

WEEKLY RESPIRATORY PATHOGENS SURVEILLANCE REPORT

SOUTH AFRICA

WEEK 44 2021



NATIONAL INSTITUTE FOR
COMMUNICABLE DISEASES

Division of the National Health Laboratory Service

CONTENTS

| | |
|--|-------|
| Surveillance programme description | 2 |
| Comments | 3 |
| Systematic Influenza-like illness (ILI) surveillance | 4-9 |
| Influenza | |
| Respiratory syncytial virus | |
| SARS-CoV-2 | |
| Influenza-like illness (ILI) Viral Watch | 10-13 |
| Influenza | |
| SARS-CoV-2 | |
| National syndromic surveillance for pneumonia | 14-19 |
| Influenza | |
| Respiratory syncytial virus | |
| SARS-CoV-2 | |
| Summary of laboratory confirmed SARS-CoV-2 cases | 20-21 |
| SARS-CoV-2 Testing Methods | 22 |

HIGHLIGHTS: WEEK 44

- The 2021 influenza season has not yet started although sustained detections of influenza continue in all surveillance programmes. In week 44, transmission is high and impact is low.

- 23 new cases of influenza from Western Cape (n=2), North West (n=12), Kwa-Zulu Natal (n=8) and Mpumalanga (n=1) surveillance sites were detected in week 44. To date, 236 influenza cases have been detected from Gauteng, Western Cape, North West, Eastern Cape, Mpumalanga and KwaZulu-Natal sentinel surveillance sites. From 1 January 2021 to date, Influenza B (Victoria) was the most commonly detected lineage in both influenza-like illness (ILI) surveillance (n=27/85, 32%) and pneumonia surveillance (n=46/112, 41%).

- RSV activity remains below seasonal threshold in both influenza-like illness (ILI) and pneumonia surveillance programmes. From 1 January 2021 to date, RSV subgroup A was the most commonly detected subgroup in both ILI surveillance (n=40/74, 54%) and pneumonia surveillance (n=212/407, 52%).

- From 2 March 2020 to date, a total of 2 903 COVID-19 cases were detected from all surveillance programmes. A sustained decline in detection rate has been noted in both ILI programme and pneumonia surveillance. Of the 2 058 hospitalised COVID-19 cases reported with available data on outcome, 346 (17%) died.

- From 1 January 2021 to date, of the 1192/1291 (92%) SARS-CoV-2 positive cases with variant type results, Delta (425/871, 49%) and Beta (157/321, 49%), were the most detected variants in pneumonia surveillance and in ILI, respectively. Delta variant predominated in both programmes (from week 22, week starting 31st May 2021 until week 38). Beta variant predominated from week 47 of 2020 to week 21 of 2021.

INFLUENZA, RESPIRATORY SYNCYTIAL VIRUS AND SARS-CoV-2 SURVEILLANCE REPORT

WEEK 44 2021

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 | GP KZ MP NW WC |
| Type of site | Primary health care clinics | General practitioners | Public hospitals |
| Case definition | <p>ILI: An acute respiratory illness with a temperature ($\geq 38^{\circ}\text{C}$) and cough, & onset ≤ 10 days</p> <p>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:</p> <ul style="list-style-type: none"> • paroxysms of coughing, • or inspiratory “whoop”, • or post-tussive vomiting • or apnoea in children < 1 year; <p>OR</p> <p>Any person in whom a clinician suspects pertussis</p> <p>Suspected SARS-CoV-2 Any person presenting with an acute (≤ 14 days) respiratory tract infection or other clinical illness compatible with COVID-19[§]</p> | <p>ILI: An acute respiratory illness with a temperature ($\geq 38^{\circ}\text{C}$) and cough, & onset ≤ 10 days</p> <p>Suspected SARS-CoV-2 Any person presenting with an acute (≤ 14 days) respiratory tract infection or other clinical illness compatible with COVID-19[§]</p> | <p>SRI: Acute (symptom onset ≤ 10 days) or chronic (symptom onset > 10) lower respiratory tract infection</p> <p>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:</p> <ul style="list-style-type: none"> • paroxysms of coughing, • or inspiratory “whoop”, • or post-tussive vomiting • or apnoea in children < 1 year; <p>OR</p> <p>Any person in whom a clinician suspects pertussis.</p> <p>Suspected SARS-CoV-2 Any person admitted with a physician-diagnosis of suspected COVID-19 and not meeting SRI case definition.</p> |
| 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 BP SARS-CoV-2 | INF RSV BP SARS-CoV-2 |
| Testing Methods | <p>INF and RSV - Fast-Track Diagnostics multiplex real-time reverse transcription polymerase chain reaction (until 31 March 2021)</p> <p>B. pertussis Multiplex real-time PCR (Tatti et al., J Clin Microbiol 2011) and culture (if PCR cycle threshold ≤ 25)</p> <p>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</p> <ul style="list-style-type: none"> • positivity assigned if PCR cycle threshold is < 40 for ≥ 1 gene targets (N, S, OR RdRp) | <p>INF and RSV - Fast-Track Diagnostics multiplex real-time reverse transcription polymerase chain reaction (until 31 March 2021)</p> <p>B. pertussis Multiplex real-time PCR (Tatti et al., J Clin Microbiol 2011) and culture (if PCR cycle threshold ≤ 25)</p> <p>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</p> <ul style="list-style-type: none"> • positivity assigned if PCR cycle threshold is < 40 for ≥ 1 gene targets (N, S, OR RdRp) | <p>INF and RSV - Fast Track Diagnostics multiplex real-time reverse transcription polymerase chain reaction (until 31 March 2021)</p> <p>B. pertussis Multiplex real-time PCR (Tatti et al., J Clin Microbiol 2011) and culture (if PCR cycle threshold ≤ 25)</p> <p>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</p> <ul style="list-style-type: none"> • positivity assigned if PCR cycle threshold is < 40 for ≥ 1 gene targets (N, S, OR RdRp) |

INFLUENZA, RESPIRATORY SYNCYTIAL VIRUS AND SARS-CoV-2 SURVEILLANCE REPORT

WEEK 44 2021

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 (Viral Watch Programme) 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.

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

**Started in 2019

***Started in November 2020

****INF: influenza virus; RSV: respiratory syncytial virus; BP: *Bordetella pertussis*; SARS-CoV-2: severe acute respiratory syndrome coronavirus 2

§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).

COMMENTS

Influenza

The 2021 influenza season has not yet started although sustained detections of influenza continue in all surveillance programmes. Since the first influenza positive case of 2021 was detected in pneumonia surveillance in week 9 of 2021 (week ending on the 07 March 2021), sporadic cases have been reported from week 16 to date. Of the 236 influenza cases detected in surveillance sites in 2021, the majority (n=95, 40%) were influenza B(Victoria). In week 44, transmission is high and impact is low.

ILI programme: In 2021 to date, specimens from 1 628 patients meeting ILI case definition were received from 4 ILI sites. Influenza was detected in 85 (5%) patients, of which 22 (26%) were influenza A(H1N1)pdm09, eight (9%) influenza A(H3N2), one (1%) influenza A(inconclusive), 15 (18%) influenza A(pending subtype results), 27 (32%) influenza B(Victoria), four (5%) influenza B(lineage inconclusive) and eight influenza B(lineage pending results). (Fig1, Table1).

Viral Watch programme: In 2021 to date, specimens were received from 191 patients from Viral Watch sites in 5 of the 8 provinces participating in surveillance. Influenza was detected in 17 (9%) patients, of which two (12%) were influenza A(H1N1)pdm09, five (29%) influenza A(pending results), five (29%) influenza B(Victoria), four (24%) influenza B(lineage inconclusive) and one (6%) influenza B(lineage pending results). (Fig7, Table5)

Pneumonia surveillance: Since the beginning of 2021, specimens from 5 338 patients with severe respiratory illness (SRI) were received from the 6 sentinel sites. Influenza was detected in 112 (2%) patients, of which 28 (25%) were influenza A(H1N1)pdm09, 18 (16%) influenza A(H3N2), two (2%) influenza A(subtype inconclusive), 10 (9%) influenza A(pending subtype results), 46 (41%) influenza B(Victoria), seven (6%) were influenza B(lineage inconclusive) and one (1%) influenza B(lineage pending results). (Fig12, Table9)

In addition, influenza was detected in 22 (3%) of 798 specimens, of which four (18%) were influenza A(H1N1)pdm09, 17 (77%) influenza B(Victoria) and one (5%) was influenza B(inconclusive) from patients who met suspected SARS-CoV-2 case definition but did not meet pneumonia/ILI surveillance case definition.

Respiratory syncytial virus

In 2021 to date, RSV detection has been reported from all surveillance programmes, activity remains below seasonal threshold. Of the 508 RSV cases detected in 2021, the majority (n=262, 52%) were RSV subgroup A.

ILI programme: In 2021 to date, 1 628 specimens from patients meeting the ILI case definition were tested and RSV was detected in 74 (5%) patients. Of which, 40 (54%) were RSV subgroup A, 31 (42%) RSV subgroup B, two (3%) RSV subgroup A and B and one (1%) was RSV (subgroup inconclusive). (Fig4, Table2)

Viral Watch programme: In 2021 to date, 191 specimens from viral watch patients were tested and RSV was detected in specimens of six (3%) patients. Of which, three (50%) were RSV subgroup A, two (33%) RSV subgroup B and one (17%) was RSV (subgroup inconclusive). (Fig9, Table6)

Pneumonia surveillance: Since the beginning of 2021, 5 338 specimens were tested and RSV was detected in specimens of 407 (8%) patients. Of which, 212 (52%) were RSV subgroup A, 190 (47%) RSV subgroup B, four (1%) RSV (subgroup inconclusive) and one (0.2%) RSV (subgroup pending). (Fig14, Table10)

In addition, RSV was detected in 21 of 798 (3%) specimens from patients who met suspected SARS-CoV-2 case definition but did not meet the pneumonia/ILI surveillance case definitions.

SARS-CoV-2 (Severe acute respiratory syndrome coronavirus 2)

ILI programme: From March 2020 to date, 2 942 patients were tested and SARS-CoV-2 was detected in 568 (19%) patients. From 1 January 2021 to date, of the 321/338 (95%) with data on variant type, majority (157/321, 49%) were Beta variant which predominated from week1 to week 24, followed by Delta (139/321, 43%) variant which predominated from week 25 to week 36. (Fig6, Table4)

Viral Watch programme: From March 2020 to date, 470 patients presenting with ILI were tested and SARS-CoV-2 was detected in 84 (18%) patients. From 1 January 2021, of the 21/84 (25%) with data on variant type, majority were (19/21, 90%) were Delta variant which dominated from week 25 to week 30. (Fig11, Table8)

Pneumonia surveillance: From March 2020 to date, 9 361 patients with severe respiratory illness (SRI) were tested and SARS-CoV-2 was detected in 2 058 (22%) patients. From 1 January 2021 to date, of the 871/954 (91%) with data on variant type, majority were (425/871, 49%) were Delta variant which dominated from week 22 to week 38 followed by Beta (401/871, 46%) variant which dominated from week 1 to 25. (Fig17, Table12)

In addition, SARS-CoV-2 was detected in 193 of 1 001 (19%) specimens from patients who met suspected SARS-CoV-2 case definition but did not meet the pneumonia/ILI surveillance case definitions.

INFLUENZA, RESPIRATORY SYNCYTIAL VIRUS AND SARS-CoV-2 SURVEILLANCE REPORT

WEEK 44 2021

INFLUENZA-LIKE ILLNESS (ILI) SURVEILLANCE PRIMARY HEALTH CARE CLINICS

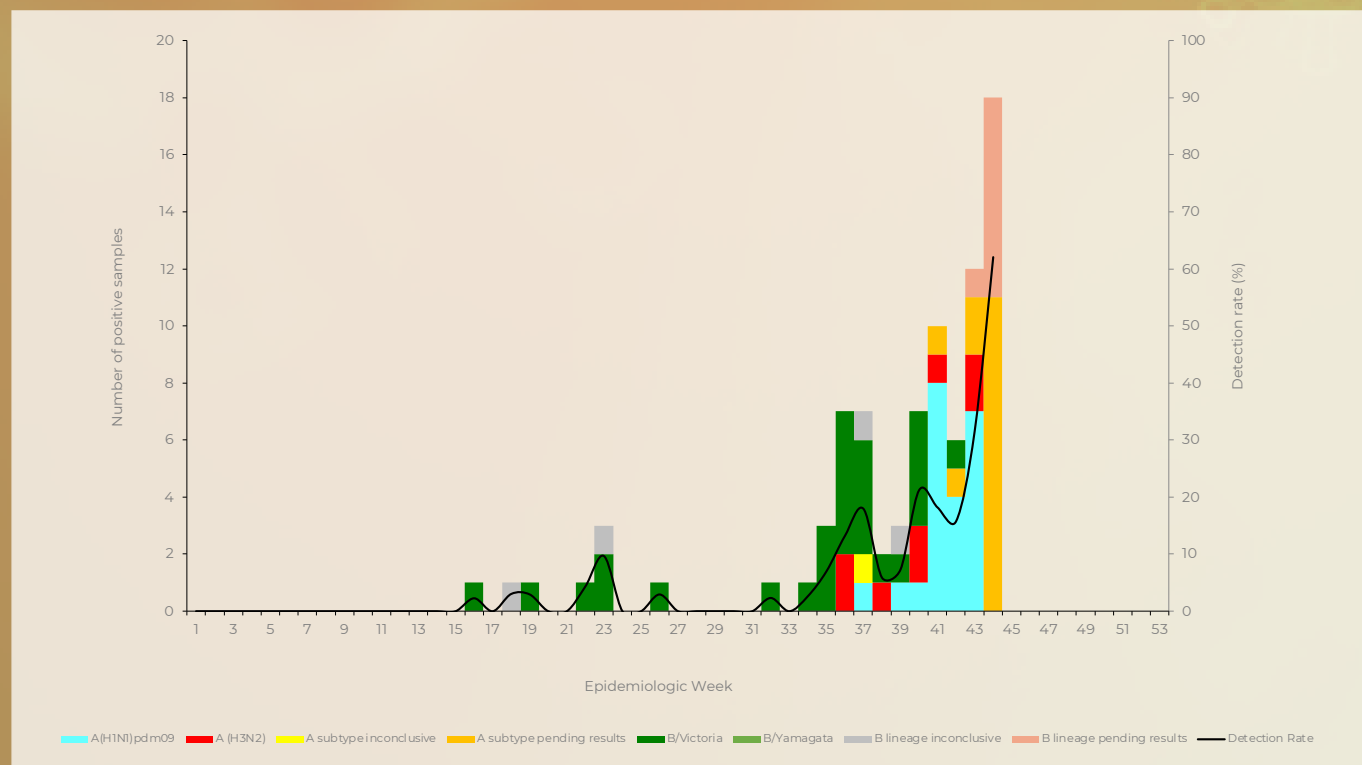


Figure 1. Number of influenza positive cases* by influenza subtype and lineage** and detection rate*** by week, Influenza-like illness (ILI) surveillance in primary health care clinics, 04/01/2021 – 07/11/2021

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

**Influenza was detected in 16 (3%) of 606 specimens, of which 2 (13%) were influenza A(H1N1)pdm09, 13 (81%) influenza B(Victoria) and one (6%) was influenza B(inconclusive) from patients who met suspected SARS-CoV-2 case definition but did not meet Influenza-like illness (ILI) case definition. These are not included in the epidemiological curve.

***Only reported for weeks with >10 specimens submitted

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, 04/01/2021 – 07/11/2021

| Clinic (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 |
|-----------------------|------------------|----------|---------------------------|--|----------------|----------------|---------------------------|--|------------------|
| Agincourt (MP) | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 218 |
| Eastridge (WC) | 0 | 4 | 0 | 0 | 6 | 0 | 0 | 0 | 219 |
| Edendale Gateway (KZ) | 0 | 1 | 0 | 0 | 6 | 0 | 2 | 8 | 225 |
| Jouberton (NW) | 20 | 3 | 1 | 13 | 12 | 0 | 1 | 0 | 736 |
| Mitchell's Plain (WC) | 1 | 0 | 0 | 2 | 3 | 0 | 1 | 0 | 230 |
| Total: | 22 | 8 | 1 | 15 | 27 | 0 | 4 | 8 | 1 628 |

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

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

**Influenza was detected in 16 (3%) of 606 specimens, of which 2 (13%) were influenza A(H1N1)pdm09, 13 (81%) influenza B(Victoria) and one (6%) was influenza B(inconclusive) from patients who met suspected SARS-CoV-2 case definition but did not meet Influenza-like illness (ILI) case definition. These are not included in the table.

[§]Influenza A subtype or B lineage results are pending

INFLUENZA, RESPIRATORY SYNCYTIAL VIRUS AND SARS-CoV-2 SURVEILLANCE REPORT

WEEK 44 2021

INFLUENZA-LIKE ILLNESS (ILI) SURVEILLANCE PRIMARY HEALTH CARE CLINICS



Figure 2. Influenza percentage detections and epidemic thresholds*, Influenza-like illness (ILI) surveillance in primary health care clinics, 04/01/2021 – 07/11/2021

*Thresholds based on 2012-2019 data

INFLUENZA, RESPIRATORY SYNCYTIAL VIRUS AND SARS-CoV-2 SURVEILLANCE REPORT

WEEK 44 2021

INFLUENZA-LIKE ILLNESS (ILI) SURVEILLANCE PRIMARY HEALTH CARE CLINICS

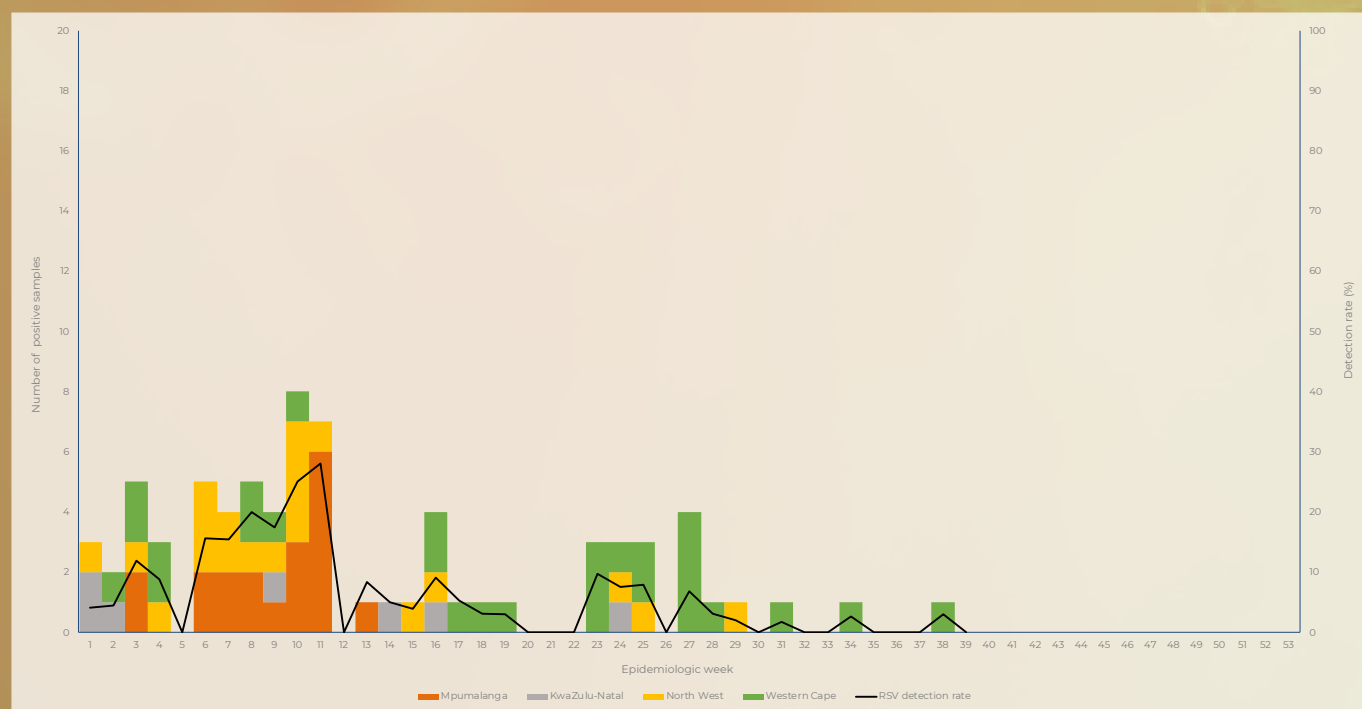


Figure 3. Number of patients testing positive for respiratory syncytial virus* by province and detection rate** by week, Influenza-like illness (ILI) surveillance in primary health care clinics, 04/01/2021 – 07/11/2021

**RSV was detected from 15 of 614 (2%) specimens from patients who met suspected SARS-CoV-2 case definition but did not meet influenza-like illness (ILI) case definition. These are not included in the epidemiological curve.

INFLUENZA, RESPIRATORY SYNCYTIAL VIRUS AND SARS-CoV-2 SURVEILLANCE REPORT

WEEK 44 2021

INFLUENZA-LIKE ILLNESS (ILI) SURVEILLANCE PRIMARY HEALTH CARE CLINICS

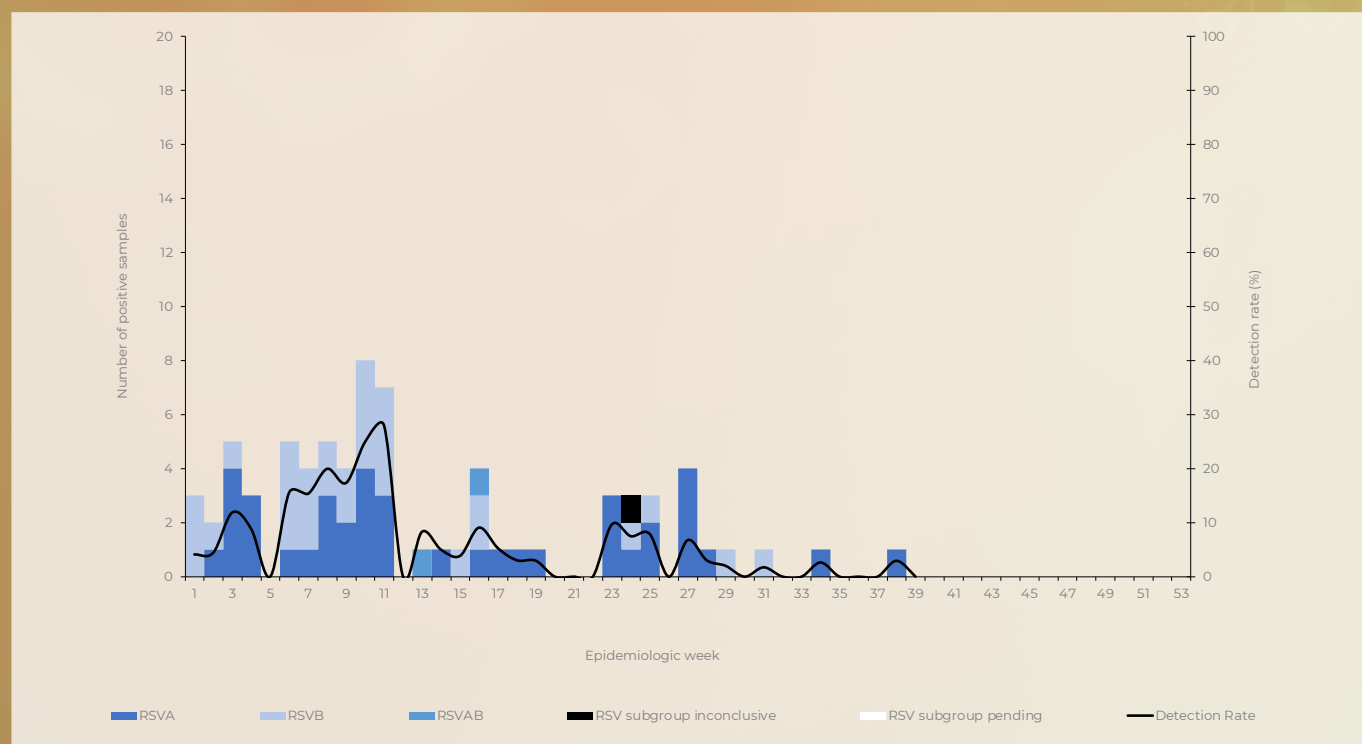


Figure 4. Number of patients testing positive for respiratory syncytial virus** by subgroup and detection rate by week, Influenza-like illness (ILI) surveillance in primary health care clinics, 04/01/2021 – 07/11/2021

Inconclusive: insufficient viral load in sample and unable to characterise further
RSV AB: Both RSV A and B subgroup identified

**RSV was detected from 15 of 614 (2%) specimens from patients who met suspected SARS-CoV-2 case definition but did not meet influenza-like illness (ILI) case definition. These are not included in the epidemiological curve.

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, 04/01/2021 – 07/11/2021

| Clinic (Province) | RSVA | RSVB | RSVAB | RSV subgroup inconclusive | RSV subgroup pending* | Total samples |
|-----------------------|-----------|-----------|----------|---------------------------|-----------------------|---------------|
| Agincourt (MP) | 11 | 7 | 1 | 0 | 0 | 218 |
| Eastridge (WC) | 23 | 1 | 1 | 0 | 0 | 219 |
| Edendale Gateway (KZ) | 1 | 6 | 0 | 0 | 0 | 225 |
| Jouberton (NW) | 2 | 17 | 0 | 1 | 0 | 736 |
| Mitchell's Plain (WC) | 3 | 0 | 0 | 0 | 0 | 230 |
| Total | 40 | 31 | 2 | 1 | 0 | 1 628 |

KZ: KwaZulu-Natal; NW: North West; WC: Western Cape; MP: Mpumalanga
Inconclusive: insufficient viral load in sample and unable to characterise further
RSV AB: Both RSV A and B subgroup identified
*RSV results for subgroups are pending

**RSV was detected from 15 of 614 (2%) specimens from patients who met suspected SARS-CoV-2 case definition but did not meet influenza-like illness (ILI) case definition. These are not included in the table.

INFLUENZA, RESPIRATORY SYNCYTIAL VIRUS AND SARS-CoV-2 SURVEILLANCE REPORT

WEEK 44 2021

INFLUENZA-LIKE ILLNESS (ILI) SURVEILLANCE PRIMARY HEALTH CARE CLINICS

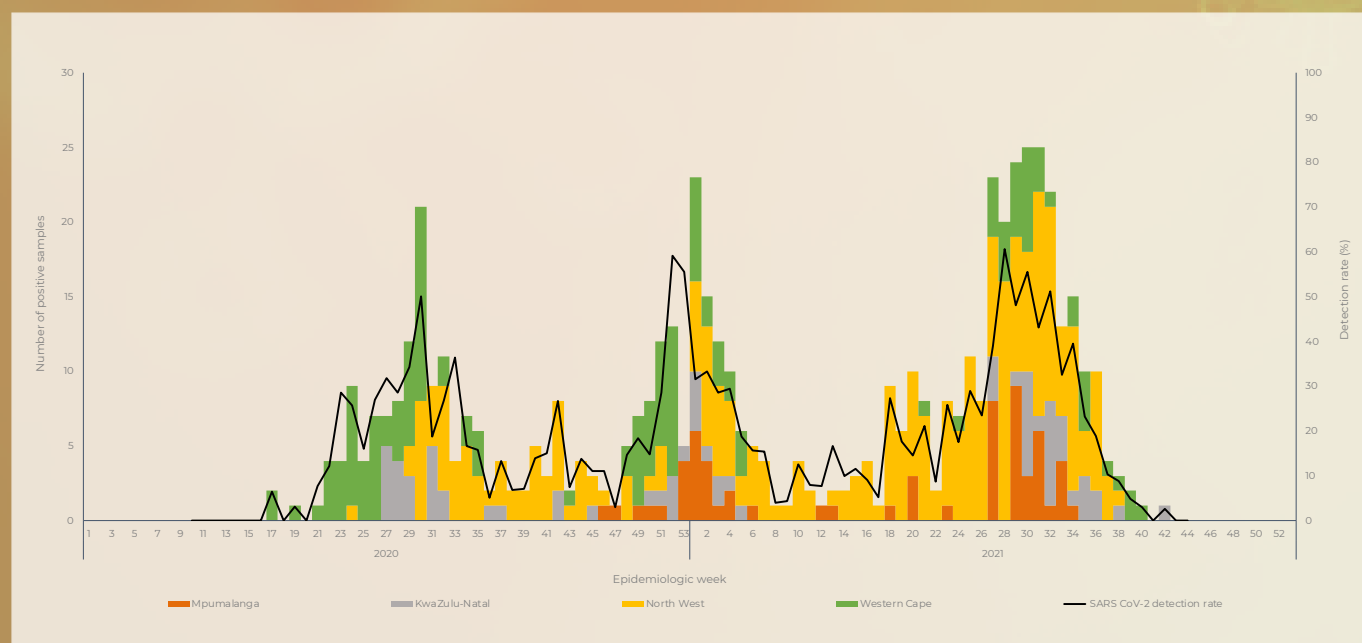


Figure 5. Number of patients testing positive for SARS-CoV-2* by province and detection rate by week, Influenza-like illness (ILI) surveillance in primary health care clinics, 02/03/2020 – 07/11/2021

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

**SARS-CoV-2 was detected in 149 of 746 (20%) specimens from patients who met suspected SARS-CoV-2 case definition but did not meet influenza-like illness (ILI) case definition. These are not included in the epidemiological curve.

Table 3. 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, 02/03/2020 – 07/11/2021

| Clinic (Province) | SARS-CoV-2 positive | Total samples tested |
|-----------------------|---------------------|----------------------|
| Agincourt (MP) | 62 | 248 |
| Eastridge (WC) | 59 | 708 |
| Edendale Gateway (KZ) | 69 | 368 |
| Jouberton (NW) | 288 | 1059 |
| Mitchell's Plain (WC) | 90 | 559 |
| Total: | 568 | 2 942 |

KZ: KwaZulu-Natal; NW: North West; WCP: Western Cape; MP: Mpumalanga (started enrolling on the 10th November 2020)

**SARS-CoV-2 was detected in 149 of 746 (20%) specimens from patients who met suspected SARS-CoV-2 case definition but did not meet influenza-like illness (ILI) case definition. These are not included in the table.

INFLUENZA, RESPIRATORY SYNCYTIAL VIRUS AND SARS-CoV-2 SURVEILLANCE REPORT

WEEK 44 2021

INFLUENZA-LIKE ILLNESS (ILI) SURVEILLANCE PRIMARY HEALTH CARE CLINICS

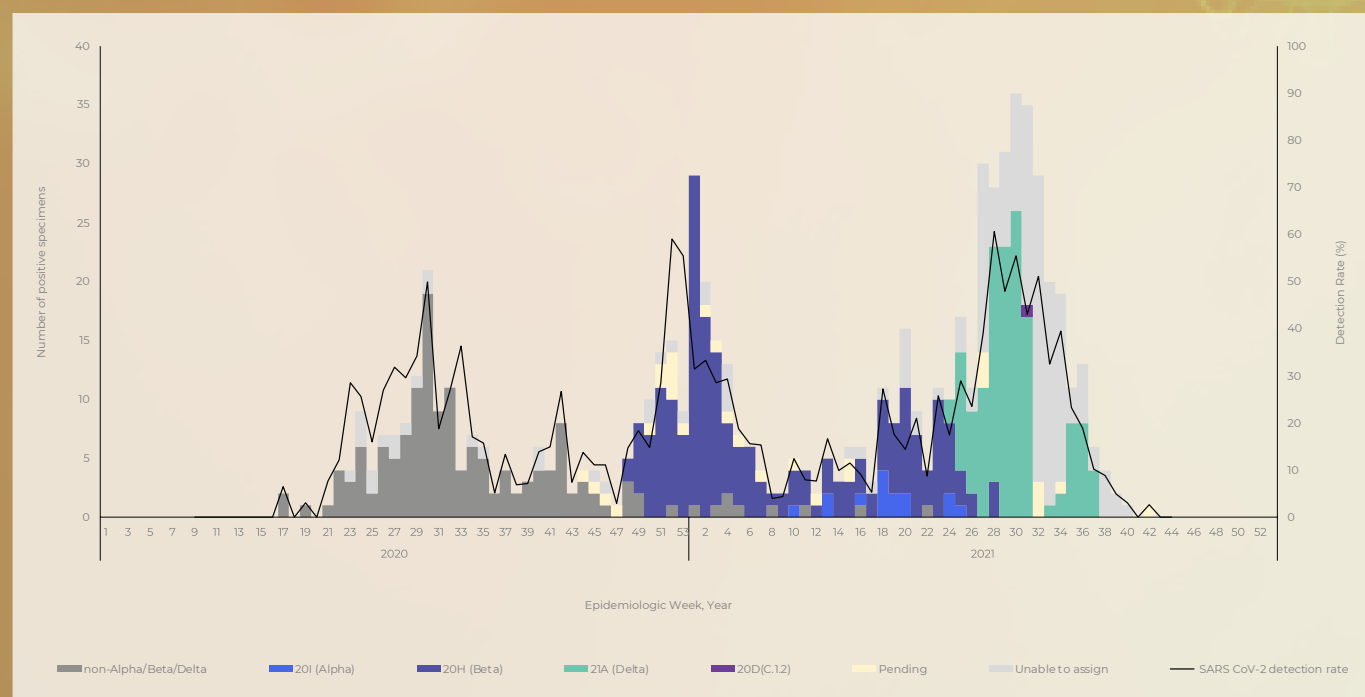


Figure 6. Number and detection rate of laboratory confirmed SARS-CoV-2* cases by variant type (variant PCR/sequencing) and week, Influenza-like illness (ILI) surveillance in primary health care clinics, 02/03/2020 – 07/11/2021

*Specimens are from patients with influenza-like illness at 5 sentinel sites in 4 provinces who met suspected SARS-CoV-2 case definition and met pneumonia (SRI) case definition as well as those that did not meet the ILI case definition.

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 4. Number of SARS-CoV-2* positive cases 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, 02/03/2020 – 07/11/2021

| Clinic (Province) | Non-Alpha/ Beta/Delta | 20I (Alpha) | 20H (Beta) | 21A (Delta) | 20D (C.1.2) | Pending | Unable to assign | Total SARS- CoV-2 positive |
|-----------------------|--------------------------|----------------|------------|----------------|----------------|-----------|---------------------|----------------------------------|
| Agincourt (MP) | 3 | 0 | 29 | 17 | 0 | 7 | 31 | 87 |
| Eastridge (WC) | 20 | 0 | 17 | 7 | 0 | 3 | 13 | 60 |
| Edendale Gateway (KZ) | 27 | 0 | 22 | 31 | 0 | 5 | 29 | 114 |
| Jouberton (NW) | 67 | 15 | 104 | 73 | 1 | 14 | 89 | 363 |
| Mitchell's Plain (WC) | 35 | 0 | 27 | 11 | 0 | 1 | 19 | 93 |
| Total: | 152 | 15 | 199 | 139 | 1 | 30 | 181 | 717 |

KZ: KwaZulu-Natal; NW: North West; WCP: Western Cape; MP: Mpumalanga (started enrolling on the 10th November 2020)

*Specimens are from patients with influenza-like illness at 5 sentinel sites in 4 provinces who met suspected SARS-CoV-2 case definition and met pneumonia (SRI) case definition as well as those that did not meet the ILI case definition.

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

INFLUENZA, RESPIRATORY SYNCYTIAL VIRUS AND SARS-CoV-2 SURVEILLANCE REPORT

WEEK 44 2021

INFLUENZA-LIKE ILLNESS (ILI) SURVEILLANCE VIRAL WATCH

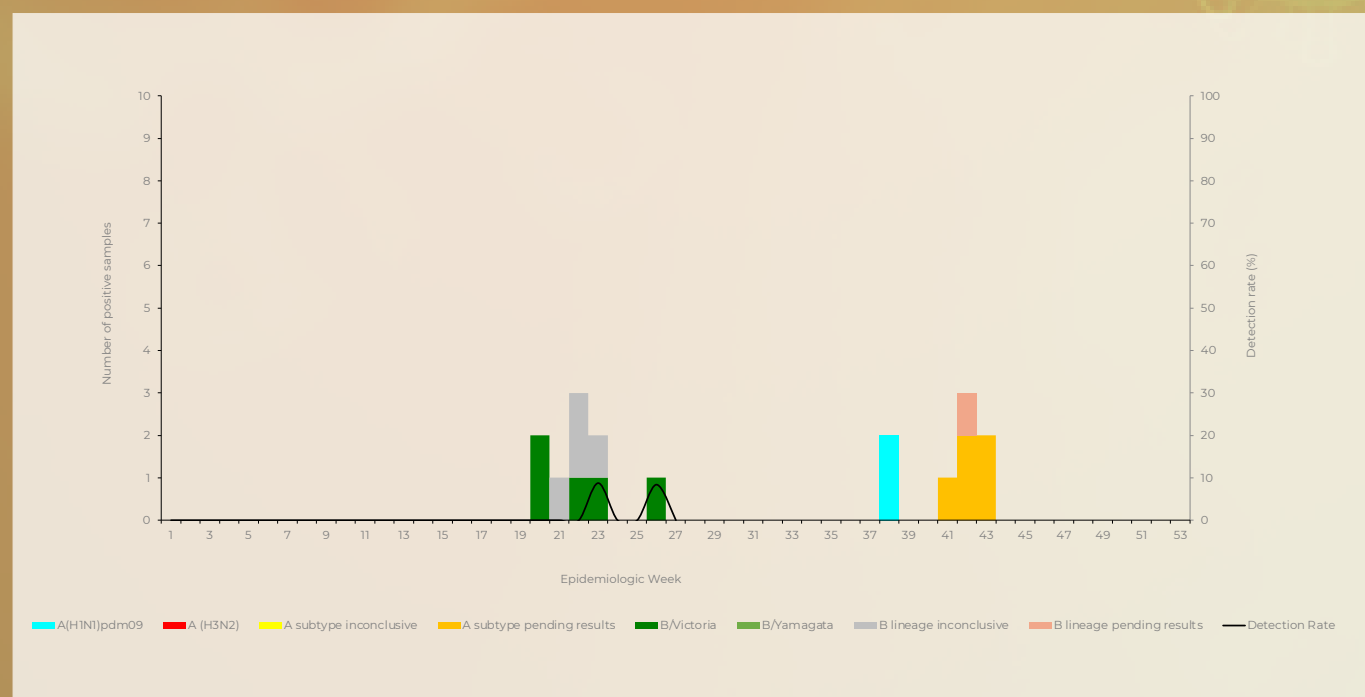


Figure 7. Number of positive patients* by influenza subtype and lineage and detection rate** by week, ILI surveillance - Viral Watch, 04/01/2021 – 07/11/2021

*Specimens from patients with Influenza-like illnesses at 90 sentinel sites in 8 provinces

** Only reported for weeks with >10 specimens submitted.

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

Table 5. Number of laboratory confirmed influenza cases by influenza subtype and lineage and total number of samples tested by province, ILI surveillance - Viral Watch, 04/01/2021 – 07/11/2021

| 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 | 0 | 0 | 0 | 1 | 0 | 0 | 1 | 4 |
| Free State | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Gauteng | 2 | 0 | 0 | 4 | 4 | 0 | 4 | 0 | 152 |
| Limpopo | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Mpumalanga | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 3 |
| North West | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 2 |
| Northern Cape | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Western Cape | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 0 | 30 |
| Total: | 2 | 0 | 0 | 5 | 5 | 0 | 4 | 1 | 191 |

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

*Influenza A subtype or B lineage results are pending

From 04 January 2021 to date, no patients were tested for influenza at the time of entry into South Africa following travel abroad.

Patients known to have acquired influenza abroad are not included in the table or epidemiological curve.

INFLUENZA, RESPIRATORY SYNCYTIAL VIRUS AND SARS-CoV-2 SURVEILLANCE REPORT

WEEK 44 2021

INFLUENZA-LIKE ILLNESS (ILI) SURVEILLANCE VIRAL WATCH

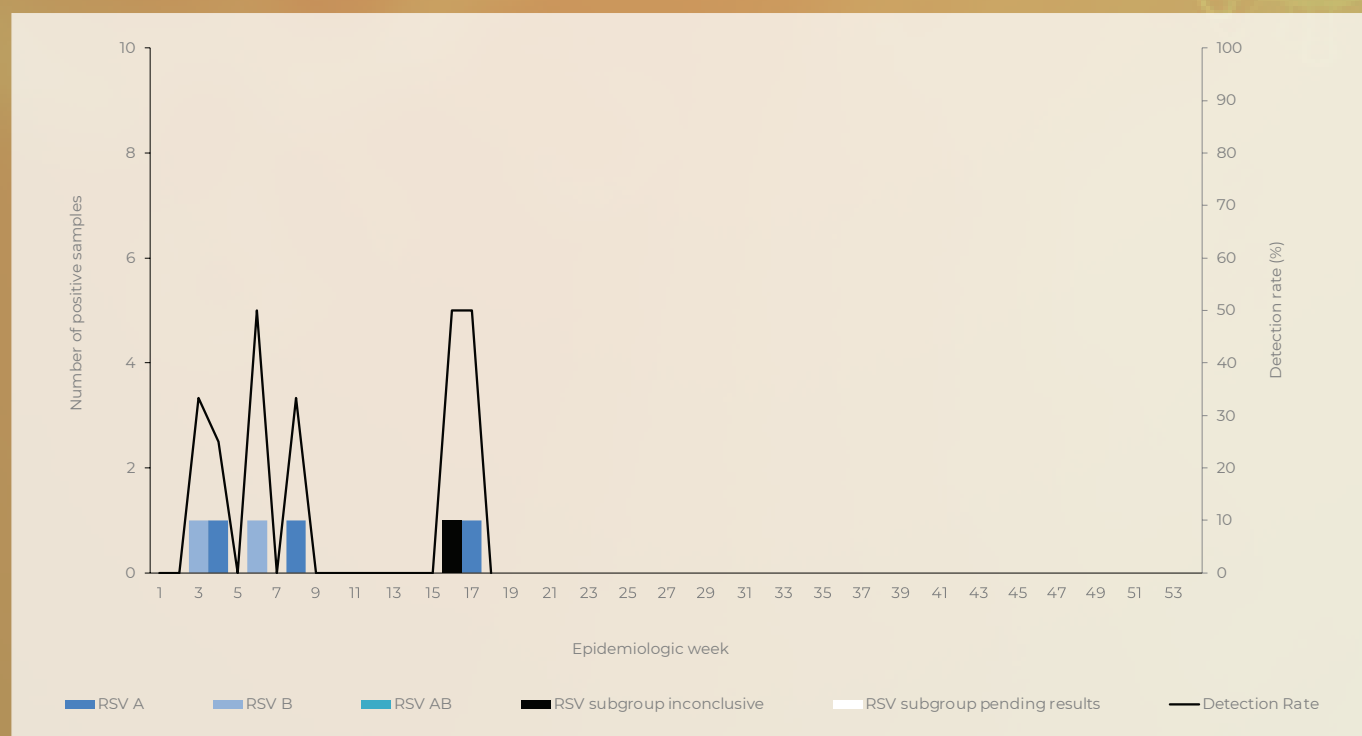


Figure 8. Number of RSV positive cases testing positive for respiratory syncytial virus (RSV)* by subgroup and detection rate by week, ILI surveillance - Viral Watch, 04/01/2021 – 07/11/2021

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

*RSV results for subgroups are pending

Table 6. Number of RSV positive cases identified and total number of samples tested by province, ILI surveillance - Viral Watch, 04/01/2021 – 07/11/2021

| Province | RSV A | RSV B | RSV AB | RSV subgroup inconclusive** | RSV subgroup pending results* | Total samples tested |
|---------------|----------|----------|----------|-----------------------------|-------------------------------|----------------------|
| Eastern Cape | 0 | 0 | 0 | 0 | 0 | 4 |
| Free State | 0 | 0 | 0 | 0 | 0 | 0 |
| Gauteng | 2 | 2 | 0 | 0 | 0 | 152 |
| Limpopo | 0 | 0 | 0 | 0 | 0 | 0 |
| Mpumalanga | 0 | 0 | 0 | 0 | 0 | 3 |
| North West | 0 | 0 | 0 | 0 | 0 | 2 |
| Northern Cape | 0 | 0 | 0 | 0 | 0 | 0 |
| Western Cape | 1 | 0 | 0 | 1 | 0 | 30 |
| Total: | 3 | 2 | 0 | 1 | 0 | 191 |

*RSV results for subgroups are pending

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

INFLUENZA, RESPIRATORY SYNCYTIAL VIRUS AND SARS-CoV-2 SURVEILLANCE REPORT

WEEK 44 2021

INFLUENZA-LIKE ILLNESS (ILI) SURVEILLANCE VIRAL WATCH

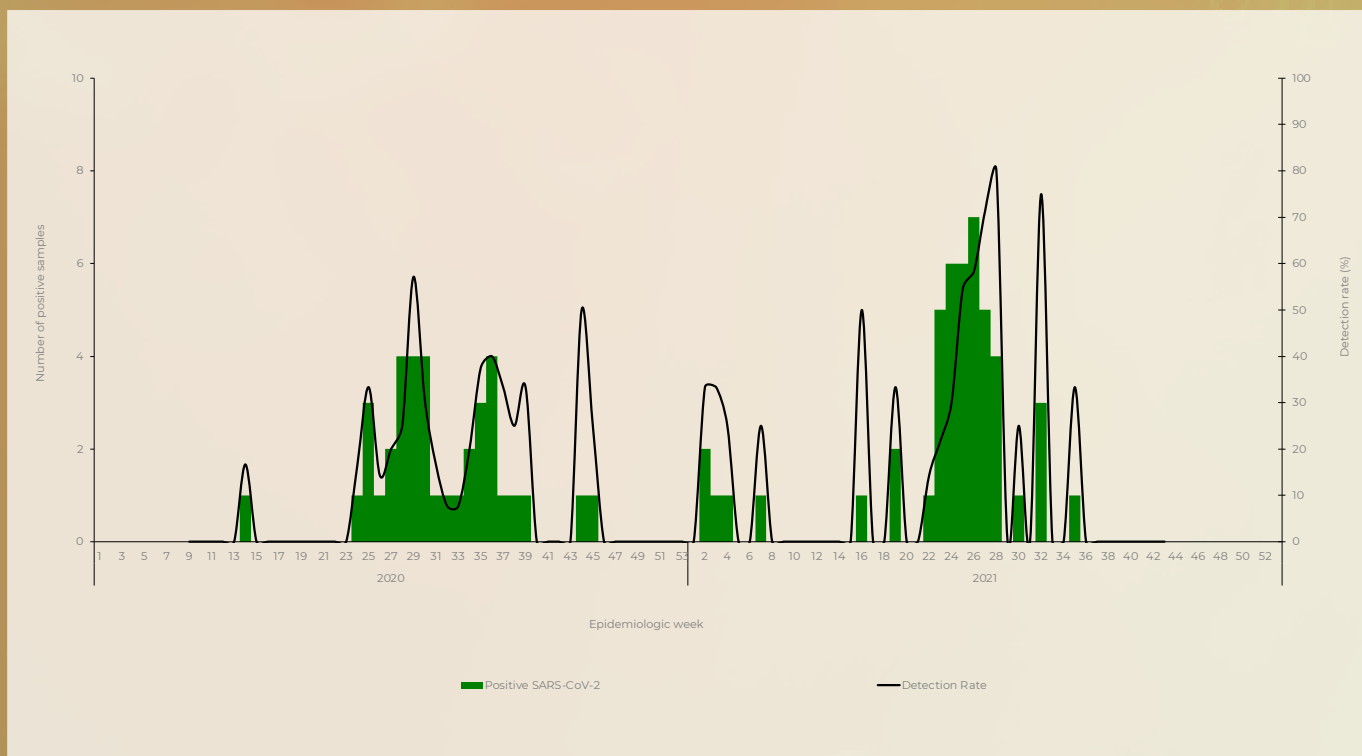


Figure 9. Number of patients testing positive for SARS-CoV-2*, by site and detection rate by week, ILI surveillance - Viral Watch, 04/01/2021 – 07/11/2021

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

Table 7. Number of SARS-CoV-2 positive cases identified and total number tested by province, ILI surveillance - Viral Watch, 04/01/2021 – 07/11/2021

| Province | SARS-CoV-2 positive | Total samples tested |
|---------------|---------------------|----------------------|
| Eastern Cape | 1 | 8 |
| Free State | 1 | 14 |
| Gauteng | 65 | 312 |
| Limpopo | 0 | 2 |
| Mpumalanga | 1 | 8 |
| North West | 0 | 2 |
| Northern Cape | 0 | 2 |
| Western Cape | 16 | 122 |
| Total: | 84 | 470 |

INFLUENZA, RESPIRATORY SYNCYTIAL VIRUS AND SARS-CoV-2 SURVEILLANCE REPORT

WEEK 44 2021

INFLUENZA-LIKE ILLNESS (ILI) SURVEILLANCE VIRAL WATCH

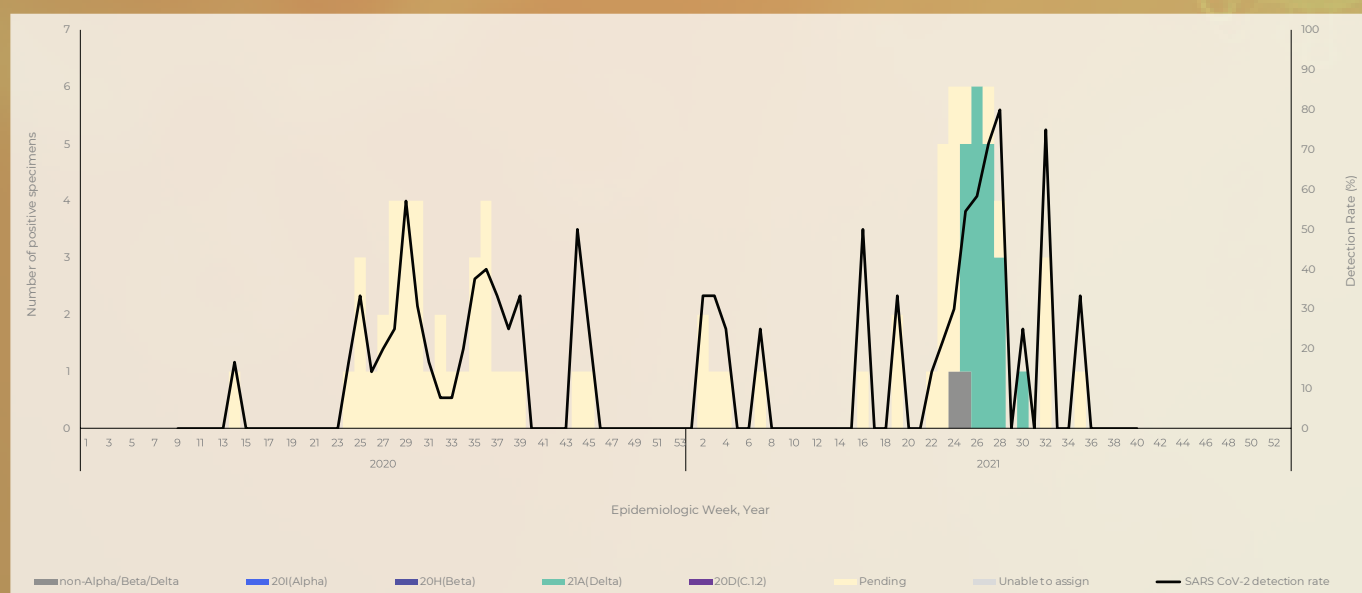


Figure 10. Number and detection rate of laboratory confirmed SARS-CoV-2* cases by variant type (variant PCR/sequencing) and week, ILI surveillance - Viral Watch, 04/01/2021 – 07/11/2021

*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 8. 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, 04/01/2021 – 07/11/2021

| Clinic (Province) | Non-Alpha/ Beta/Delta | 20I (Alpha) | 20H (Beta) | 21A (Delta) | 20D (C.1.2) | Pending | Unable to assign | Total SARS- CoV-2 positive |
|-------------------|--------------------------|----------------|---------------|----------------|----------------|-----------|---------------------|-------------------------------|
| Eastern Cape | 0 | 0 | 0 | 0 | 0 | 1 | 0 | 1 |
| Free State | 0 | 0 | 0 | 0 | 0 | 1 | 0 | 1 |
| Gauteng | 2 | 0 | 0 | 18 | 0 | 45 | 0 | 65 |
| Limpopo | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Mpumalanga | 0 | 0 | 0 | 0 | 0 | 1 | 0 | 1 |
| North West | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Northern Cape | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Western Cape | 0 | 0 | 0 | 1 | 0 | 15 | 0 | 16 |
| Total: | 2 | 0 | 0 | 19 | 0 | 63 | 0 | 84 |

*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

INFLUENZA, RESPIRATORY SYNCYTIAL VIRUS AND SARS-CoV-2 SURVEILLANCE REPORT

WEEK 44 2021

NATIONAL SYNDROMIC SURVEILLANCE FOR PNEUMONIA

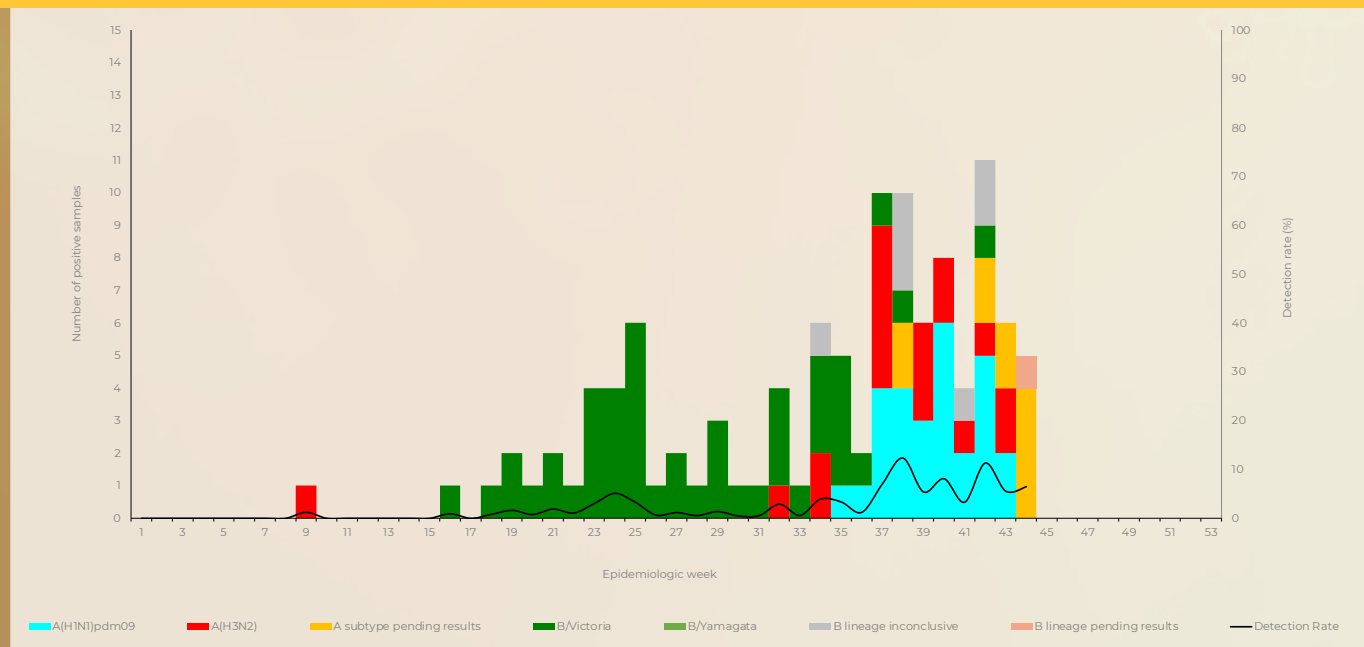


Figure 11. Number of positive influenza positive cases* by influenza subtype and lineage** and detection rate*** by week, pneumonia surveillance public hospitals, 04/01/2021 – 07/11/2021

*Specimens from patients hospitalised with pneumonia at 7 sentinel sites in 5 provinces

**Influenza was detected in six (3%) of 184 specimens, of which two (33%) were influenza A(H1N1)pdm09 and four (67%) were influenza B(Victoria) from patients who met suspected SARS-CoV-2 case definition but did not meet pneumonia (SRI) case definition. These are not included in the epidemiological curve.

***Only reported for weeks with >10 specimens submitted

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

Table 9. Number of laboratory confirmed influenza cases by subtype and lineage** and total number of samples tested by hospital, pneumonia surveillance public hospitals, 04/01/2021 – 07/11/2021

| 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 | 0 | 0 | 0 | 9 | 0 | 0 | 1 | 824 |
| Helen Joseph-Rahima Moosa (GP) | 20 | 12 | 1 | 4 | 17 | 0 | 5 | 0 | 1407 |
| Klerksdorp-Tshepong (NW) | 4 | 1 | 0 | 3 | 4 | 0 | 2 | 0 | 852 |
| Mapulaneng-Matikwana (MP) | 2 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 523 |
| Red Cross (WC) | 1 | 2 | 0 | 1 | 12 | 0 | 0 | 0 | 690 |
| Mitchell's Plain (WC) | 1 | 2 | 0 | 1 | 2 | 0 | 0 | 0 | 821 |
| Tintswalo (MP) | 0 | 0 | 1 | 1 | 1 | 0 | 0 | 0 | 221 |
| Total: | 28 | 18 | 2 | 10 | 46 | 0 | 7 | 1 | 5 338 |

GP: Gauteng; KZ: KwaZulu-Natal; NW: North West; MP: Mpumalanga (Tintswalo started enrolling on the 10th Feb 2021); WC: Western Cape

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

**Influenza was detected in six (3%) of 184 specimens, of which two (33%) were influenza A(H1N1)pdm09 and four (67%) were influenza B(Victoria) from patients who met suspected SARS-CoV-2 case definition but did not meet pneumonia (SRI) case definition. These are not included in the table.

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

INFLUENZA, RESPIRATORY SYNCYTIAL VIRUS AND SARS-CoV-2 SURVEILLANCE REPORT

WEEK 44 2021

NATIONAL SYNDROMIC SURVEILLANCE FOR PNEUMONIA

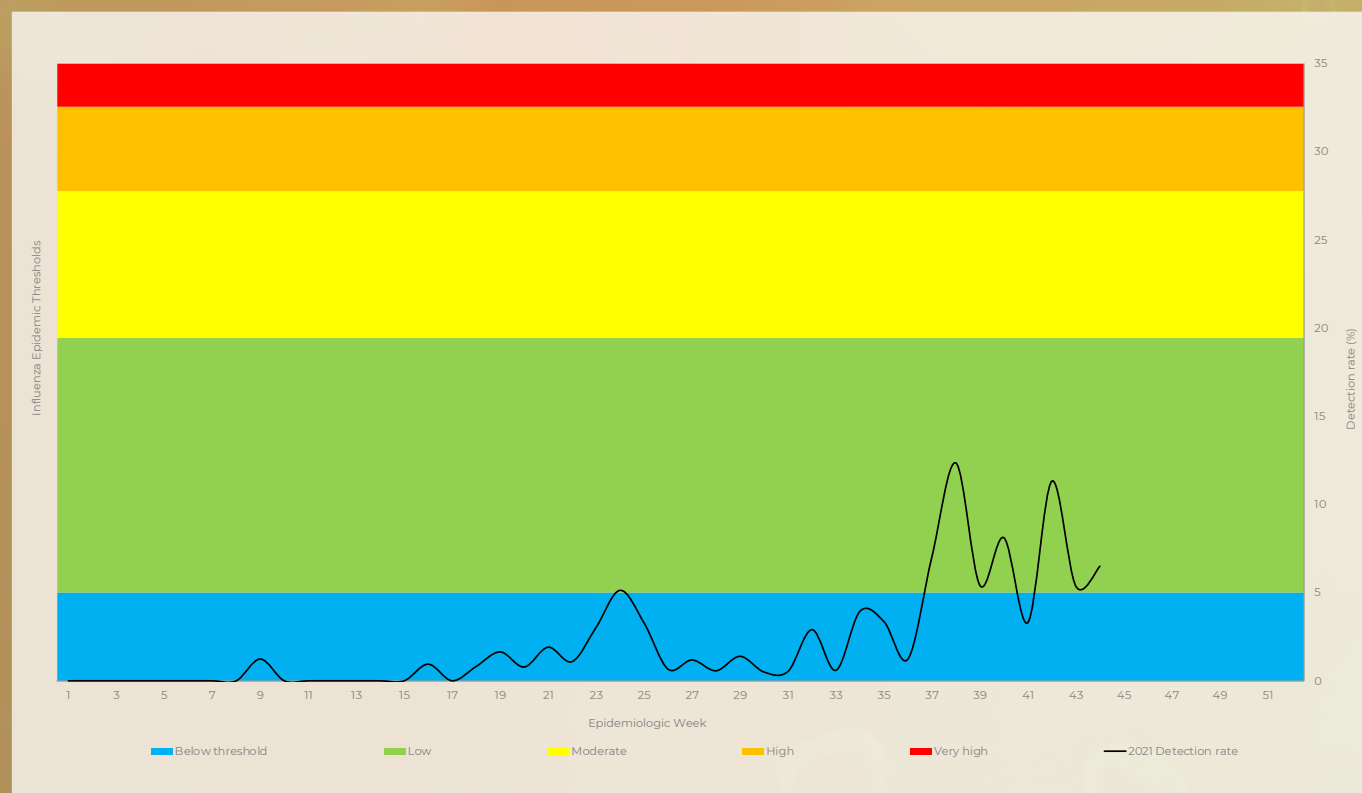


Figure 12. Influenza percentage detections and epidemic thresholds*, pneumonia surveillance public hospitals, 04/01/2021 – 07/11/2021

*Thresholds based on 2010-2019 data

INFLUENZA, RESPIRATORY SYNCYTIAL VIRUS AND SARS-CoV-2 SURVEILLANCE REPORT

WEEK 44 2021

NATIONAL SYNDROMIC SURVEILLANCE FOR PNEUMONIA

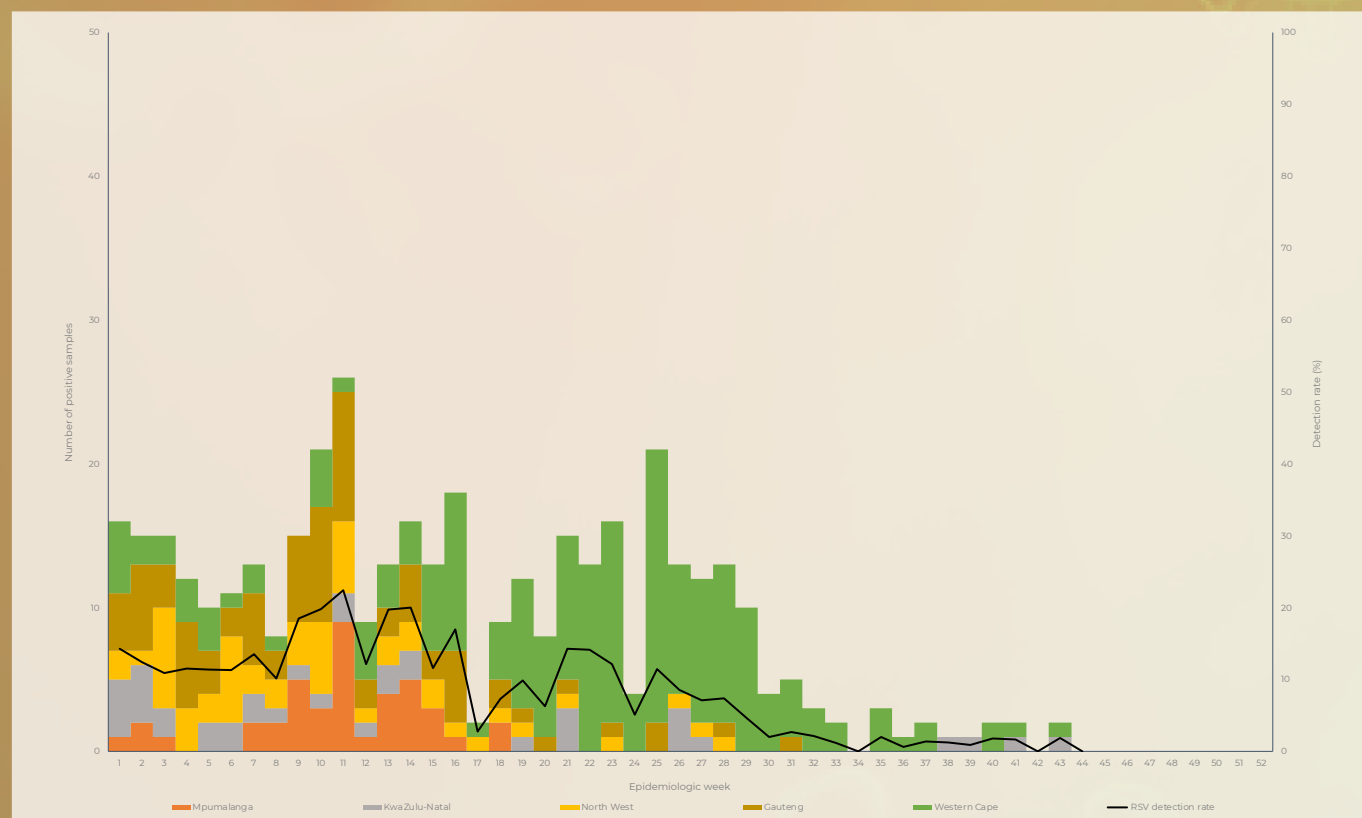


Figure 13. Number of patients testing positive for respiratory syncytial virus* by province and detection rate by week, pneumonia surveillance public hospitals, 04/01/2021 – 07/11/2021

*RSV was detected in six of 184 (3%) specimens from patients who met suspected SARS-CoV-2 case definition but did not meet pneumonia (SRI) case definition. These are not included in the epidemiological curve.

INFLUENZA, RESPIRATORY SYNCYTIAL VIRUS AND SARS-CoV-2 SURVEILLANCE REPORT

WEEK 44 2021

NATIONAL SYNDROMIC SURVEILLANCE FOR PNEUMONIA

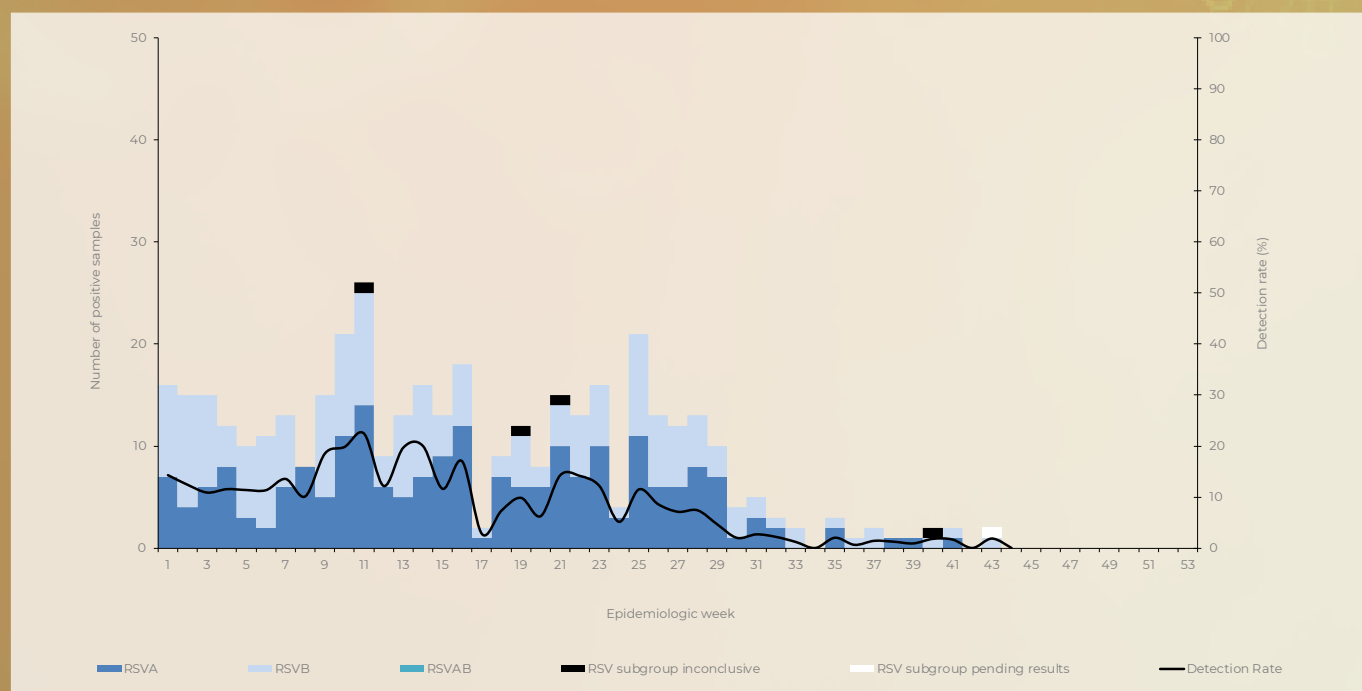


Figure 14. Number of patients testing positive for respiratory syncytial virus* by subgroup and detection rate by week, pneumonia surveillance public hospitals, 04/01/2021 – 07/11/2021

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

*RSV was detected in six of 184 (3%) specimens from patients who met suspected SARS-CoV-2 case definition but did not meet pneumonia (SRI) case definition. These are not included in the epidemiological curve.

Table 10: Number of patients positive for respiratory syncytial virus subgroups** by subgroups identified and total number of samples tested by hospital, pneumonia surveillance public hospitals, 04/01/2021 – 07/11/2021

| Hospital (Province) | RSVA | RSVB | RSVAB | RSV subgroup inconclusive | RSV subgroup pending* | Total samples |
|--------------------------------|------------|------------|----------|---------------------------|-----------------------|---------------|
| Edendale (KZ) | 7 | 29 | 0 | 0 | 1 | 824 |
| Helen Joseph-Rahima Moosa (GP) | 37 | 42 | 0 | 0 | 0 | 1407 |
| Klerksdorp-Tshepong (NW) | 8 | 44 | 0 | 2 | 0 | 852 |
| Mapulaneng-Matikwana (MP) | 21 | 7 | 0 | 0 | 0 | 523 |
| Red Cross (WC) | 87 | 50 | 0 | 2 | 1 | 690 |
| Mitchell's Plain (WC) | 41 | 12 | 0 | 1 | 0 | 821 |
| Tintswalo (MP) | 10 | 3 | 0 | 0 | 0 | 221 |
| Total: | 211 | 187 | 0 | 5 | 2 | 5 338 |

GP: Gauteng; KZ: KwaZulu-Natal; NW: North West; MP: Mpumalanga (Tintswalo started enrolling on the 10th Feb 2021); WC: Western Cape

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

RSV AB: Both RSV A and B subgroup identified

*RSV results for subgroups are pending

**RSV was detected in six of 184 (3%) specimens from patients who met suspected SARS-CoV-2 case definition but did not meet pneumonia (SRI) case definition. These are not included in the table

INFLUENZA, RESPIRATORY SYNCYTIAL VIRUS AND SARS-CoV-2 SURVEILLANCE REPORT

WEEK 44 2021

NATIONAL SYNDROMIC SURVEILLANCE FOR PNEUMONIA

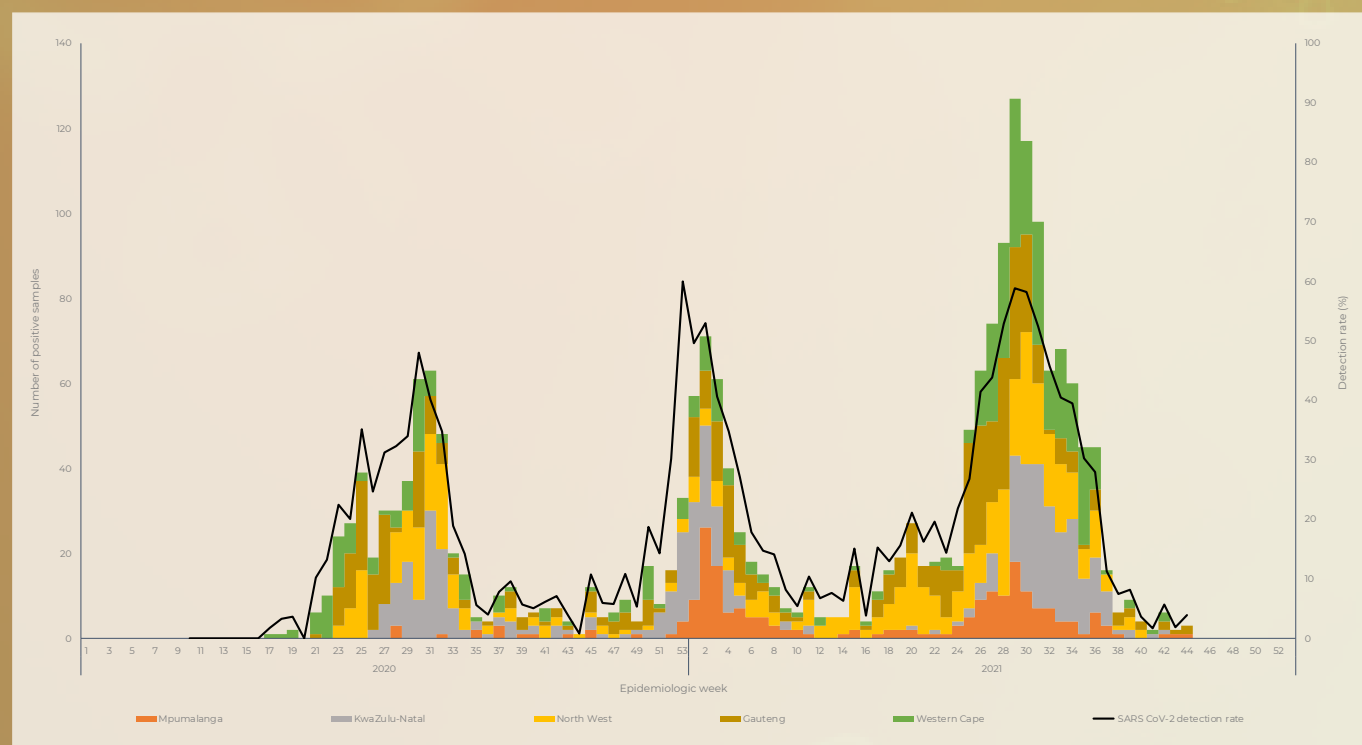


Figure 15. Number of patients testing positive for SARS-CoV-2*[§] by province and detection rate by week, pneumonia surveillance public hospitals, 02/03/2020 – 07/11/2021

*Specimens from patients hospitalized with pneumonia at 6 sentinel sites in 5 provinces

[§]SARS-CoV-2 was detected in 44 of 255 (19%) specimens from patients who met suspected SARS-CoV-2 case definition but did not meet pneumonia (SRI) case definition. These are not included in the epidemiological curve.

Table 11. Number of patients positive for SARS-CoV-2** and total number of samples tested by hospital, pneumonia surveillance public hospitals, 02/03/2020 – 07/11/2021

| Hospital (Province) | SARS-CoV-2 positive | Total samples tested |
|--------------------------------|---------------------|----------------------|
| Edendale (KZ) | 460 | 1 674 |
| Helen Joseph-Rahima Moosa (GP) | 491 | 2 252 |
| Klerksdorp-Tshepong (NW) | 476 | 1 445 |
| Mapulaneng-Matikwana (MP) | 167 | 825 |
| Red Cross (WC) | 61 | 1 654 |
| Mitchell's Plain (WC) | 351 | 1 290 |
| Tintswalo (MP) | 52 | 221 |
| Total: | 2 058 | 9 361 |

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

**SARS-CoV-2 was detected in 44 of 255 (19%) specimens from patients who met suspected SARS-CoV-2 case definition but did not meet pneumonia (SRI) case definition. These are not included in the table.

INFLUENZA, RESPIRATORY SYNCYTIAL VIRUS AND SARS-CoV-2 SURVEILLANCE REPORT

WEEK 44 2021

NATIONAL SYNDROMIC SURVEILLANCE FOR PNEUMONIA

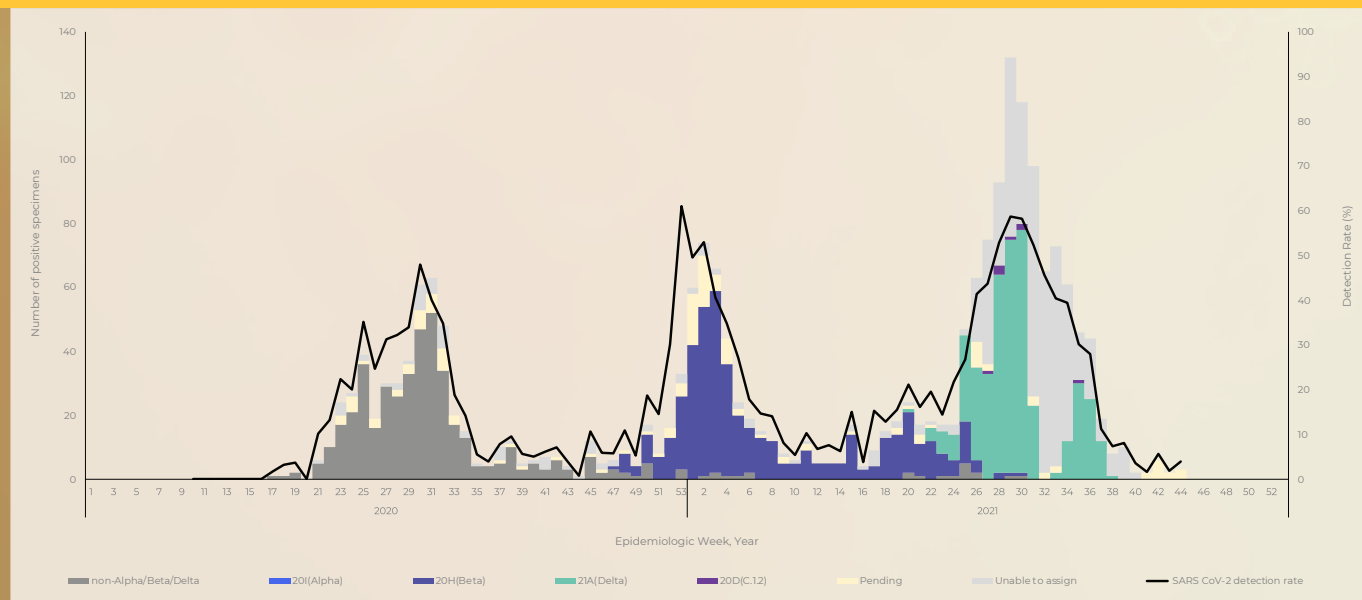


Figure 16. Number and detection rate of laboratory confirmed SARS-CoV-2 cases* by variant type (variant PCR/sequencing), pneumonia surveillance public hospitals, 02/03/2020 – 07/11/2021

Specimens are from hospitalized patients at 7 sentinel sites in 5 provinces who met suspected SARS-CoV-2 case definition and met pneumonia (SRI) case definition as well as those that did not meet the SRI case definition.

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 12. 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, 02/03/2020 – 07/11/2021

| Hospital (Province) | Non-Alpha/ Beta/Delta | 20I (Alpha) | 20H (Beta) | 21A (Delta) | 20D (C.1.2) | Pending | Unable to assign | Total SARS- CoV-2 positive |
|-----------------------------------|--------------------------|----------------|---------------|----------------|----------------|------------|---------------------|-------------------------------|
| Edendale (KZ) | 102 | 1 | 75 | 83 | 2 | 59 | 145 | 467 |
| Helen Joseph-Rahima Moosa (GP) | 134 | 5 | 120 | 101 | 4 | 27 | 101 | 492 |
| Klerksdorp-Tshepong (NW) | 130 | 9 | 112 | 94 | 2 | 13 | 116 | 476 |
| Mapulaneng- Matikwana (MP) | 16 | 0 | 90 | 29 | 0 | 20 | 47 | 202 |
| Red Cross (WC) | 15 | 0 | 5 | 7 | 0 | 7 | 27 | 61 |
| Mitchell's Plain (WC) | 50 | 0 | 49 | 96 | 0 | 17 | 140 | 352 |
| Tintswalo (MP) | 0 | 1 | 12 | 15 | 0 | 6 | 18 | 52 |
| Total: | 447 | 16 | 463 | 425 | 8 | 149 | 594 | 2 102 |

GP: Gauteng; KZ: KwaZulu-Natal; NW: North West; MP: Mpumalanga (Tintswalo started enrolling on the 10th Feb 2021); WC: Western Cape

*Specimens are from hospitalized patients at 7 sentinel sites in 5 provinces who met suspected SARS-CoV-2 case definition and met pneumonia (SRI) case definition as well as those that did not meet the SRI case definition.

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

INFLUENZA, RESPIRATORY SYNCYTIAL VIRUS AND SARS-CoV-2 SURVEILLANCE REPORT

WEEK 44 2021

SUMMARY OF LABORATORY CONFIRMED SARS-COV-2 CASES

Table 13. Characteristics of individuals with laboratory-confirmed SARS-CoV-2, enrolled in influenza-like illness (ILI) and pneumonia surveillance programmes, South Africa, 2 March 2020 - 07 November 2021

| Characteristic | Influenza-like illness (ILI), public-sector, n=717 (%) | Pneumonia, n=2 102 (%) |
|-----------------------------|--|------------------------|
| Age group | | |
| 0-9 | 52/717 (7) | 138/2102 (7) |
| 10-19 | 53/717 (7) | 15/2102 (1) |
| 20-39 | 334/717 (47) | 383/2102 (18) |
| 40-59 | 223/717 (31) | 787/2102 (37) |
| 60-79 | 53/717 (7) | 701/2102 (33) |
| ≥80 | 2/717 (<1) | 78/2102 (4) |
| Sex-female | 433/717 (60) | 1282/2102 (61) |
| Province* | | |
| Gauteng | N/A | 492/2102 (23) |
| KwaZulu-Natal | 114/717 (16) | 467/2102 (22) |
| Mpumalanga** | 87/717 (12) | 254/2102 (12) |
| North West | 363/717 (51) | 476/2102 (23) |
| Western Cape | 153/716 (21) | 412/2092 (20) |
| Race | | |
| Black | 540/710 (76) | 1659/2088 (79) |
| Coloured | 140/710 (20) | 327/2088 (16) |
| Asian/Indian | 4/710 (1) | 52/2088 (2) |
| White | 22/710 (3) | 41/2088 (2) |
| Other | 4/710 (1) | 9/2088 (<1) |
| Variant^{§§} | | |
| Non-Alpha/Beta/Delta | 152/535 (28) | 447/1497 (30) |
| 20I(Alpha) | 15/535 (3) | 16/1497 (1) |
| 20H(Beta) | 199/535 (37) | 463/1497 (31) |
| 21A(Delta) | 139/535 (26) | 425/1497 (28) |
| 20D(C.1.2) | 1/535 (0.2) | 8/1497 (1) |
| Pending results | 29/535 (5) | 138/1497 (9) |

INFLUENZA, RESPIRATORY SYNCYTIAL VIRUS AND SARS-CoV-2 SURVEILLANCE REPORT

WEEK 44 2021

| Characteristic | Influenza-like illness (ILI), public-sector, n=717 (%) | Pneumonia, n=2 102 (%) |
|------------------------------|--|------------------------|
| Presentation | | |
| Fever | 556/710 (78) | 968/2088 (46) |
| Cough | 691/710 (97) | 2002/2088 (96) |
| Shortness of breath | 259/710 (36) | 1639/2088 (79) |
| Chest pain | 312/710 (44) | 851/2088 (41) |
| Diarrhoea | 54/710 (8) | 133/2088 (6) |
| Underlying conditions | | |
| Hypertension [§] | 17/691 (2) | 249/1896 (13) |
| Cardiac | 2/710 (<1) | 48/2088 (2) |
| Lung disease | 0/710 (0) | 3/2088 (<1) |
| Diabetes | 17/710 (2) | 438/2088 (21) |
| Cancer | 2/710 (<1) | 13/2088 (1) |
| Tuberculosis | 11/710 (2) | 56/2088 (3) |
| HIV-infection | 134/710 (19) | 460/2088 (22) |
| Other *** | 12/710 (2) | 90/2088 (4) |
| Management | | |
| Oxygen therapy | 9/710 (1) | 1635/2088 (78) |
| ICU admission | N/A | 50/2088 (2) |
| Ventilation | N/A | 55/2088 (3) |
| Outcome*** | | |
| Died | 0/707 (0) | 346/2058 (17) |

*ILI surveillance not conducted in Gauteng province

**Mpumalanga (ILI site started enrolling on the 10th November 2020 and an additional SARI site started enrolling on the 10th February 2021)

***Chronic lung, liver and kidney disease, organ transplant, pregnancy, malnutrition, obesity, tracheostomy, prematurity, seizure, stroke, anaemia, asplenia, burns, Systemic lupus erythematosus, seizures

****Outcome includes patients who are still hospitalised, have been discharged or referred, and those who died

[§]Data on hypertension was not collected on all cases.

^{§§} These individuals with unassigned sequence are not included, ILI (n=181) and SRI (n=594)

Note: Children may be over-represented amongst hospitalised patients due to the inclusion of a large paediatric hospital in Cape Town.

Of the 346 patients who died, three were in <20 age group, 25 in the 20-39 year age group, 117 in the 40-59 year age group, and 201 were ≥60 years; 199/346 (57%) were female.

Of the 1194 hospitalised cases with COVID19 vaccine status, 97 (8%) received vaccine, 13 (13%) received Johnson & Johnson, 75 (77%) received the first dose of Pfizer-BioNTech, 1 (3%) Astra Zeneca and 8 (8%) could not remember the vaccine type received. 20 (21%) of the 97 cases had full vaccination (n=13, J&J and n=7, Pfizer-BioNTech). Cases age varied from 35 years to >60 years old and the majority (72%, 70/97) were >60 years old. Females were a majority 57% (55/97) and 11% (11/97) cases were HIV infected.

INFLUENZA, RESPIRATORY SYNCYTIAL VIRUS AND SARS-CoV-2 SURVEILLANCE REPORT

WEEK 44 2021

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 (Ct) <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 (Ct) 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.

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 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 cleanup 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).