Molecular Epidemiology of SARS-CoV-2 in Healthcare Workers at Chris Hani Baragwanath Hospital in South Africa

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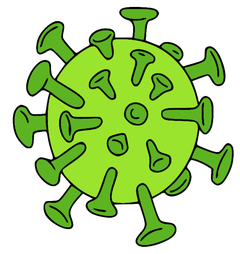
INTRODUCTION

- The SARS-CoV-2 pandemic, starting in December 2019, caused over 3.29 billion cases and 28.9 million deaths by May 2024. Ongoing virus mutations led to new variants, requiring constant monitoring to address challenges in vaccine effectiveness and control measures.
- Healthcare workers (HCWs) face elevated risks of infection due to close patient contact and contaminated environments. HCWs can also contribute to hospital-based transmission, highlighting the need for stringent infection control in healthcare settings.
- Identifying transmission clusters among HCWs is crucial for understanding disease spread within healthcare facilities. Such insights help guide targeted interventions to reduce transmission risks and protect both HCWs and patients.
- This study sequenced the genomes of SARS-CoV-2 infecting HCWs in South Africa, aiming to identify potential transmission clusters during the three waves of the COVID-19 pandemic.

METHODS

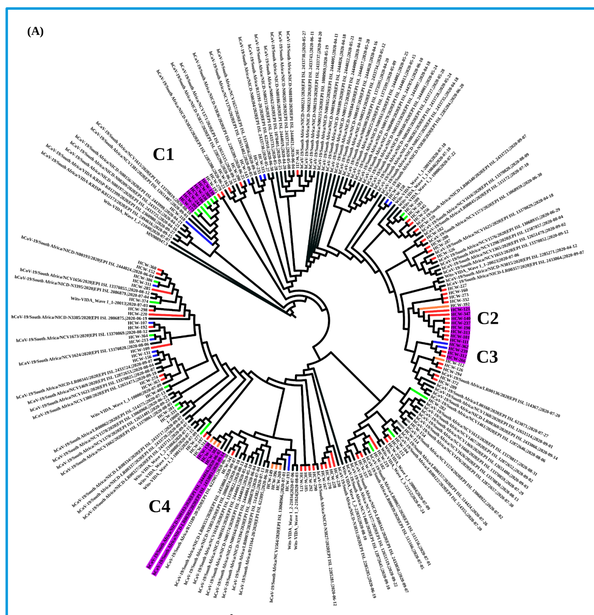
April 2020- May 2022: HCW from 5 departments at CHBAH routinely screened for SARS-CoV-2 irrespective of symptomology as part of a longitudinal surveillance study.
Viral RNA was extracted from nasal swabs and subjected to confirmatory SARS-CoV-2 nucleic acid amplification test.
Nucleic acid with a cycle threshold of <30 were selected for whole genome sequencing on the Illumina iSeq platform



Viral sequences (>90% coverage) were uploaded onto GISAID and analysed using bioinformatic pipelines.
Contemporaneous SARS-CoV-2 sequences extracted from GISAID originating from Johannesburg, South Africa (Africa / South Africa / Johannesburg) were used to contextualize the clustering of HCW sequences with that of the surrounding community.

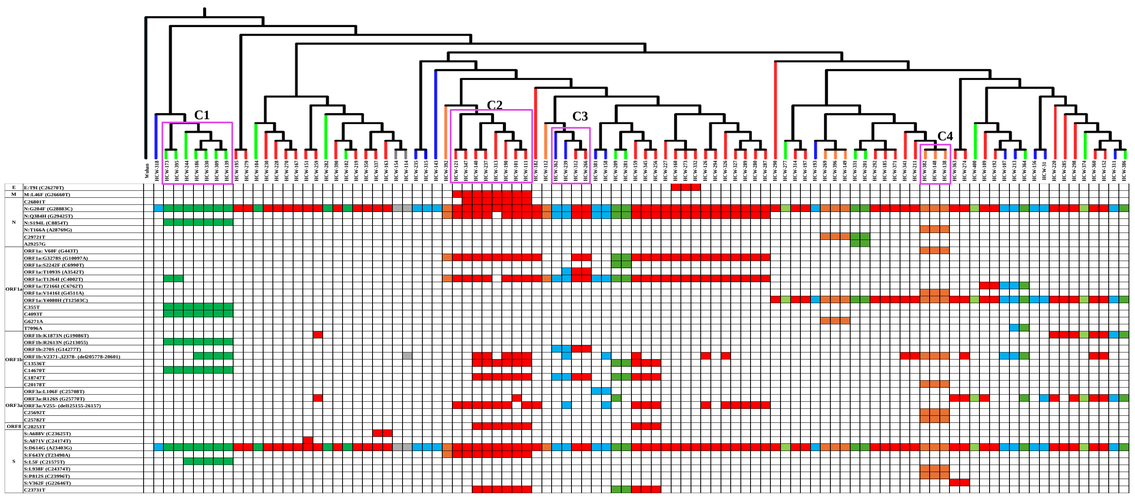
Putative transmission clusters: 10-day window between positive result in cases where HCWs were from the same or related departments
Isogenic sequences confirmed viral sequence similarity in clusters.
Common source of infection: HCWs test positive with 0-2days
HCW-HCW transmission: positive tests occurred between 3-10 days

RESULTS: WAVE 1 PHYLOGENETIC CHARACTERISATION



(B)

Sample ID	Infection Timeline 1 (Days)	Infection Timeline 2 (Days)
Cluster 1		
HCW-309	0	
HCW-139	0	
HCW-244	1	
HCW-330	2	
HCW-186	5	0
HCW-173	14	9
HCW-395	14	9
Cluster 2		
HCW-101	0	
HCW-121	0	
HCW-347	2	
HCW-111	9	
HCW-140	10	
HCW-237	10	
HCW-313	10	
HCW-190	10	
Cluster 3		
HCW-362	0	
HCW-239	7	
HCW-266	12	0
HCW-312	21	9
Cluster 4		
HCW-382	0	
HCW-138	0	
HCW-148	8	

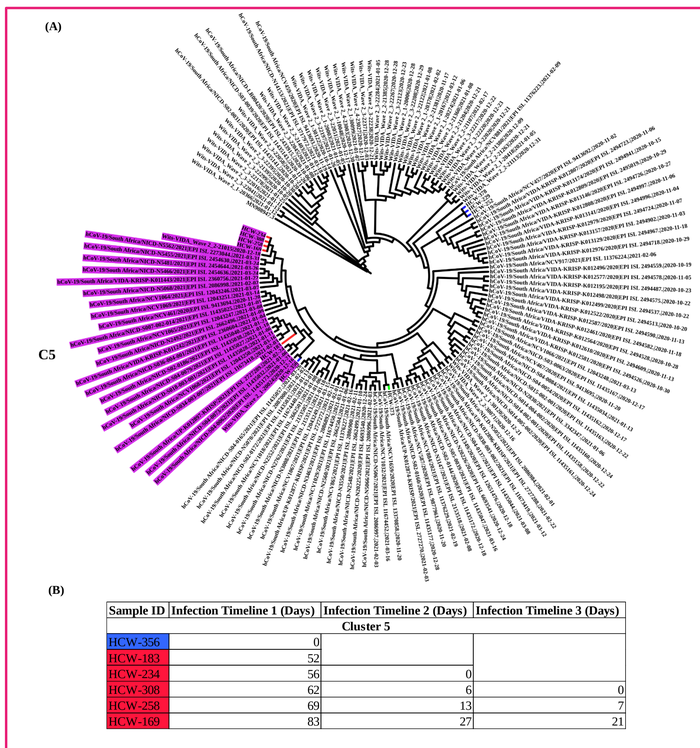


(C)

Figure 1: Phylogenetic characterisation of sequences isolated from viruses detected in the study community and HCW during the first infections wave. The branches of the tree and mutations are coloured according to department from which the SARS-CoV-2 specimen was identified; Internal Medicine (Red), Pediatrics (Green), Laboratory (Blue), Intensive care (Orange) and Obstetrics and Gynecology (Grey). Clusters are indicated in purple (C1-C4). Panel A represents a maximum likelihood phylogenetic tree from the database of HCW, the in-house COVID-19 vaccine trial (RSA, Black) and GISAID (EPI_ISL numbered, Black) sequences for SARS-CoV-2 sequences isolated in Johannesburg, South Africa. Two infection timelines (days) for the sequences in each cluster are shown in Panel B. Mutation, SNP and deletion summary for each sequence from the first wave is represented in Panel C. Abbreviations: SNP: Single Nucleotide Polymorphism, ORF: Open-reading frame, M: membrane, N: nucleocapsid, E: envelope, S: spike and del: deletion, RSA: South Africa

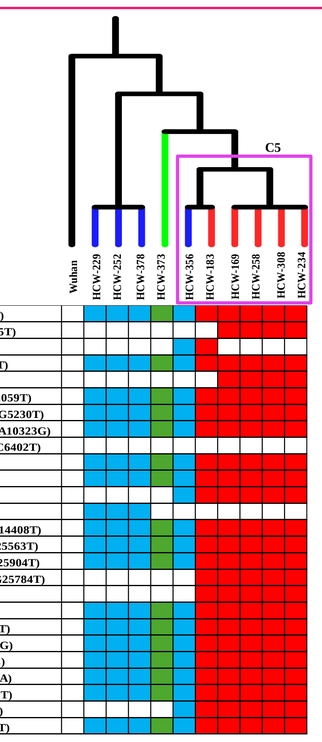
C1 (Pediatrics Department): Seven HCWs formed this cluster, with early cases testing positive within 48 hours, sharing five specific SNPs, indicating a common source of infection. Further HCW-HCW transmissions were identified over the following nine days based on shared genomic profiles.
C2 (Internal Medicine Department): Eight HCWs were part of this cluster. Initial infections on D0 and D2 shared key SNPs, with later cases occurring 9-10 days after and sharing a similar genomic profile, suggesting HCW-HCW transmission from the first positive case.
C3 (Laboratory and Internal Medicine): Four HCWs from two departments formed this cluster, with separate transmission timelines identified. Laboratory HCWs shared one SNP, while Internal Medicine HCWs shared two SNPs, indicating possible HCW-HCW transmission within each department.
C4 (Intensive Care Department): Three HCWs in this cluster likely contracted the virus from a patient, with isogenic viral sequences. HCW-HCW transmission occurred over an 8-day period, based on infection timing and sequence data.

RESULTS: WAVE 2 PHYLOGENETIC CHARACTERISATION



(B)

Sample ID	Infection Timeline 1 (Days)	Infection Timeline 2 (Days)	Infection Timeline 3 (Days)
Cluster 5			
HCW-356	0		
HCW-183	52		
HCW-234	56	0	
HCW-308	62	6	0
HCW-258	69	13	7
HCW-169	83	27	21

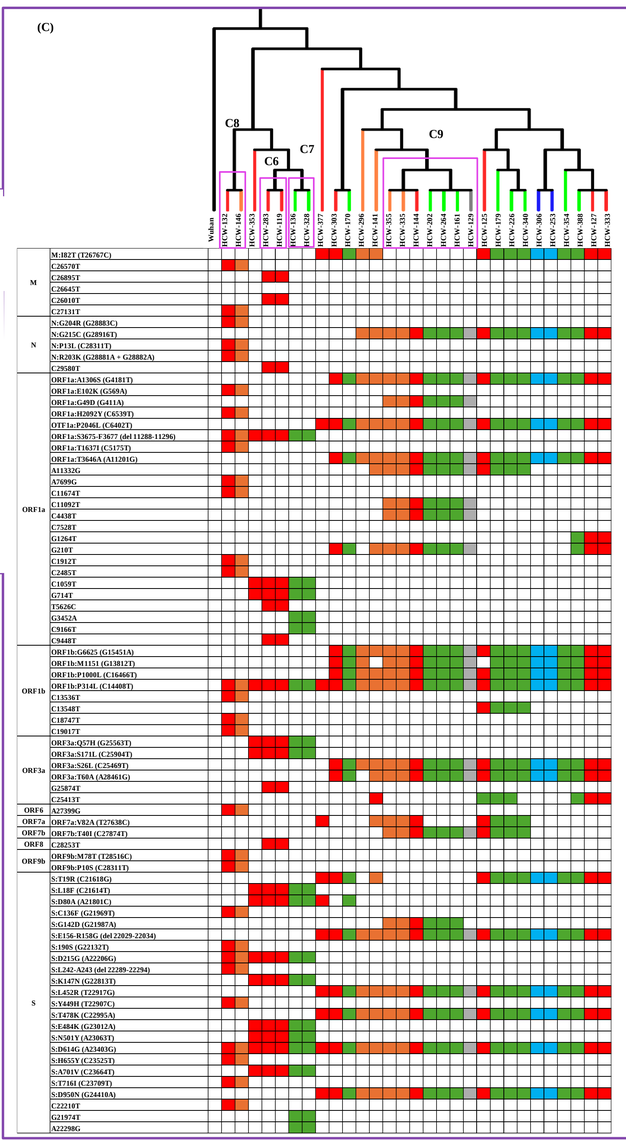


(C)

Figure 2: Phylogenetic characterisation of sequences isolated from viruses detected in the study community and HCW during the second infection wave. The branches of the tree and mutations are coloured according to department from which the SARS-CoV-2 specimen was identified; Internal Medicine (Red), Pediatrics (Green), Laboratory (Blue), Intensive care (Orange) and Obstetrics and Gynecology (Grey). Clusters are indicated in purple (C4). Panel A represents a maximum likelihood phylogenetic tree from the database of HCW, the in-house COVID-19 vaccine trial (RSA, Black) and GISAID (EPI_ISL numbered, Black) sequences for SARS-CoV-2 sequences isolated in Johannesburg, South Africa during the second wave. Three infection timelines (days) for the sequences in each cluster are shown in Panel B. Mutation, SNP and deletion summary for each sequence from the second wave is represented in Panel C

Cluster Composition: Six HCWs, five from Internal Medicine and one from the laboratory, formed C5, with sequences interspersed among 23 community samples, sharing key mutations.
Transmission Events: Two HCWs (HCW-356 and HCW-183) shared an additional SNP but tested positive 52 days apart, suggesting different infection sources.
Internal Medicine Transmission: HCW-HCW transmission was likely within Internal Medicine, as HCW-308 tested positive 6 days after the first case, followed by further transmission to HCW-258.
Community Infection: Despite sharing genomic similarities with the cluster, HCW-234 likely contracted the infection from the community rather than through internal transmission.

RESULTS: WAVE 3 PHYLOGENETIC CHARACTERISATION



(A)

(B)

Sample ID	Infection Timeline 1 (Days)	Infection Timeline 2 (Days)
Cluster 6		
HCW-283	0	
HCW-119	3	
Cluster 7		
HCW-136	0	
HCW-328	2	
Cluster 8		
HCW-146	0	
HCW-132	5	
Cluster 9		
HCW-335	0	
HCW-355	1	
HCW-264	14	
HCW-129	28	0
HCW-161	31	3
HCW-202	45	
HCW-144	40	

Figure 3. Phylogenetic characterisation of sequences isolated from viruses detected in the study community and HCW during the third infection wave. The branches of the tree and mutations are coloured according to department from which the SARS-CoV-2 specimen was identified; Internal Medicine (Red), Pediatrics (Green), Laboratory (Blue), Intensive care (Orange) and Obstetrics and Gynecology (Grey). Clusters are indicated in purple (C6-C9). Panel A represents a maximum likelihood phylogenetic tree from the database of HCW, the in-house COVID-19 vaccine trial (RSA, Black) and GISAID (EPI_ISL numbered, Black) sequences for SARS-CoV-2 sequences isolated in Johannesburg, South Africa during the third wave. Two infection timelines (days) for the sequences in each cluster are shown in Panel B. Mutation, SNP and deletion summary for each sequence from the second wave is represented in Panel C.

C6: Two HCWs from Internal Medicine shared isogenic SARS-CoV-2 sequences, suggesting a likely HCW-HCW transmission event, with HCW-119 testing positive 3 days after HCW-283.
C7: Two HCWs from Pediatrics had isogenic sequences, and the timeline suggests both contracted the virus from the same infected patient, with HCW-328 testing positive 2 days after HCW-136.
C8: One HCW from Intensive Care and another from Internal Medicine shared isogenic sequences, suggesting HCW-HCW transmission, with HCW-132 testing positive 5 days after HCW-146.
C9: Seven HCWs from multiple departments shared some common SNPs, but the timelines and limited department crossover suggest community importation rather than HCW-HCW transmission, except for a potential transmission event between HCW-129 and HCW-161 from Obstetrics and Pediatrics, respective

DISCUSSION AND CONCLUSION

The study analysed 164 SARS-CoV-2 samples from HCWs at CHBAH. Phylogenetic analysis identified 9 putative transmission clusters involving 41 HCWs, with 25 showing evidence of HCW-HCW transmission and 16 linked to unidentified common sources.
Only **13%** of SARS-CoV-2 infections among HCWs were due to **HCW-HCW transmission**, contrasting with higher rates reported in other studies.
Most HCWs were likely **infected through community exposure** or contact with patients rather than fellow HCWs.
This lower rate might be due to **rigorous weekly testing** and self-quarantining protocols, which **reduced HCW-HCW transmission opportunities**.
This study highlights that routine surveillance and stringent infection control are vital for protecting HCWs, who were more often infected by patients than colleagues, underscoring the need for ongoing genomic surveillance to safeguard these frontline defenders against severe disease.