

Weekly respiratory pathogens report Week 48 of 2023

Highlights

- This is the final report for 2023. The next respiratory pathogens report will be published in week 3 (week starting 22 January 2024). However, we will continue to monitor the trends and report more frequently if necessary.
- The 2023 influenza season ended in week 27 (week starting 10 July 2023), with sporadic cases of influenza A and influenza B detected from week 35 (week starting 28 August 2023).
- In 2023 to date, 1038 influenza cases have been detected from all surveillance programmes. Of those with typing information available 91% (920/1007) were influenza A(H3N2), 1% (12/1007) were influenza A(H1N1)pdm09 and 7% (75/1007) were influenza B Victoria.
- In 2023 to date 815 respiratory syncytial virus (RSV) cases have been detected from all surveillance programmes. The RSV season ended in week 21 (week starting 22 May 2023). The detection rate remains below seasonal threshold.
- In 2023 to date 208 cases of *Bordetella pertussis* have been detected, of which 32% (66/208) were from Gauteng Province, 21% (44/208) North West Province, 16% (34/208) from Mpumalanga Province, 14% (29/208) from KwaZulu-Natal Province, 13% (28/208) from Western Cape Province, and 3% (7/208) from Eastern Cape Province.
- In 2023 to date 506 COVID-19 cases have been detected from all surveillance programmes. Of the 453 specimens sequenced to date, a variant could be assigned in 76% (343/453). Of these, 99% (339/343) were assigned the Omicron variant, majority of which 43% (146/339) were Omicron (23A/XBB.1.5) followed by 13% (45/339) Omicron (23D/XBB.1.9). The rest of Omicron lineages contributed <10% each, including seven Omicron (23F/EG.5.1.1) sequences which belong to the latest added variant of interest. Variant could not be assigned for 24% (110/453) due to a low viral load or insufficient sample.

Programme Descriptions

Programme	Influenza-like illness (ILI)	Viral Watch	National Syndromic Surveillance for Pneumonia
Start year	2012	1984	2009
Provinces*	KZ	EC	EC
	NW	FS	GP
	wc	GP	KZ
	MP	LP	MP
	1411	MP	NW
		NC	WC
		NW	
		WC	
Type of site	Primary health care clinics	General practitioners	Public hospitals
Case definition	ILI: An acute respiratory illness with a	ILI: An acute respiratory illness with a	SRI: Acute (symptom onset≤10 days) or
case demination	temperature (≥38°C) and cough, & onset	temperature (≥38°C) and cough, & onset	chronic (symptom onset >10) lower
		· · · · · · · · · · · · · · · · · · ·	
	≤10 days	≤10 days	respiratory tract illness
	Suspected pertussis		Suspected pertussis
	Any person with an acute cough illness		Any person with an acute cough illness
			· · ·
	lasting ≥14 days (or cough illness of any		lasting ≥14 days (or cough illness of any
	duration for children <1 year), without a		duration for children <1 year), without a
	more likely diagnosis AND one or more of		more likely diagnosis AND one or more of
	the following signs or symptoms:		the following signs or symptoms:
	 paroxysms of coughing, 		 paroxysms of coughing,
	 or inspiratory "whoop", 		 or inspiratory "whoop",
	 or post-tussive vomiting 		 or post-tussive vomiting
	 or apnoea in children <1 year; 		 or apnoea in children <1 year;
	OR		OR
	Any person in whom a clinician suspects		Any person in whom a clinician suspects
	pertussis		pertussis.
	Suspected SARS-CoV-2	Constant CARC CAVA	Suspected SARS-CoV-2
	Any person presenting with an acute	Suspected SARS-CoV-2	Any person admitted with a physician-
	(≤14 days) respiratory tract infection or	Any person presenting with an acute	diagnosis of suspected COVID-19 and
	other clinical illness compatible with	(≤14 days) respiratory tract infection or	not meeting SRI case definition.
	COVID-19**	other clinical illness compatible with	not meeting 3M case definition.
		COVID-19**	
Specimens collected	Oropharyngeal & nasopharyngeal swabs	Throat and/or nasal swabs or Nasopharyngeal swabs	Oropharyngeal & nasopharyngeal swabs
Main pathogens	INF	INF	INF
tested***	RSV	RSV	RSV
icaicu	BP	SARS-CoV-2	BP
	SARS-CoV-2	JANJ-CUV-2	SARS-CoV-2
Tosting Mothods	INF and RSV	INF and RSV	INF and RSV
Testing Methods			
	- Fast-Track Diagnostics multiplex real-	- Fast-Track Diagnostics multiplex real-	- Fast Track Diagnostics multiplex real-
	time reverse transcription polymerase	time reverse transcription polymerase	time reverse transcription polymerase
	chain reaction (until 31 March 2021)	chain reaction (until 31 March 2021)	chain reaction (until 31 March 2021)
	B. pertussis	B. pertussis	B. pertussis
	Multiplex real-time PCR (Tatti et al., J Clin	Multiplex real-time PCR (Tatti et al., J Clin	Multiplex real-time PCR (Tatti et al., J Clin
	Microbiol 2011) and culture (if PCR cycle	Microbiol 2011) and culture (if PCR cycle	Microbiol 2011) and culture (if PCR cycle
	threshold ≤25)	threshold ≤25)	threshold ≤25)
	SARS-CoV-2	SARS-CoV-2	SARS-CoV-2
	1 April 2020 – 31 March 2021: Roche E	1 April 2020 – 31 March 2021: Roche E	1 April 2020 – 31 March 2021: Roche E
	gene real-time PCR essay (Corman et al.,	gene real-time PCR essay Corman et al.,	gene real-time PCR essay (Corman et al.,
	Euro Surv 2020)	Euro Surv 2020)	Euro Surv 2020)
	1 April 2021 to date: Allplex™ SARS-CoV-	1 April 2021 to date: Allplex™ SARS-CoV-	1 April 2021 to date: Allplex™ SARS-CoV-
	2/FluA/FluB/RSV PCR kit	2/FluA/FluB/RSV PCR kit	2/FluA/FluB/RSV PCR kit
	- positivity assigned if PCR cycle	- positivity assigned if PCR cycle	- positivity assigned if PCR cycle
	threshold is <40 for ≥1 gene targets	threshold is <40 for ≥1 gene targets	threshold is <40 for ≥1 gene targets
	(N, S, OR RdRp)	(N, S, OR RdRp)	(N, S, OR RdRp)
	(N, J, ON NUMP)	(N, J, OK NUNP)	(N, J, OK NUNP)

Epidemic Threshold

Thresholds are calculated using the Moving Epidemic Method (MEM), a sequential analysis using the R Language, available from: http://CRAN.R-project.org/web/package=mem) designed to calculate the duration, start and end of the annual influenza epidemic. MEM uses the 40th, 90th and 97.5th percentiles established from available years of historical data to calculate thresholds of activity. Thresholds of activity for influenza and RSV are defined as follows: Below seasonal threshold, Low activity, Moderate activity, High activity, For influenza, thresholds from outpatient influenza like illness (ILI in primary health care clinics) are used as an indicator of disease transmission in the community and thresholds from pneumonia surveillance are used as an indicator of impact of disease. For RSV, thresholds from pneumonia surveillance, using data from children aged < 5 years are used to define the start and end of the season.

^{*} EC: Eastern Cape; FS: Free State; GP: Gauteng; KZ: KwaZulu-Natal; LP: Limpopo; MP: Mpumalanga: NC: Northern Cape; NW: North West; WC: Western Cape

^{**}Symptoms include ANY of the following respiratory symptoms: cough, sore throat, shortness of breath, anosmia (loss of sense of smell) or dysgeusia (alteration of the sense of taste), with or without other symptoms (which may include fever, weakness, myalgia, or diarrhoea). Testing for SARS-CoV-2 was initiated in all three surveillance programmes in week 10 of 2020 (week starting 2 March 2020).***INF: influenza virus; RSV: respiratory syncytial virus; BP: Bordetella pertussis; SARS-CoV-2: severe acute respiratory syndrome coronavirus 2

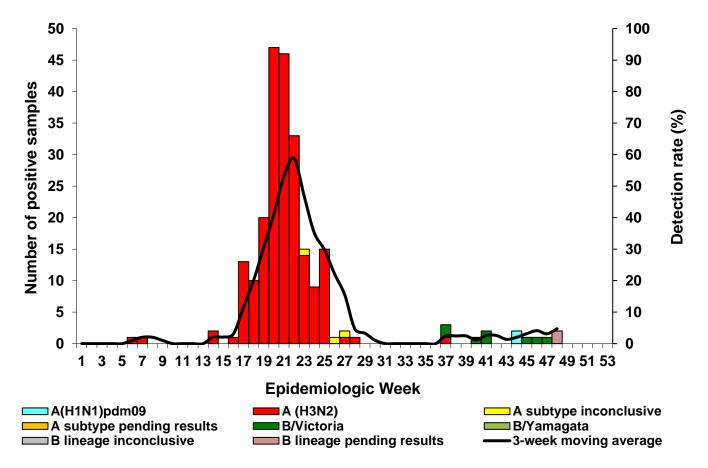


Figure 1. Number of influenza positive cases* by influenza subtype and lineage** and 3-week moving average by week, influenza-like illness (ILI) surveillance in primary health care clinics, 01/01/2023 – 03/12/2023

Inconclusive: insufficient viral load in sample and unable to characterise further

Table 1. Number of laboratory-confirmed influenza* cases by subtype and lineage and total number of samples tested by clinic and province, influenza-like illness (ILI) surveillance in primary health care clinics, 01/01/2023 – 03/12/2023

Clinic (Province)	A(H1N1) pdm09	A(H3N2)	A subtype in- conclusiv e**	A subtype pending results***	B/ Victoria	B/ Yamagata	B lineag e in- conclu sive*	B lineage pending results* **	Total sample s
Agincourt (MP)	0	41	0	0	0	0	0	0	296
Eastridge (WC)	2	35	1	0	1	0	0	1	321
Edendale Gateway (KZ)	0	53	2	0	6	0	0	1	525
Jouberton (NW)	0	76	0	0	0	0	0	0	365
Mitchell's Plain (WC)	0	10	0	0	1	0	0	0	177
Total:	2	215	3	0	8	0	0	2	1684

KZ: KwaZulu-Natal; NW: North West; WC: Western Cape; MP: Mpumalanga

^{*}Specimens from patients with influenza-like illnesses at 5 sentinel sites in 4 provinces

^{**} Influenza A(H3N2) was detected in 10/84, 12% of specimens from patients who met suspected SARS-CoV-2 or *B. pertussis* case definition but did not meet influenza-like illness (ILI) case definition.

^{*} Influenza A(H3N2) was detected in 10/84, 12% of specimens from patients who met suspected SARS-CoV-2 or *B. pertussis* case definition but did not meet influenza-like illness (ILI) case definition.

^{**}Inconclusive: insufficient viral load in sample and unable to characterise further

^{***}Influenza A subtype or B lineage results are pending

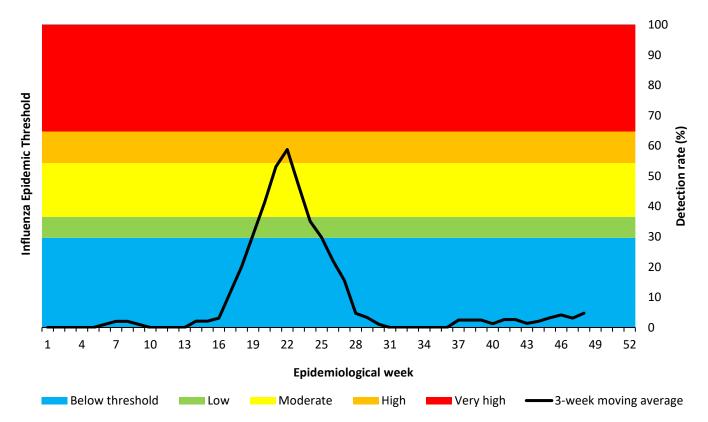


Figure 2. Influenza percentage detections and epidemic thresholds* among cases of all ages, influenza-like illness (ILI) surveillance in primary health care clinics, 01/01/2023 –03/12/2023

*Thresholds based on 2012-2019 data

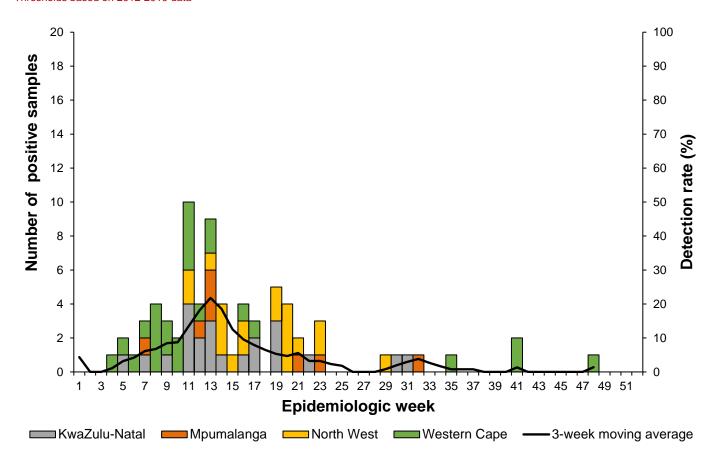


Figure 3. Number of patients testing positive for respiratory syncytial virus* by province and 3-week moving average by week, influenza-like illness (ILI) surveillance in primary health care clinics, 01/01/2023 -03/12/2023

*RSV was detected in 1/84, 1.2% of specimens of patients who met suspected SARS-CoV-2 or *B. pertussis* case definition but did not meet influenza-like illness (ILI) case definition.

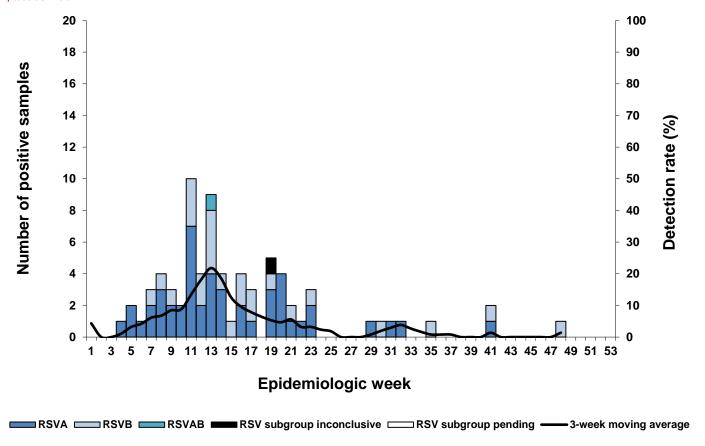


Figure 4. Number of patients testing positive for respiratory syncytial virus* by subgroup and 3-week moving average by week, influenza-like illness (ILI) surveillance in primary health care clinics, 01/01/2023 – 03/12/2023

RSV AB: Both RSV A and B subgroups identified.

Inconclusive: insufficient viral load in sample and unable to characterise further

Table 2. Number of patients testing positive for respiratory syncytial virus (RSV)* by subgroups identified and total number of samples tested by clinic and province, influenza-like illness (ILI) surveillance in primary health care clinics, 01/01/2023 - 03/12/2023

Clinic (Province)	RSVA	RSVB	RSVAB**	RSV subgroup inconclusive*	RSV subgroup pending** **	Total samples
Agincourt (MP)	3	4	1	0	0	296
Eastridge (WC)	19	5	0	0	0	321
Edendale Gateway (KZ)	9	12	0	1	0	525
Jouberton (NW)	15	4	0	0	0	365
Mitchell's Plain (WC)	1	0	0	0	0	177
Total	47	25	1	1	0	1684

KZ: KwaZulu-Natal; NW: North West; WC: Western Cape; MP: Mpumalanga

^{*}RSV was detected in 1/84, 1.2% of specimens of patients who met suspected SARS-CoV-2 or *B. pertussis* case definition but did not meet influenza-like illness (ILI) case definition.

^{*}RSV was detected in 1/84, 1.2% of specimens of patients who met suspected SARS-CoV-2 or *B. pertussis* case definition but did not meet influenza-like illness (ILI) case definition.

^{**}RSV AB: Both RSV A and B subgroups identified

^{***}Inconclusive: insufficient viral load in sample and unable to characterise further

^{****}RSV results for subgroups are pending

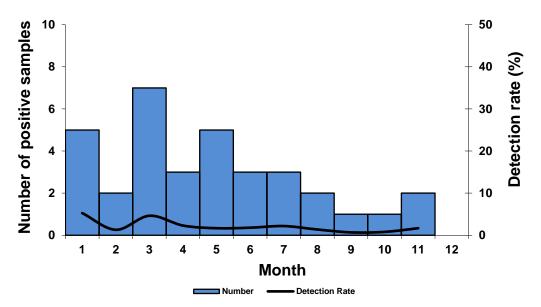


Figure 5. Number of patients testing positive for *B. pertussis** and detection rate by month, influenza-like illness (ILI) surveillance primary health care clinics**, 01/01/2023 –03/12/2023

Table 3. Number of patients testing positive for *B. pertussis** identified and total number of samples tested by province, influenza-like illness (ILI) surveillance primary health care clinics, 01/01/2023 –03/12/2023

Clinic (Province)	<i>B. pertussis</i> Positive	Total samples
Agincourt (MP)	6	292
Eastridge (WC)	3	319
Edendale Gateway (KZ)	10	519
Jouberton (NW)	15	364
Mitchell's Plain (WC)	0	172
Total:	34	1666

KZ: KwaZulu-Natal; NW: North West; WC: Western Cape; MP: Mpumalanga

^{*}B. pertussis was detected in 1/84, 1.2% of specimens of patients who met the suspected SARS-CoV-2 or B. pertussis case definition but did not meet influenza-like illness (ILI) case definition. These are not included in the epidemiological curve.

^{**} Specimens from patients with influenza-like illnesses at 5 sentinel sites in 4 provinces

^{*}B. pertussis was detected in 1/84, 1.2% of specimens of patients who met the suspected SARS-CoV-2 or B. pertussis case definition but did not meet influenzalike illness (ILI) case definition. These are not included in the epidemiological curve.

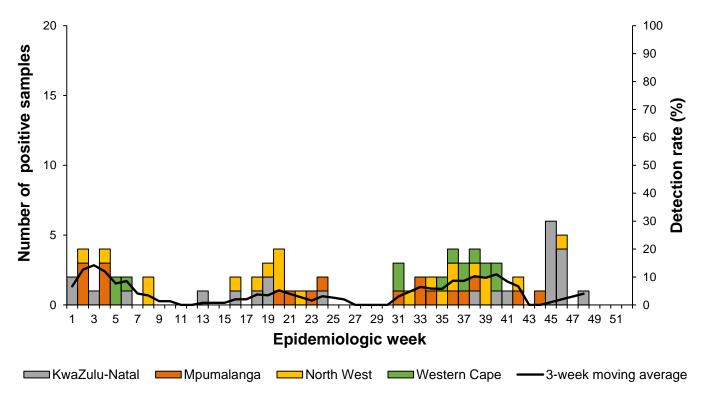


Figure 6. Number of patients* testing positive for SARS-CoV-2** by province and 3-week moving average by week, influenza-like illness (ILI) surveillance in primary health care clinics, 01/01/2023 – 03/12/2023

Table 4. Number of patients positive for SARS-CoV-2* identified and total number of samples tested by clinic and province, influenza-like illness (ILI) surveillance primary health care clinics, 01/01/2023 – 03/12/2023

Clinic (Province)	SARS-CoV-2 positive	Total samples tested
Agincourt (MP)	19	296
Eastridge (WC)	9	321
Edendale Gateway (KZ)	24	525
Jouberton (NW)	21	365
Mitchell's Plain (WC)	4	177
Total:	77	1684

KZ: KwaZulu-Natal; NW: North West; WCP: Western Cape; MP: Mpumalanga

^{*}Specimens from patients with influenza-like illnesses at 5 sentinel sites in 4 provinces

^{**}SARS-CoV-2 was detected in 1/84, 1.2% of specimens from patients who met suspected SARS-CoV-2 or *B. pertussis* case definition but did not meet influenzalike illness (ILI) case definition. These are not included in the epidemiological curve.

^{*}SARS-CoV-2 was detected in 1/84, 1.2% of specimens from patients who met suspected SARS-CoV-2 or *B. pertussis* case definition but did not meet influenzalike illness (ILI) case definition. These are not included in the epidemiological curve.

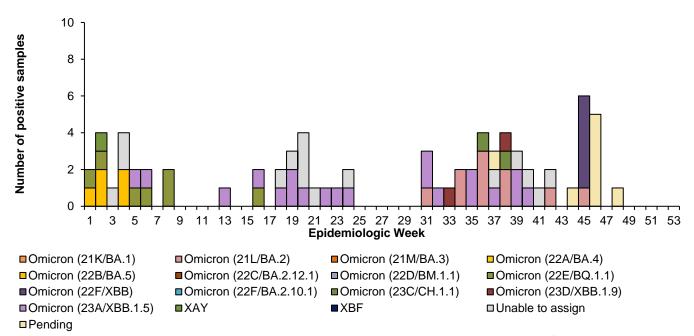


Figure 7. Number of laboratory-confirmed SARS-CoV-2* cases by variant type (variant PCR/sequencing) and week, influenza-like illness (ILI) surveillance in primary health care clinics, 01/01/2023 – 03/12/2023

Unable to assign: no lineage assigned due to poor- sequence quality OR low viral load (Ct≥35) OR variant PCR could not assign variant and no sequencing result Pending: outstanding variant results

Table 5. Number of cases positive for SARS-CoV-2* by variant** (variant PCR and/or sequencing) identified and total number of samples tested by clinic and province, influenza-like illness (ILI) surveillance primary health care clinics, 01/01/2023 – 03/12/2023

Province	Omicron (21L/BA.2)	Omicron (21M/ BA.3)	Omicron (22A/BA.4)	Omicron (22B/ BA.5)	Omicron (22C/BA.2.12.	Omicron (22D/BM.1.1)	Omicron (22E/BQ.1.1)	Omicron (22F/XBB)	Omicron (22F/XXB.1)	Omicron (23A/XBB.1.5	Omicron (23B/XBB.1.16)	Omicron (23D/XBB.1.9	Omicron (23C/CH.1.1)	хау	Unable to assign**	Pending***	SARS-CoV-2 positive	Total samples tested
Agincourt Clinic (MP) Eastridge	5	0	0	4	0	0	0	0	0	3	1	1	0	1	3	1	19	296
Clinic (WC)	0	0	0	0	0	0	0	0	0	5	0	1	1	0	2	0	9	321
Edendale Clinic (KZ) Jouberto	1	0	0	1	0	0	3	5	0	6	0	0	1	0	3	5	25	595
n Clinic (NW) Mitchell's	4	0	0	0	0	0	3	0	0	5	0	0	0	0	7	2	21	379
Plain Clinic (WC)	0	0	0	0	0	0	1	0	0	1	1	1	0	0	0	0	4	177
Total:	10	0	0	5	0	0	7	5	0	20	2	3	2	1	15	8	78	1768

KZ: KwaZulu-Natal; NW: North West; WCP: Western Cape; MP: Mpumalanga

Unable to assign: no lineage assigned due to poor- sequence quality **OR** low viral load ($C_t \ge 35$) **OR** variant PCR could not assign variant and no sequencing result **Pending**: outstanding variant results

^{*}Specimens are from patients with influenza-like illness at 5 sentinel sites in 4 provinces who met influenza-like illness (ILI), suspected SARS-CoV-2 or *B. pertussis* case definition

^{*}Specimens are from patients with influenza-like illness at 5 sentinel sites in 4 provinces who met influenza-like illness (ILI), suspected SARS-CoV-2 or *B. pertussis* case definition

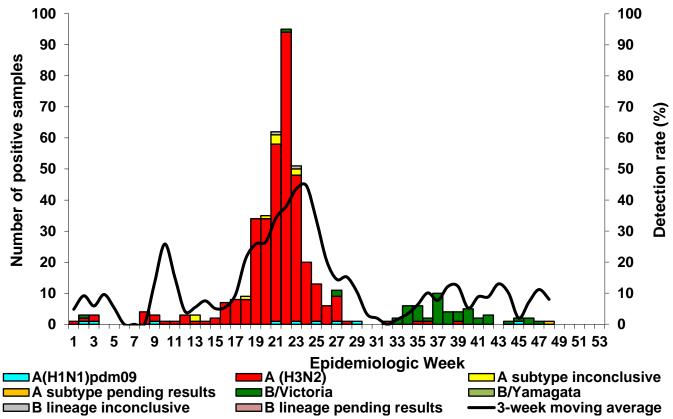


Figure 8a. Number of positive patients* by influenza subtype and lineage and 3-week moving average by week, influenza-like illness (ILI) surveillance - Viral Watch, 01/01/2023 – 03/12/2023

^{*}Specimens from patients with influenza-like illnesses at 92 sentinel sites in 8 provinces Inconclusive: insufficient viral load in sample and unable to characterise further

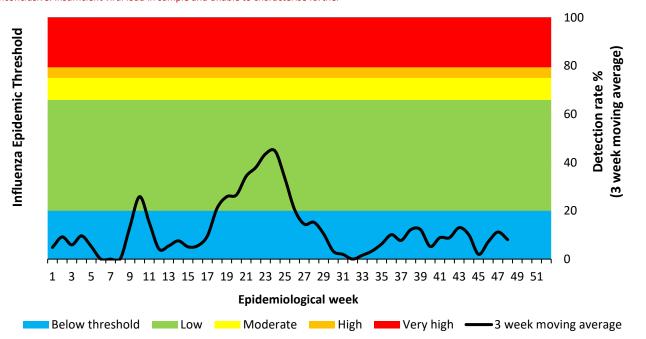


Figure 8b. Influenza percentage detections and epidemic thresholds* among cases of all ages, Influenza-like Illness (ILI) surveillance - Viral Watch, 01/01/2023 – 03/12/2023

^{*}Thresholds based on 2015-2019 data

Table 6. Number of laboratory-confirmed influenza cases by influenza subtype and lineage and total number of samples tested by province, ILI surveillance - Viral Watch, 01/01/2023 – 03/12/2023

Province	A(H1N1) pdm09	A(H3N2)	A subtype inconclusive	A subtype pending results*	B/Victori a	B/Yamaga ta	B lineage inconclusi ve	B lineage pending results*	Total samples
Eastern Cape	0	17	0	0	8	0	0	0	58
Free State	0	2	0	0	0	0	0	0	2
Gauteng	4	129	4	0	12	0	1	0	897
Limpopo	0	2	0	0	0	0	0	0	6
Mpumalanga	1	19	2	0	1	0	0	0	61
North West	0	3	0	0	0	0	0	0	3
Northern Cape	0	0	0	0	0	0	0	0	0
Western Cape	4	187	3	1	29	0	1	0	530
Total:	9	359	9	1	50	0	2	0	1557

^{*}Inconclusive: insufficient viral load in sample and unable to characterise further

^{**}Influenza A subtype or B lineage results are pending

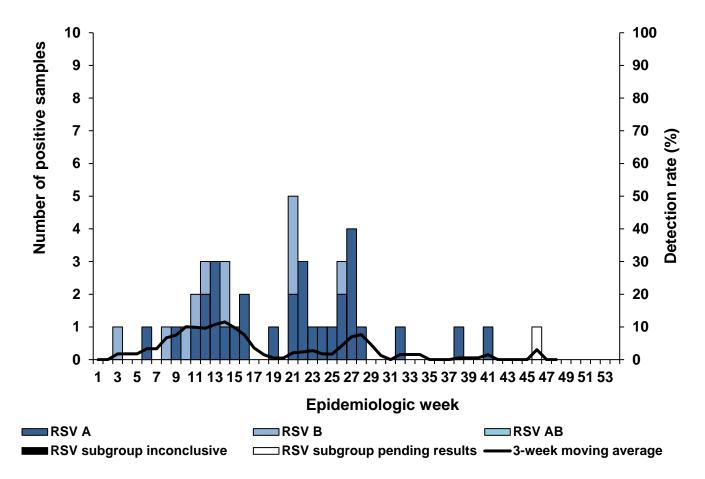


Figure 9. Number of RSV positive cases testing positive for respiratory syncytial virus (RSV)* by subgroup and 3-week moving average by week, Influenza-like Illness (ILI) surveillance - Viral Watch, 01/01/2023 – 03/12/2023

^{*}Specimens from patients with Influenza-like illnesses at 92 sentinel sites in 8 provinces

Table 7. Number of RSV positive cases identified and total number of samples tested by province, influenza-like illness surveillance - Viral Watch, 01/01/2023 – 03/12/2023

Province	RSV A	RSV B	RSV AB*	RSV subgroup inconclusive **	RSV subgroup pending results***	Total samples tested
Eastern Cape	1	1	0	0	0	58
Free State	0	0	0	0	0	2
Gauteng	19	7	0	0	1	897
Limpopo	0	0	0	0	0	6
Mpumalanga	2	0	0	0	0	61
North West	0	0	0	0	0	3
Northern Cape	0	0	0	0	0	0
Western Cape	9	3	0	0	0	530
Total:	31	11	0	0	1	1557

^{*}RSV AB: Both RSV A and B subgroup identified

^{***}RSV results for subgroups are pending

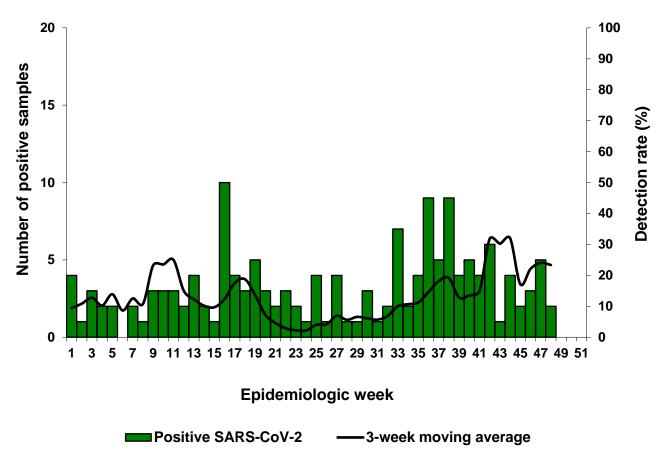


Figure 10. Number of patients testing positive for SARS-CoV-2*, by site and 3-week moving average** by week, influenza-like illness(ILI) surveillance - Viral Watch, 01/01/2023 – 03/12/2023

^{**}Inconclusive: insufficient viral load in sample and unable to characterise further

^{*}Specimens from patients with influenza-like illnesses at 92 sentinel sites in 8 provinces

Table 8. Number of SARS-CoV-2 positive cases identified and total number tested by province, influenza-like illness (ILI) surveillance - Viral Watch, 01/01/2023 – 03/12/2023

Province	SARS-CoV-2 positive	Total samples tested
Eastern Cape	5	58
Free State	0	2
Gauteng	95	897
Limpopo	0	6
Mpumalanga	8	61
North West	0	3
Northern Cape	0	0
Western Cape	47	530
Total:	155	1557

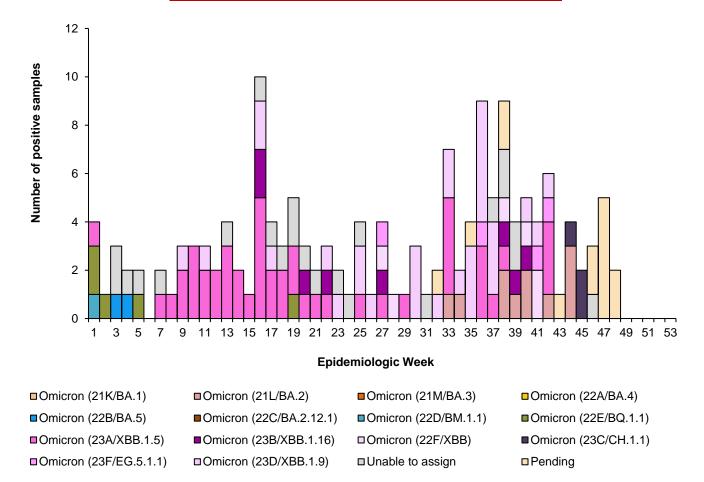


Figure 11. Number of laboratory confirmed SARS-CoV-2* cases by variant type (variant PCR/sequencing) and week, influenza-like illness (ILI) surveillance - Viral Watch, 01/01/2023 – 03/12/2023

^{*}Specimens from patients with influenza-like illnesses at 92 sentinel sites in 8 provinces

Unable to assign: no lineage assigned due to poor- sequence quality OR low viral load (Ct ≥35) OR variant PCR could not assign variant and no sequencing result

Pending: outstanding variant results

Table 9. Number of SARS-CoV-2* positive cases by variant (variant PCR and/or sequencing) identified and total number of samples tested by province, ILI surveillance - Viral Watch, 01/01/2023 – 03/12/2023

Clinic (Province)	Omicron (21L/BA.2)	Omicron (21M/BA.3)	Omicron (22B/BA.5)	Omicron (22D/BM.1.1)	Omicron (22E/BQ.1.1)	Omicron (23A/XBB.1.5)	Omicron (23B/XBB.1.16)	Omicron (22F/XBB)	Omicron (23F/EG.5.1)	Omicron (23D/XBB.1.9)	Omicron (23C/CH.1.1)	Unable to assign**	Pending***	Total SARS- CoV-2 positive	Total samples tested
Eastern Cape	0	0	0	0	0	2	0	0	0	2	0	1	0	5	58
Free State	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2
Gauteng	4	0	2	1	3	32	4	5	1	21	0	10	12	95	897
Limpopo	0	0	0	0	0	0	0	0	0	0	0	0	0	0	6
Mpumalan ga	3	0	0	0	0	1	0	1	0	1	0	2	0	8	61
North West	0	0	0	0	0	0	0	0	0	0	0	0	0	0	3
Northern Cape	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Western Cape	4	0	0	0	2	12	4	0	3	7	3	10	2	47	530
Total:	11	0	2	1	5	47	8	6	4	31	3	23	14	155	1557

^{*}Specimens from patients with influenza-like illnesses at 92 sentinel sites in 8 provinces

Unable to assign: no lineage assigned due to poor- sequence quality OR low viral load (C₁≥35) OR variant PCR could not assign variant and no sequencing result Pending: outstanding variant results

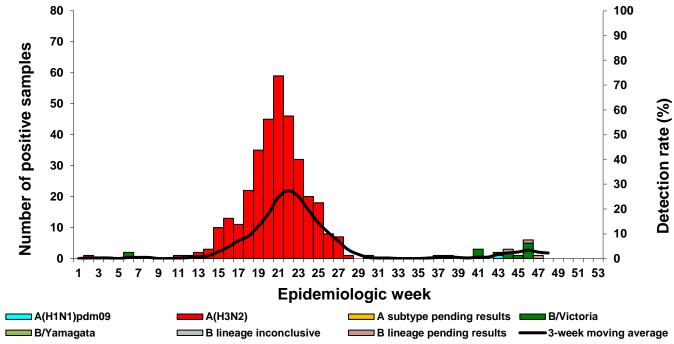


Figure 12. Number of positive influenza positive cases* by influenza subtype and lineage** and 3-week moving average by week, pneumonia surveillance public hospitals, 01/01/2023 – 03/12/2023

Inconclusive: insufficient viral load in sample and unable to characterise further

^{**}No cases of Alpha, Beta or 20D (C.1.2) variants detected.

^{*}Specimens from patients hospitalised with pneumonia at 15 sentinel sites in 6 provinces

^{**}No cases who met suspected the SARS-CoV-2 or B. pertussis case definition but did not meet pneumonia (SRI) case definition.

Table 10. Number of laboratory confirmed influenza cases by subtype and lineage* and total number of samples tested by hospital, pneumonia surveillance public hospitals, 01/01/2023-03/12/2023

Hospital (Province)	A(H1N1)pd m09	A(H3N2)	A subtype inconclusive	A subtype pending results***	B/Victoria	B/Yamagata	B lineage inconclusive	B lineage pending results***	Total samples
Edendale (KZ)	0	29	1	0	3	0	0	0	706
Helen Joseph-Rahima Moosa (GP)	1	65	1	0	5	0	0	0	1312
Khayelitsha (WC)	0	29	1	0	1	0	0	1	587
Klerksdorp-Tshepong (NW)	0	47	1	0	0	0	0	0	534
Livingstone (EC)	0	22	1	0	0	0	0	0	609
Mapulaneng- Matikwana (MP)	0	22	2	0	0	0	0	0	474
Mitchell's Plain (WC)	0	16	0	0	2	0	0	0	438
Red Cross (WC)	0	26	1	0	3	0	0	0	944
Tambo Memorial (GP)	0	33	1	0	2	0	0	0	555
Tembisa (GP)	0	27	3	0	1	0	0	1	611
Tintswalo (MP)	0	11	0	0	0	0	0	0	275
Tygerberg (WC)	0	9	0	0	0	0	0	0	122
Total:	1	336	12	0	17	0	0	2	7167

^{*} No cases who met suspected SARS-CoV-2 or B. pertussis case definition but did not meet pneumonia (SRI) case definition.

^{***}Influenza A subtype or B lineage results are pending

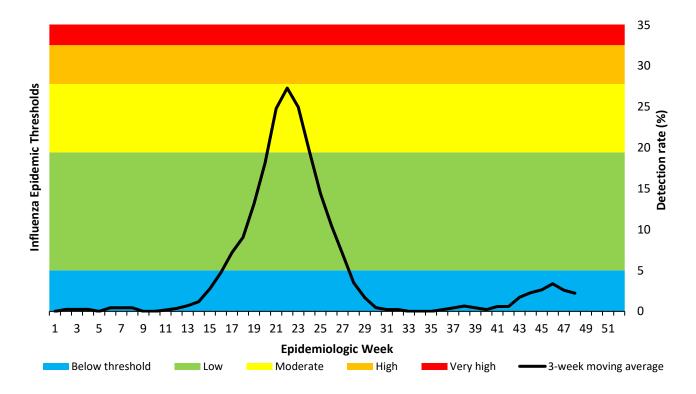


Figure 13. Influenza percentage detections and epidemic thresholds* among cases of all ages, pneumonia surveillance public hospitals, 01/01/2023 - 03/12/2023

^{**}Inconclusive: insufficient viral load in sample and unable to characterise further

^{*}Thresholds based on 2010-2019 data

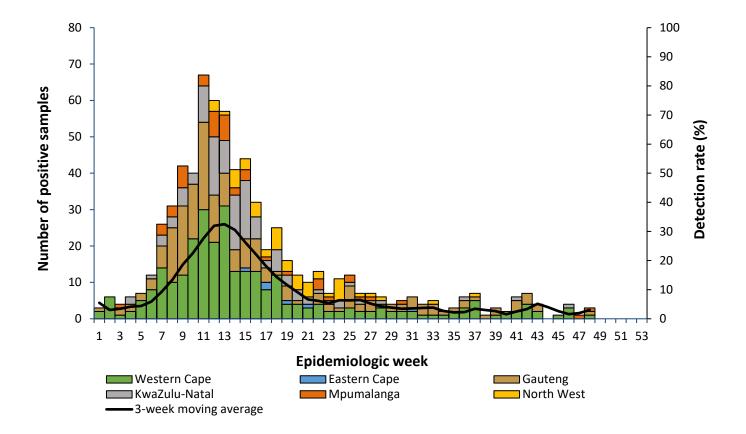


Figure 14. Number of patients (all ages) testing positive for respiratory syncytial virus* by province and 3-week moving average by week, pneumonia surveillance public hospitals, 01/01/2023 – 03/12/2023

Specimens from patients hospitalised with pneumonia at 15 sentinel sites in 6 provinces.

^{*}No cases who met suspected SARS-CoV-2 or B. pertussis case definition but did not meet pneumonia (SRI) case definition.

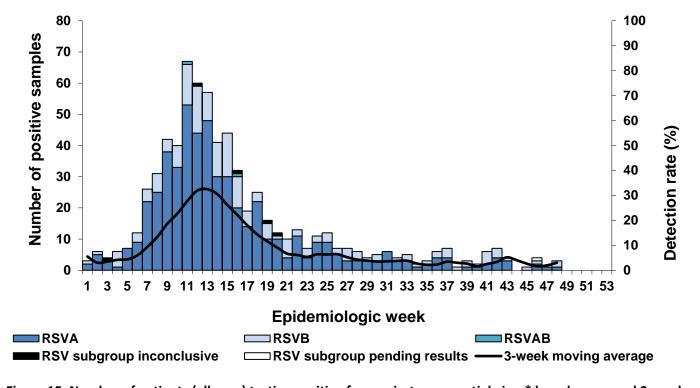


Figure 15. Number of patients (<u>all ages</u>) testing positive for respiratory syncytial virus* by subgroup and 3-week moving average by week, pneumonia surveillance public hospitals, 01/01/2023 – 03/12/2023

Specimens from patients hospitalised with pneumonia at 15 sentinel sites in 6 provinces. Inconclusive: insufficient viral load in sample and unable to characterise further RSV AB: Both RSV A and B subgroup identified RSV subgroup pending: RSV results for subgroups are pending

Table 11. Number of patients (all ages) positive for respiratory syncytial virus subgroups* by subgroups identified and total number of samples tested by hospital, pneumonia surveillance public hospitals, 01/01/2023 – 03/12/2023

Hospital (Province)	RSVA	RSVB	RSVAB**	RSV subgroup inconclusive**	RSV subgroup pending** **	Total samples
Edendale (KZ)	39	74	1	1	0	706
Helen Joseph-Rahima Moosa (GP)	155	25	0	1	0	1312
Khayelitsha (WC)	5	4	0	0	0	587
Klerksdorp-Tshepong (NW)	45	3	0	1	0	534
Livingstone (EC)	4	2	0	0	0	609
Mapulaneng-Matikwana (MP)	13	7	1	0	0	474
Mitchell's Plain (WC)	69	12	0	0	0	438
Red Cross (WC)	155	37	0	2	1	944
Tambo Memorial (GP)	4	3	0	0	0	555
Tembisa (GP)	2	2	0	0	0	611
Tintswalo (MP)	26	1	0	0	0	275
Tygerberg (WC)	1	1	0	0	0	122
Total:	518	171	2	5	1	7167

^{*}No cases who met suspected SARS-CoV-2 or B. pertussis case definition but did not meet pneumonia (SRI) case definition.

^{****}RSV results for subgroups are pending

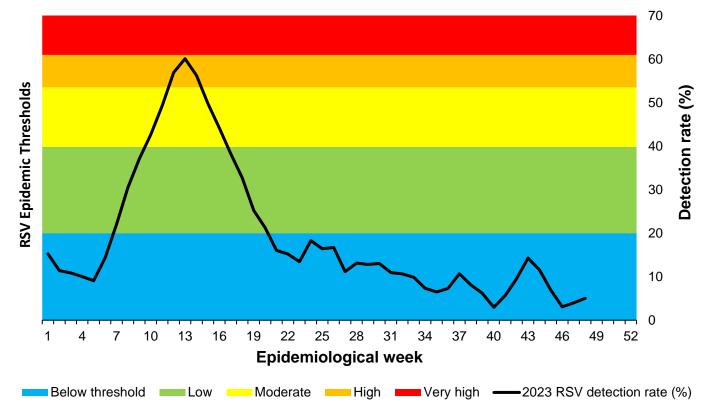


Figure 16. RSV percentage 3-week moving average and epidemic thresholds* among children aged < 5 years, pneumonia surveillance public hospitals, 01/01/2023 - 03/12/2023

^{**}RSV AB: Both RSV A and B subgroup identified

^{***}Inconclusive: insufficient viral load in sample and unable to characterise further

^{*}Thresholds based on 2010-2019 data

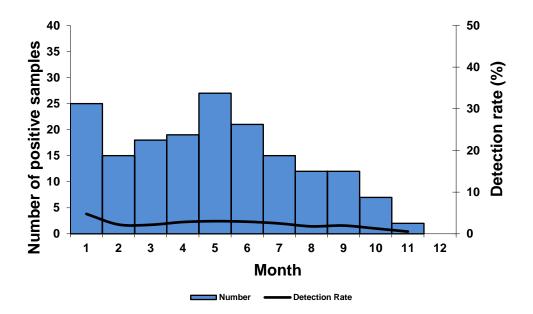


Figure 17. Number of patients testing positive for *B. pertussis** and 3-week moving average by month, pneumonia surveillance public hospitals**, 01/01/2023 – 03/12/2023

Table 12. Number of patients testing positive for *B. pertussis** identified and total number of samples tested by hospital and province, pneumonia surveillance public hospitals, 01/01/2023 – 03/12/2023

Hospital (Province)	<i>B. pertussis</i> Positive	Total samples				
Edendale (KZ)	19	699				
Helen Joseph-Rahima Moosa (GP)	49	1301				
Khayelitsha (WC)	4	587				
Klerksdorp-Tshepong(NW)	28	531				
Livingstone (EC)	7	605				
Mapulaneng-Matikwana (MP)	23	469				
Mitchell's Plain (WC)	3	438				
Red Cross (WC)	16	935				
Tambo Memorial (GP)	9	551				
Tembisa (GP)	8	605				
Tintswalo (MP)	5	275				
Tygerberg (WC)	2	122				
Total:	173	7118				

^{*}No cases who met the suspected SARS-CoV-2 or *B. pertussis* case definition but did not meet the pneumonia (SRI) case definition. These are not included in the table.

^{*}No cases who met the suspected SARS-CoV-2 or *B. pertussis* case definition but did not meet Pneumonia Surveillance case definition.

^{*}Specimens from patients hospitalised with pneumonia at 15 sentinel sites in 6 provinces.

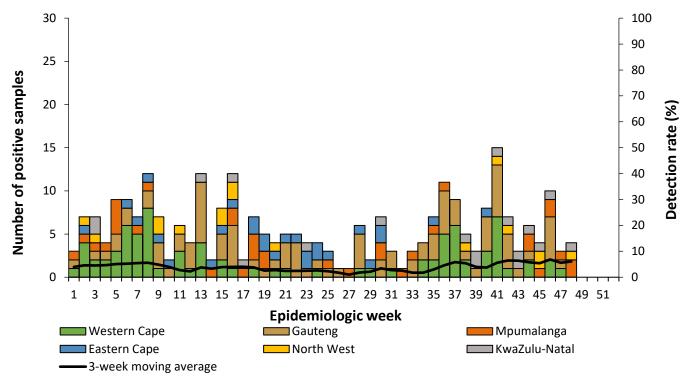


Figure 18. Number of patients testing positive for SARS-CoV-2** by province and 3-week moving average by week, pneumonia surveillance public hospitals, 01/01/2023 – 03/12/2023

Table 13. Number of patients positive for SARS-CoV-2* and total number of samples tested by hospital, pneumonia surveillance public hospitals, 01/01/2023 – 03/12/2023

Hospital (Province)	SARS-CoV-2	Total samples				
. , ,	positive	tested				
Edendale (KZ)	18	706				
Helen Joseph-Rahima Moosa (GP)	42	1312				
Khayelitsha (WC)	25	587				
Klerksdorp-Tshepong (NW)	16	534				
Livingstone (EC)	27	609				
Mapulaneng-Matikwana (MP)	23	474				
Mitchell's Plain (WC)	22	438				
Red Cross (WC)	32	944				
Tambo Memorial (GP)	30	555				
Tembisa (GP)	22	611				
Tintswalo (MP)	12	275				
Tygerberg (WC)	4	122				
Total:	273	7167				

^{*} No cases who met suspected SARS-CoV-2 or B. pertussis case definition but did not meet pneumonia (SRI) case definition.

^{*}Specimens from patients hospitalized with pneumonia at 15 sentinel sites in 6 provinces.

^{**}No cases met suspected SARS-CoV-2 or B. pertussis case definition but did not meet pneumonia (SRI) case definition.

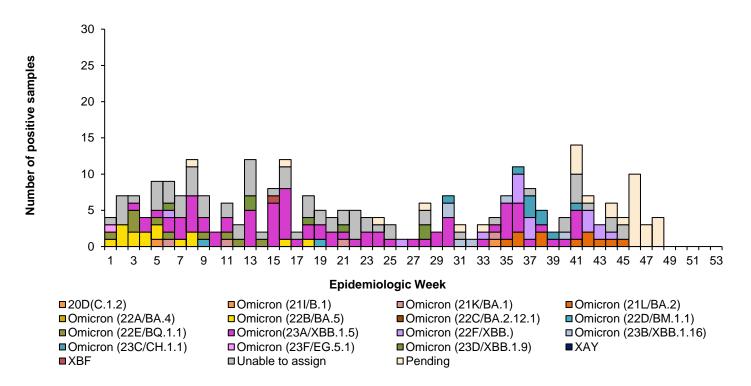


Figure 19. Number and 3-week moving average of laboratory-confirmed SARS-CoV-2 cases* by variant type (variant PCR/sequencing), pneumonia surveillance public hospitals, 01/01/2023 – 03/12/2023

Table 14. Number of SARS-CoV-2 positive cases* by variant (variant PCR and/or sequencing) identified and total number of samples tested by hospital, pneumonia surveillance public hospitals, 01/01/2023 – 03/12/2023

Hospital (Province)	20D (C.1.2)	Omicron (211/B.1)	Omicron (21L/BA.2)	Omicron (21K/BA.1)	Omicron (22B/BA.5)	Omicron (22D/BM.1.1	Omicron (22E/BQ.1.1)	Omicron(23 A/XBB.1.5)	Omicron (23B/XBB.1.	Omicron (22F/XBB.)	Omicron (23F/EG.5.1)	Omicron (23C/CH.1.1)	Omicron (23D/XBB.1.	XBF	XBL	Unable to assign	Pending	SARS-CoV-2 positive	Total samples
Edendale (KZ)	0	0	1	0	0	0	1	4	0	1	0	1	1	0	0	4	5	18	706
Helen Joseph- Rahima Moosa (GP)	0	1	0	1	1	0	1	13	0	7	2	0	3	0	0	8	5	42	1312
Khayelitsha (WC)	0	0	1	2	1	0	1	9	0	0	0	0	0	0	0	8	3	25	587
Klerksdorp- Tshepong (NW)	0	0	2	0	1	0	2	7	0	0	0	0	0	0	0	3	1	16	534
Livingstone (EC)	0	0	0	1	1	2	0	13	0	0	0	2	0	1	0	7	0	27	609
Mapulaneng- Matikwana (MP)	0	0	1	0	3	0	1	3	1	0	0	0	0	0	0	9	5	23	474
Mitchell's Plain (WC)	0	0	1	0	1	0	0	7	0	4	0	2	1	0	0	5	1	22	438
Red Cross (WC)	0	0	0	0	4	0	0	10	3	1	1	4	0	0	1	2	5	32	944
Tambo Memorial (GP)	1	0	2	0	0	0	3	8	0	1	0	0	5	0	0	9	2	30	555
Tembisa (GP)	0	0	2	0	1	0	1	1	0	0	0	0	1	0	0	12	4	22	611
Tintswalo (MP)	0	0	1	0	1	0	0	2	2	2	0	0	0	0	0	4	0	12	275
Tygerberg (WC)	0	0	0	0	1	0	0	2	0	0	0	0	0	0	0	1	0	4	122
Total:	1	1	11	4	15	2	10	79	6	16	3	9	11	1	1	72	31	273	7167

^{*}Specimens are from hospitalized patients at 15 sentinel sites in 6 provinces who met the pneumonia (SRI), suspected SARS-CoV-2 or *B. pertussis* case definition **Unable to assign**: no lineage assigned due to poor- sequence quality **OR** low viral load (C₁≥35) **OR** variant PCR could not assign variant and no sequencing result **Pending**: outstanding variant results

Page **19** of **20**

^{*}Specimens are from hospitalized patients at 15 sentinel sites in 6 provinces who met the pneumonia (SRI), suspected SARS-CoV-2 or *B. pertussis* case definition **Unable to assign**: no lineage assigned due to poor- sequence quality **OR** low viral load (C₁≥35) **OR** variant PCR could not assign variant and no sequencing result **Pending**: outstanding variant results

Methods

SARS-CoV-2 Testing

March 2020 – March 2021: SARS-CoV-2 was detected using the Roche E gene real-time PCR assay (Corman et al. *Euro Surveillance* 2020) with cycle threshold (C_t) <40 interpreted as positive for SARS-CoV-2. From April 2021 to date the laboratory changed to the AllplexTM SARS-CoV-2/FluA/FluB/RSV kit (Seegene Inc., Seoul, South Korea), with positivity assigned if the PCR cycle threshold (C_t) was <40 for \geq 1 gene targets (N, S or RdRp).

A confirmed SARS-CoV-2 case is a person of any age enrolled in surveillance with laboratory confirmation of SARS-CoV-2 infection by PCR. Only positive SARS-CoV-2 specimens on PCR are further tested to determine variant/lineage type by variant PCR or genomic sequencing.

Allplex™ SARS-CoV-2 Variants I PCR detects Alpha and Beta/Gamma variants. The assay was conducted on all SARS-CoV-2-positive samples from 1 March 2020 – 30 June 2021.

Allplex™ SARS-CoV-2 Variants II PCR detects Delta variant and distinguishes Beta from Gamma. The assay was conducted on SARS-CoV-2-positive samples from 1 Jan to 30 June 2021.

Extraction: Total nucleic acids were extracted from 200µl NP/OP samples in universal or viral transport medium using a MagNA Pure 96 automated extractor and DNA/Viral NA Small Volume v2.0 extraction kit (Roche Diagnostics, Mannheim, Germany).

SARS-CoV-2 genomic surveillance

SARS-CoV-2 Whole-Genome Sequencing and Genome Assembly RNA Extraction

RNA was extracted either manually or automatically in batches, using the QIAamp viral RNA mini kit (QIAGEN, CA, USA) or the Chemagic 360 using the CMG-1049 kit (PerkinElmer, MA, USA). A modification was done on the manual extractions by adding 280 μ l per sample, in order to increase yields. 300 μ l of each sample was used for automated magnetic bead-based extraction using the Chemagic 360. RNA was eluted in 60 μ l of the elution buffer. Isolated RNA was stored at -80 °C prior to use.

PCR and Library Preparation

Sequencing was performed using the Illumina COVIDSeq protocol (Illumina Inc., CA, USA) or nCoV-2019 ARTIC network sequencing protocol v3 (https://artic.network/ncov-2019). These are amplicon-based next-generation sequencing approaches. Briefly, for the nCoV-2019 ARTIC network sequencing protocol, the first strand synthesis was carried out on extracted RNA samples using random hexamer primers from the SuperScript IV reverse transcriptase synthesis kit (Life Technologies, CA, USA) or LunaScript RT SuperMix Kit (New England Biolabs (NEB), MA, USA). The synthesized cDNA was amplified using multiplex polymerase chain reactions (PCRs) using ARTIC nCoV-2019 v3 primers. For the COVIDSeq protocol, the first strand synthesis was carried out using random hexamer primers from Illumina and the synthesized cDNA underwent two separate multiplex PCR reactions.

For Illumina sequencing using the nCoV-2019 ARTIC network sequencing protocol, the pooled PCR products underwent bead-based tagmentation using the Nextera Flex DNA library preparation kit (Illumina Inc., CA, USA). The adapter-tagged amplicons were cleaned up using AmpureXP purification beads (Beckman Coulter, High Wycombe, UK) and amplified using one round of PCR. The PCRs were indexed using the Nextera CD indexes (Illumina Inc., CA, USA) according to the manufacturer's instructions. For COVIDSeq sequencing protocol, pooled PCR amplified products were processed for tagmentation and adapter ligation using IDT for Illumina Nextera UD Indexes. Further enrichment and clean-up was performed as per protocols provided by the manufacturer (Illumina Inc., CA, USA). Pooled samples from both COVIDSeq protocol and nCoV-2019 ARTIC network protocol were quantified using Qubit 3.0 or 4.0 fluorometer (Invitrogen Inc., MA, USA) using the Qubit dsDNA High Sensitivity assay according to manufacturer's instructions. The fragment sizes were analyzed using TapeStation 4200 (Invitrogen Inc., MA, USA). The pooled libraries were further normalized to 4nM concentration and 25 µl of each normalized pool containing unique index adapter sets were combined in a new tube. The final library pool was denatured and neutralized with 0.2 N sodium hydroxide and 200 mM Tris-HCL (pH7), respectively. 1.5 pM sample library was spiked with 2% PhiX. Libraries were loaded onto a 300-cycle NextSeq 500/550 HighOutput Kit v2 and run on the Illumina NextSeq 550 instrument (Illumina Inc., CA, USA).

Assembly, Processing and Quality Control of Genomic Sequences

Raw reads from Illumina sequencing were assembled using the Exatype NGS SARS-CoV-2 pipeline v1.6.1, (https://sars-cov-2.exatype.com/). The resulting consensus sequence was further manually polished by considering and correcting indels in homopolymer regions that break the open reading frame (probably sequencing errors) using Aliview v1.27, (https://ormbunkar.se/aliview/) (Larsson, 2014). Mutations resulting in mid-gene stop codons and frameshifts were reverted to wild type. All assemblies determined to have acceptable quality (defined as having at least 1 000 000 reads and at least 40 % 10 X coverage) were deposited on GISAID (https://www.gisaid.org/) (Elbe & Buckland-Merrett, 2017; Shu & McCauley, 2017).

Classification of Lineage, Clade and Associated Mutations

Assembled genomes were assigned lineages using the 'Phylogenetic Assignment of Named Global Outbreak Lineages' (PANGOLIN) software suite (https://github.com/hCoV-2019/pangolin) (Rambaut et al., 2020), a tool used for dynamic SARS-CoV-2 lineage classification. The SARS-CoV-2 genomes in our dataset were also classified using the clade classification proposed by NextStrain (https://nextstrain.org/), a tool built for real-time tracking of the pathogen evolution (Hadfield et al., 2018).