

Application of Whole Genome Sequencing – COVID-19 and beyond

Department of Health Technology and Informatics Dr. Gilman K H SIU Associate Professor, Limin Endowed Young Scholar in Medical Laboratory Science The Hong Kong Polytechnic University

IDIC Theme Based Training Seminar Nov 2022

Opening Minds • Shaping the Future 啟迪思維 • 成就未來

Global phylogeny of COVID-19



GISAID: https://gisaid.org/phylodynamics/global/nextstrain/ (as of 7 Nov 2022)

Rapid sequencing and automatic analysis pipeline





Health Bureau The Government of the Hong Kong Special Administrative Region of the People's Republic of China HMRF commissioned project

Total number of SARS-CoV-2 samples sequenced

No. of genomes sequenced in Hong Kong

(as of Nov 8, 2022)

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No. of genomes sequenced by PolyU (as of Nov 8, 2022)

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(~52.0% of the sequenced cases in HK)

SARS-CoV-2 Phylogeny (HK local cases)



The Impact – Preventing the importation of cases (外防輸入)



The Impact – Preventing the importation of cases (外防輸入)



The Impact – Halting the spread of the virus in the community (內防擴散)

G

Centers for Disease Control and Prevention CDC 24/7: Saving Lives, Protecting People™

(IF: 16.126) Search

EMERGING INFECTIOUS DISEASES

EID Journal > Volume 27 > Number 1—January 2021 > Main Article

Volume 27, Number 1—January 2021

Research

Territorywide Study of Early Coronavirus Disease Outbreak, Hong Kong, China

Kenneth Siu-Sing Leung, Timothy Ting-Leung Ng, Alan Ka-Lun Wu, Miranda Chong-Yee Yau, Hiu-Yin Lao, Ming-Pan Choi, Kingsley King-Gee Tam, Lam-Kwong Lee, Barry Kin-Chung Wong, Alex Yat Man Ho, Kam-Ton Yip, Kwok-Cheung Lung, Raymond Wai-To Liu, Eugene Yuk-Keung Tso, Wai-Shing Leung, Man-Chun Chan, Yuk-Yung Ng, Kit-Man Sin, Kitty Sau-Chun Fung, Sandy Ka-Yee Chau, Wing-Kin To, Tak-Lun Que, David Ho Keung Shum, Shea Ping Yip, Wing Cheong Yam, and Gilman Kit-Hang Siu

Wave 1

Time point: *Jan Feb* 2020 Major Lineages : *B.43* Source: *Returnees from China*





Leung et al. *Emerg Infect Dis* 2020

- Cases in Eastern District were caused by a single source → Hidden transmission chain
- Identified the potential source of the Buddha Worship Hall cluster
- Demonstrated the possibility of "hidden spreader" as a source of COVID-19 community outbreak.



MAJOR ARTICLE



Nosocomial Outbreak of Coronavirus Disease 2019 by Possible Airborne Transmission Leading to a Superspreading Event

Vincent Chi-Chung Cheng,^{1,2,a} Kitty Sau-Chun Fung,^{3,a} <u>Gilman Kit-Hang Siu,^{4,a} Shuk-Ching Wong</u>,^{1,a} Lily Shui-Kuen Cheng,³ Man-Sing Wong,⁵ Lam-Kwong Lee,⁴ Wan-Mui Chan,⁶ Ka-Yee Chau,³ Jake Siu-Lun Leung,⁴ Allen Wing-Ho Chu,⁶ Wai-Shan Chan,³ Kelvin Keru Lu,⁴ Kingsley King-Gee Tam,⁶ Jonathan Daniel Ip,⁶ Kenneth Siu-Sing Leung,⁶ David Christopher Lung,⁷ Herman Tse,⁸ Kelvin Kai-Wang To,⁶ and Kwok-Yung Yuen⁶









 T470N is specific to the transmission chains disseminating in Kowloon East District.

THE LANCET Regional Health Western Pacific

Complementation of contact tracing by mass testing for successful containment of beta COVID-19 variant (SARS-CoV-2 VOC B.1.351) epidemic in Hong Kong

Vincent Chi-Chung Cheng^{*},^{1,2} <u>Gilman Kit-Hang Siu^{*}</u>,³ Shuk-Ching Wong^{*},¹ Albert Ka-Wing Au,⁴ Cecilia Suk-Fun Ng,⁴ Hong Chen,⁴ Xin Li,² Lam-Kwong Lee,³ Jake Siu-Lun Leung,³ Kelvin Keru Lu,³ Hazel Wing-Hei Lo,³ Evelyn Yin-Kwan Wong,³ Shik Luk,⁵ Bosco Hoi-Shiu Lam,⁵ Wing-Kin To,⁵ Rodney Allan Lee,⁶ David Christopher Lung,⁷ Mike Yat-Wah Kwan,⁸ Herman Tse,⁹ Shuk Kwan Chuang,⁴ Kelvin Kai-Wang To,¹⁰ Kwok-Yung Yuen¹⁰





30 Apr 2021

(D23) d

5 May 2021 (D28)





- First team to identify the transmission link between Filipino domestic helpers and an Indian businessman
- Reported to CHP within 12 hours after sample receival
- Facilitated tracing the entire transmission chain and their close contacts.
- Avoid another episode of community outbreak

Current research gap – Difficult to understand and lack of spatiotemporal data



Current research gap – Lack of phylogenetic relatedness and transmission pathway



("Home - Johns Hopkins Coronavirus Resource Center", 2022)

("Latest Situation of Novel Coronavirus Infection in Hong Kong", 2022)

Conventional phylogeny and Evolution rate



Construction of the "Time Tree"

- The number of mutation can be used as a 'molecular clock' to date the divergence between sequences.
- The tree was re-rooted to infer the internal nodes (ancestors).
- This tree demonstrates the phylogenetic relatedness and divergence of SARS-CoV-2 cases in Hong Kong



Construction of the "Time Tree"

- The dates of case confirmation were added
- Branch lengths correspond directly to elapsed time
- Each node is placed such that its position reflects its known or inferred date.





Frequencies (colored by Lineage)



https://nextstrain.org/groups/ncovHK/ncov/Overview_

Colour the cases with different attributes

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Genomic epidemiology of novel coronavirus - Hk-focused subsampling

Built with nextstrain.com/groups/novHK/. Maintained by Gilman Siu's research team.

Showing 365 of 365 genomes sampled between Dec 2019 and Oct 2021.



Filter out the cases with Delta variant Lineage AY.23

60%

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Genomic epidemiology of novel coronavirus - Hk-focused subsampling

Built with nextstrain.com/groups/novHK/. Maintained by Gilman Siu's research team.

Showing 365 of 365 genomes sampled between Dec 2019 and Oct 2021.



Filter out the cases in Tai Po



Early stage of 5th wave of outbreak

5th wave

Time point: 29 Dec 2021 – Present Major Lineages : *AY.127, BA.1, BA.1.1, BA.2* Source: *Aircrew exempted from quarantine, Imported case from Nepal and Kwai Chung Estate Cluster, Hamster pet shop*



Omicron Lineage BA.2.2

Omicron Lineage BA.1

Omicron Lineage BA.1.1

Delta Lineage AY.127

Early stage of 5th wave of outbreak



Aircrew exempted from quarantine (CHP#12611) → BA.1.1 Transmission



Aircrew exempted from quarantine (CHP#12676) – BA.1 Transmission



Maple Garder

 Revealed the transmission routes of cases sourced from infected aircrews → Successfully contained the "aircrew clusters"

Hamster-to-human transmission (CHP#13047) – Delta AY.127 Transmission



Hamster-to-human transmission (CHP#13047) – Delta AY.127 Transmission

Taylor & Francis

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Emerging Microbes & Infections

ISSN: (Print) (Online) Journal homepage: https://www.tandfonline.com/loi/temi20

Co-circulation of two SARS-CoV-2 variant strains within imported pet hamsters in Hong Kong

Kin-Hang Kok, Shuk-Ching Wong, Wan-Mui Chan, Lei Wen, Allen Wing-Ho Chu, Jonathan Daniel Ip, Lam-Kwong Lee, Ivan Tak-Fai Wong, Hazel Wing-Hei Lo, Vincent Chi-Chung Cheng, Alex Yat-Man Ho, Bosco Hoi-Shiu Lam, Herman Tse, David Lung, Ken Ng Ho-Leung Ng, Albert Ka-Wing Au, Gilman Kit-Hang Siu & Kwok-Yung Yuen







Imported case from Nepal (CHP#12891,#13045) → BA.2.2

frontiers Frontiers in Public Health (IF: 6.461)

Understanding spatiotemporal symptom onset risk of Omicron BA.1, BA.2 and hamster-related Delta AY.127

Chengzhuo Tong¹, Wenzhong Shi^{1*}, Gilman Kit-Hang Siu², Anshu Zhang¹ and Zhicheng Shi^{3*}

¹Department of Land Surveying and Geo-Informatics, Otto Poon Charitable Foundation Smart Cities Research Institute, The Hong Kong Polytechnic University, Kowloon, Hong Kong SAR, China, ²Department of Health Technology and Informatics, The Hong Kong Polytechnic University, Kowloon, Hong Kong SAR, China, ²Research Institute for Smart Cities, School of Architecture and Urban Planning, Shenzhen University, Shenzhen, China





- Rapid sequencing identified the import of a new Omicron variant BA.2
- First team to identify the sources of the outbreak in Kwai Chung Estate, the start of the 5th wave

Transmission chains of four BA.2.2 amino acid mutants



Genotyping of Imported SARS-CoV-2 variants identified at HK Airport





No. and proportion of local SARS-CoV-2 vs Daily death



Long TJ et al (Unpublished study, 2022)

- More emerging lineages are expected in local cases in the coming winter
- XBB.1 is the most mutated variant so far (64 aa changes and 87 nucleotide changes, i.e. >99.7% genomic similarity, when compared to the original strain in Wuhan)



 Introduction of new Omicron variants is unlike to cause an upsurge of death and hospitalization rate

XB characteristic mutations

Application of High-Throughput Sequencing – Beyond COVID-19



Application of High-Throughput Sequencing – Beyond COVID-19





Melioidosis Outbreak in Kowloon West Cluster in 2022

- From January to July 2022, 4 cases of melioidosis were diagnosed;
- from August to October 2022, an unusual upsurge of 18 cases of melioidosis were detected in the Kowloon West Cluster



Melioidosis Outbreak in Kowloon West Cluster in 2022

Burkholderia pseudomallei in the soil and muddy water may expose to the ground after typhoons
 → spread easier with strong wind and storms.



Air sampling near Pak Tin Construction site

 B.pseudomallei was cultured from one of the air samples, which was collected on 15 Aug, 2022 (5 days after typhoon Mulan).



Core-Genome Phylogeny of clinical and environmental isolates



Wu WGV et al (Unpublished study, 2022)

Soil sampling at the construction site on 5 Oct 2022





Report our finding to CHP



Satellite image of the Pak Tin Estate in 2017 and 2022











Analysis of proportion of urban vegetation by multispectral satellite imagery



Wu WGV et al (Unpublished study, 2022)

- Continuous reduction in urban vegetation in Shum Shui Po District since 2016
- Unvegetated soil is more likely to be picked up by the wind
- Leading to soil erosion during typhoon → facilitate airborne transmission of bacteria embedded in soil

Application of High-Throughput Sequencing – Beyond COVID-19



Routine Surveillance of Foodborne Antimicrobial Resistance (AMR)



Proportion of MDRO in food samples



- MDRO were isolated in around 20% of food samples
- ESBL-PE and CRE represents the most encountered MDRO in raw meat
- Carbapenem resistant Acinetobacter spp. is more common in sashimi

Spatial distribution of foodborne MDRO

Overall MDRO (ESBL-PE, CRE, VRE, CRA, CRPA)





Resistome in different food products and their import origins



Phylogenetic Relatedness between foodborne and clinical VRE



Application of High-Throughput Sequencing – Beyond COVID-19



Current workflow for diagnosis bloodstream infection



Proposed workflow of nanopore sequencing-based assays for rapid diagnosis of BSI



The concordance of bacterial taxa between ONT sequencing and culture method



Clinical utility of metagenomic and amplicon-based sequencing



Application of High-Throughput Sequencing – Beyond COVID-19



Targeted amplicon sequencing for drug resistant TB

Food and Health Bureau



Funding supported by

Nanopore MinION-based assay for HIV resistance testing







DeepChek Patient / Sample Information Physician / Project Information Sample Patient Alternative ID Date Collected Name 19/07/2019 Institution Date Reported Name 24/07/2019 Address DOB Sequencing platform Gende Software version 2.0 Viral Expert system 2.2 Load Sample, barcode08_500_Long_Takara_Barcoding_LongAmp_1step ID. COVERED POSITION PROT 02_AG (96.25 %) RT 0206 (91.71 %) **PROT** 1-99 RT 1-440 INT 09 CPX (95.03 %) GP120 G (78.93 %) INT 1-289 GP41 1-20,71-141 PROT, INT, GPS20, GP41; - Subtype B x00405 was a allowing in the Third of 7 GP120 1-511 ANTIRETROVIRAL DRUG DEEPCHEK* HIV (ANRS 29) Resistance mutations between >3% and <20.009 Generic name Assessment Resistance mutations >20.00% Enfuvirtide S Fostemsavir S Atazanavir/r S G16E (4.38%), L33I (3.08%) Darunavir/r 600/100 mg S BID Darunavir/r 800/100 mg ā QD K20M (3.38%), K20R (4.7%), L24I (7.86%), F53L Lopinavir/r (5.16%) M36I (82.96%), H69K (49.65%), F53L (5.16%), L89I (6.85%) Tipranavir/r L89M (81.54%) K65R (7.73%), L210W (4.39%) Abacavir K65R (7.73%) Emtricitabin K65R (7.73%) Lamiyudine K65R (7.73%), K65E (10.54%), L210W (4.39%) Tenofovir Zidovudine L210W (4.39%) Doravirine L100I (5.6%) Efavirenz L100I (5.6%), K101E (17%) Etravirine L100I (5.6%), K101E (17%), V179I (6.94%) Nevirapine L100I (5.6%), K101E (17%) Rilpivirine L100I (5.6%), K101E (17%), K103R (6.75%) E157Q (86.19%) Bictegravir Cabotegravir E157Q (86.19%) Dolutegravir 50mg BID Dolutegravir 50mg QD E157Q (86.19%) T66I (4.42%), T97A (3.16%), Y143H (3.56%), E157Q (86.19%) Elvitegravin P145S (4.46%) E157Q (86.19%) Y143H (3.56%) Raltegravir Susceptible Possible resistance R Resistance 1

For the selected protein and drug, at least one amino acid mutation defined in the related drug resistance guideline and located in the expected range of covered positions (between amino acids 40 and 348 tor region RT, 1 and 36 tor region RT, 1 a determined.

Funding supported by



Identify drug resistance mutation and differentiate quasispecies



Nanopore MinION-based assay for HIV resistance testing

clusters compositions

20 27 25

25

The agreement between ONT and NGS in high AF
Number of sample: 42
AF > 0.8 in both ONT and NGS





of INDEL

29 25

26

Heatmap of gene and drug resistance(s) in subtypes



*Resistance level: HL: High-Level Resistance, IL: Intermediate Resistance, LL: Low-Level Resistance, PLL: Potential Low-Level Resistance

Acknowledgement



Dr. Franklin CHOW Dr. Annie LEE Mr. Lam Kwong LEE Mr. Timothy NG Mr Ivan WONG Mr. Jake LEUNG Ms. Chloe CHAN Ms. Hiu Yiu LAO Mr. Kelvin LU Ms. Sharon FUNG Mr. Jain NG Mr. Sky WONG

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> > Dr. Kingsley TAM

Prof. Honglin CHEN

Dr. Bobo MOK

Dr. Teng LONG

Dr. Tommy LAM

Dr. Marcus HUNG

Dr. Vincent CHENG Prof.. KY YUEN