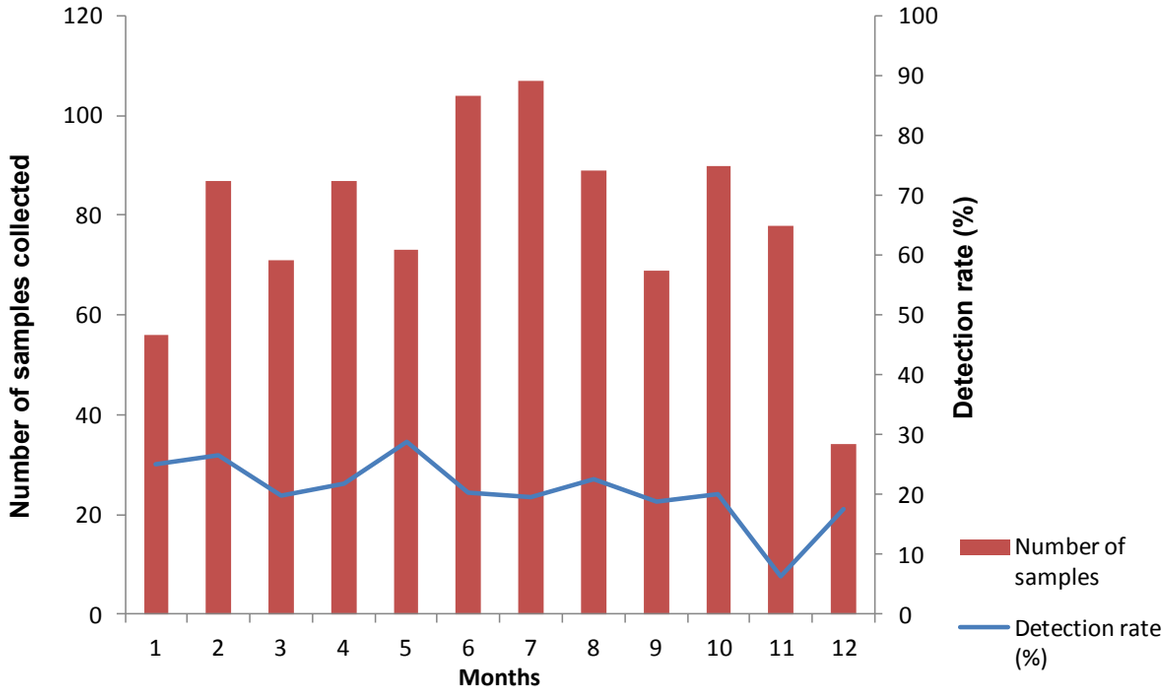


Figure 7: Numbers of samples collected for tuberculosis testing and TB detection rate for patients fitting the severe acute respiratory illness (SRI) case definition, pneumonia surveillance, South Africa, 2014.

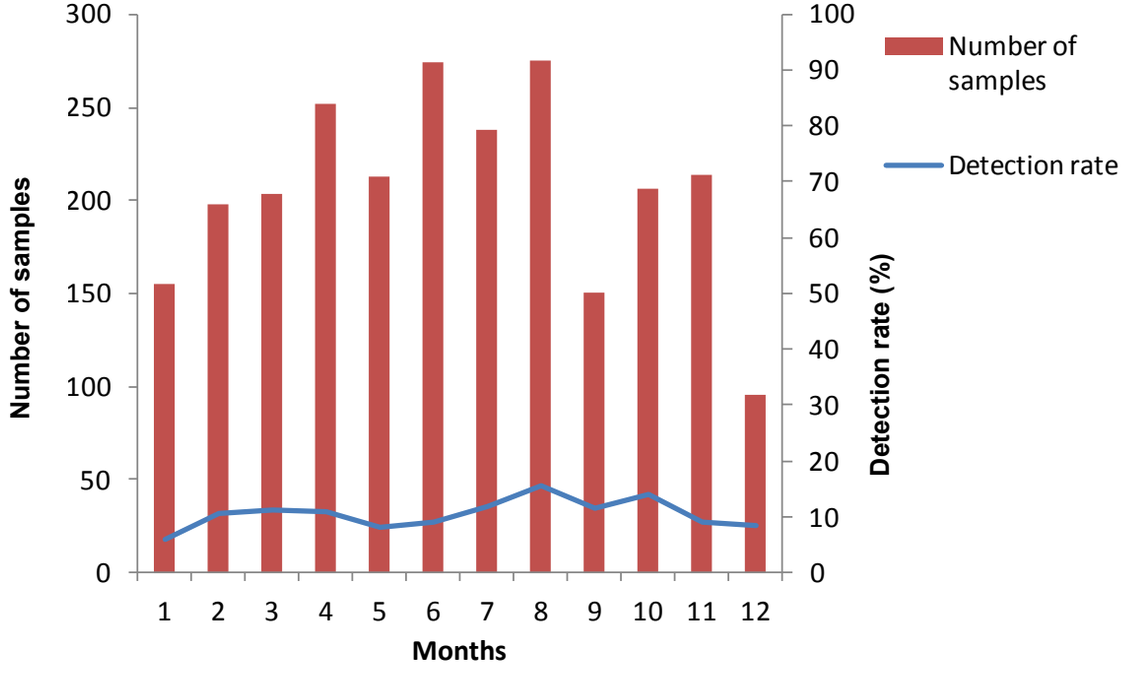


Figure 8: Numbers of samples collected for *Pneumocystis jirovecii* and detection rate for patients fitting the severe respiratory illness (SRI) case definition, pneumonia surveillance, South Africa, 2014.

Systematic ILI surveillance at public health clinics

Influenza and viral pathogens

During 2014, 1585 patients with ILI were enrolled at the two clinics and 1558 (98%) upper respiratory samples were tested. The overall detection rate for influenza was 13% (n=202). Excluding non-subtyped samples, 80% (156/195) were influenza A(H3N2), 15% (30/195) were influenza B and 5% (9/195) were influenza A(H1N1) pdm09. There were no dual infections. Influenza positive samples were detected from week 19. The detection rate reached 10% in week 24 and remained above 10% until week 38 (figure 9).

Of the 1557 samples tested, 71 (5%) tested positive for parainfluenza 1-3, 63 (4%) for RSV and 49 (3%) for human metapneumovirus. Only RSV demonstrated a defined seasonality which preceded the influenza season. The detection rate for RSV rose above 10% in week 5, peaked in week 12 and remained above 10% until week 17 (figure 10). In the group of patients with ILI: 1% (16/1463) tested positive for *B. pertussis,* 0.5% (7/1460) for *M. pneumoniae* and 0.5% (7/1460) for *C. pneumonia.* The highest numbers of positive samples for *B. pertussis* were in the <5 and 25 to 44 year age groups. Similarly, in *C. pneumonia* the highest number of cases was in the <5 year age group. The highest number of cases of *M. pneumonia* was in the 5 to 14 year age group (table 7). There were no positive tests for *Legionella spp.* There was no seasonality for bacterial pathogens (figure 11).

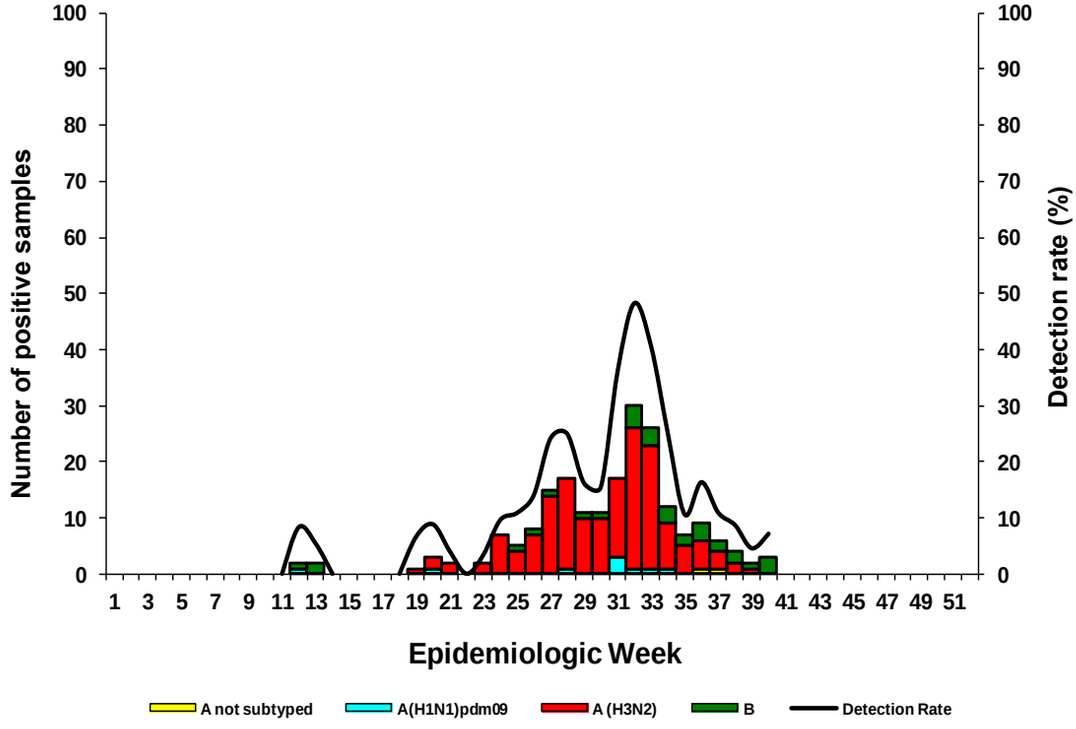


Figure 9: Influenza detection rate by subtype and week in patients with influenza-like illness (ILI) at public health clinics at two sites, South Africa, 2014.

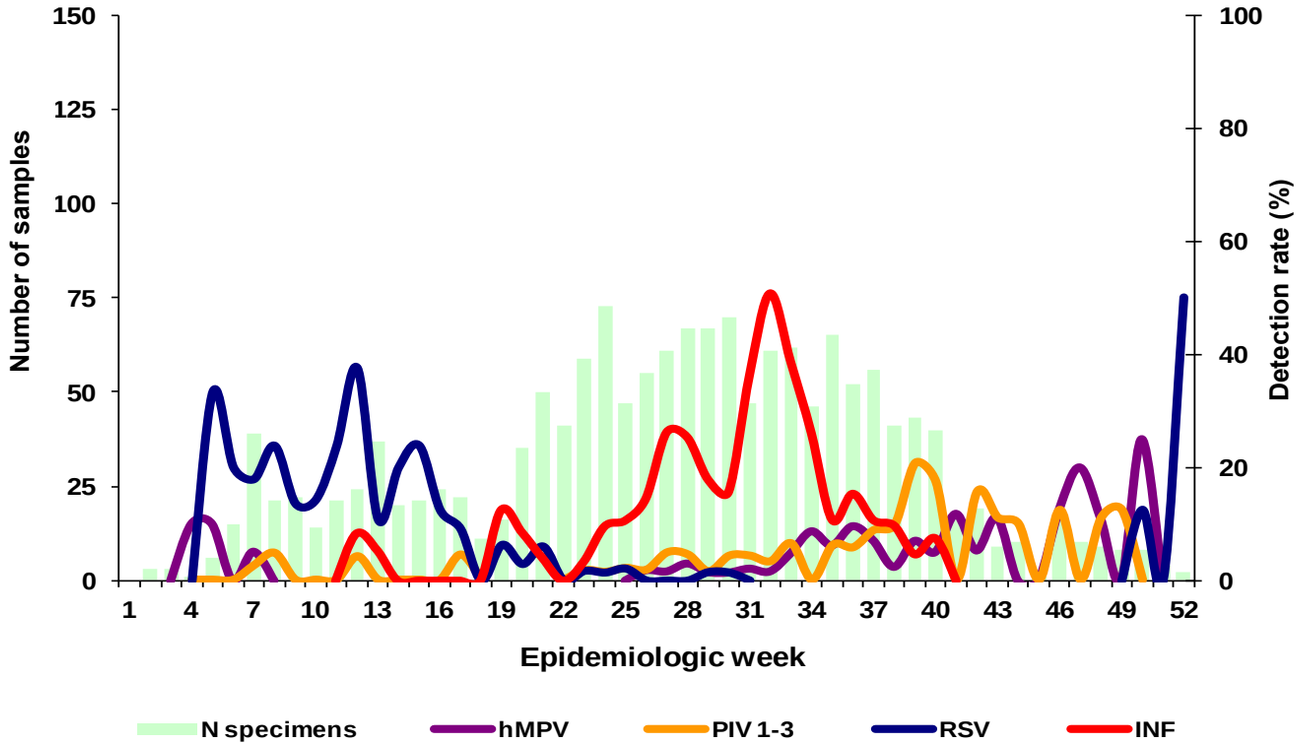


Figure 10: Detection rates of human metapneumonvirus (hMPV), parainfluenza (PIV) 1-3, respiratory syncytial virus (RSV) and influenza by week in patients fitting the influenza-like illness (ILI) case definition at public health clinics, South Africa, 2014.

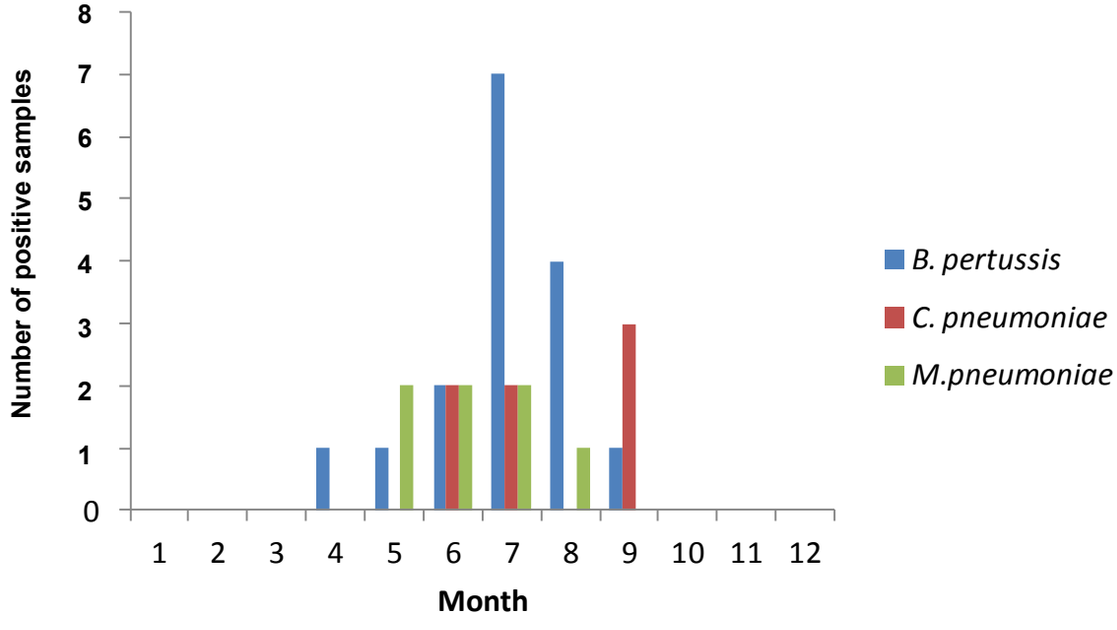


Figure 11: Numbers of positive samples of bacterial pathogens by week and pathogen, in patients fitting the influenza-like illness (ILI) case definition, South Africa, 2014.

Table 7: Detection rate and characteristics of patients with influenza-like illness who were tested for bacterial pathogens at public health clinics at two sites, South Africa, 2014.

	Bordetella pertussis **n/N (%)**	***Mycoplasma pneumoniae*** **n/N (%)**	***Chlamydia pneumoniae*** **n/N (%)**
Age group, years			
0-4	4/16 (25)	2/7 (29)	4/7 (57)
5-14	5/16 (31)	3/7 (43)	3/7 (43)
15-24	3/16 (19)	0/7	0
25-44	4/16 (25)	1 (14)	0
45-64	0/16	0 /7	0
≥ 65	0/16	1/7 (14)	0
Female gender	9/16 (56)	5/7(71)	6/7(86)
Site			
Edendale Gateway clinic	6/16 (38)	5/7(71)	5/7(71)
Jouberton clinic	10/16 (62)	2/7(29)	2/7 (29)

Additional surveillance activities

Viral watch (VW)

In 2014, 117 general practitioners across South Africa's 9 provinces participated in the VW programme. A total of 1054 samples was tested for influenza; of these 515 (49%) tested positive for influenza. The season was dominated by influenza A(H3N2) in which 351/515 (68%) of samples tested positive for influenza A(H3N2), 19% tested positive for influenza B and 12% (64/515) tested positive for influenza A(H1N1)pdm09. The season started in week 22 and the detection rate remained above 10% until week 41 (figure 12).

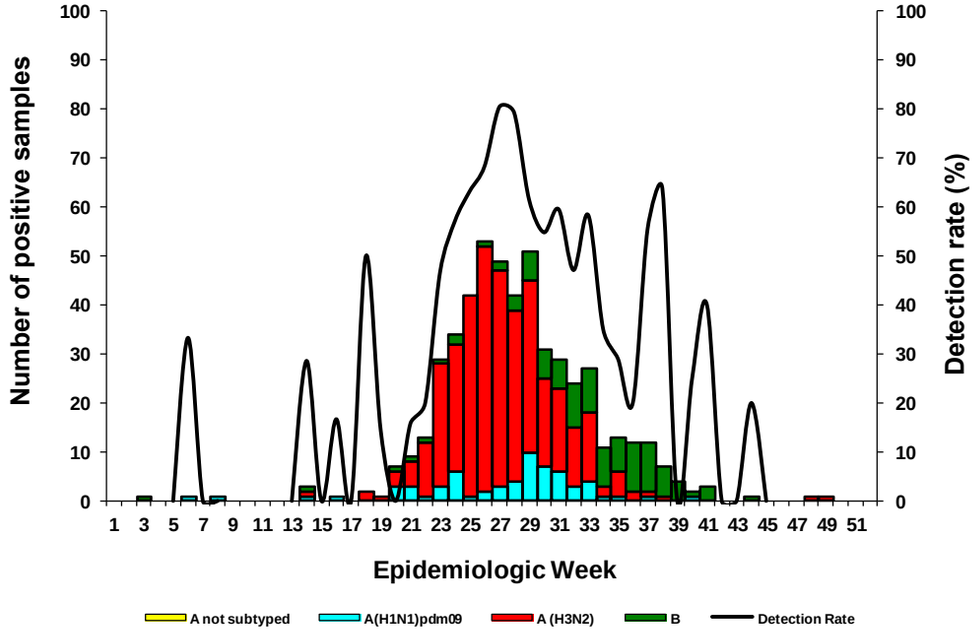


Figure 12: Numbers of samples and influenza detection rates by viral subtype and week in patients fitting the case definition of ILI enrolled into the Viral Watch programme, South Africa, 2014.

Respiratory Morbidity Surveillance

In order to describe the influence of the influenza season on the number of pneumonia and influenza hospitalizations, the NICD reviewed anonymized data from a private hospital group. The numbers of hospitalizations for pneumonia and influenza during the influenza season were compared to those for the periods preceding and following the season. During 2014 there were 672 598 consultations reported to the NICD through the respiratory morbidity data mining surveillance system. Of these, 29 558 (4%) were due to pneumonia or influenza (P&I) (International Classification of Diseases 10 codes J10-18). There were 22 059 (75%) inpatients and 7499 (25%) outpatients with P&I discharge data.

An increase in P&I consultations and admission was observed during the period with a higher number of seasonal influenza virus isolations reported to the Viral Watch, ILI and SARI surveillance programmes respectively (figures 13, 14, and cross reference figure 3; ILI influenza figure 9; ILI Viral Watch influenza figure 12). A second lower peak preceded the influenza season, corresponding to the circulation of respiratory syncytial virus (figures 13, 14 and cross reference figure 4; SARI viruses, figure 10; ILI viruses).

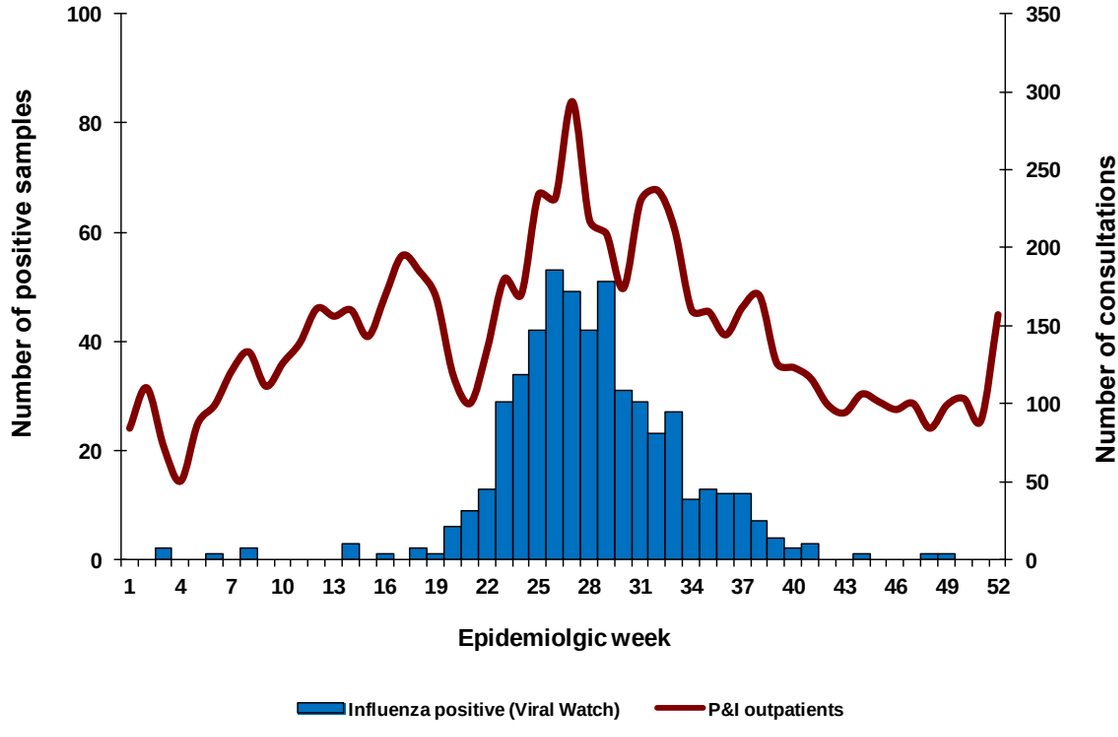


Figure 13: Numbers of private hospital outpatient consultations with a discharge diagnosis of pneumonia and influenza (P&I), and numbers of influenza positive viral isolates (Viral Watch) by week, South Africa, 2014.

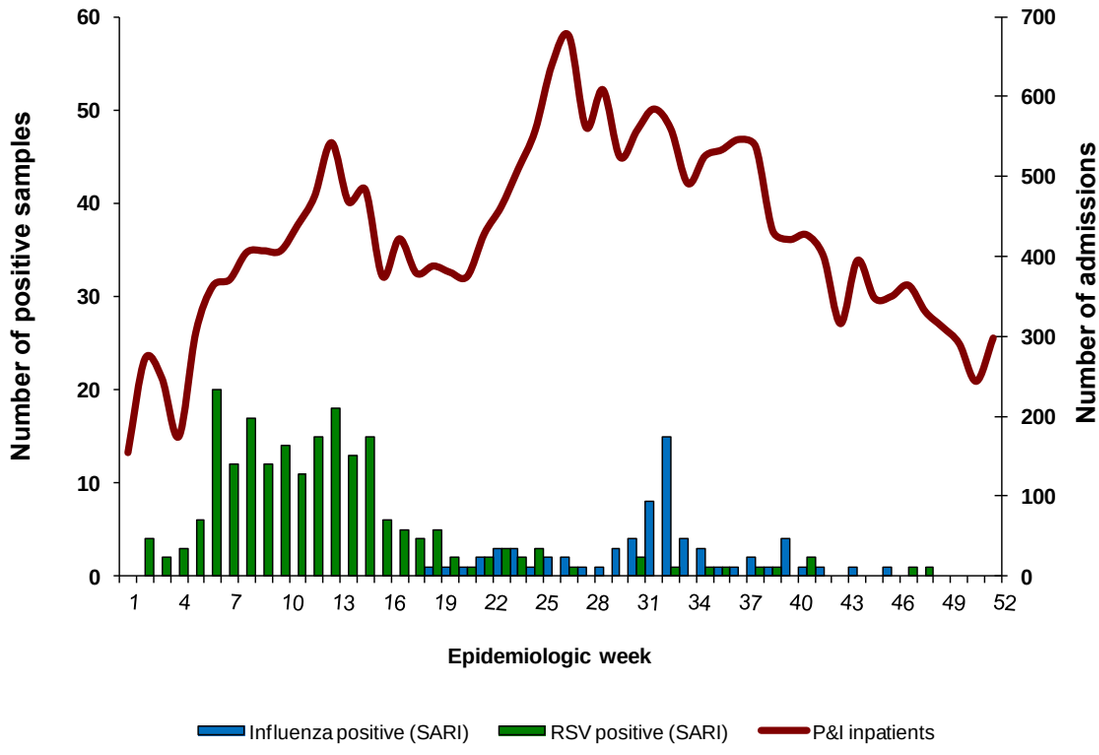


Figure 14: Numbers of admissions for pneumonia and influenza, as well as numbers of influenza positive viral isolates (Viral watch) and respiratory syncytial virus (RSV) positive isolates (SARI) by week, South Africa, 2014.

Molecular characterizations of influenza

During the influenza season of 2014, influenza virus isolation was attempted on clinical samples that tested positive for influenza on a real time multiplex PCR assay with a crossing point value of 30 or less. Madin-Darby Canine Kidney (MDCK) cells were used for virus isolations and about 67% (50/75) were successful. Of the 50 influenza virus isolations obtained, 43 were from influenza A viruses and 7 from influenza B viruses. The majority of influenza A virus isolates (n= 35) were influenza A/H3N2 which dominated the season. Compared to the 3 previous seasons this represents a great success in the isolation of H3N2 viruses in cell cultures. Changes in the phenotypic characteristics of H3N2 viruses were reported by the WHO Collaborating Centres. The embryonic egg isolations attempted were not successful. On investigating the failure of isolates, it was established that the collaborating centres recently used slightly older chicken embryos than normally used.

Antigenic characterization of influenza virus isolates

Turkey red blood cells were used in the hemagglutination and hemagglutination inhibition assays to determine the antigenic reactivity of the influenza virus isolates. A total of 46 virus isolates could be characterised antigenically by hemagglutination inhibition assay (HAI) of which 70 % (32/46) were influenza A(H3N2). Of the influenza A(H3N2) viruses serotyped 69% (22/32) showed normal reactivity to the A/Texas/50/2012 vaccine strain reference antiserum. Five influenza A(H3N2) isolates reacted with a ≥2-fold lower titre than the control or reference antiserum, 3 reacted with a 4-fold lower titre and 2 with 8-fold lower titres. All seven A(H1N1)pdm09 isolates showed normal reactivity to the corresponding reference antiserum. Seven influenza B virus isolates were characterized for reactivity to reference antisera raised against vaccine or reference antigens. Five influenza B isolates reacted to the B/Yamagata lineage reference antisera of which 1

reacted with a 2-fold lower titre. Two reacted with similar titres as the reference B/Victoria lineage strain. However, these 2 samples were collected outside of the influenza season from individuals with travel history.

Genetic characterisation of influenza A (H3N2), A (H1N1)pdm09 and influenza B strains was carried out by sequencing and phylogenetic analysis of the hemagglutinin (HA) genes. Lineages were identified by specific amino acid mutations relative to a designated reference strain as described by the WHO Vaccine Consultation Meeting team.

Influenza A(H3N2)

H3N2 HA gene sequences were generated from 35 clinical specimens selected from the 2014 season for both the ILI and SARI surveillance programs. All 2014 strains are within the genetic group 3 of the seven lineages (WHO CC, London) identified, specifically subgroup 3C.3. Subgroup 3C is characterised by the following amino acid mutations: Q33R, N145S and N278K relative to A/Perth/16/2009 as reference. One strain is in the sub-subgroup 3C.2a identified in February 2014 as one of two emerging lineages with low reactivity to the current vaccine The 2014 vaccine strain A/Texas/50/2012 is in subgroup 3C.1 (CDC nomenclature). The deduced amino acid alignment showing amino acid mutations compared to the A/ Perth/16/2009 reference strain.

The neuraminidase (NA) gene was sequenced for 30 influenza A(H3N2) positive samples and 3/30 had the mutations Y155F and D251V which divide the subgroup 3C. Two sequences showed an additional mutation at position 315 (S>G). All viruses had the N402D mutation that results in the loss of a potential N-linked glycosylation site which, together with the S367N and K369T mutations, results in a shift in potential of the N-linked glycosylation site.

In the 2014 season the HA gene from only 5 influenza A (H1N1)pdm09 positive clinical samples was sequenced. Three of these samples were in subgroup 6B as compared to 2013 when it was in 6C. Subgroups 6B and 6C are characterised by the following mutations: D97N, S185T, S203T and K283E, E3474K, S451N and E499K in HA1 and 2. The amino acid mutations K163Q and A256T characterise subgroup 6B.

Influenza B

The HA1 region of the HA genes from a total of 7 clinical samples positive for influenza B was sequenced and characterised. No B/Victoria lineage strains were sequenced as those identified were from outside of South Africa's influenza season.

B/Yamagata lineage

Seven viruses sequenced belong to clade 2 of B/ Yamagata lineage viruses in reference to the B/ Florida/4/2006 strain. This is on the deduced amino acid alignments showing the identified amino acid changes and the following are characteristic of clade 2 to which the current vaccine strain belongs: R48K, P108A and T181A.

Discussion

The influenza season in South Africa in 2014 was predominated by influenza A(H3N2), followed by influenza B and influenza A(H1N1)pdm09. The season started in week 22 at the ILI sites but the detection rate in the SARI programme only remained constantly above 10% from week 31. In 2014 some changes to the SARI surveillance programme were made - in particular the change of surveillance site in Gauteng Province. As a result of this the total numbers of samples tested for 2014 were slightly lower than previous years.

Other commonly detected pathogens in patients with pneumonia were pneumococcus, tuberculosis, RSV and PCP. Atypical bacterial pathogens and *B. pertussis* were detected in <5% of individuals. However, an increase in

B. pertussis cases was observed in July and August of 2014, followed by a reduction in case numbers without any specific intervention. The increase was investigated and contamination was excluded. The increase likely reflects winter seasonality and disease periodicity which has been reported in some countries. [12,13] A full assessment of this observation is difficult as systematic surveillance for pertusiss has only been implemented since 2012 and baseline data on pertussis epidemiology from South Africa are limited.[14,15] Ongoing systematic pertussis surveillance is needed to provide robust baseline data on disease burden and epidemiology and to monitor for future increases in disease.

The most commonly identified pathogens in patients with ILI were influenza and RSV. A total of 115 A(H3N2) positive clinical samples, mainly from patients with ILI, were tested for the presence of the E119V mutation associated with reduced susceptibility to oseltamivir by real-time RT-PCR. Only the wild-type E119 variant was detected in all samples. Representative cell culture and egg isolates as well as clinical samples were sent to the WHO Collaborating Centres in London and Australia for further characterisation.

The CRDM is working towards comprehensive surveillance for the clinical syndromes of ILI and pneumonia. This is the first report to combine the viral pathogens with the additional testing for bacterial pathogens and some of the atypical causes of pneumonia in our setting. Additional work is being done on the interaction between these pathogens and the risk factors for severe disease which will assist clinicians and policy makers to improve health care and implement prevention strategies such as vaccines.

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References

1. Cohen C, Moyes J, Tempia S, Groom M, Walaza S, Pretorius M, Dawood H, Chagan M, Haffejee S, Variava E, Kahn K, Tshangela A, von Gottberg A, Wolter N, Cohen AL, Kgokong B, Venter M, Madhi SA. Severe influenza -associated respiratory infection in high HIV prevalence setting, South Africa, 2009-2011. *Emerg Infect Dis.* 2013; 19(11):1766-74.
2. McAnerney JM, Cohen C, Moyes J, Besselaar TG, Buys A, Schoub BD, Blumberg L. Twenty-five years of outpatient influenza surveillance in South Africa, 1984-2008. *J Infect Dis.* 2012; 15; 206 Suppl 1:S153-8
3. Surveillance for *Bordetella pertussis*, atypical bacterial causes of pneumonia, *Haemophilus influenzae* and *Streptococcus pneumoniae* within the Severe Respiratory Illness Surveillance Programme, 2013. *Communicable Diseases Surveillance Bulletin* 2014; 12: 135-146. November2014:http://www.nicd.ac.za/?page=communicable_diseases_surveillance_bulletin&id=45
4. Centers for Disease Control and Prevention (CDC). CDC real-time RT-PCR (rRT-PCR) protocol for detection and characterization of influenza 2009 A(H1N1)pdm virus—RUO International. Atlanta,
5. Respiratory virus surveillance report. *Communicable Diseases Surveillance Bulletin* 2014; 12: 9–18. http://www.nicd.ac.za/assets/files/CommDisBull%2012(1)-April%202014_Fin.pdf
6. Thurman KA, Warner AK, Cowart KC, Benitez AJ, Winchell JM. Detection of *Mycoplasma pneumoniae*, *Chlamydia pneumoniae* and *Legionella* spp. in clinical specimens using a single-tube multiplex real-time PCR assay. *Diagn Microbiol Infect Dis* 2011; 70: 1-9.
7. Tatti KM, Sparks KN, Boney KO, Tondella ML. Novel multitarget real-time PCR assay for rapid detection of *Bordetella* species in clinical specimens. *J Clin Microbiol* 2011; 49: 4059-66.
8. Carvalho M, Tondella ML, McCaustland K et al. Evaluation and improvement of real-time PCR assays targeting lytA, ply, and psaA genes for detection of pneumococcal DNA. *J Clin Microbiol* 2007; 45: 2460-6.
9. http://www.cepheid.com/administrator/components/com_productcatalog/library-files/2cc115cd6ba267207c04661a3d0fcfc8-Xpert-MTB-RIF-ENGLISH-Package-Insert-301-1404-Rev-B-February-2015.pdf
10. http://www.hain-lifescience.de/en/products/microbiology/mycobacteria/genotype-mtbdrplus.html
11. Dini L, Du Plessis M, Frean J, Fernandez V. High prevalence of dihydropteroate synthase mutations in *Pneumocystis jirovecii* isolated from patients with Pneumocystis pneumonia in South Africa. *J Clin Microbiol* 2010;48: 2016-21.
12. Crowcroft NS, Peebody RG. Recent developments in pertussis. *Lancet* 2006 10;367(9526):1926-36.
13. http://www.cdc.gov/nip/publications/pink/pert.pdf
14. Hallbauer UM, Goosen Y, Pieters M. An outbreak of pertussis in Bloemfontein, South Africa, 2008-2009. *S Afr Fam Pract* 2011; 16;53(5):495-500.
15. Rensburg MA, Esser M, de Beer C. The seroprevalence of *Bordetella pertussis* antibodies in adolescents in the Western Cape. *South Afr J Epidemiol Infect* 2014; 28(4):202-6.

NATIONAL SURVEILLANCE OF TRANSMITTED HIV-1 DRUG RESISTANCE IN 2012

Gillian Hunt[1], Johanna Ledwaba[1], Monalisa Kalimashe[1], Anna Salimo[1], Siyabonga Cibane[1], Beverly Singh[1], Adrian Puren[1], Simbarashe Takuva[1], Natalie Exner Dean[2], Michael R. Jordan[3] & Lynn Morris[1]

[1]Centre for HIV and STIs, NICD
[2]Department of Biostatistics, College of Public Health & Health Professions, University of Florida, Gainesville FL, USA
[3]Tufts University School of Medicine, Boston MA, USA

Background

Sentinel site surveillance of transmitted HIV drug resistance (TDR) is recommended by the World Health Organization to assess levels of transmitted resistance among recently infected populations within a country or region. Results from the analysis of specimens collected as part of the 2012 National Antenatal Sentinel HIV & Herpes Simplex Type-2 Prevalence Survey in South Africa are reported.[1] In the survey, national HIV prevalence among antenatal women was estimated at 29.5%. Previously published TDR estimates from two provinces (Gauteng and KwaZulu-Natal) indicated that while TDR was low prior to 2008, these levels were moderate (>5%) in KwaZulu-Natal in 2009-2011.[2]

Methods

Remnant serum specimens were obtained from primagravid HIV-infected women <21 years of age attending any one of 1497 sentinel primary health care clinics in all 9 provinces (table 1). Genotyping was performed by sequencing the protease and reverse transcriptase genes using population sequencing methodologies. Mutations were interpreted using CPR v6.0 (http://cpr.stanford.edu/), which analyses data according to the 2009 Surveillance of Drug Resistance Mutations (SDRM) list.[3] For drug level testing (DLT), a semi-quantitative Liquid Chromatography MS/MS method was used to screen serum samples for the presence of various antiretroviral drugs (AZT, EFV, FTC, LPV, NVP, TNF). Specificity was ensured by the use of deuterated internal standards for all analytes. Point prevalence estimates of TDR levels with 95% CI were calculated using STATA. National estimates were weighted by number of eligible specimens in each province as a contribution to the total survey.

Table 1: Distribution of remnant serum specimens obtained by province for estimates of transmitted HIV drug resistance (TDR), South Africa, 2012.

Province	Total number of specimens collected in ANSUR 2012	Provincial contribution to ANSUR	Provincial HIV prevalence estimate	Number of eligible specimens	Number of specimens not available for testing	Number removed - unresolved phylogenetic linkage	Number removed - DLT positive	Number of specimens included in TDR analysis
Eastern Cape	4625	13.5%	29.1%	127	10	3	2	112
Free State	2325	6.8%	32.0%	75	3	0	1	71
Gauteng	6862	20.0%	29.9%	121	25	0	2	94
KwaZulu-Natal	7011	20.5%	37.4%	327	15	0	8	304
Limpopo	3579	10.4%	22.3%	56	14	0	0	42
Mpumalanga	2201	6.4%	35.6%	70	8	0	3	59
North West	2457	7.2%	29.7%	61	14	0	0	47
Northern Cape	1190	3.5%	17.8%	14	7	0	0	7
Western Cape	4010	11.7%	16.9%	35	1	0	0	34
National	34260	100.0%	29.5%	886	97	3	16	770

Results

A total of 886 specimens met inclusion criteria. From this, 97 had insufficient volume for testing. Of 789 available specimens, 542 were successfully amplified and sequenced. Three sequences were excluded due to unresolved phylogenetic linkage. Thirty nine sequences had SDRMs, of which 36 had sufficient volume to be tested for antiretroviral therapy drug levels, as well as a sample set of 77 specimens that were not amplifiable by PCR. Four (11%) specimens with SDRMs and 12 (16%) no-amps were positive for detection of drug levels, indicating recent drug exposure in these women. All 16 specimens positive for DLT were removed from further analysis.

From the remaining 770 specimens included, 532 (81%) were genotyped. The majority of specimens (99%) clustered with Subtype C; 5 specimens were found to be non-C. No regional clustering of sequences was noted.

National TDR point prevalence was estimated at 5.3% (95% CI 3.7 – 7.5%) for the non-nucleoside reverse transcriptase inhibitors (NNRTI) drug class, 1.1% (95% CI 0.5 – 2.4%) for nucleoside reverse transcriptase inhibitors (NRTI) and 0.6% (95% CI 0.1 – 1.6%) for protease inhibitors (PI) (table 2). Four provinces had NNRTI point prevalence estimates >5%, and 1 province had a PI point prevalence estimate of >5%, although most provincial analyses were limited by small sample sizes. The predominant mutations detected were K103N (57%), V106M (23%) and K101E (11%), whereas M184V/I was the predominant NRTI mutation (9%). Two specimens had dual NNRTI and NRTI class resistance.

Table 2: Point prevalence estimates of transmitted HIV drug resistance (TDR), South Africa, 2012.

Province	Number of specimens amplifiable by genotyping PCR	Genotyping amplification rate	Number of sequences with PI mutations	PI Point Prevalence (95% CI)	Number of sequences with NRTI mutations	NRTI Point Prevalence (95% CI)	Number of sequences with NNRTI mutations	NNRTI Point Prevalence
Eastern Cape	99	88.4%	0	0% (0 - 3.7)	0	0% (0 - 3.7)	3	3% (1.0 - 8.5)
Free State	54	76.1%	0	0% (0 - 6.6)	1	1.9% (0.3 - 9.8)	4	7.4% (2.9 - 17.6)
Gauteng	65	69.1%	1	1.5% (0.3 - 8.2)	0	0% (0 - 5.6)	6	9.2% (4.3 - 18.7)
KwaZulu-Natal	196	64.5%	0	0% (0 - 1.9)	4	2% (0.8 - 5.1)	8	4.1% (2.1 - 7.8)
Limpopo	20	47.6%	0	0% (0 - 16.1)	0	0% (0 - 16.1)	2	10% (2.8 - 30.1)
Mpumalanga	45	76.3%	0	0% (0 - 7.9)	1	2.2% (0.4 - 11.6)	2	4.4% (1.2 - 14.8)
North West	21	44.7%	2	9.5% (2.7 - 28.9)	0	0% (0 - 15.5)	1	4.8% (0.8 - 22.7)
Northern Cape	4	57.1%	0	0% (0 - 49.0)	0	0% (0 - 49.0)	0	0% (0 - 49.0)
Western Cape	28	82.4%	0	0% (0 - 12.1)	0	0% (0 - 12.1)	2	7.1% (2.0 - 22.6)
National	532	69.1%	3	0.6% (0.1 - 1.6)	6	1.1% (0.5 - 2.4)	28	5.3% (3.7 - 7.5)

NRTI = nucleoside reverse transcriptase inhibitors; NNRTI = non-nucleoside reverse transcriptase inhibitors; PI = protease inhibitors

Conclusion

These data provide the first national TDR estimate for South Africa. They indicate that levels of TDR are low to moderate for the NNRTI drug class and are low for NRTI and PI. The detection of antiretroviral drugs in women epidemiologically predicted to be recently infected suggests that screening criteria for inclusion in TDR surveys are less reliable in countries such as South Africa with a mature epidemic and an extensive ARV treatment program. Future analysis should include the use of testing algorithms for recent infection to exclude women who are chronically infected. Efforts should be intensified in-country to review sources of resistance including assessing early warning indicator data and monitor levels of acquired drug resistance in treated populations.

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References

1. The 2012 National Antenatal Sentinel HIV and Herpes Simplex Type-2 Prevalence Survey in South Africa, 2014. Department of Health: Pretoria.
2. Hunt G, et al., Surveillance of transmitted HIV drug resistance in five provinces in South Africa in 2011. Communicable Diseases Surveillance Bulletin 2013; 11:122.
3. Bennett DE, et al., Drug resistance mutations for surveillance of transmitted HIV-1 drug-resistance: 2009. PLoS One 2009; 4(3): p. e4724

ANTIMICROBIAL RESISTANCE SURVEILLANCE FROM SENTINEL PUBLIC HOSPITALS, SOUTH AFRICA, 2014

Olga Perovic,[1,2] Verushka Chetty[1] & Samantha Iyaloo[1]

[1] National Institute for Communicable Diseases, NHLS
[2]Department of Clinical Microbiology and Infectious Diseases, University of the Witwatersrand

Introduction

Antimicrobial resistance (AMR) is a key public health concern that threatens effective treatment of severe infections, both locally and globally. Surveillance is conducted to determine the extent and pattern of resistance amongst the most important pathogens causing infections in humans.[1] Integrated data on resistance in bacteria are obtained from electronic reports generated by public laboratories in South Africa. The objectives of the AMR surveillance programme are to determine the number of cases reported from selected hospitals by month for selected pathogens and to describe antimicrobial susceptibility to the most important treatment regimens by pathogen and by hospital.

Methods

All data for this report were sourced from the National Health Laboratory Service (NHLS) Corporate Data Warehouse (CDW). This is a national repository for laboratories serving all public health hospitals in South Africa and contains archived data from two laboratory information systems (LISs), either DISA or TrakCare.[2]

Bloodstream infections over the period January-December 2014 were extracted for the following

pathogens: *Acinetobacter baumannii* complex, *Enterobacter cloacae* complex, *Escherichia coli, Enterococcus faecalis, Enterococcus faecium, Klebsiella pneumoniae, Pseudomonas aeruginosa* and *Staphylococcus aureus.* Routine electronic data were collected from sentinel sites (mostly tertiary academic hospitals) (table 1).

Due to two different LISs, each with its own coding system for organisms and antibiotics, as well as a lack of standardisation across NHLS laboratories on how data were captured, extensive cleaning and recoding of data was necessary. Cleaning of the data involved creating unique patient identifiers, which enabled de-duplication and the generation of patient-level data.

Antimicrobial susceptibility reporting was based on Clinical Laboratory Standards Institute (CLSI) guidelines.[3] The various laboratory methods used included Microscan, Vitek, E test and disk diffusion. Vancomycin resistance is not reported for *Staphylococcus aureus* due to the lack of confirmatory test methods (pending agreement with the South African Society for Clinical Microbiology (SASCM)). Data were omitted for those sites that tested fewer than 30 organisms for a particular antibiotic.

Table 1: Hospitals participating in antimicrobial resistance surveillance by province, South Africa, and their characteristics.

Hospital Site	Province	Academic Hospital	No of beds
Charlotte Maxeke Johannesburg Academic Hospital (CMJAH)	Gauteng	Yes	1088
Chris Hani Baragwanath Hospital (CHBH)	Gauteng	Yes	3200
Dr George Mukhari Hospital (DGMH)	Gauteng	Yes	1200
Grey's Hospital (GH)	KwaZulu-Natal	Yes	530
Groote Schuur Hospital (GSH)	Western Cape	Yes	893
Helen Joseph Hospital (HJH)	Gauteng	Yes	700
Inkosi Albert Luthuli Central Hospital (IALCH)	KwaZulu-Natal	Yes	846
King Edward VIII Hospital (KEH)	KwaZulu-Natal	Yes	922
Mahatma Gandhi Hospital (MGH)*	KwaZulu-Natal	No	350
Nelson Mandela Academic Hospital/Mthatha Tertiary (NMAH)	Eastern Cape	Yes	520
RK Khan Hospital (RKKH)	KwaZulu-Natal	No	543
Steve Biko Academic Hospital (SBAH)	Gauteng	Yes	832
Tygerberg Hospital (TH)	Western Cape	Yes	1310
Universitas Hospital (UH)	Free State	Yes	650

Results

Data for bloodstream infections and antimicrobial susceptibility tests are summarised for *Acinetobacter baumannii* complex (figure 1), *Enterobacter cloacae* complex (figure 2), *Enterococcus faecalis* (figure 3), *Enterococcus faecium* (figure 4), *Escherichia coli (*figure 5*), Klebsiella pneumoniae* (figure 6), *Pseudomonas aeruginosa* (figure 7) and *Staphylococcus aureus* (figure 8). For each organism, the total number of cases, as well as their susceptibility profiles and percentage susceptibility to selected antimicrobial agents by site were analysed (figures 1-8).

***Acinetobacter baumannii* complex**

Acinetobacter baumannii was resistant to most of the antimicrobial agents tested. This is due to its ability to harbour multiple mechanisms of resistance, such as the loss of outer membrane porins resulting in reduced permeability, efflux systems, *ampC* β-lactamases and others. The proportions of isolates resistant to imipenem, cefepime and ceftazidime were high at 77%, 79% and 75% respectively, whereas resistance proportions were 67% to ciprofloxacin, 43% to amikacin and 51% to tobramycin. Resistance to colistin was estimated to be 5%. Resistance to most agents have not changed in comparison with the previous year, except for the increase in resistance to tobramycin and colistin. AST testing and breakpoints for colistin are lacking and these results should be treated with caution.

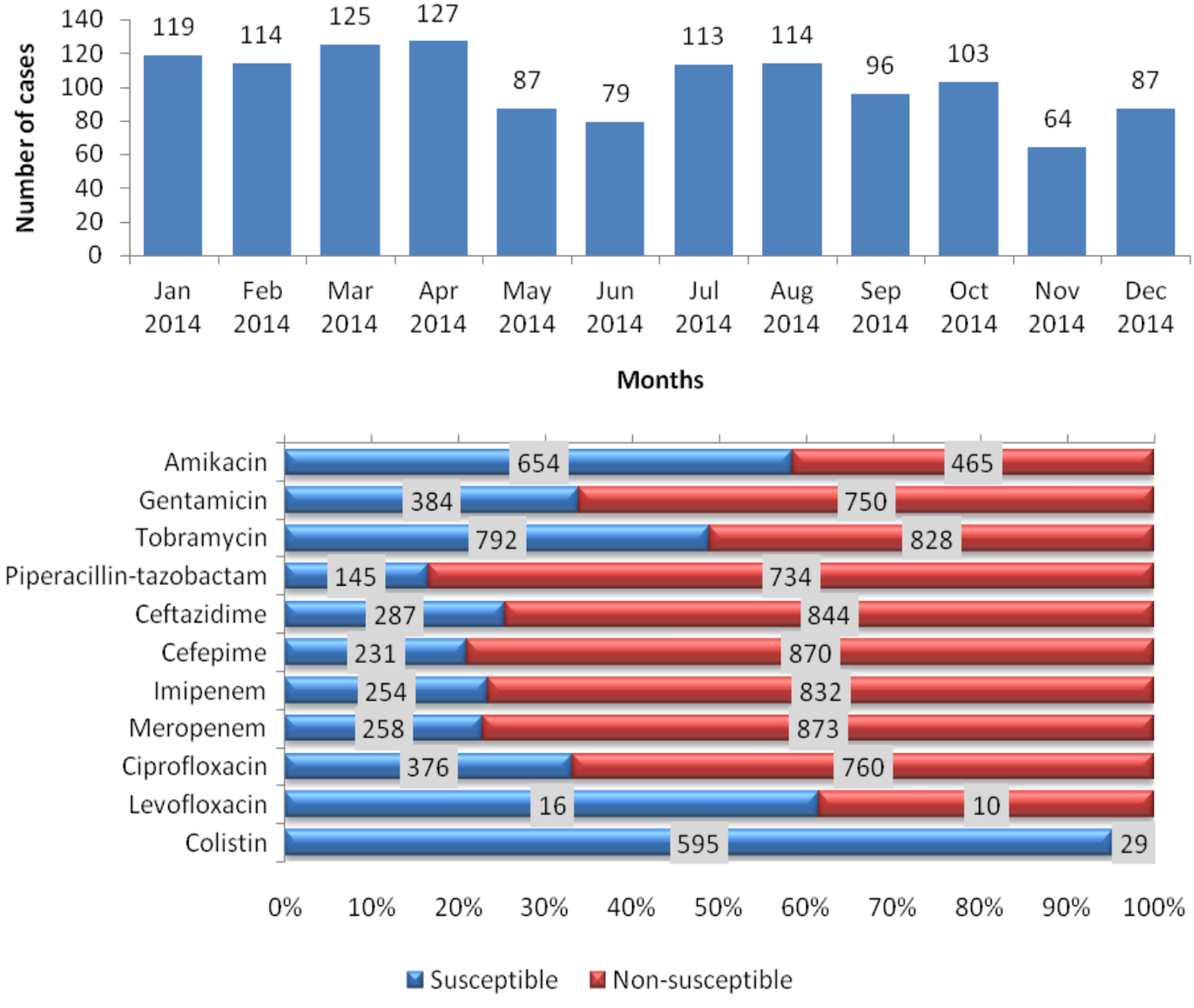


Figure 1: *Acinetobacter baumannii* cases by month, and numbers and percentages of susceptible and resistant *A. baumannii* complex isolates from blood cultures at public-sector sentinel sites, 2014. Total number of isolates analyzed =1228.

***Enterobacter cloacae* complex**

The high level of *Enterobacter cloacae* complex isolates resistant to ertapenem (12%) should be taken with reservation (refer to the limitations discussed earlier), although resistance to imipenem and meropenem has remained stable (2%). Resistance to cefepime (35%) is indicative of *ampC* β-lactamase hyper-production in combination with porin loss, which may confer resistance to cephalosporins.

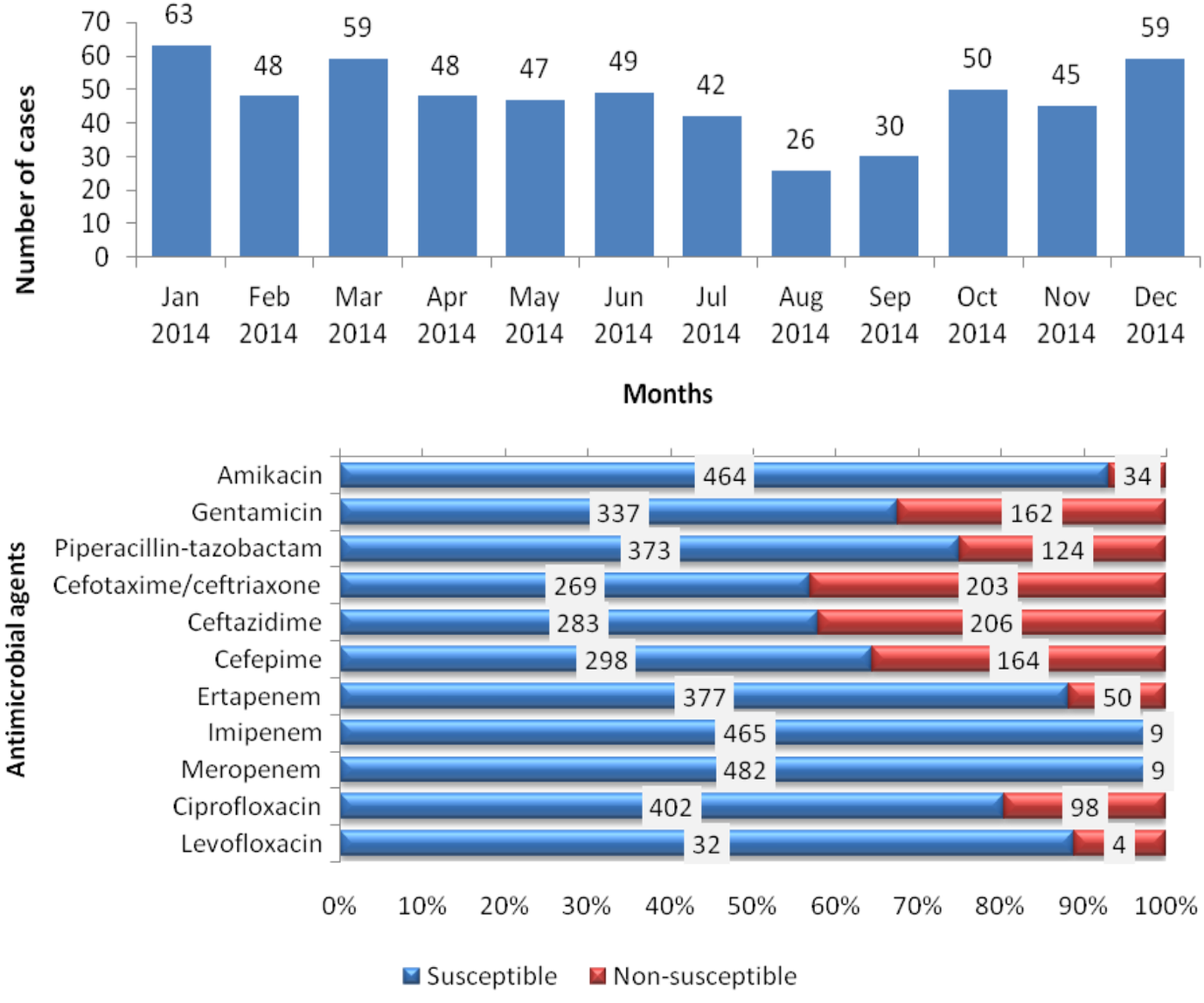


Figure 2: *Enterobacter cloacae* cases by month, and numbers and percentages of susceptible and resistant *E. cloacae* complex isolates from blood cultures at public-sector sentinel sites, 2014. Total number of isolates analyzed = 566.

Enterococcus faecalis

Enterococcus faecalis exhibited 17% resistance to penicillins and 2% (non-confirmed) resistance to vancomycin. There were no significant changes in comparison to the previous year. Results obtained from phenotypic methods for linezolid-intermediate or resistant *Enterococcus* spp. should be interpreted with caution since the gold standard for confirmation and quantification of linezolid resistance in enterocci is detection of the G2576T mutation.

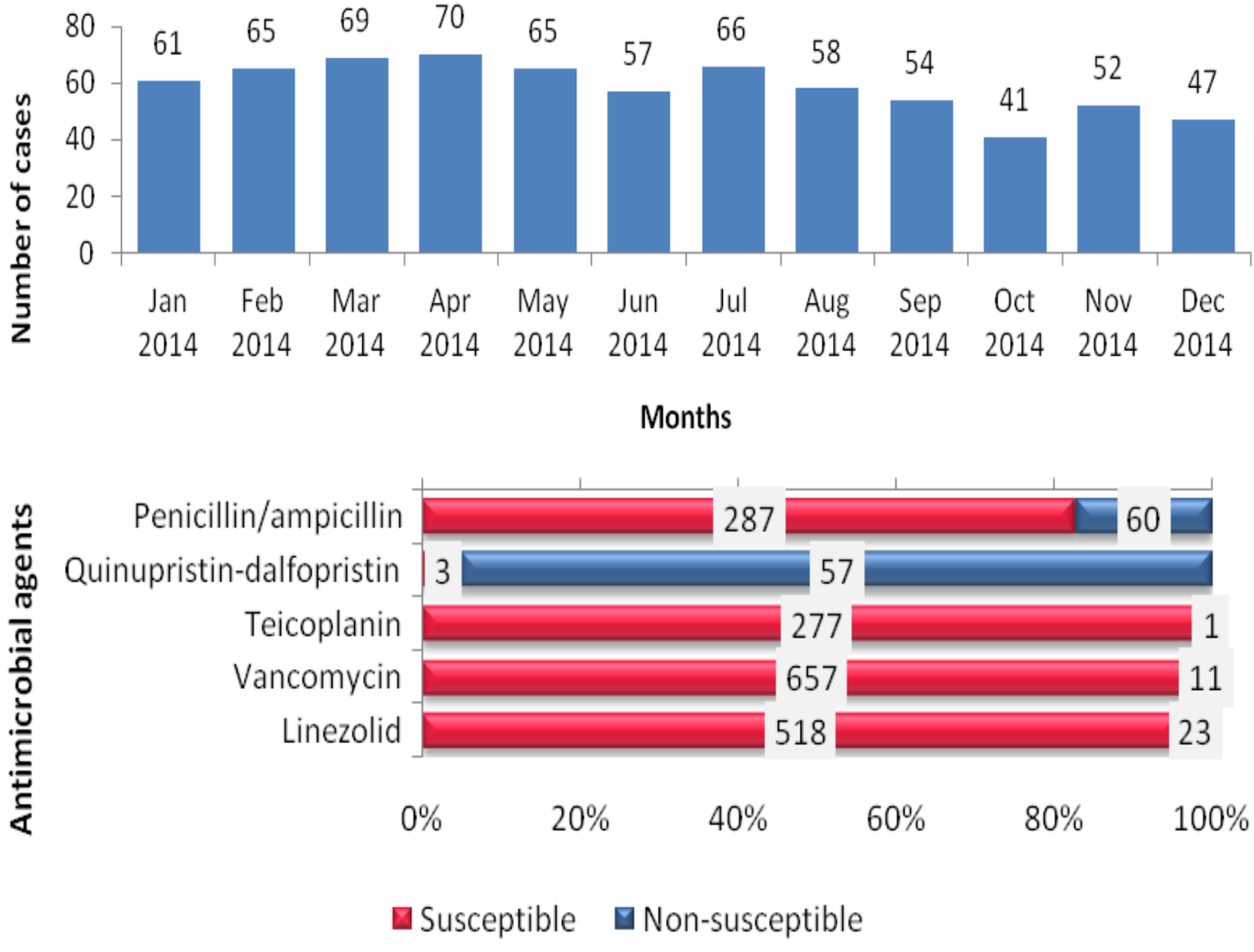


Figure 3: *Enterococcus faecalis* cases by month, and numbers and percentages of susceptible and resistant *E. faecalis* isolates from blood cultures at public-sector sentinel sites, 2014. Total number of isolates analyzed = 705.

Enterococcus faecium

Enterococcus faecium is inherently resistant to β-lactam agents. There was a decrease in resistance to vancomycin from 13% in 2013 to 5% in 2014, which may be explained by the containment of outbreaks in a few hospitals.

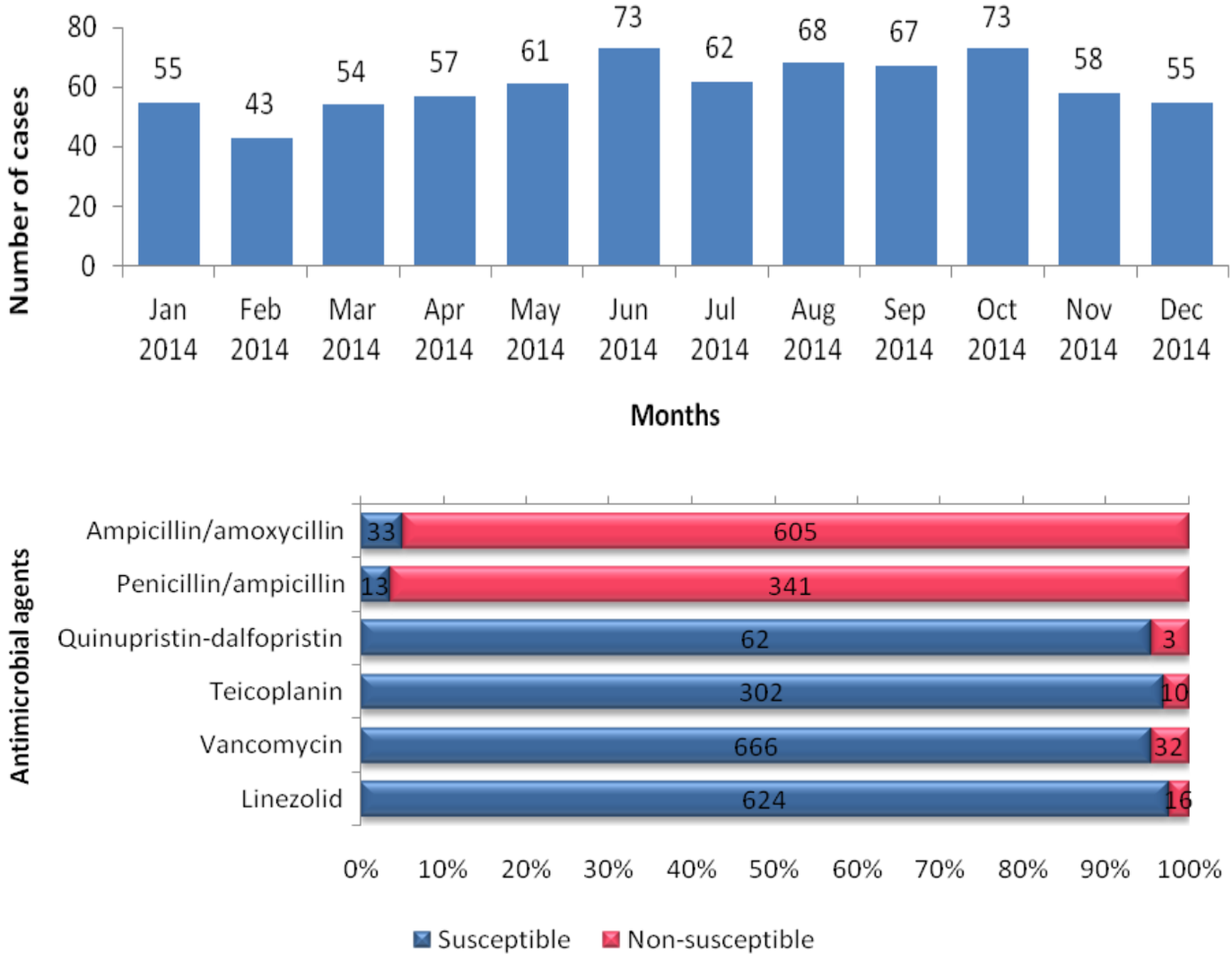


Figure 4: *Enterococcus faecium* cases by month, and numbers and percentages of susceptible and resistant *E. faecium* isolates from blood cultures at public-sector sentinel sites, 2014. Total number of isolates analyzed = 726.

Escherichia coli

Escherichia coli showed a minor increase in resistance to almost all β-lactams, whereas no change in resistance to ciprofloxacin over a two-year period was noted. Resistance to 3rd generation cephalosporins indicates the presence of extended spectrum β-lactamases (ESBLs) and was recorded in 25% of all isolates.

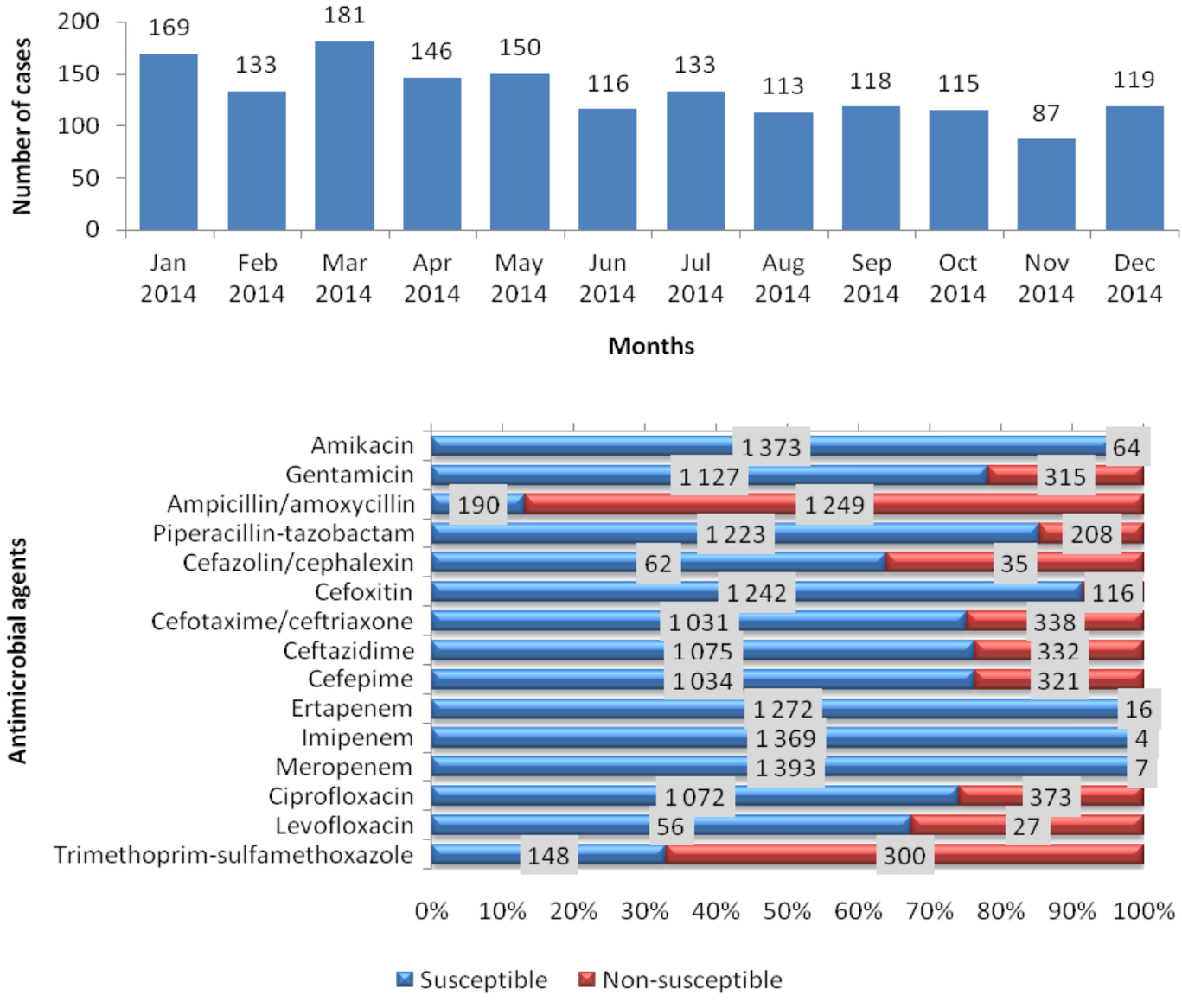


Figure 5: *Escherichia coli* cases by month, and numbers and percentages of susceptible and resistant *E. coli* isolates from blood cultures at public-sector sentinel sites, 2014. Total number of isolates analyzed = 1580.

Klebsiella pneumoniae

Klebsiella pneumoniae was resistant to multiple antimicrobials, including 3rd and 4th generation cephalosporinsthat indicate production of ESBLs (70%), ciprofloxacin (39%) and piperacillin-tazobactam (48%). The proportion of isolates resistant to ertapenem was low. Although resistance to other carbapenems was low, the rapid emergence of strains with carbapenemase production threatens the efficacy and use of this class of antimicrobials as a therapeutic option. Thus, continuous monitoring of resistance needs to be implemented. In hospitals where resistance is ≥10%, a nosocomial outbreak should be considered and investigated.

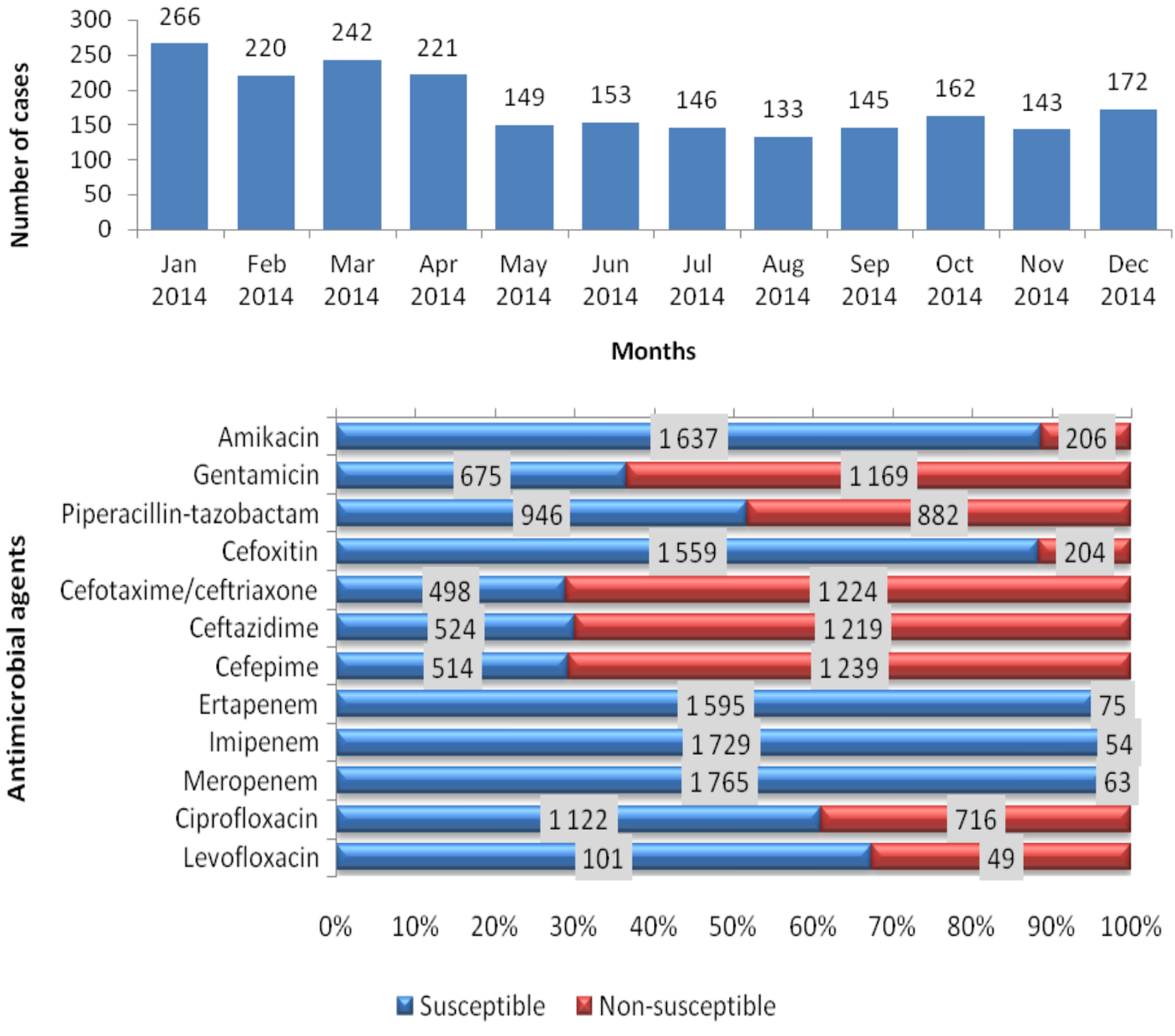


Figure 6: *Klebsiella pneumoniae* cases by month, and numbers and percentages of susceptible and resistant *K. pneumoniae* isolates from blood cultures at public-sector sentinel sites, 2014. Total number of isolates analyzed = 2152.

Pseudomonas aeruginosa

Compared to *A. baumannii, Pseudomonas aeruginosa* isolates displayed greater susceptibility to the antimicrobial agents tested. Resistance to piperacillin-tazobactam was high at 33%. There appeared to be modest decrease in resistance in 2014 compared to 2013 for the majority of antimicrobial agents, which may be explained by reasons listed in the limitations. Colistin resistance was the lowest.

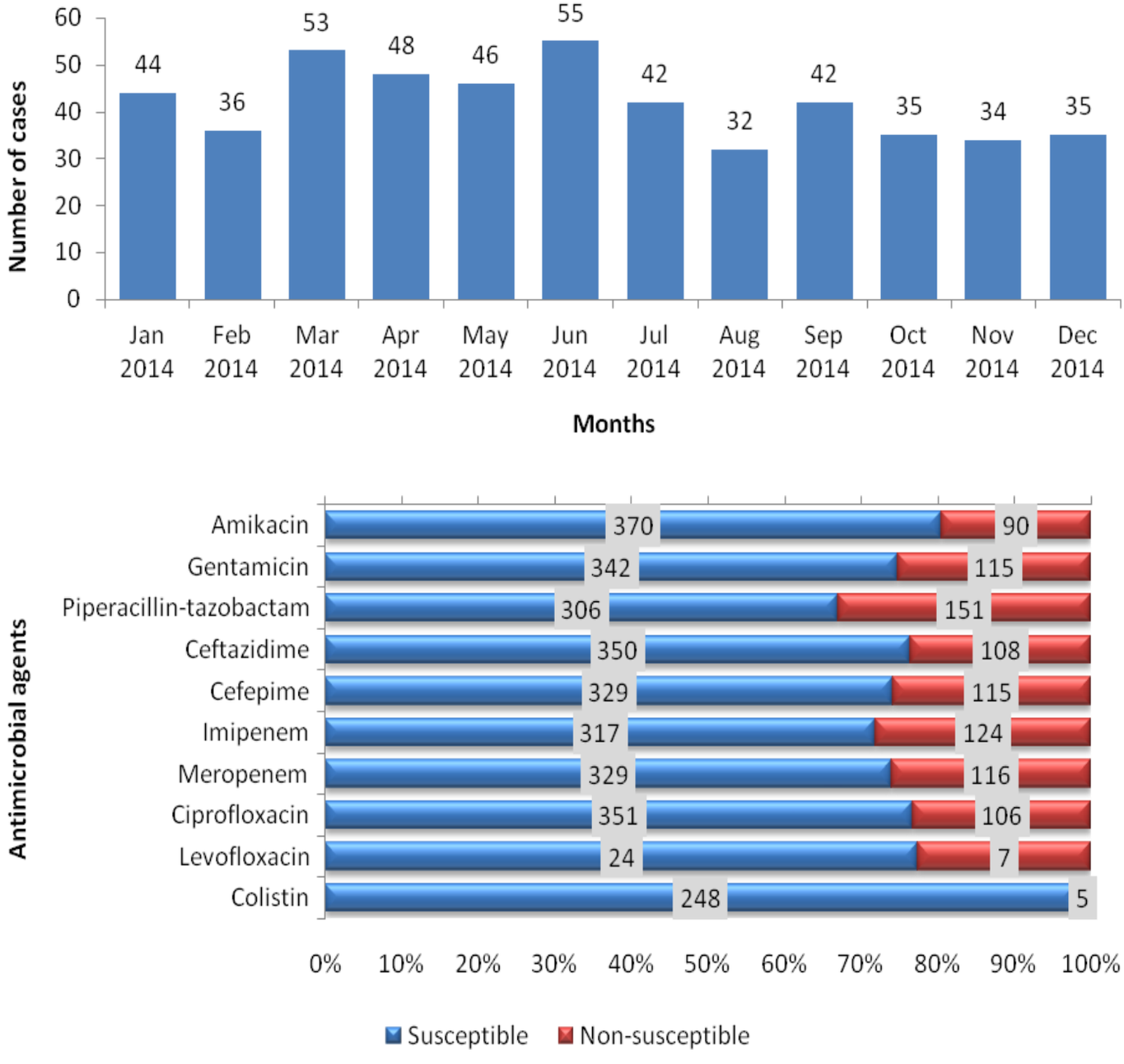
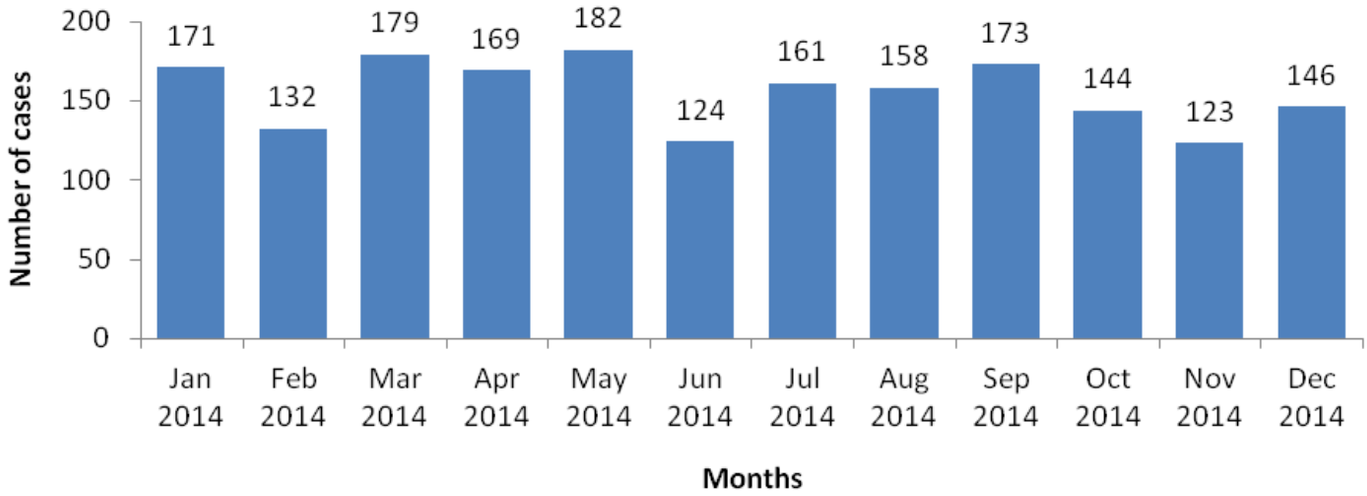


Figure 7: *Pseudomonas aeruginosa* cases by month, and numbers and percentages of susceptible and resistant *P. aeruginosa* isolates from blood cultures at public-sector sentinel sites, 2014. Total number of isolates analyzed = 502.

Staphylococcus aureus

Nine *S. aureus* isolates were reported to be vancomycin resistant. However, this was not confirmed and these data should be treated with caution as vancomycin resistance is exceptionally rare. Confirmatory phenotypic gold standard methods are available internationally and should be performed on each isolate flagged as resistant. Resistance to methicillin/oxacillin and all other β-lactams have decreased compared to the previous year. Cefoxitin resistance indicated methicillin-resistant *Staphylococcus aureus* (MRSA). Resistances to erythromycin and clindamycin have marginally decreased compared to 2013.



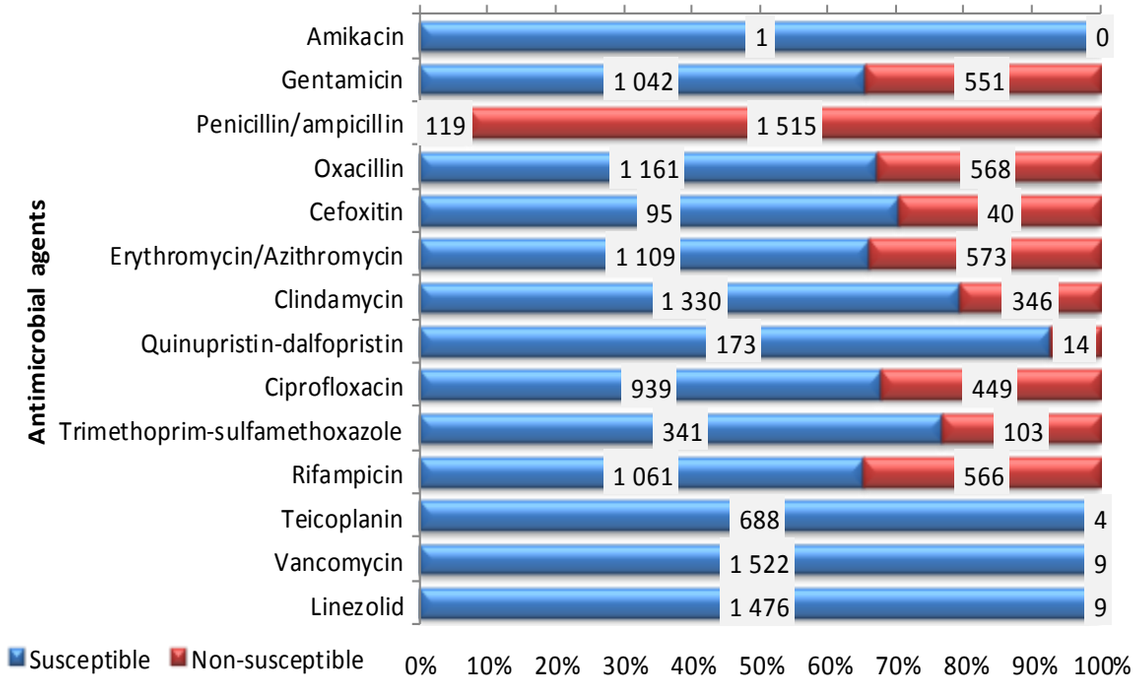


Figure 8: *Staphylococcus aureus* cases by month, and numbers and percentages of susceptible and resistant *S. aureus* isolates from blood cultures at public-sector sentinel sites, 2014. Total number of isolates analyzed = 1862.

Carbapenemase-producing Enterobacteriaceae (CPE)

The Antimicrobial Resistance Laboratory of the NICD (including the Cape Town satellite unit) analyzed the occurrence of CPE genes on all referred isolates from public laboratories based on phenotypic CLSI criteria for carbapenem resistance (table 2).[3] Isolates were sent to the reference labs on a voluntary basis. Few organisms presented with more than one carbapenemase gene.

Table 2: Numbers of confirmed carbapenemase-producing Enterobactericeae by species and genoptype. Percentages in parentheses represent proportions positive for the CPE genotype.

Carbapenemase-producing Enterobacteriaceae	No. of isolates
Species	
Klebsiella pneumoniae	186
Serratia marcescens	4
Enterobacter cloacae	87
Citrobacter freundii	5
Escherichia coli	9
Morganella morganii	3
Others	23
Total	**317**
Genotype	
OXA-48	43 (24%)
VIM	43 (24%)
NDM	85 (47%)
GES	3 (1,5%)
KPC	5 (3%)
IMP	1 (0.5%)
Total	**180**

Discussion and conclusion

Certain limitations are inherent in the data presented. Data may be incomplete due to missing cases not captured on the LIS or non-standardised coding of pathogens and antibiotics. Testing methods and microbiological practice vary between sites and this could account for variation in the results presented. Confirmatory antimicrobial susceptibility test (AST) methods were not performed for any of these organisms and results presented here are reported as captured on the LIS. Thus, while some results may suggest the occurrence of an outbreak, it is not possible to confirm this. For certain sites, not all organisms are represented. This may be due to organisms not being identified at a particular site for 2014. Nevertheless, the data presented in this report highlight the importance of surveillance for antimicrobial resistance patterns. Active surveillance needs to be ongoing in order to identify trends as well as possible outbreaks.

Disclaimer

Data are reported as received through the Central Data Warehouse. No clinical data or molecular data were available to distinguish between hospital-associated and community-acquired infections.

Acknowledgements

The NHLS CDW team is acknowledged for cleaning the data and preparing the tables and figures. Ashika Singh-Moodley and Diane Rip for CPE are thanked for gene identification.

References

1. Langmuir AD. The surveillance of communicable diseases of national importance. *N Engl J Med* 1963; 268: 182-92.
2. Garner JS, et al. CDC definitions for nosocomial infections. *Am J Infect Control* 1988; 16: 128-140.
3. Performance Standards for Antimicrobial Susceptibility Testing. Clinical and Laboratory Standards Institute (CLSI), 2014; M 100-S24, Vol. 34 No.3

Table 1: Provisional number of laboratory confirmed cases of diseases under surveillance reported to the NICD - South Africa, corresponding periods 1 January - 31 December 2013/2014*

Disease/Organism	1 Jan to 31 Dec, year	EC	FS	GA	KZ	LP	MP	NC	NW	WC	South Africa
Anthrax	2013	0	0	0	0	0	0	0	0	0	0
	2014	0	0	0	0	0	0	0	0	0	0
Botulism	2013	0	0	0	0	0	0	0	0	0	0
	2014	0	0	0	0	0	0	0	0	0	0
Cryptococcus spp.	2013	707	247	2137	1712	153	369	56	259	604	6244
	2014	722	204	1353	1477	125	286	40	238	848	5293
Haemophilus influenzae, invasive disease, all serotypes	2013	26	17	111	49	2	14	5	3	105	332
	2014	32	16	96	48	0	20	5	8	91	316
Haemophilus influenzae, invasive disease, < 5 years											
Serotype b	2013	5	1	11	5	0	2	0	0	6	30
	2014	2	2	11	3	0	0	1	0	10	29
Serotypes a,c,d,e,f	2013	0	1	5	2	0	1	1	0	8	18
	2014	1	1	6	2	0	0	1	0	3	14
Non-typeable (unencapsulated)	2013	0	2	20	7	0	0	1	1	24	55
	2014	1	1	13	5	0	1	1	0	20	42
No isolate available for serotyping	2013	1	5	23	9	1	5	1	0	7	52
	2014	5	1	25	13	0	3	0	3	24	74
Measles	2013	2	0	3	1	0	0	0	1	1	8
	2014	2	1	16	6	0	3	35	0	6	69
Neisseria meningitidis, invasive disease	2013	45	14	72	39	1	4	2	6	50	233
	2014	36	5	58	25	0	2	0	2	66	194
Novel Influenza A virus infections	2013	0	0	0	0	0	0	0	0	0	0
	2014	0	0	0	0	0	0	0	0	0	0
Plague	2013	0	0	0	0	0	0	0	0	0	0
	2014	0	0	0	0	0	0	0	0	0	0
Rabies	2013	0	2	0	1	3	1	0	0	0	7
	2014	4	0	0	0	1	0	0	1	0	6
Salmonella typhi	2013	1	2	24	11	0	11	0	1	14	64
	2014	4	4	51	18	1	10	0	0	21	109
Streptococcus pneumoniae, invasive disease, all ages	2013	301	191	1003	496	53	133	80	130	479	2866
	2014	230	184	982	499	35	121	44	106	535	2736
Streptococcus pneumoniae, invasive disease, < 5 years	2013	44	41	215	74	6	10	7	32	69	498
	2014	29	19	203	85	8	15	8	19	80	466
Vibrio cholerae O1	2013	0	0	0	0	1	0	0	0	0	1
	2014	0	0	2	0	0	0	0	0	0	2
Viral Haemorrhagic Fever (VHF)											
Crimean Congo Haemorrhagic Fever (CCHF)	2013	0	2	0	0	0	2	0	1	0	5
	2014	0	2	0	0	0	0	4	0	0	6
Other VHF (not CCHF)	2013	0	0	0	0	0	0	0	0	0	0
	2014	0	0	0	0	0	0	0	0	0	0

Footnotes

*Numbers are for cases of all ages unless otherwise specified. Data presented are provisional cases reported to date and are updated from figures reported in previous bulletins.

Provinces of South Africa: EC – Eastern Cape, FS – Free State, GA – Gauteng, KZ – KwaZulu-Natal, LP – Limpopo, MP – Mpumalanga, NC – Northern Cape, NW – North West, WC – Western Cape

0 = no cases reported

Table 2: Provisional laboratory indicators for NHLS and NICD, South Africa, corresponding periods 1 January - 31 December 2013/2014*

Programme and Indicator	1 Jan to 31 Dec, year	EC	FS	GA	KZ	LP	MP	NC	NW	WC	South Africa
Acute Flaccid Paralysis Surveillance											
Cases < 15 years of age from whom specimens received	2013	69	21	68	83	55	40	7	24	43	410
	2014	62	21	86	78	49	44	14	27	42	423

Footnotes

*Numbers are for all ages unless otherwise specified. Data presented are provisional numbers reported to date and are updated from figures reported in previous bulletins.

Provinces of South Africa: EC – Eastern Cape, FS – Free State, GA – Gauteng, KZ – KwaZulu-Natal, LP – Limpopo, MP – Mpumalanga, NC – Northern Cape, NW – North West, WC – Western Cape

Monitoring for the presence of polio in a country is based on AFP (acute flaccid paralysis) surveillance – the hallmark clinical expression of paralytic poliomyelitis. The clinical case definition of AFP is an acute onset of flaccid paralysis or paresis in any child under 15 years of age. AFP is a statutory notifiable disease and requires that 2 adequate stool specimens are taken as soon as possible, 24 to 48 hours apart, but within 14 days after onset of paralysis, for isolation and characterisation of polio virus. The differential diagnosis of AFP is wide, the most common cause of which is Guillain-Barre Syndrome. The incidence of AFP in a population has been studied in a number of developing countries and WHO have determined, as a result of these studies, that the criterion for adequate surveillance of AFP is 2 cases per 100 000 population of children less than 15 years of age (it was formerly 1 per 100,000 but this was thought to be inadequately sensitive).

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