NATIONAL INSTITUTE FOR COMMUNICABLE DISEASES

Division of the National Health Laboratory Service

Weekly respiratory pathogens report

Week 29 of 2023

Highlights

- The 2023 influenza season started in week 17 (week starting 24 April 2023) when the influenza detection rate (3-week moving average) breached the seasonal threshold and peaked in week 22 (week starting on 4 June 2023). Influenza transmission and impact is currently below seasonal threshold, and circulation continues to decline.

- In 2023 to date, 941 influenza cases have been detected from all surveillance programmes, of which 99% (847/857) of those with typing information available were influenza A(H3N2). The majority of cases were reported from Western Cape 34% (320/941), followed by Gauteng 28% (259/941), North West 14% (128/941), KwaZulu-Natal 10% (95/941), Mpumalanga 10% (96/941), Eastern Cape 4% (39/941), Limpopo <1% (2/941) and Free State <1% (2/941) sentinel surveillance sites.

- In 2023 to date, 735 respiratory syncytial virus (RSV) cases have been detected from all surveillance programmes. The RSV season ended in week 21 (week starting 22 May 2023), although circulation of RSV continues.

- In 2023 to date, 142 cases of *Bordetella pertussis* were detected, of which 26% (37/142) were from Gauteng Province, 22% (31/142) North West Province, 17% (24/142) from Mpumalanga Province 17% (24/142) Western Cape Province, 14% (20/142) from KwaZulu-Natal Province and 4% (6/142) from Eastern Cape Province.

- In 2023 to date, 268 COVID-19 cases were detected from all surveillance programmes. Of the 251 specimens sequenced, a variant could be assigned in 67% (169/251). Of these, 98% (166/169) were assigned the Omicron variant, of which 57% (94/166) were Omicron (23A/XBB.1.5), 13% (22/166) were Omicron (22B/BA.5), 13% (22/166) were Omicron (22E/BQ.1.1), 11% (18/166) were Omicron (22F/BA.2.10.1), 2% (4/166) Omicron (23B/XBB.1.16), and 2% (3/166) each Omicron (21K/BA.1) and Omicron (22D/BM.1.1). One (0.4%, 1/251) was assigned XAY, XBF and XBL each respectively, while for the remaining 33% (82/251), a variant could not be assigned due to a low viral load or insufficient sample.
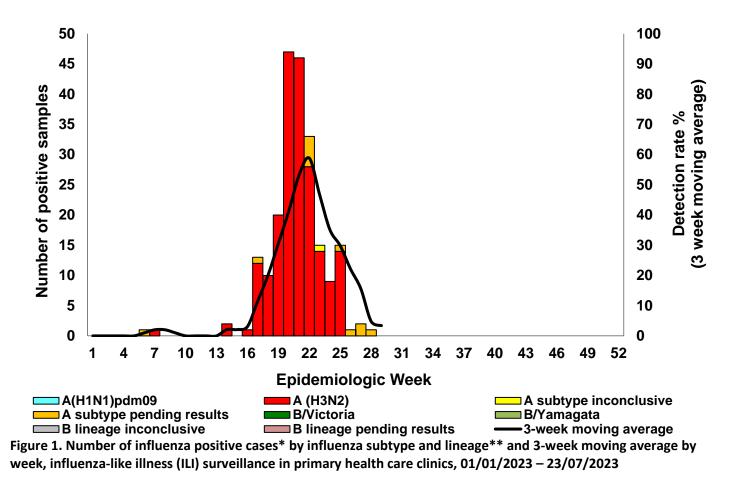
Programme Descriptions

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

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, Very 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

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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 – 23/07/2023**

*Specimens from patients with influenza-like illnesses at 5 sentinel sites in 4 provinces
** Influenza A(H3N2) was detected in 10/24, 42% 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

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 – 23/07/2023

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	39	0	2	0	0	0	0	172
Eastridge (WC)	0	30	0	5	0	0	0	0	201
Edendale Gateway (KZ)	0	51	1	3	0	0	0	0	363
Jouberton (NW)	0	75	0	1	0	0	0	0	253
Mitchell's Plain (WC)	0	9	0	1	0	0	0	0	94
Total:	**0**	**204**	**1**	**12**	**0**	**0**	**0**	**0**	**1083**

KZ: KwaZulu-Natal; NW: North West; WC: Western Cape; MP: Mpumalanga
* Influenza A(H3N2) was detected in 10/24, 42% 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

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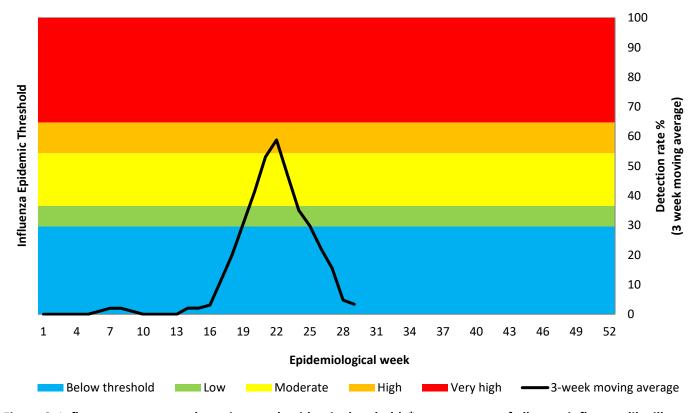
Figure 2. Influenza percentage detections and epidemic thresholds* among cases of all ages, influenza-like illness (ILI) surveillance in primary health care clinics, 01/01/2023 –23/07/2023
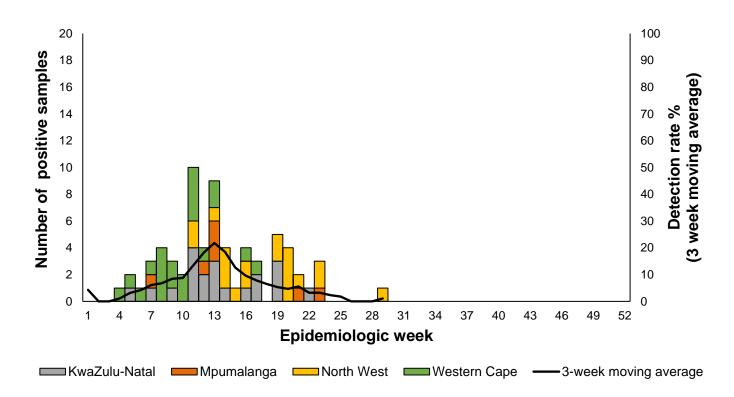*Thresholds based on 2012-2019 data

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 –23/07/2023
*RSV was detected in 1/24, 4% 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.

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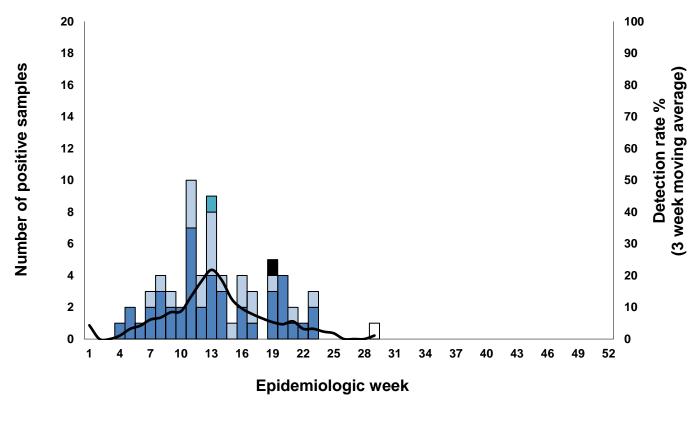
Figure 4. Number of patients testing positive for respiratory syncytial virus* by subgroup and 3-week moving average by week, influenza-like illness (ILI) surveillance in primary health care clinics, 01/01/2023 –23/07/2023

*RSV was detected in 1/24, 4% 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

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 –23/07/2023

Clinic (Province)	RSVA	RSVB	RSVAB**	RSV subgroup inconclusive***	RSV subgroup pending****	Total samples
Agincourt (MP)	2	4	1	0	0	172
Eastridge (WC)	18	2	0	0	0	201
Edendale Gateway (KZ)	8	11	0	1	0	363
Jouberton (NW)	14	4	0	0	1	253
Mitchell's Plain (WC)	1	0	0	0	0	94
Total	43	21	1	1	1	1083

KZ: KwaZulu-Natal; NW: North West; WC: Western Cape; MP: Mpumalanga
*RSV was detected in 1/24, 4% 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

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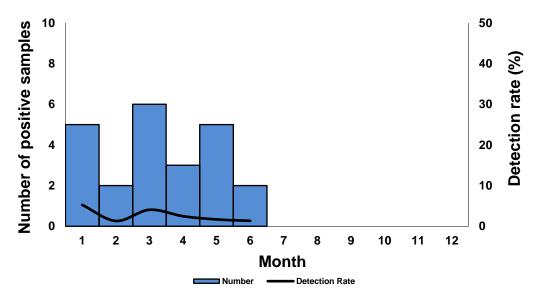
Figure 5. Number of patients testing positive for *B. pertussis** and detection rate by month, influenza-like illness (ILI) surveillance primary health care clinics**, 01/01/2023 –23/07/2023

**B. pertussis* was detected in 1/24, 4% 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

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 –23/07/2023

Clinic (Province)	*B. pertussis* Positive	Total samples
Agincourt (MP)	4	157
Eastridge (WC)	2	179
Edendale Gateway (KZ)	6	318
Jouberton (NW)	11	230
Mitchell's Plain (WC)	0	78
Total:	23	962

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

**B. pertussis* was detected in 1/24, 4% 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.
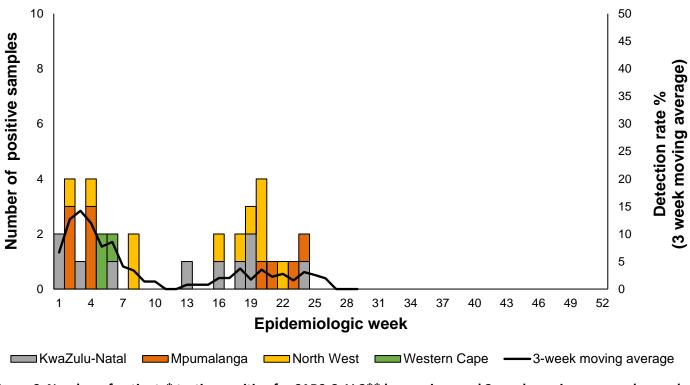
NB: Results pending for 49 samples.

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 –23/07/2023

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

**SARS-CoV-2 was not detected in 24 specimens from patients who met 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.

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 –23/07/2023

Clinic (Province)	SARS-CoV-2 positive	Total samples tested
Agincourt (MP)	10	172
Eastridge (WC)	1	201
Edendale Gateway (KZ)	10	363
Jouberton (NW)	11	253
Mitchell's Plain (WC)	2	94
Total:	**34**	**1083**

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

*SARS-CoV-2 was not detected in 24 specimens from patients who met 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.

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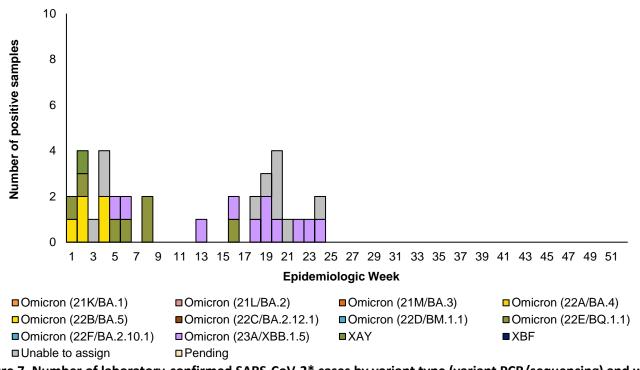
Figure 7. Number of laboratory-confirmed SARS-CoV-2* cases by variant type (variant PCR/sequencing) and week, influenza-like illness (ILI) surveillance in primary health care clinics, 01/01/2023-23/07/2023

*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

Unable to assign: no lineage assigned due to poor- sequence quality **OR** low viral load ($C_t \geq 35$) **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, 01/01/2023-23/07/2023**

Province	Omicron (21L/BA.2)	Omicron (21M/ BA.3)	Omicron (21A/BA.4)	Omicron (22B/ BA.5)	Omicron (22C/BA.2.12.1)	Omicron (22D/BM.1.1)	Omicron (22E/BQ.1.1)	Omicron (22F/BA.2.10.1)	Omicron (23A/XBB.1.5)	XAY	Unable to assign**	Pending***	SARS-CoV-2 positive	Total samples tested
Agincourt Clinic (MP)	0	0	0	4	0	0	0	0	3	1	2	0	10	172
Eastridge Clinic (WC)	0	0	0	0	0	0	0	0	1	0	0	0	1	201
Edendale Clinic (KZ)	0	0	0	1	0	0	3	0	4	0	2	0	10	382
Jouberton Clinic (NW)	0	0	0	0	0	0	3	0	2	0	6	0	11	258
Mitchell's Plain Clinic (WC)	0	0	0	0	0	0	1	0	1	0	0	0	2	94
Total:	0	0	0	5	0	0	7	0	11	1	10	0	34	1107

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

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

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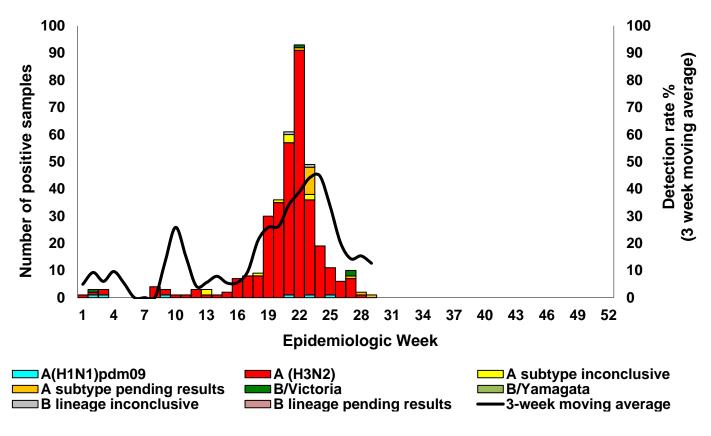
Figure 8. Number of positive patients* by influenza subtype and lineage and 3-week moving average by week, ILI surveillance - Viral Watch, 01/01/2023-23/07/2023

*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, ILI surveillance - Viral Watch, 01/01/2023-23/07/2023
*Thresholds based on 2015-2019 data

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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, 01/01/2023-23/07/2023

Province	A(H1N1) pdm09	A(H3N2)	A subtype inconclusive	A subtype pending results*	B/Victoria	B/Yamagata	B lineage inconclusive	B lineage pending results*	Total samples
Eastern Cape	0	17	0	0	0	0	0	0	36
Free State	0	2	0	0	0	0	0	0	2
Gauteng	3	120	4	4	1	0	0	1	553
Limpopo	0	2	0	0	0	0	1	0	6
Mpumalanga	1	17	2	0	0	0	0	0	37
North West	0	3	0	0	0	0	0	0	3
Northern Cape	0	0	0	0	0	0	0	0	0
Western Cape	2	171	3	11	3	0	1	0	346
Total:	**6**	**332**	**9**	**15**	**4**	**0**	**2**	**1**	**983**

*Inconclusive: insufficient viral load in sample and unable to characterise further
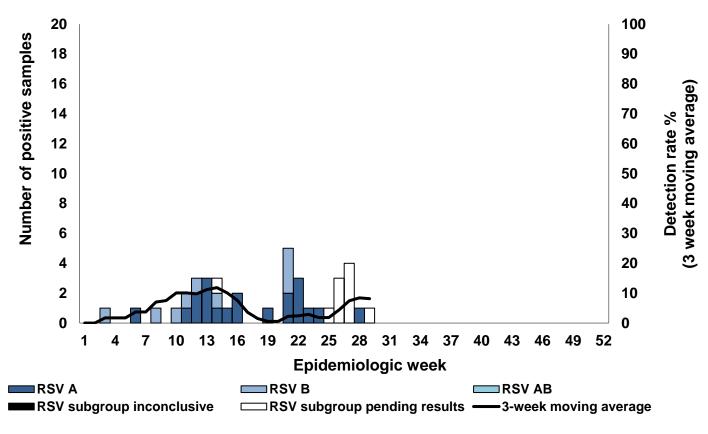**Influenza A subtype or B lineage results are pending

Figure 9. Number of RSV positive cases testing positive for respiratory syncytial virus (RSV)* by subgroup and 3-week moving average by week, ILI surveillance - Viral Watch, 01/01/2023-23/07/2023
*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, ILI surveillance - Viral Watch, 01/01/2023-23/07/2023

Province	RSV A	RSV B	RSV AB*	RSV subgroup inconclusive **	RSV subgroup pending results***	Total samples tested
Eastern Cape	1	1	0	0	0	36
Free State	0	0	0	0	0	2
Gauteng	10	5	0	0	9	553
Limpopo	0	0	0	0	0	6
Mpumalanga	1	0	0	0	1	37
North West	0	0	0	0	0	3
Northern Cape	0	0	0	0	0	0
Western Cape	9	3	0	0	0	346
Total:	21	9	0	0	10	983

*RSV AB: Both RSV A and B subgroup identified
**Inconclusive: insufficient viral load in sample and unable to characterise further
***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, ILI surveillance - Viral Watch, 01/01/2023-23/07/2023**
*Specimens from patients with influenza-like illnesses at 92 sentinel sites in 8 provinces

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Table 8. Number of SARS-CoV-2 positive cases identified and total number tested by province, ILI surveillance - Viral Watch, 01/01/2023-23/07/2023

Province	SARS-CoV-2 positive	Total samples tested
Eastern Cape	4	36
Free State	0	2
Gauteng	49	553
Limpopo	0	6
Mpumalanga	3	37
North West	0	3
Northern Cape	0	0
Western Cape	20	346
Total:	**76**	**983**

Legend:
- Omicron (21K/BA.1)
- Omicron (21L/BA.2)
- Omicron (21M/BA.3)
- Omicron (22A/BA.4)
- Omicron (22B/BA.5)
- Omicron (22C/BA.2.12.1)
- Omicron (22D/BM.1.1)
- Omicron (22E/BQ.1.1)
- Omicron (22F/XBB.1.9.1)
- Omicron (23A/XBB.1.5)
- Omicron (23B/XBB.1.16)
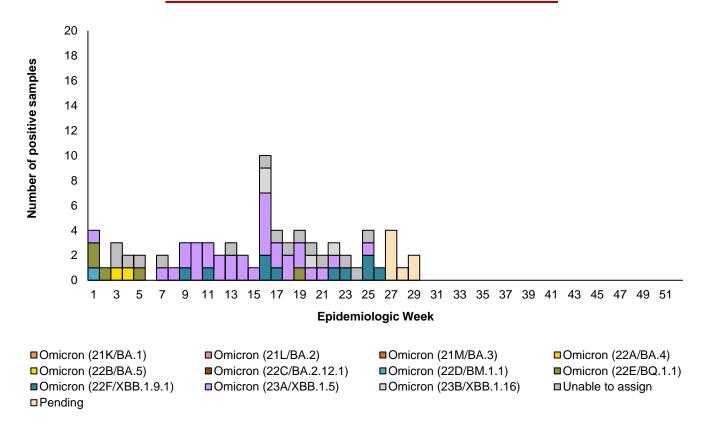- Unable to assign
- Pending

Figure 11. Number of laboratory confirmed SARS-CoV-2* cases by variant type (variant PCR/sequencing) and week, ILI surveillance - Viral Watch, 01/01/2023-23/07/2023

*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 (C$_t$ ≥35) **OR** variant PCR could not assign variant and no sequencing result
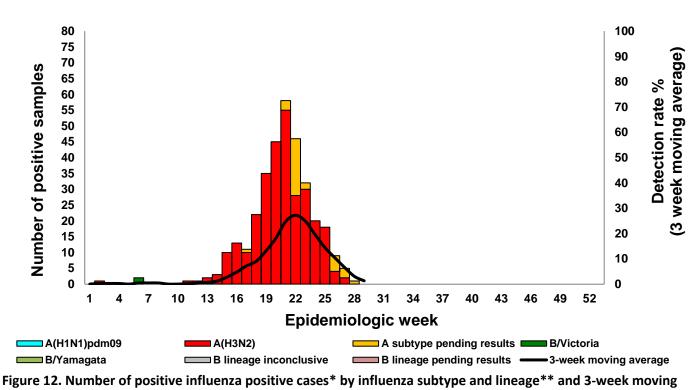**Pending**: outstanding variant results

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, 01/01/2023-23/07/2023

Clinic (Province)	Omicron (21L/BA.2)	Omicron (21M/BA.3)	Omicron (22A/BA.4)	Omicron (22B/BA.5)	Omicron (22C/BA.2.12.1)	Omicron (22D/BM.1.1)	Omicron (22E/BQ.1.1)	Omicron (22F/XBB.1.9.1)	Omicron (23A/XBB.1.5)	Omicron (23B/XBB.1.16)	Unable to assign**	Pending***	Total SARS-CoV-2 positive	Total samples tested
Eastern Cape	0	0	0	0	0	0	0	1	2	0	1	0	4	36
Free State	0	0	0	0	0	0	0	0	0	0	0	0	0	2
Gauteng	0	0	0	2	0	1	3	7	24	2	5	5	49	553
Limpopo	0	0	0	0	0	0	0	0	0	0	0	0	0	6
Mpumalanga	0	0	0	0	0	0	0	0	1	0	1	1	3	37
North West	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
Western Cape	0	0	0	0	0	0	2	2	5	2	8	1	20	346
Total:	0	0	0	2	0	1	5	10	32	4	15	7	76	983

*Specimens from patients with influenza-like illnesses at 92 sentinel sites in 8 provinces
**No cases of Alpha, Beta or 20D (C.1.2) variants detected.
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

Figure 12. Number of positive influenza positive cases* by influenza subtype and lineage and 3-week moving average by week, pneumonia surveillance public hospitals, 01/01/2023-23/07/2023**

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

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Table 10. Number of laboratory confirmed influenza cases by subtype and lineage* and total number of samples tested by hospital, pneumonia surveillance public hospitals, 01/01/2023-23/07/2023

Hospital (Province)	A(H1N1)pdm09	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	1	0	0	0	467
Helen Joseph-Rahima Moosa (GP)	0	57	1	7	0	0	0	0	847
Khayelitsha (WC)	0	25	1	4	1	0	0	0	401
Klerksdorp-Tshepong (NW)	0	46	1	1	0	0	0	0	350
Livingstone (EC)	0	21	1	0	0	0	0	0	436
Mapulaneng-Matikwana (MP)	0	17	1	6	0	0	0	0	306
Mitchell's Plain (WC)	0	12	0	4	0	0	1	0	307
Red Cross (WC)	0	24	1	2	0	0	0	0	609
Tambo Memorial (GP)	0	27	1	6	0	0	0	0	350
Tembisa (GP)	0	25	2	1	0	0	0	0	355
Tintswalo (MP)	0	10	0	1	0	0	0	0	194
Tygerberg (WC)	0	8	0	1	0	0	0	0	105
Total:	0	301	10	33	2	0	1	0	4727

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

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

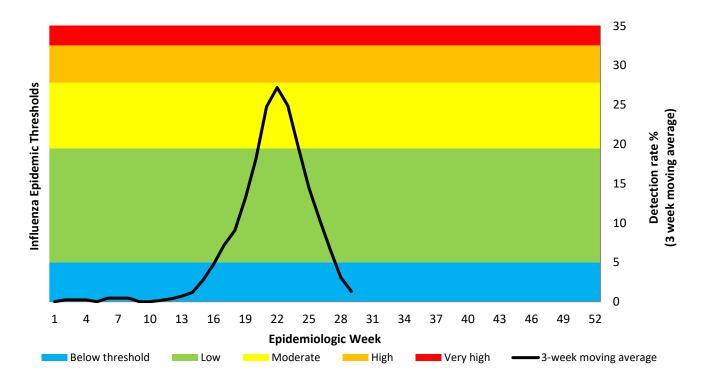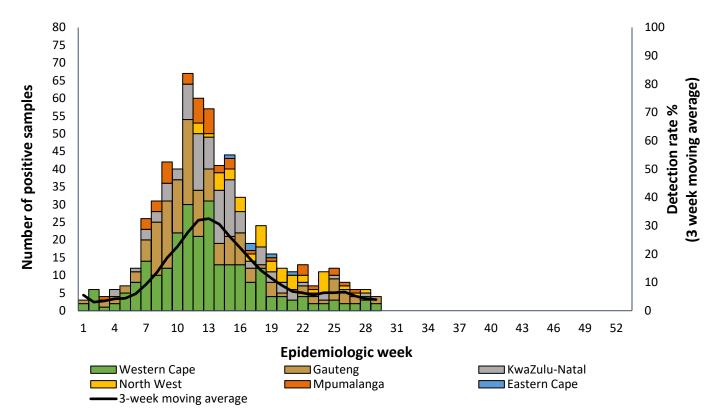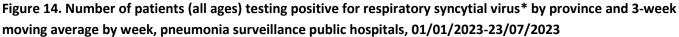***Influenza A subtype or B lineage results are pending

Figure 13. Influenza percentage detections and epidemic thresholds* among cases of all ages, pneumonia surveillance public hospitals, 01/01/2023-23/07/2023

*Thresholds based on 2010-2019 data

Data are provisional as reported to date (Data for this report drawn on 26/07/2023). Number of consultations/specimens are reported/analysed by date of consultation/specimen collection. Data cleaning is ongoing and this may result in some changes in subsequent reports.

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, 01/01/2023-23/07/2023

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.
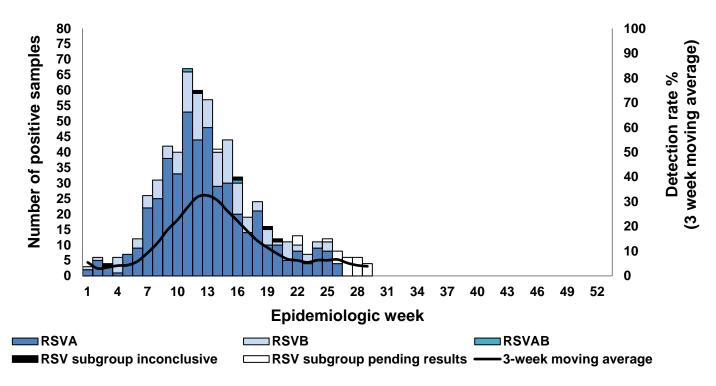

Figure 15. Number of patients (<u>all ages</u>) testing positive for respiratory syncytial virus* by subgroup and 3-week moving average by week, pneumonia surveillance public hospitals, 01/01/2023-23/07/2023

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

Data are provisional as reported to date (Data for this report drawn on 26/07/2023). Number of consultations/specimens are reported/analysed by date of consultation/specimen collection. Data cleaning is ongoing and this may result in some changes in subsequent reports.

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, 01/01/2023-23/07/2023

Hospital (Province)	RSVA	RSVB	RSVAB**	RSV subgroup inconclusive***	RSV subgroup pending****	Total samples
Edendale (KZ)	37	71	1	1	2	467
Helen Joseph-Rahima Moosa (GP)	138	10	0	1	7	847
Khayelitsha (WC)	4	3	0	0	2	401
Klerksdorp-Tshepong (NW)	42	2	0	1	2	350
Livingstone (EC)	3	2	0	0	0	436
Mapulaneng-Matikwana (MP)	13	5	1	0	0	306
Mitchell's Plain (WC)	63	10	0	0	4	307
Red Cross (WC)	133	25	0	2	6	609
Tambo Memorial (GP)	2	2	0	0	0	350
Tembisa (GP)	1	2	0	0	1	355
Tintswalo (MP)	25	0	0	0	1	194
Tygerberg (WC)	1	1	0	0	0	105
Total:	**462**	**133**	**2**	**5**	**25**	**4727**

*No cases who met suspected SARS-CoV-2 or *B. pertussis* case definition but did not meet pneumonia (SRI) case definition.
**RSV AB: Both RSV A and B subgroup identified
***Inconclusive: insufficient viral load in sample and unable to characterise further
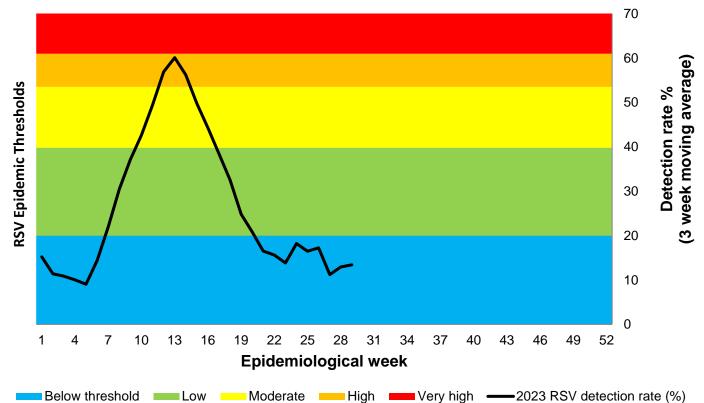****RSV results for subgroups are pending

Figure 16. RSV percentage 3-week moving average and epidemic thresholds* among children aged < 5 years, pneumonia surveillance public hospitals, 01/01/2023-23/07/2023
*Thresholds based on 2010-2019 data

Data are provisional as reported to date (Data for this report drawn on 26/07/2023). Number of consultations/specimens are reported/analysed by date of consultation/specimen collection. Data cleaning is ongoing and this may result in some changes in subsequent reports.
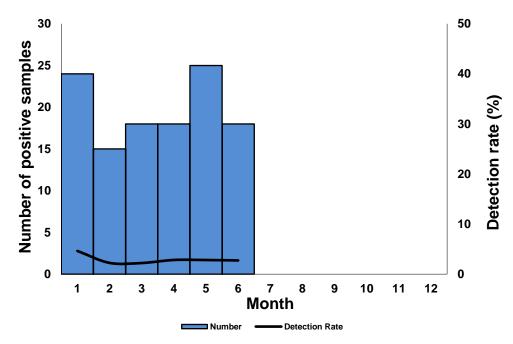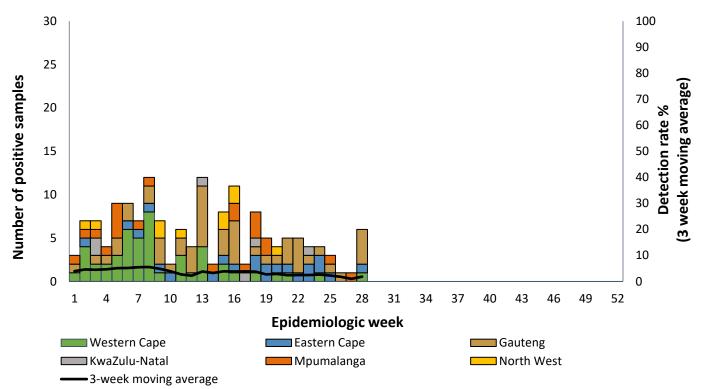
Figure 17. Number of patients testing positive for *B. pertussis** and 3-week moving average by month, pneumonia surveillance public hospitals**, 01/01/2023-23/07/2023
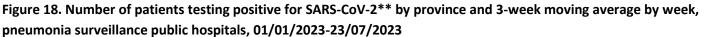
*No cases who met the suspected SARS-CoV-2 or *B. pertussis* case definition but did not meet Pneumonia Surveillance case definition.
*Specimens from patients hospitalised with pneumonia at 15 sentinel sites in 6 provinces.

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, 01/01/2023-23/07/2023

Hospital (Province)	*B. pertussis* Positive	Total samples
Edendale (KZ)	14	422
Helen Joseph-Rahima Moosa (GP)	25	730
Khayelitsha (WC)	3	340
Klerksdorp-Tshepong(NW)	19	316
Livingstone (EC)	6	387
Mapulaneng-Matikwana (MP)	17	266
Mitchell's Plain (WC)	3	290
Red Cross (WC)	14	558
Tambo Memorial (GP)	6	295
Tembisa (GP)	6	302
Tintswalo (MP)	3	176
Tygerberg (WC)	2	94
Total:	118	4176

*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.

Data are provisional as reported to date (Data for this report drawn on 26/07/2023). Number of consultations/specimens are reported/analysed by date of consultation/specimen collection. Data cleaning is ongoing and this may result in some changes in subsequent reports.

Figure 18. Number of patients testing positive for SARS-CoV-2** by province and 3-week moving average by week, pneumonia surveillance public hospitals, 01/01/2023-23/07/2023

*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.

Table 13. Number of patients positive for SARS-CoV-2* and total number of samples tested by hospital, pneumonia surveillance public hospitals, 01/01/2023-23/07/2023

Hospital (Province)	SARS-CoV-2 positive	Total samples tested
Edendale (KZ)	6	467
Helen Joseph-Rahima Moosa (GP)	19	847
Khayelitsha (WC)	16	401
Klerksdorp-Tshepong (NW)	10	350
Livingstone (EC)	22	436
Mapulaneng-Matikwana (MP)	16	306
Mitchell's Plain (WC)	15	307
Red Cross (WC)	13	609
Tambo Memorial (GP)	20	350
Tembisa (GP)	12	355
Tintswalo (MP)	5	194
Tygerberg (WC)	4	105
Total:	**158**	**4727**

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

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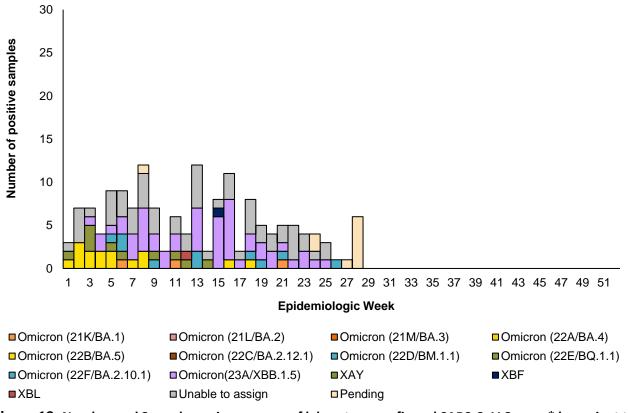
Figure 19. Number and 3-week moving average of laboratory-confirmed SARS-CoV-2 cases* by variant type (variant PCR/sequencing), pneumonia surveillance public hospitals, 01/01/2023-23/07/2023

*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 (C$_t$≥35) **OR** variant PCR could not assign variant and no sequencing result
Pending: outstanding variant results

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, 01/01/2023-23/07/2023

Hospital (Province)	Omicron (21K/BA.1))	Omicron (22A/BA.4)	Omicron (22B/BA.5)	Omicron (22C/BA.2.12.1)	Omicron (22D/BM.1.1)	Omicron(22E/BQ.1.1)	Omicron (22F/BA.2.10.1)	Omicron(23A/XBB.1.5)	XBF	XBL	Unable to assign**	Pending***	Total SARS-CoV-2 positive	Total samples tested
Edendale (KZ)	0	0	0	0	0	1	1	1	0	0	3	0	6	467
Helen Joseph-Rahima Moosa (GP)	1	0	1	0	0	1	3	9	0	0	4	0	19	847
Khayelitsha (WC)	1	0	1	0	0	1	0	5	0	0	8	0	16	401
Klerksdorp-Tshepong (NW)	0	0	1	0	0	2	0	5	0	0	2	0	10	350
Livingstone (EC)	1	0	1	0	2	0	0	5	1	0	9	3	22	436
Mapulaneng-Matikwana (MP)	0	0	3	0	0	1	0	2	0	0	9	1	16	306
Mitchell's Plain (WC)	0	0	1	0	0	0	2	7	0	0	4	1	15	307
Red Cross (WC)	0	0	4	0	0	0	1	4	0	1	2	1	13	609
Tambo Memorial (GP)	0	0	0	0	0	3	0	10	0	0	4	3	20	350
Tembisa (GP)	0	0	1	0	0	1	1	1	0	0	7	1	12	355
Tintswalo (MP)	0	0	1	0	0	0	0	0	0	0	4	0	5	194
Tygerberg (WC)	0	0	1	0	0	0	0	2	0	0	1	0	4	105
Total:	3	0	15	0	2	10	8	51	1	1	57	10	158	4727

*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 (C$_t$≥35) **OR** variant PCR could not assign variant and no sequencing result
Pending: outstanding variant results

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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 Allplex™ 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 ≥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.
Variant PCR

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.

<u>Extraction:</u> 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 µl per sample, in order to increase yields. 300 µl of each sample was used for automated magnetic bead-based extraction using the Chemagic 360. RNA was eluted in 60 µl 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, (http://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).

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