Whole-genome sequencing to track SARS-CoV-2 transmission in nosocomial outbreaks

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Summary

Whole virus genome sequencing is an effective tool enabling the high resolution characterisation of SARS-CoV-2 strains introduced into a hospital and provides evidence for transmission of SARS-CoV-2 between healthcare workers and patients and between healthcare workers themselves.

Abstract

Background: During the first wave of the coronavirus disease 2019 (COVID-19) pandemic, outbreaks of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) in healthcare institutions posed a significant problem. Due to limited evidence, guidance on appropriate infection prevention and control (IPC) measures such as the wearing of face masks varied. Here, we applied whole virus genome sequencing (WvGS) to analyse transmission routes of SARS-CoV-2 in hospital-acquired (HA) COVID-19.

Methods: An investigation was undertaken for all HA cases of COVID-19 from March to April 2020. Fifty SARS-CoV-2 samples were analysed by WvGS and their phylogenetic relationship established.

Results: WvGS identified transmission events previously undetected by epidemiological analysis and provided evidence for SARS-CoV-2 transmission between healthcare workers (HCW) and patients and among HCW themselves. The majority of HA COVID-19 cases occurred in patients highly dependent on nursing care, suggesting the likely route of transmission was by close contact or droplet, rather than aerosol, transmission. Mortality among HA COVID-19 infections was recorded as 33%.

Conclusions: This study provides evidence that SARS-CoV-2 transmission occurs from symptomatic and asymptomatic HCWs to patients. Interventions including comprehensive screening of HCWs for COVID-19 symptoms, PCR testing of asymptomatic HCWs upon identification of HA cases and implementation of universal use of surgical masks for all clinical care is indicated to prevent viral transmission. Our study highlights the importance of close collaboration between guidance bodies and frontline IPC experts for developing control measures in an emergency pandemic situation caused by a virus with undefined transmission modus.

Key words

Whole virus genome sequencing, nosocomial outbreaks, severe acute respiratory syndrome coronavirus 2, infection prevention and control

Introduction

The coronavirus disease 2019 (COVID-19) caused by the novel virus SARS-CoV-2 (severe acute respiratory syndrome coronavirus 2) was first identified in Wuhan, Hubei, China following an unusual cluster of pneumonia in the region.[1] The risk of SARS-CoV-2 transmission within healthcare institutions has been well described.[2,3] China, the USA and several European countries have reported high rates of COVID-19 infection amongst healthcare workers (HCWs).[3–5] In Ireland, the Health Protection Surveillance Centre reported 8,215 positive cases amongst HCWs (up to the 1st of July), amounting to 32% of all of cases diagnosed nationally. Over 1,687 outbreaks/clusters have occurred in Ireland secondary to local transmission, of which hospital outbreaks comprise 10% .[6]

Whole genome sequencing (WGS) has been utilized for the investigation of outbreaks of multi-drug resistant bacteria such as carbapenemase-producing *Enterobacterales* and other clinically significant pathogens.[7–10] The WGS amplicon strategy has been used to obtain high-quality consensus sequences of SARS-CoV-2.[11,12] Application of this approach, allows for higher resolution analysis, enhancing our understanding of transmission patterns, whilst guiding policies designed to prevent disease spread.

At the start of the pandemic, recommendations on usage of personal protective equipment (PPE), particularly masks, varied between countries.[13] Initial infection prevention and control guidance recommended the use of surgical masks only for HCWs involved in the care of COVID-19 patients.[14] Environmental contamination with SARS-CoV-2 in hospital was perceived to be lower risk than person-to-person transmission[15] Due to fears regarding PPE shortages, mask use was not advised when caring for asymptomatic persons or for HCWs in non-clinical areas.[16]

In this report, we apply the amplicon-based Illumina sequencing protocol to the investigation of nosocomial outbreaks of SARS-CoV-2 and demonstrate how knowledge of transmission networks can better inform IPC practices.

Methods

Study setting

This study was undertaken in a tertiary referral centre. As per national policy, a hospital-acquired (HA) COVID-19 case was defined as a patient admitted without symptoms suggestive of COVID-19 who had SARS-CoV-2 RNA detected at least seven days after admission.[17] In order to apply greater precision to our inclusion criteria we further categorised patients by length of admission greater than 14 days which took into consideration the full incubation period of the virus. Symptoms suggestive of COVID-19 were fever, cough, myalgia, fatigue, headache, anosmia and diarrhoea.

HA COVID-19 cases were identified by laboratory records from March 7th to May 10th, 2020. An outbreak was classified as two or more cases of HA COVID-19 on a ward. Visiting of patients was restricted to compassionate care only from March 13th. Masks, gowns and gloves were worn when assessing suspected or confirmed cases of COVID-19. Positive cases were transferred to a designated COVID ward. Close contacts were isolated with droplet precautions for 14 days.

During March 2020, HCWs were advised by occupational medicine to remain home and attend community testing services if they experienced fever and cough or shortness of breath. On March 24th, in-house SARS-CoV-2 PCR for HCWs was established; the screening criteria for COVID-19 were expanded on April 6th to include sore throat, headache, myalgia, fever, cough, anosmia or ageusia. Epidemiological criteria collected on SARS-CoV-2 infected HCWs were date of PCR test and the ward they had been working on.

A close contact was defined as a HCW or patient who spent more than 15 minutes face-to-face within 2 metres of a confirmed case or patients who shared a multi-bedded room with a confirmed case for more than 2 hours.

PCR

Viral RNA was purified from nasopharyngeal swabs using the MagNA Pure 96 system (Roche Diagnostics). Quantitative RT-PCR was performed using the COVID-19 RealStar® SARS-CoV-2 RT-PCR Kit (Altona Diagnostics) and the Genesig® Real-Time_PCR assay (Primerdesign Ltd.) on the LightCycler®480 II (Roche) according to the manufacturer's instructions.

Sequencing and bioinformatics analysis

Complementary DNA was obtained from isolated RNA through reverse transcription and multiplex PCR according to the protocol provided by the Artic Network initiative.[18] Libraries were prepared using the NEBNext Ultra II kit (New England Biolabs) and sequenced on an Illumina MiSeq using 300-cycle v2 reagent kits (Illumina). Bowtie 2 was used for aligning the sequencing reads to the reference genome for SARS-CoV-2 (GenBank number, MN908947.3) and SAMtools for manipulating the alignments.[19] SNPs were used to define clusters and a median-joining network was generated including these data from this study and an additional 1,000 strains collected from GISAID available on May 22nd. Clade annotation was included for the Pangolin, GISAID and NextStrain systems. [20,21]

Ethical approval

Approval was granted by the institution's ethics committee.

Results

Epidemiology of nosocomial COVID-19

The first COVID-19 patient presented to our hospital on March 7th. In Ireland the number of reported cases increased steadily from February 29th, plateauing at the end of March, with a sustained peak until mid-April (Figure S1). Consistent with this, the number of COVID-19 cases admitted daily to our hospital peaked on March 27th. The number of infected HCWs increased in parallel with community transmission levels, also peaking at the end of March. (Figure 1)

The first HA case was diagnosed March 23rd 2020 (Figure 1). Patients associated with the first two outbreaks (wards 2 & 10) were in shared 6-bed rooms and transmission by HCWs was not established. Beds were over one metre apart with one bathroom per room. The first symptomatic HCW with an epidemiological link to a HA case occurred in outbreak 3 (ward 3). Three patients cared for by this HCW tested positive. The remaining five patients in the outbreak had no established contact with a positive HCW. No additional positive staff members were identified during this outbreak.

A fourth outbreak occurred on medical wards 6 and 8 between the 2nd and 9th of April. The index case on ward 8 was originally considered a single hospital-acquired case, but when two additional patients were diagnosed simultaneously three days later on the neighbouring ward 6, all other inpatients on ward 6 and ward 8 were tested. All patients were in single *en-suite* rooms. The two wards shared nursing teams and clinical teams and several staff members working on the ward had been diagnosed with COVID-19. Six additional SARS-CoV-2 infected patients were detected by screening.

At the same time two outbreaks occurred on surgical wards 7 and adjacent ward 9 involving 7 patients. Similar to the medical wards, the first two patients became symptomatic and additional cases were detected following testing of all the other inpatients: Five patients were in single *en-suite* rooms and two patients were sharing a room. At the time no obvious epidemiological link with the concomitant outbreak on the medical ward could be established.

In response to the increase in HA cases the occupational health testing criteria were changed. A concern that COVID-19 positive staff members experiencing milder symptoms, such as headache and sore throat, were being missed, led to a decision to broaden the testing criteria to include any other symptom possibly indicating SARS-CoV-2 infection (see methods).

At this point national IPC policy recommended the use of masks only for the management of COVID-19 patients. Prompted by the increasing number of HA COVID-19 cases suggesting transmission from asymptomatic/ pre-symptomatic HCWs, the hospital IPC team mandated surgical masks for all HCW/patient interactions. Masks were not required in non-clinical areas. No new hospital acquired cases were identified from April 18th to April 27th.

On April 28th a new hospital acquired case emerged. The patient suffered from agitation and would wander up and down the corridor accompanied by his designated carer. He was unable to maintain physical distancing and characteristically would shout directly into faces of staff members passing by. An agency nurse designated to his care became symptomatic and was excluded from work. One week later the patient developed a dry cough and tested positive for SARS-CoV-2. Seven additional HCWs on the ward developed symptoms and six tested positive. Due to a high level of patient immobility on this ward, only a small number of patients would have been considered close contacts of this index case. Despite this, eleven additional patients were identified as positive on screening. Upon questioning staff reported that surgical masks were frequently removed in the nurses station when communicating with each other and replaced for patient interactions.

Whole virus genome sequencing and outbreak investigation

SARS-CoV-2 RNA was extracted from nasopharyngeal swabs obtained from COVID-19 cases and their corresponding HCWs were sequenced to completion. The average sequence quality per samples was > 99% for 46 samples, and between 92 and 94% for 4 samples (Table S1). Sequenced SARS-CoV-2 with tiles average quality < 89% were excluded in the study. Phylogenetic analysis identified six independent groups of which clusters 1-3 were related to 39 patients. The combination of the phylogenetic cluster data (Figure 2), the ward, sampling dates, and classification as HCW or patient, uncovered a hitherto unrecognised association among samples distributed over time (Figure 3). In a global context, cases clustered in distinct areas of the phylogenetic tree suggesting a diverse genetic origin of the virus (Figure 4).

Cluster 1 contained the majority of specimens associated with the outbreak on ward 1 at the end of April (Figure 2). This provides evidence of HCW-patient transmission as 5 HCWs were positive prior to the ward screening on April 29th. Many patients on this ward did not have direct contact with one another due to immobility.

Cluster 2 (wards 2, 3, 4, and 5) demonstrated the ability of WvGS to link new HA cases in distinct locations with previous outbreaks. This occurred on two separate occasions (ward 4 and ward 6). The patient diagnosed on ward 4 had no established epidemiological link at the time of diagnosis and the second patient was assumed to be part of the ward 6 outbreak. Sequencing revealed both patients carried the ward 3 strain and ward history confirmed they had been discharged from there the previous week.

Cluster 3 as shown on the tree illustrates the outbreaks on wards 6/8 and 7/9, which were considered distinct at the time as these wards are geographically separate employing different HCWs. The phylogenetic tree reveals indistinguishable strains on these wards. One of the HCWs identified in this cluster was a ‘floater’. ‘Floaters’ are responsible for the personal care of highly dependent patients and are assigned to different wards on a daily basis. Two ‘floaters’ working between these 4 wards tested positive, providing an epidemiological link between outbreaks. A positive HCW in this cluster had no direct patient contact indicating that transmission may occur in non-clinical areas between HCWs.

All three clusters stem from one main branch and are broadly the same hospital strain with minor sequence variations per ward. The fourth and fifth clusters are suggestive of different strains, originating from distinct branches. A patient in the fourth cluster was identified epidemiologically as part of the ward 1 outbreak on April

29th. However, this patient's virus sequence aligns closely with a patient diagnosed on April 7th and both patients overlapped on the same ward for a number of days.

Clinical characteristics of hospital-acquired COVID-19

52 patients were infected with SARS-CoV-2 during their hospital admission from March 23rd to May 7th. Clinical characteristics are described in Table 1. The majority of patients required assistance with mobility (65%) and self-care (77%). The crude mortality rate amongst HA cases was calculated at 33%. As the majority had a ceiling of care established, (i.e. not for cardiopulmonary resuscitation or mechanical ventilation) only 3 patients were admitted to ICU. Overall, 14 patients (27%) died from complications of COVID-19.

Discussion

HA COVID-19 presents an ongoing threat to vulnerable patients with a high level of dependency and comorbidity. To prevent hospital outbreaks of COVID-19 it is essential to identify modes of viral transmission in order to implement effective IPC measures.

Our study highlights for the first time the analytical strength of WvGS specifically for the investigation of hospital outbreaks of SARS-CoV-2 and how these data can facilitate improved IPC practices.

A study of nosocomial infection amongst HCWs and their families was undertaken in Wuhan. Nine separate clusters were identified and WvGS was utilised to sequence 12 strains revealing a high level of dissemination across transmission chains. Only HCW strains were sequenced and therefore genomic links between HCW and patients were not established.[22] WvGS accurately identified dissemination routes of SARS-CoV-2 across Australia. Provisional contact tracing suggested an epidemiological link between shared HCWs and 54 cases of COVID-19. However, phylogenetic analysis revealed that the cases were divided into 4 distinct clusters with 15 cases linked through WvGS to a social event unrelated to the health-care setting.[23]

Unlike other studies published to date, we sought to use WvGS to enhance our knowledge of hospital transmission and to provide supportive evidence for IPC interventions introduced during the outbreak.[24] Through WvGS, links between outbreaks were established and points of both HCW-to-patient and HCW-to-HCW transmission identified. The majority of HA clusters were broadly linked to the same strain suggesting a high level of intra-hospital transmission. Prior to sequencing, clear epidemiological links between cases were often not apparent. WvGS data suggested the majority of HA cases were spread *via* HCWs as many patients

were either immobile or in single *en-suite* rooms. Furthermore, phylogenetic analysis highlighted the potential of WvGS to distinguish separate strains introduced into the hospital.

A number of changes brought about a decline in HA cases starting with a change to staff testing allowing testing of HCW with mild viral symptoms. This was the first step in minimising HCW-patient transmission.

At the start of the pandemic, decisions around PPE were particularly influenced by a lack of supply supporting the use of masks only for activity involving COVID-19 patients.[16] On the 31st of March the European Centre for Disease Prevention and Control stated that the use of surgical masks should be considered for all HCWs.[25] Despite an increasing number of hospitals reporting outbreaks, national guidance advised using masks only for the care of COVID-19 patients until the end of April.[26]

By April 13th, five separate ward outbreaks had occurred in our centre, with a significant number of patients epidemiologically linked to positive staff contacts. In addition, clear evidence of pre-symptomatic transmission reported in the literature added to concerns regarding asymptomatic spread of infection between HCWs and patients.[27] Masks were mandated for all HCW-patient interactions in clinical areas and no HA cases were diagnosed for ten days. On June 5th the WHO issued guidance supporting the continuous use of masks by HCWs in areas with community transmission or large-scale outbreaks.[28]

Despite the introduction of universal mask use in clinical care, an outbreak occurred at the end of April. Restricted space in non-clinical areas posed a challenge for social distancing. The reported removal of masks upon entry made it a likely point of transmission between staff. In addition, we described an agitated wandering patient positive for COVID-19. A number of studies highlighted the phenomenon of a "superspreader".[29–31] It is possible that this patient amplified the transmission of COVID-19 *via* close contact with HCWs. This particular outbreak suggests that masks should be worn at all times, including in administrative areas and particularly where social distancing cannot be observed. The transmission of SARS-CoV-2 via environmental contamination has been hypothesised as a potential source of hospital outbreaks.[32] Transmission via contaminated surfaces or other fomites to hands of HCW or to patients was felt to be unlikely in this study, as hand hygiene and environmental cleaning compliance remained above 90% in regular audits.Our study identified common characteristics amongst patients across all hospital disciplines with HA COVID-19. Most of the hospital outbreaks described to date are confined to specific patient cohorts.[33,34] The average age was higher than studies describing both community and HA cases.[35] Hence a large proportion of patients required assistance with mobility and self-care, thereby putting them at greater risk of infection from asymptomatic staff.

The overall mortality rate of 33% amongst HA cases is consistent with other studies describing hospital outbreaks and is not a reflection of overall COVID-19 deaths but rather the frailty of this patient population.[32] A nosocomial outbreak study of SARS-CoV-2 in a London hospital assessed the impact of various IPC measures. The study reported a reduction in HA cases once PPE (gloves, gowns and surgical masks), were introduced for all hospital interactions. Unlike our study, the majority of cases were attributed to patient-patient transmission in a shared ward. The study acknowledged that in the absence of sequencing data, all putative transmission routes were merely inferred.[32]

The majority of HCW-patient exposure occurred before the introduction of universal surgical masks. A minority of patients constituted contacts of one another in multi-bedded rooms. However not all patients in six-bedded rooms became infected. This suggests that the majority of transmission is spread *via* droplets and close contact, as patients with high levels of dependency were shown to be at greater risk than independent patients occupying the same room. Outside of aerosol generating procedures, it is likely that aerosolization occurs in rare circumstances, such as in superspreading events described above. The infectivity of SARS-CoV-2 containing aerosols has been demonstrated in tissue-culture assays.[36] This may provide an explanation for the extent of the outbreak which occurred at the end of April.

Conclusions

The focus of our epidemiological analysis was to identify specific patterns of SARS-CoV-2 transmission within our hospital. We demonstrate the role of WvGS for the purpose of outbreak investigation and show how sequencing informs our understanding of transmission patterns. Our study suggests that in nosocomial outbreaks, apart from aerosol generating procedures, the majority of transmission is spread *via* droplets and close contact. By doing so we provide evidence supporting the use of surgical masks in both clinical and non-clinical areas and demonstrate the need to restrict HCW movement across different wards. It is clear from our study that in a novel viral pandemic, when the exact mode of viral transmission remains unclear, ongoing communication between advisory organisations and front-line workers is paramount in shaping public health policy.

Notes

Author contributions

KS designed and co-ordinated the study. GM performed whole genome sequencing of isolates and bioinformatic analysis of sequences. SF guided the sequencing analysis and bioinformatic analyses of whole genome sequencing data. GG and SC provided viral sequences and assisted in sequence interpretation. ML, NM, USF collected and analysed epidemiological information on all cases. LF and AP collected and analysed epidemiological information on healthcare workers infected with Covid-19. ML wrote the original draft of the manuscript. KS co-ordinated the writing and revising of this paper. All authors commented on the draft manuscript and contributed to the final version.

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Conflict of interest disclosures

KS and LF report advisory board or *ad-hoc* consultancy fees from Menarini, Pfizer, Novartis, MSD, Astellas, Astra-Zeneca and Gilead. The remaining authors have no conflict of interest to declare.

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Figure legends

Figure 1: COVID-19 cases among inpatients and healthcare workers

COVID-19 patients admitted via the emergency department are shown in grey, nosocomial COVID-19 cases are shown in red. COVID-19 cases among healthcare workers are represented by the blue line. Before the 24th of March HCWs with fever or cough were asked to attend one of the community testing centres via referral from their GP. Due to the significant delay in obtaining test results for HCWs, occupational health in-house testing began on the 24th of March.

Figure 2. Maximum likelihood phylogenetic tree, including other Irish SARS-CoV-2 sequences. The six clusters are annotated based on the average distance among the sequences and the tree is rooted with the reference MN908947 from Wuhan, China. Virus samples are coloured according to the wards and are reported date of the isolation. HCW=health care worker; PAT= patient.

Figure 3. SARS-CoV-2 cases and temporal onset. Phylogenetic cluster data is shown in combination with ward location, sampling dates, and classification as HCW or patient.

Figure 4. HA outbreak and global SARS-CoV-2 sequences. Phylogenetic tree representing 1,000 sequences collected from GISAID/NextStrain from all over the world. The figure shows the distribution of the Irish clusters (in red) presented in this study in the context of the global genomes available

Table 1: Clinical characteristics of patients with hospital acquired COVID-19

	All patients (n=52)	Group 1 (≥ 14 days) (n=38)	Group 2 (7-13 days) (n=14)
Characteristics			
Median age, years	77	77	77
>70	36 (69%)	25 (66%)	11 (79%)
Sex			
Male	32 (62%)	23 (61%)	8 (57%)
Female	20 (38%)	14 (37%)	6 (43%)
Comorbidities			
Cardiovascular disease	31 (60%)	23 (61%)	8 (57%)
Chronic Pulmonary disease	8 (15%)	7 (18%)	1 (7%)
Diabetes	13 (25%)	10 (26%)	3 (21%)
Obesity	5 (10%)	4 (11%)	1 (7%)
Immunosuppressed[1]	12 (23%)	10 (26%)	2 (14%)
Chronic renal failure	10 (19%)	7 (18%)	3 (21%)
Chronic neurological disorders [2]	13 (25%)	11 (29%)	2 (14%)
Dementia	8 (15%)	7 (18%)	1 (7%)
Malignancy	21 (40%)	14 (37%)	7 (50%)
Symptoms			
Fever	31 (60%)	22 (58%)	9 (64%)
Hypothermia	1 (2%)	1 (3%)	0
Dyspnoea	6 (12%)	3 (8%)	3 (21%)
Cough	18 (35%)	10 (26%)	8 (57%)
Vomiting, diarrhoea	3 (6%)	3 (8%)	0
Anosmia	1 (2%)	1 (3%)	0
Asymptomatic	9 (17%)	7 (18%)	2 (14%)
Clinical outcome			
Oxygen requirement	27 (52%)	21 (55%)	6 (43%)
Non-invasive ventilation	1 (2%)	1 (3%)	0
ICU admission	3 (6%)	2 (6%)	1 (7%)
Intubation	3 (6%)	2 (6%)	1 (7%)
Death	17 (33%)	13 (34%)	4 (29%)
Premorbid functional status			
Assistance with mobility	34 (65%)	26 (68%)	8 (57%)
Assistance with self-care	40 (77%)	30 (79%)	10 (71%)
Physiotherapy	34 (65%)	25 (66%)	9 (64%)
Additional care needs			
Dressing changes	12 (23%)	9 (24%)	3 (21%)
Drain management	12 (23%)	8 (21%)	4 (29%)
Intra-venous therapy	41 (79%)	30 (79%)	11 (79%)
Exposure			
Close contact of positive patient	15 (27%)	11 (29%)	4 (29%)
Close contact of positive staff	33 (63%)	24 (63%)	9 (64%)
Contact status unknown	12 (23%)	9 (24%)	3 (21%)
Single room	20 (38%)	14 (37%)	6 (43%)
Multiple bed	32 (62%)	24 63%)	8 (57%)

1. Patients on chemotherapy for cancer, medications such as steroids, methotrexate and those with HIV
2. Neurological conditions including epilepsy, multiple sclerosis, spinal cord injury and Parkinson's disease

Figure 1

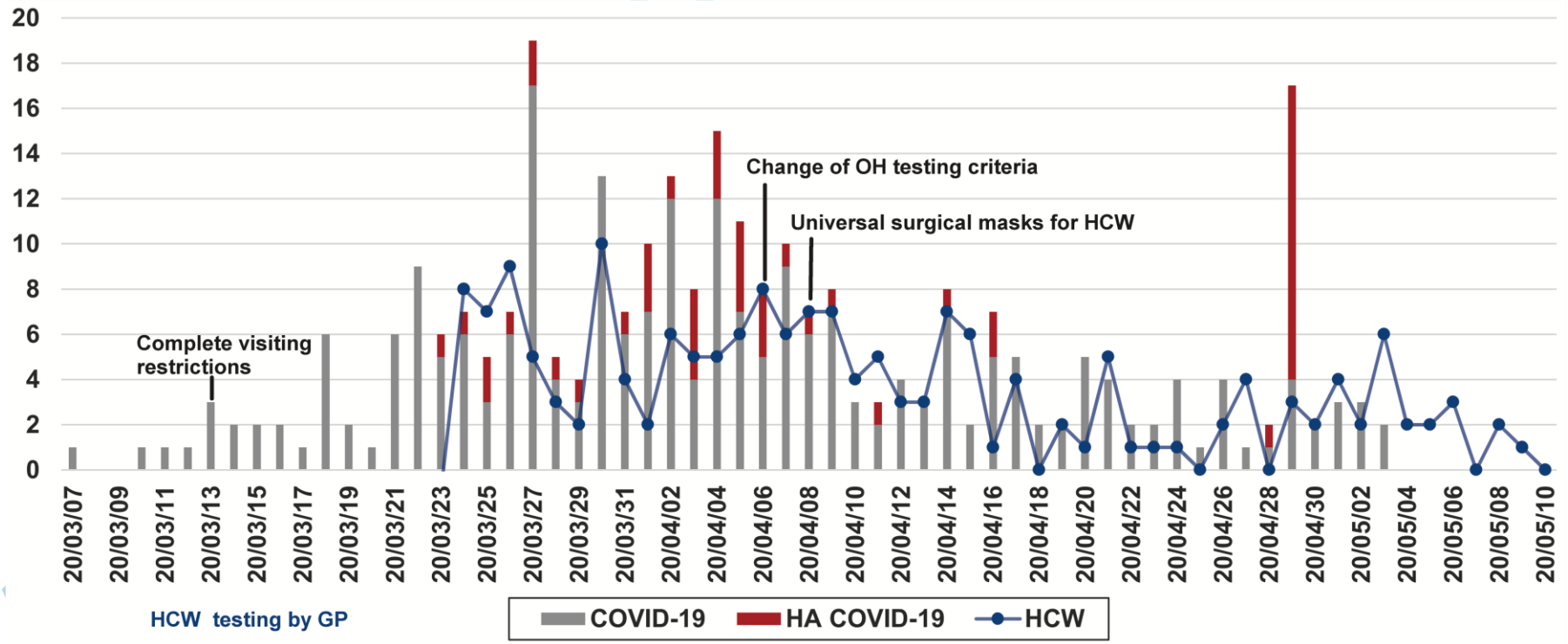
20
18
16
14
12
10
8
6
4
2
0
Change of OH testing criteria
Universal surgical masks for HCW
Complete visiting restrictions
20/03/07
20/03/09
20/03/11
20/03/13
20/03/15
20/03/17
20/03/19
20/03/21
20/03/23
20/03/25
20/03/27
20/03/29
20/03/31
20/04/02
20/04/04
20/04/06
20/04/08
20/04/10
20/04/12
20/04/14
20/04/16
20/04/18
20/04/20
20/04/22
20/04/24
20/04/26
20/04/28
20/04/30
20/05/02
20/05/04
20/05/06
20/05/08
20/05/10
HCW testing by GP
COVID-19
HA COVID-19
HCW

Figure 2

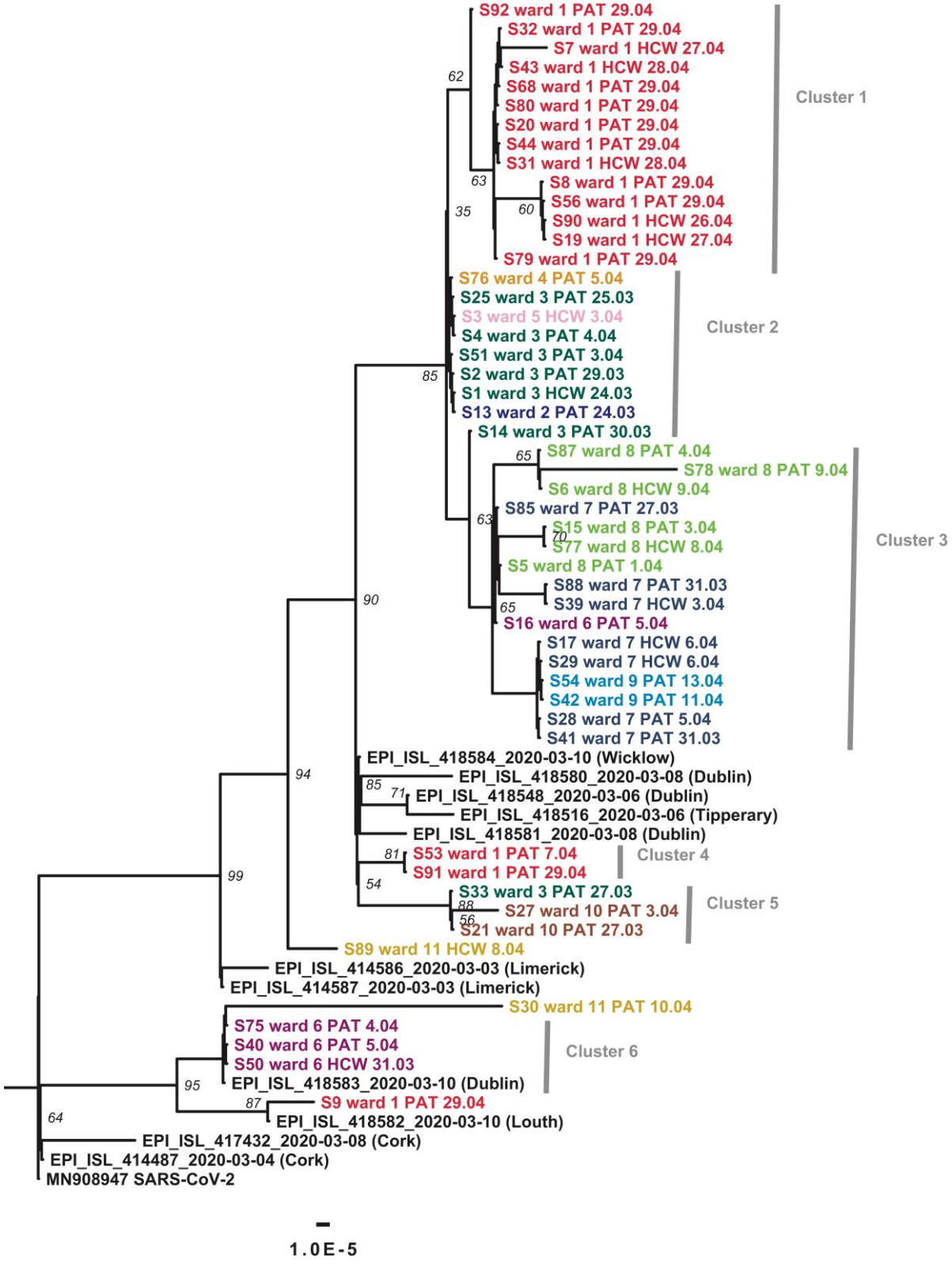
S92 ward 1 PAT 29.04
S32 ward 1 PAT 29.04
S7 ward 1 HCW 27.04
S43 ward 1 HCW 28.04
S68 ward 1 PAT 29.04
S80 ward 1 PAT 29.04
S20 ward 1 PAT 29.04
S44 ward 1 PAT 29.04
S31 ward 1 HCW 28.04
S8 ward 1 PAT 29.04
S56 ward 1 PAT 29.04
S90 ward 1 HCW 26.04
S19 ward 1 HCW 27.04
S79 ward 1 PAT 29.04
Cluster 1
S76 ward 4 PAT 5.04
S25 ward 3 PAT 25.03
S3 ward 5 HCW 3.04
S4 ward 3 PAT 4.04
S51 ward 3 PAT 3.04
S2 ward 3 PAT 29.03
S1 ward 3 HCW 24.03
S13 ward 2 PAT 24.03
S14 ward 3 PAT 30.03
Cluster 2
S87 ward 8 PAT 4.04
S78 ward 8 PAT 9.04
S6 ward 8 HCW 9.04
S85 ward 7 PAT 27.03
S15 ward 8 PAT 3.04
S77 ward 8 HCW 8.04
S5 ward 8 PAT 1.04
S88 ward 7 PAT 31.03
S39 ward 7 HCW 3.04
S16 ward 6 PAT 5.04
S17 ward 7 HCW 6.04
S29 ward 7 HCW 6.04
S54 ward 9 PAT 13.04
S42 ward 9 PAT 11.04
S28 ward 7 PAT 5.04
S41 ward 7 PAT 31.03
Cluster 3
EPI_ISL_418584_2020-03-10 (Wicklow)
EPI_ISL_418580_2020-03-08 (Dublin)
EPI_ISL_418548_2020-03-06 (Dublin)
EPI_ISL_418516_2020-03-06 (Tipperary)
EPI_ISL_418581_2020-03-08 (Dublin)
S53 ward 1 PAT 7.04
S91 ward 1 PAT 29.04
Cluster 4
S33 ward 3 PAT 27.03
S27 ward 10 PAT 3.04
S21 ward 10 PAT 27.03
Cluster 5
S89 ward 11 HCW 8.04
EPI_ISL_414586_2020-03-03 (Limerick)
EPI_ISL_414587_2020-03-03 (Limerick)
S30 ward 11 PAT 10.04
S75 ward 6 PAT 4.04
S40 ward 6 PAT 5.04
S50 ward 6 HCW 31.03
EPI_ISL_418583_2020-03-10 (Dublin)
Cluster 6
S9 ward 1 PAT 29.04
EPI_ISL_418582_2020-03-10 (Louth)
EPI_ISL_417432_2020-03-08 (Cork)
EPI_ISL_414487_2020-03-04 (Cork)
MN908947 SARS-CoV-2
62
63
35
60
85
65
63
70
65
90
94
85
71
81
54
88
56
99
95
87
64
1.0E-5

Figure 3

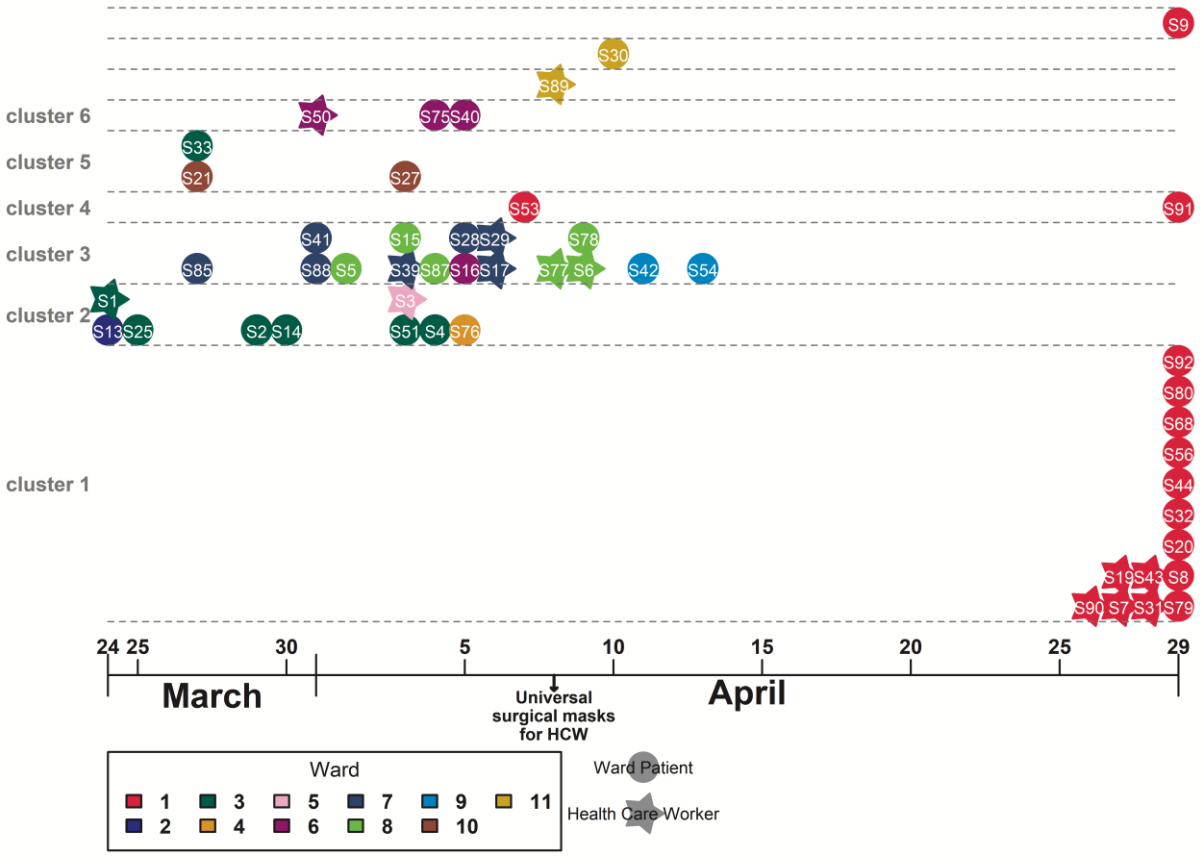

Figure 4


Cluster 6
B.2 / V / 19A
Africa
Asia
Europe
North America
Oceania
South America
Current Study
B.1 / G / 20A
Cluster 5
Cluster 1-3
Cluster 4
B.1.1 / GR / 20B