LKS Faculty of Medicine
HKU-Pasteur Research Pole
香港大學-巴斯德研究中心

Annual Report
2021

Roberto Bruzzone, Co-Director
Leo Poon, Co-Director
Malik Peiris, Honorary Director

HKU-Pasteur Research Pole
7/F Jockey Club Building for Interdisciplinary Research
5, Sassoon Road, Hong Kong SAR

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Summary

Mission

The HKU-Pasteur Research Pole (HKU-PRP) is a joint laboratory, established by The University of Hong Kong (HKU) and the Institut Pasteur (IP), under the School of Public Health (SPH) of the Li Ka Shing Faculty of Medicine of the University of Hong Kong. HKU-PRP aims to confront the challenges posed by microbes by developing research and education programs that willcontribute to mitigate the impact of infectious diseases.

Research

We have organized our activity around Group Leaders who are engaged in competitive research projects aligned with the scientific priorities of HKU and IP. We focus on emerging and re-emerging infectious diseases (respiratory and mosquito-borne viruses) antimicrobial resistance (AMR) and computational approaches to investigate diversity and evolution of pathogens. The pandemic caused by the new SARS-CoV-2 led us to re-orient research priorities, producing an impressive research output in collaboration with the School of Public Health, the Center for Immunology & Infection and and other laboratories worldwide. HKU-Pasteur has remained involved in sharing methods and data with laboratories worldwide. We have contributed to filling knowledge gaps on the biology of SARS-CoV-2 by producing data on sero-epidemiology, immune response in adults and children, developing experimental animal models, and probing the systems biology of the host response to this novel pathogen. The demonstration that surveillance of sewage waters with sensitive molecular tests could provide early warning of COVID-19 outbreaks, in collaboration with the Department of Engineering at HKU has received the Gold Medal at the 2021 Inventions Geneva Evaluation Days. HKU-Pasteur has published over 60 papers since January 2021.

Teaching

HKU-PRP has pioneered a unique course series in Hong Kong and in the region that provides state of the art lectures and practical workshops in a "Master class" setting to outstanding postgraduate students and postdoctoral fellows coming from countries with markedly different resources. Unfortunately, the ongoing pandemic led to cancellation of all courses in 2021 and 2022. We hope that our educational program will resume, gradually, in 2023, when travel restrictions are lifted. All our courses have obtained the Pasteur International Courses "PIC label", which has been established to increase the visibility and branding of the educational programs of the Pasteur Network. The new PIC Labelling Program will ensure that, in an increasingly competitive environment, the "Institut Pasteur" excellence will be recognized as best-in-class for research and training, attracting students and researchers, as well as laboratory technicians and health professionals.

Perspectives

We have developed a strong identity to promote the missions of HKU, IP and the Institut Pasteur International Network, through research, teaching and public health activities. We have opened the Center for Immunology & Infection (C2I), the new collaborative project of HKU and the Institut Pasteur, funded with a major grant from the Innovation & Technology Commission. This translational research laboratory will address significant public healthcare challenges through novel technology platforms for biomarker discovery and the development of new vaccine and therapeutic strategies. We have recruited a new Group Leader, who joined us at the beginning of 2022, to maintain our focus on understanding the immunological and virological determinants of antibody responses after respiratory virus infection and vaccination at a population as well as at the individual level. An external scientific review was undertaken in June 2021. The reviewers noted that HKU-Pasteur has evolved into a world-renowned research center, exemplified by continuing high-impact scientific output, provision of highly rated international courses and acquisition of ample extramural funding over the last two decades. Its embedment within the HKU School of Public Health in 2013 further facilitated HKU-Pasteur to be a productive regional hub for the Pasteur Network.

2. Overview of the Programs

2.1 Research

The scientific activity of HKU-PRP is organized around core research questions that meet the overarching goals of Internationalization, Innovation, Interdisciplinarity and Impact (HKU's 3+1). We combine wet lab and computational approaches to understand in mechanistic terms the interactions between microbes (pathogenic or commensals), hosts and the environment. The three main pillars of our research are: *Understanding how do viruses invade, replicate and escape infected cells*; *Understanding what makes a microbe pathogenic*; *Understanding how do microbes deal with the host immune response and the environment*.

Research in **the lab of Vijaykrishna Dhanasekaran** aims to elucidate the evolution and epidemioly of rapidly evolving pathogens through **disease surveillance in the Asia-Pacific, sequencing, genomics and integrative phylodynamics and population genetics experiments**. Since establishment in Hong Kong in September 2020, the new lab has rapidly characterised the genomic epidemiology of SARS-CoV-2 during the first four waves in Hong Kong, and described the effects of disruptions due to COVID-19 on animal and human influenza and other respiratory viruses.

Leo Poon was appointed as Co-Director in July 2020. His lab studies viruses at the animal and human interface, with a particular interest on influenza virus and coronaviruses. The overarching goal is to generate experimental evidences to develop evidence-based control measures by developing three interrelated research areas: Emerging Infectious Diseases; Basic Virology and vaccinology; and Molecular diagnosis. **Research on COVID-19 resulted in 26 publications in this reporting period**. Major scientific achievements included monitoring SARS-CoV-2 variant introductions and their transmission dynamic in Hong Kong, which led to revising governmental policy against COVID-19 (e.g. setting up quarantine hotels for incoming travelers and banning importation of Syrian hamsters for commercial purposes); conducting a serological study of vaccine-induced antibody response against Omicron variant , which led to the governmental recommendation of using the mRNA vaccine Comirnaty as a booster, as well as assessing several new materials for developing "smart surfaces" to inactivate infectious SARS-CoV-2. For these and other contributions on COVID-19 control, **the Poon lab has received an award (Outstanding Project Team on COVID-19 Research Award) from Food and Health Bureau, Hong Kong Government**.

The group of Hein Min Tun uses conventional microbiology and molecular biology techniques, cutting-edge sequencing technologies, coupled with bioinformatics, statistical and epidemiological approaches with the goal to contribute to improving scientific understanding of the impact of microbiome and AMR in public health. Briefly, the Tun lab is 1) studying the composition, function, and dynamics of human and animal microbiomes in health and disease; and 2) monitoring antimicrobial resistance (AMR) bacteria and resistome in humans, animals, and the environment using a holistic One Health approach. **His lab is one of the teams behind the multidisciplinary project "Innovative Sewage Testing Tool for SARS-CoV-2", which has been awarded a Gold Medal at the Special Edition of the 2021 Inventions Geneva Evaluations Day**.

The main objectives of the group of Sophie Valkenburg are to define immune correlates of protection for influenza viruses from infection and vaccination. These approaches were adapted to address the COVID-19 pandemic. The lab's primary focus is

to study adaptive immunity to these viruses, and how this could be harnessed and optimized by vaccination to improve protection from infection. A vaccine which ultimately combines antibody and T cell-based immunity will provide a full-proof immunological barrier to infection and variants, which their studies, aimed at elucidating how cross-reactive T and B cell responses provide broad immunity, will ultimately help develop. **Sophie Valkenburg left HKU-Pasteur in December 2021 to take up a position as Associate Professor at the Peter Doherty Institute, University of Melbourne, Australia**, but is actively involved in several funded projects that are supporting a smaller lab still working at HKU-Pasteur.

Sook-San Wong was appointed Assistant Professor in January 2022. Her lab is focused on understanding the immunological and virological determinants of robust antibody responses after respiratory virus infection and vaccination at a population as well as at the individual level. **The laboratory research platform is based on immunological assays focusing on humoral immunity, using samples from clinical and epidemiological cohorts as well as from animal models**. This research is critical to our understanding of respiratory viruses' vaccine efficacy and pathogenesis and how that affects the population's susceptibility to respiratory viruses. The lab is also studying population immunity to zoonotic viruses and how that contributes to our risk assessment process of such pathogens.

Malik Peiris and Leo Poon have expanded their efforts to confront the SARS-CoV-2 pandemic, which resulted in an impressive research output produced by scientists in the School of Public Health and HKU-Pasteur, with the following major outcomes: **Leo Poon has successfully secured** (as the coordinator) **a 5-year grant to continue research on COVID-19, funded through the Theme-based Research Scheme**. This program features 10 principal investigators, with **two other HKU-Pasteur Grtoup Leaders as co-principal investigators**.

2.2 Teaching and Education

HKU-PRP has pioneered in Hong Kong a teaching program of excellence that has been established to train in biomedical sciences a selected group students coming from all over the world. We offer three major international courses on an annual/biennial basis – Cell Biology, Virology and Immunology – which feature lectures from leading scientists and have received increasing support from extramural funding. All HKU-Pasteur courses have been approved by the Research Postgraduate Committee of HKU for inclusion in the coursework curriculum of MPhil/PhD students. The coronavirus pandemic led to the cancellation of all HKU-Pasteur courses, as well as the Public Health Workshop series at the Pasteur Institute of Ho Chi Minh City**,** in 2021 and 2022. All Group Leaders are actively engaged in our international courses as well as in the undergraduate and postgraduate curriculum of HKU.

We have hosted 1 international student for laboratory placement, **from the University of Chicago (USA), 2 interns from the French International School**, and **six training students from the Hong Kong Institute of Vocational Education**.

2.3 Other Major Activities

We retain leadership roles in a number of global projects. **Roberto Bruzzone** is the Chair of the Board of Directors of the International Severe Acute Respiratory and Emerging Infection Consortium (https://ww.isaric.org), a network of networks which aims at ensuring that clinical researchers have the open access protocols and data-sharing processes needed to facilitate a rapid response to emerging diseases. **Malik Peiris** continues to serve on a number of WHO working groups in relation to both avian and swine origin influenza virus and is the Co-Director of the WHO H5 Reference Laboratory at HKU. **Leo Poon** currently is a committee member of the Coronavirus study group, ICTV, IUMS, and is an Advisor to the Hong Kong SAR for Food and Environmental Hygiene. He is also an Ad Hoc Expert of the WHO Influenza Molecular Diagnosis Working Group, and of the WHO Expert group for COVID-19 for clinical diagnosis and virus evolution. He has been involved in developing guidelines for these organizations to control COVID-19 (e.g. Guidelines for molecular surveillance of COVID-19 for WHO).

We have inaugurated the Center for Immunology & Infection (C2I), funded by the Innovation and Technology Commission in April 2021**, in the presence of Mr Alexandre Giorgini, Consul General of France in Hong Kong and Macau** and **Mr Leo Kung, member of the Advisory Committee of HKU-Pasteur**. C2I leverages highly multi-disciplinary expertise, developed by the parent institutions, to tackle public health challenges through four major programmes: *The Healthy Human Global Project*; *Development of novel vaccine platforms for influenza*; *Mosquito borne viruses: Epidemiology, pathogenesis and interventions*; *Platform technologies for responding to lethal respiratory virus infections*. **C2I is co-directed by Malik Peirs (Co-Director and Managing Director) and Roberto Bruzzone (Co-Director)**. C2I will contribute to Hong Kong's transformation into an international innovation and technology hub of the Greater Bay Area of Guangdong, Hong Kong and Macau.

3. Progress Report

3.1 Vijaykrishna Dhanasekaran Lab

Main Objectives and Strategy

Our research is to identify factors that shape the emergence, evolution, and spread of rapidly evolving pathogens. To do this we conduct disease surveillance in animal, humans, and the environment, characterise virus genomes and virus-host interactions using next-generation sequencing methods, and apply computational methods to link the sequence data with clinical, epidemiological and immunological data that are generated from disease surveillance and laboratory experiments. These include, to investigate factors affecting the diversity and evolution of pathogens in their natural reservoirs such as birds and bats; in livestock production systems; to elucidate origins of human pandemics; and to infer factors affecting the transmission of human pathogens as driven by host innate and adaptive immune systems. Our primary organism of study is influenza and coronavirus, although we have ongoing projects in other respiratory and gastroenteric pathogens such as RSV and Enterovirus 71, vector-borne pathogens such as Dengue and Ross River, and more recently on the genomics of bacterial pathogens such as *Klebsiella pneumoniae*. The Vijay lab moved to HKU-Pasteur in September 2020.

Achievements and Ongoing Research

INFLUENZA

Human seasonal influenza under COVID-19 and the potential consequences of influenza lineage elimination [Funding: US National Institute of Allergy and Infectious Diseases (NIAID) (HHSN272201400006C)]

Annual epidemics of seasonal influenza cause hundreds of thousands of deaths, high levels of morbidity, and substantial economic loss. Yet, global influenza circulation has been heavily suppressed by public health measures and travel restrictions since the onset of the COVID-19 pandemic (**Figures 1, 2**). Notably, the influenza B/Yamagata lineage has not been conclusively detected since April 2020, and A(H3N2), A(H1N1), and B/Victoria viruses have since circulated with considerably less genetic diversity (**Figure 3**). Travel restrictions have largely confined regional outbreaks of A(H3N2) to South and Southeast Asia, B/Victoria to China, and A(H1N1) to West Africa (**Figure 4**). Seasonal influenza transmission lineages continue to perish globally, except in these select hotspots, which will likely seed future epidemics. Waning population immunity and sporadic case detection will further challenge influenza vaccine strain selection and epidemic control. We offer a perspective on the potential short- and long-term evolutionary dynamics of seasonal influenza and discuss potential consequences and mitigation strategies as global travel gradually returns to pre-pandemic levels.

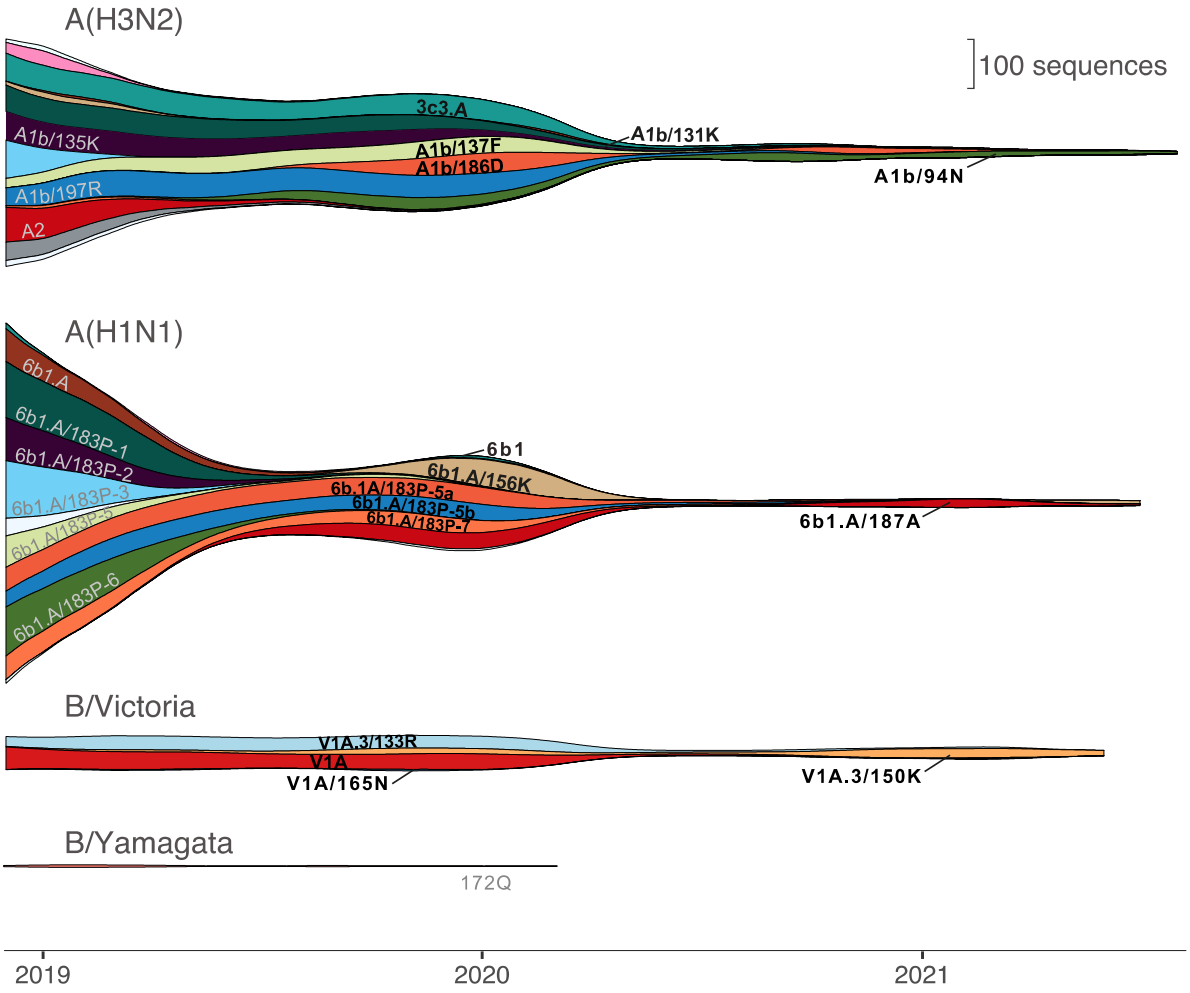

Figure 1. Streamgraph showing temporal changes in influenza lineage circulation. Lineage prevalence was estimated using sample collection dates of all sequences submitted to the Global Initiative for Sharing All Influenza Data (GISAID) from December 2018 to August 2021. Lineages detected since April 2020 are labelled in black; lineages that have not been detected since April 2020 are labelled in gray.

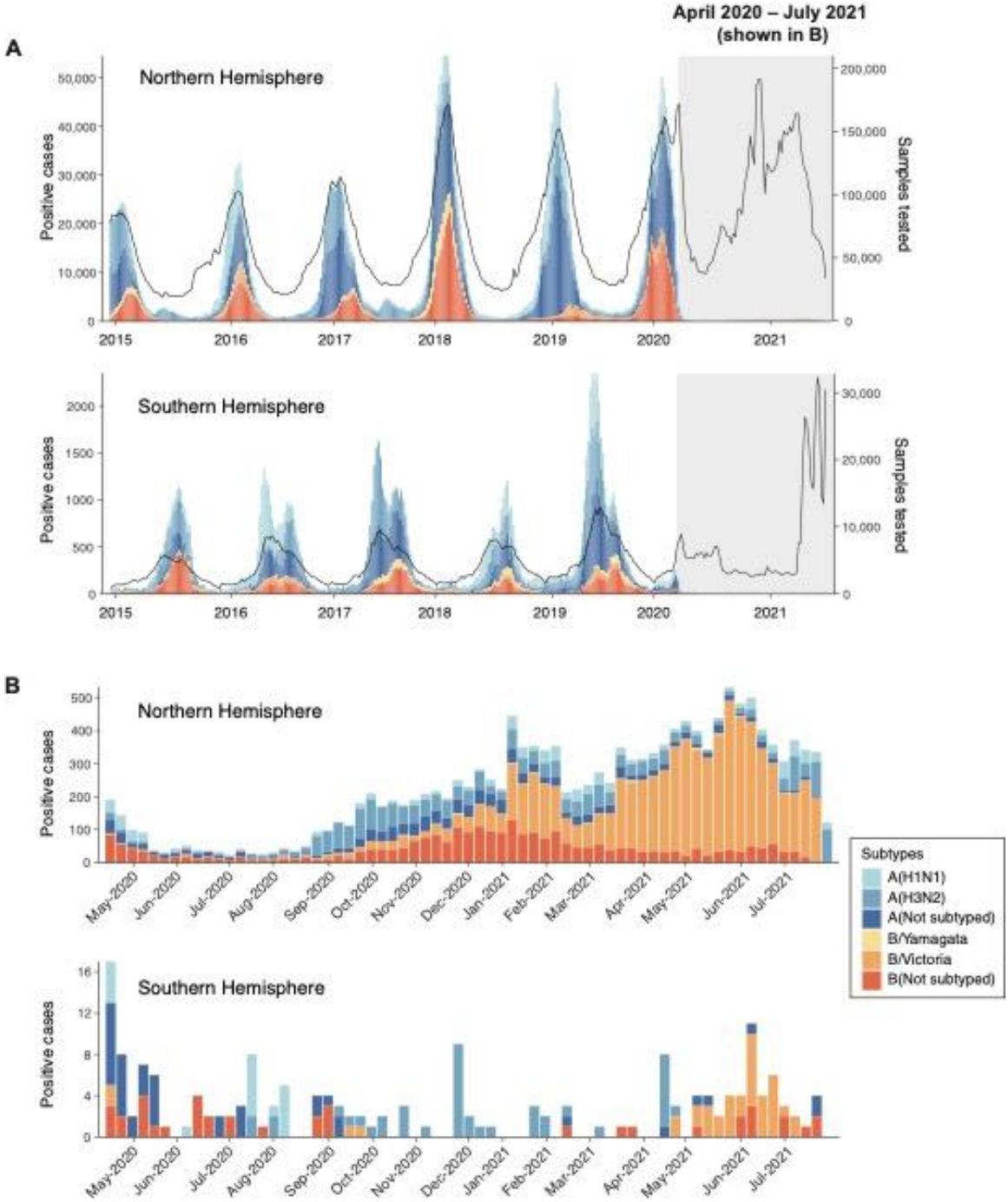

Figure 2. Virological surveillance of seasonal influenza viruses. **A**, Time series comparing FluNet data on seasonal influenza activity in the Northern and Southern Hemispheres from 2015 to July 2021, with the COVID-19 pandemic shaded in gray. Stacked bar chart (left-hand y-axis) represents the number of influenza-positive cases per week colored by subtype. Black trend-line (right hand y-axis) shows the number of specimens tested per week. **B**, Magnified view of the gray-shaded bar charts in A showing influenza-positive specimens from April 2020 to July 2021. Note: y-axis scales differ in each panel.

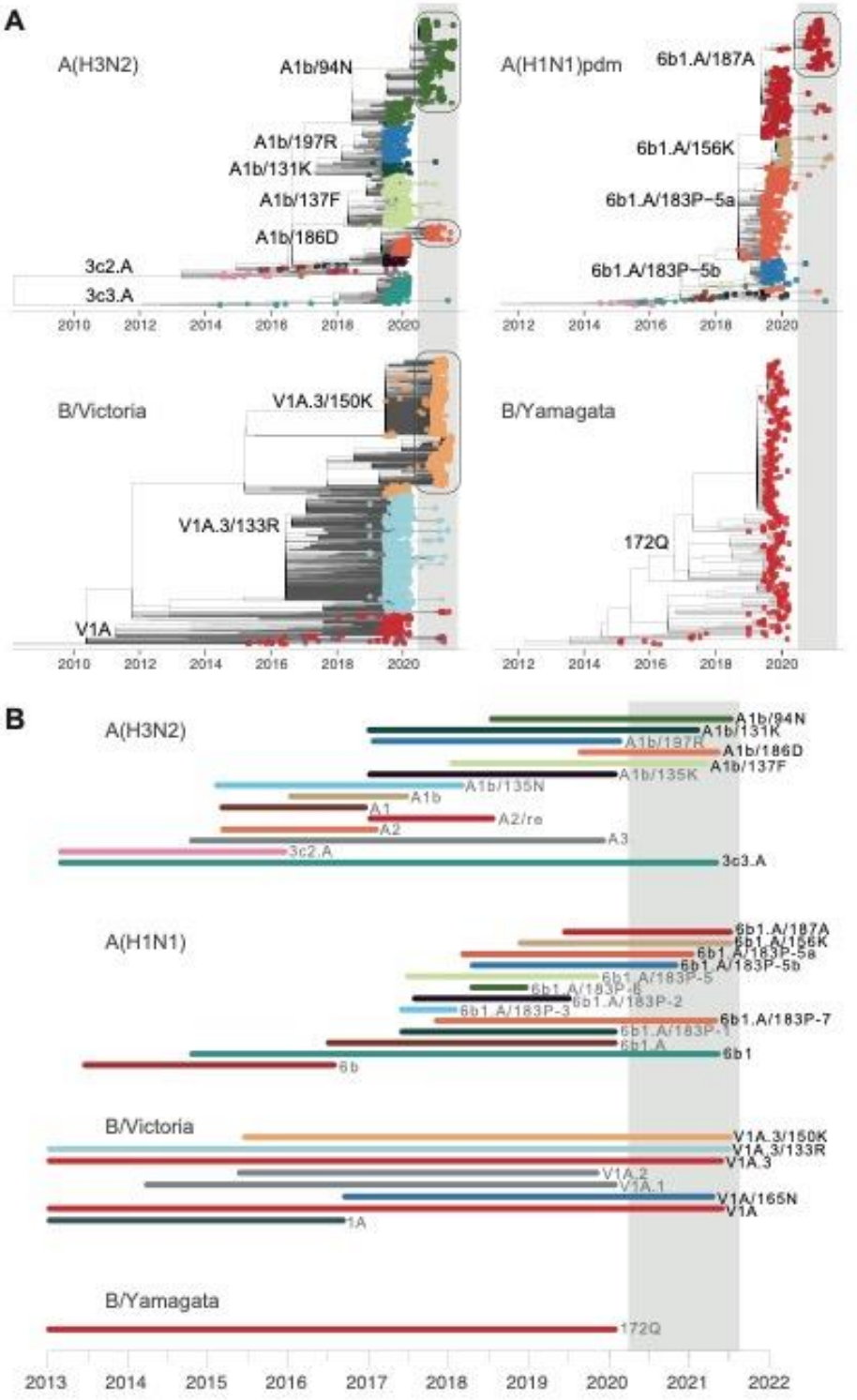

Figure 3. Comparison of seasonal influenza virus evolution before and since COVID-19 emergence. A, Evolutionary relationships and divergence times of HA genes inferred using maximum likelihood (see Online Methods). Tips are colored by WHO clade designations. Phylogenetic trees of major clades that continued to circulate during 2020/2021 (marked in A) are shown in Supplementary Figures 1–4, including A(H3N2) clades A1b/94N and A1b/186D; A(H1N1) clade 6b1.A/187A lineages in West Africa; and B/Victoria lineages from several regions around the world. **B**, Timeline of recently circulating seasonal influenza virus HA clades from mean estimated divergence time to most recent sequence. Time since the onset of the COVID-19 pandemic is shaded in gray.

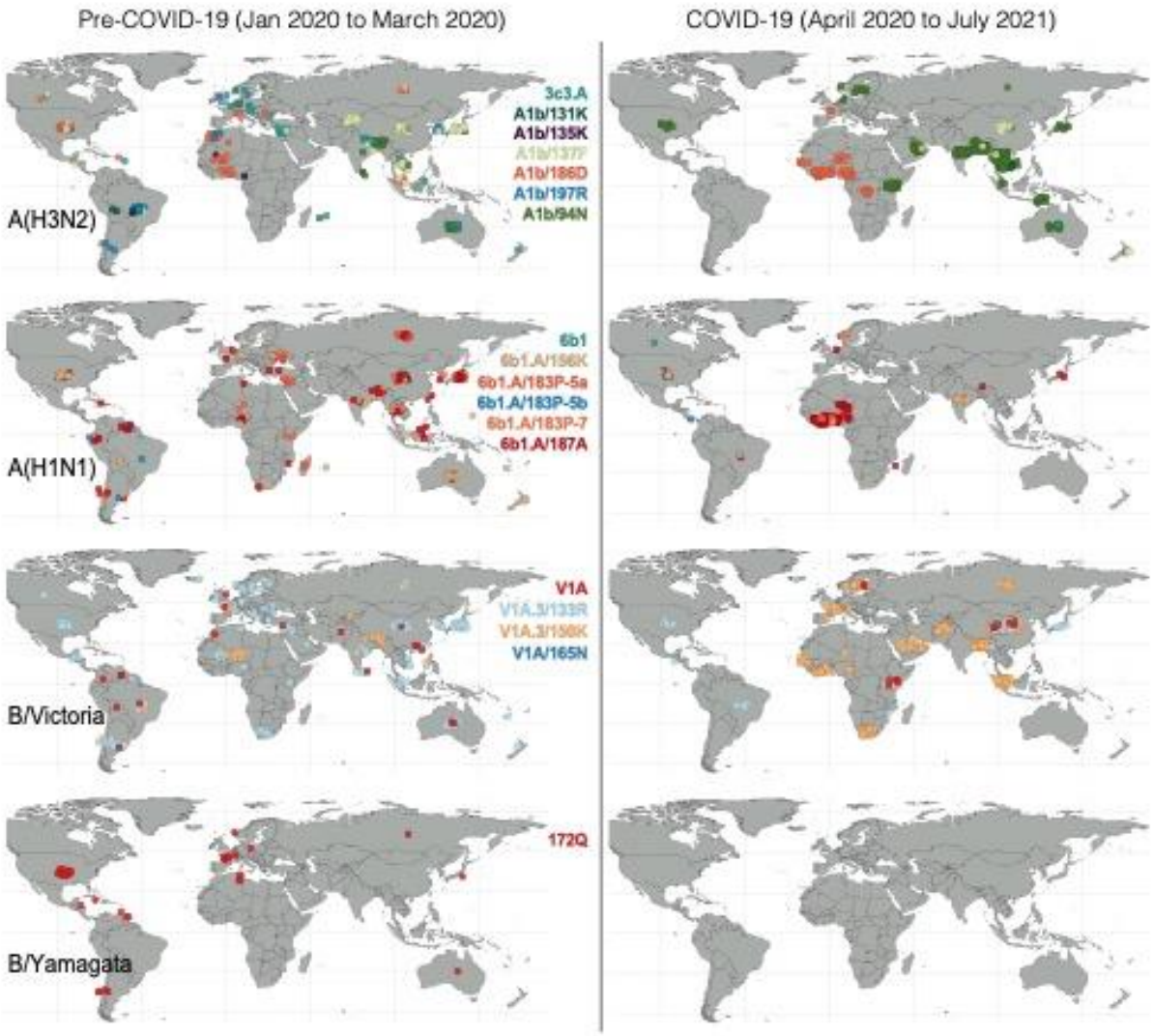

Figure 4. Geographic distribution of influenza HA sequences before (left) and after (right) COVID-19 emergence. From April 2020 to July 2021, A(H3N2) (590 sequences from 32 countries), A(H1N1) (254 sequences from 18 countries), and B/Victoria (834 sequences from 34 countries) were available for analysis. In comparison to the 16-month period before April 2020 (December 2018 to March 2020), there was a reduction in sequences of 97% for A(H3N2), 99% for A(H1N1), 92% for B/Victoria, and 100% for B/Yamagata. Last updated on August 24th, 2021.

Lineage-specific protection and immune imprinting shape the age distributions of influenza B cases [Funding: US National Institute of Allergy and Infectious Diseases (NIAID) (HHSN272201400006C)]

How a history of influenza virus infections contributes to protection is not fully understood, but such protection might explain the contrasting age distributions of cases of the two lineages of influenza B, B/Victoria and B/Yamagata (**Figure 5**). Fitting a statistical model (**Figure 6**) to those distributions using surveillance data from New Zealand, we found they could be explained by historical changes in lineage frequencies combined with cross-protection between strains of the same lineage (**Figure 7**). We found additional protection against B/Yamagata in people for whom it was their first influenza B infection, similar to the immune imprinting observed in influenza A (**Figure 8**). While the data were not informative about B/Victoria imprinting, B/Yamagata imprinting could explain the fewer B/Yamagata than B/Victoria cases in cohorts born in the 1990s and the bimodal age distribution of B/Yamagata cases. Longitudinal studies can test if these forms of protection inferred from historical data extend to more recent strains and other populations.

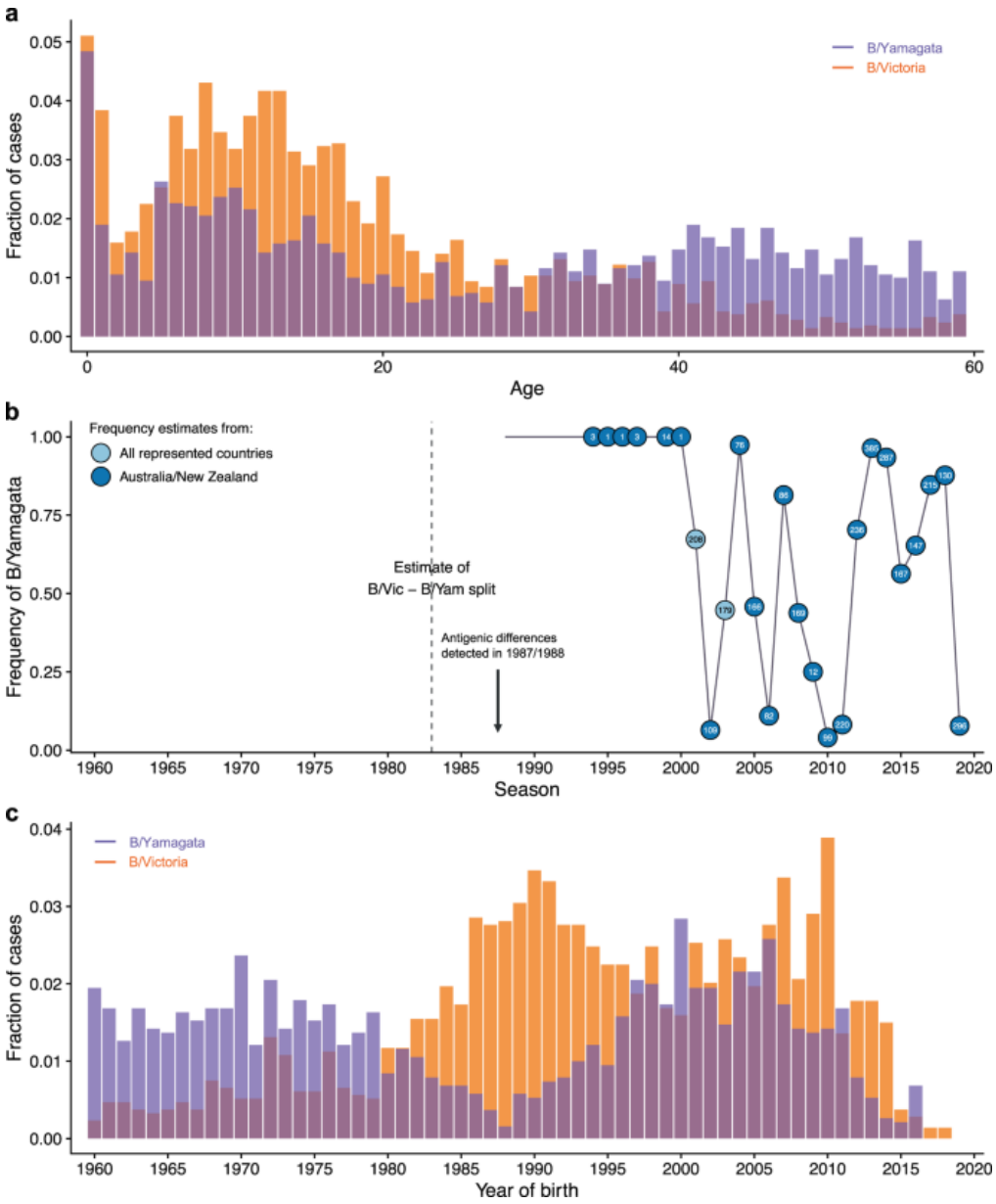

Figure 5. Historical frequencies and age distributions of the influenza B lineages. **A**, Distribution of medically attended influenza B cases in New Zealand in 2001–2019 by age (in years) of the infected person. **B**, Frequency of B/Yamagata estimated from sequences deposited on GISAID and the NCBI Influenza Virus Database. Frequencies were estimated for New Zealand and Australia combined to increase power. The circles are annotated with the number of isolates used to estimate frequencies in each season. In 2001 and 2003, when both lineages were known to be circulating in Australia and New Zealand but the number of isolates from those countries combined were small, we estimated lineage frequencies using isolates from all countries represented in the sequence databases. **C**, Distribution of medically attended influenza B cases in New Zealand in 2001–2019 by birth year of the infected person. In **A** and **C**, the fraction of cases was calculated relative to all cases observed for each lineage (including ages and birth years not shown in the figure).

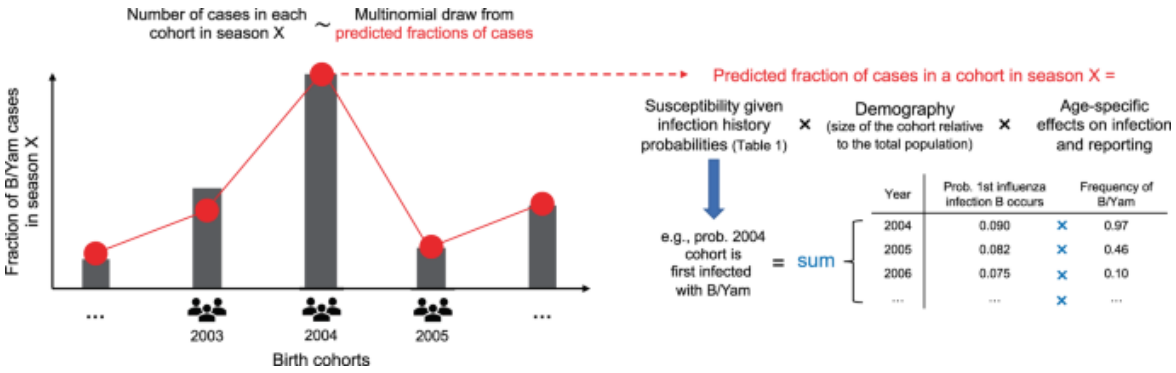

Figure 6. Statistical model of influenza B infections by birth year. Extending the model developed by Gostic et al.10, for each influenza B season we predicted the fraction of cases observed in each cohort based on cohort-specific infection histories and on additional factors. The probabilities of the different infection histories in Table 1 are derived in the "Methods" using a discrete-time probabilistic model of infection. As an example, the diagram shows how the probability that a person is first infected with B/Yamagata depends on the frequency of B/Yamagata in the years after birth. Given a person's first influenza B infection occurs in a particular year, we assumed that the probability this infection was caused by a particular lineage is equal to the frequency of the lineage in that year. We obtained the total probability of being first infected with a particular lineage by summing across all possible years when the first influenza B infection might have occurred. The susceptibility of each cohort is then calculated as a weighted sum of the susceptibilities associated with each infection history. In addition to infection history, other factors that affect the fraction of cases observed in a birth cohort include the cohort's size relative to the total population and the effect of age itself (rather than year of birth) on infection risk and on the probability that an infection receives medical attention and becomes a case. We fitted the model by maximum likelihood assuming the distributions of cases by birth year in different seasons were independent multinomial draws.

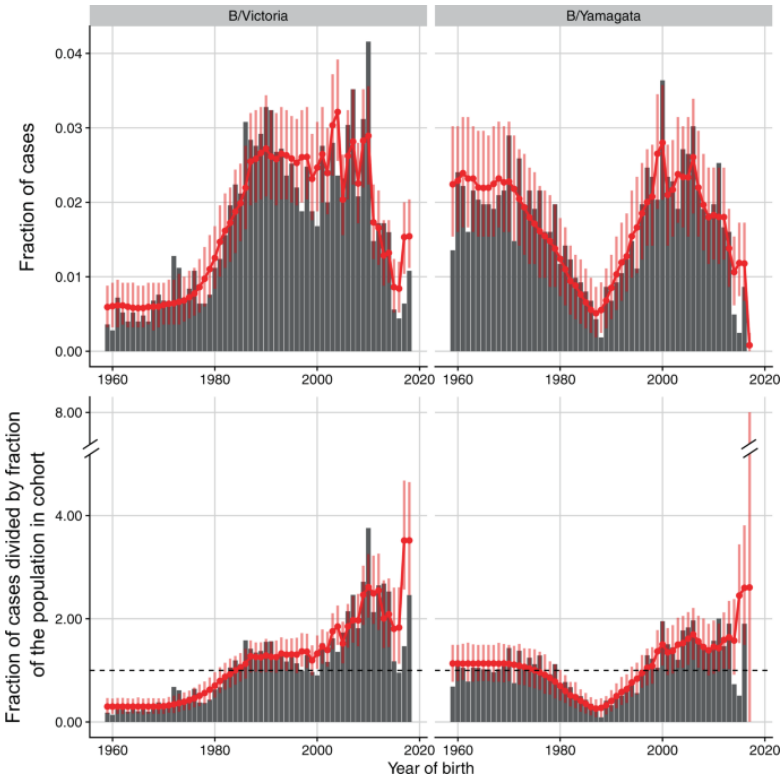

Figure 3. Observed and predicted distributions of influenza B cases in New Zealand by birth year. The model was simultaneously fitted to the age distributions in each observation year from 2001 to 2019, accounting for uncertainty in the birth year of each reported case given the patient's age. For plotting, we pooled observed and predicted numbers of cases across observation years for each birth year, assuming the earliest possible birth year for each age (e.g., an age of 10 years in 2000 was assumed to correspond to the birth year 1989). Red lines and dots show the fraction of cases in each birth cohort as predicted by the model. Vertical bars are 95% bootstrap confidence intervals based on n = 1000 multinomial draws from the predicted distributions indicated by the dots. In the bottom row, predicted and observed fractions of cases were normalized by dividing by the fraction of the population born in that birth year (i.e., the null expectation if all birth years were infected at the same rate).

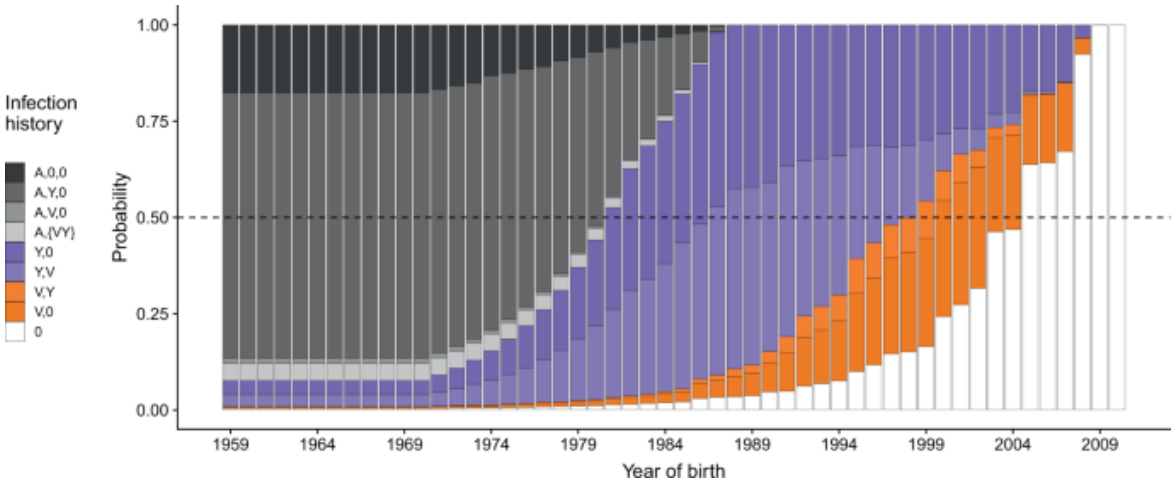

Figure 4. Probabilities of different infection histories with influenza B in New Zealand for people born between 1959 and 2010, and observed in 2010. Infection histories consist of the lineage of first infection and lineages encountered later regardless of their order. Probabilities were estimated by fitting the model to case data from New Zealand. (A,0,0): first infection before 1988 and no subsequent infections with either B/Victoria or B/Yamagata. (A,Y,0) and (A,V,0): first infection before 1988 followed by B/Yamagata but not B/Victoria and by B/Victoria but not B/Yamagata, respectively. (A,{V,Y}): first infection before 1988 followed by infections with both B/Victoria and B/Yamagata in any order. (Y,V) and (Y,0): first infection with B/Yamagata, with and without a subsequent B/Victoria infection. (V,Y) and (V,0): first infection with B/Victoria, with and without a subsequent B/Yamagata infection. (0): fully naive to influenza B.

SARS-CoV-2

Genomic epidemiology of SARS-CoV-2 under an elimination strategy in Hong Kong [Funding: US National Institute of Allergy and Infectious Diseases (NIAID) (HHSN272201400006C)]

Hong Kong employed a strategy of intermittent public health and social measures alongside increasingly stringent travel regulations to eliminate domestic SARS-CoV-2 transmission. By analyzing 1899 genome sequences (>18% of confirmed cases) from 23-January-2020 to 26-January-2021, we reveal the effects of fluctuating control measures on the evolution and epidemiology of SARS-CoV-2 lineages in Hong Kong (**Figures 9, 10**). Despite numerous importations, only three introductions were responsible for 90% of locally-acquired cases. Community outbreaks were caused by novel introductions rather than a resurgence of circulating strains. Thus, local outbreak prevention requires strong border control and community surveillance, especially during periods of less stringent social restriction. Non-adherence to prolonged preventative measures may explain sustained local transmission observed during wave four in late 2020 and early 2021. We also found that, due to a tight transmission bottleneck, transmission of low-frequency single nucleotide variants between hosts is rare.

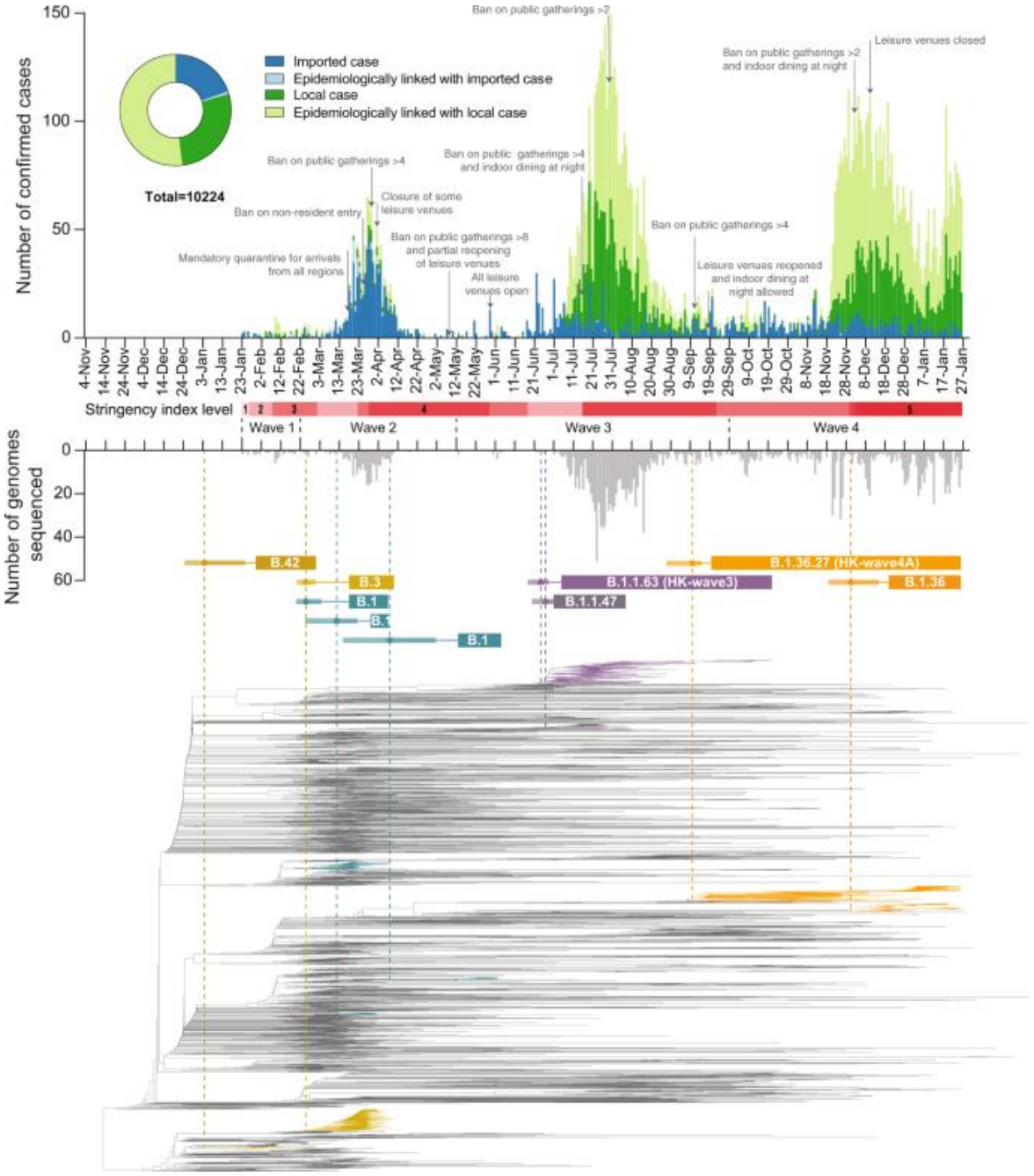

Figure 9. Epidemiological summary and time-scaled phylogeny of SARS-CoV-2 in Hong Kong. Confirmed cases (above) and sequenced genomes (below) are shown as bar charts across the four pandemic waves. Control-measure stringency applied in Hong Kong is based on the Oxford COVID-19 Government Response Tracker17. Time-scaled phylogeny of representative genomes from Hong Kong (n=610) and overseas regions (n=1,538) shows monophyletic clades containing at least five community cases in Hong Kong. The two largest Hong Kong lineages during HK-wave3 and HK-wave4A, B.1.1.63 and B.1.36.27, were subsampled to 100 and 65 sequences, respectively.

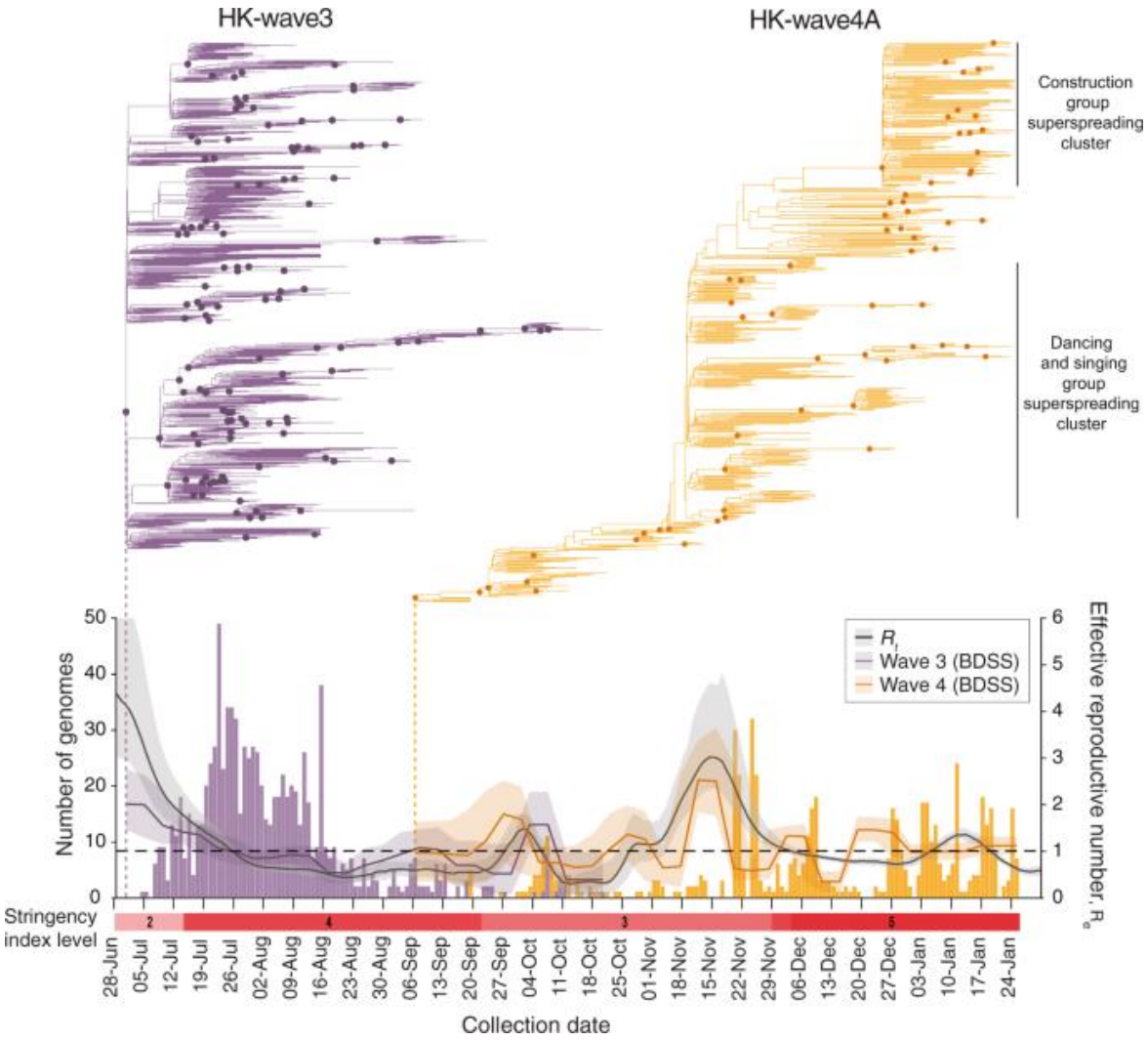

Figure 10. Phylodynamics of waves three and four in Hong Kong. Evolutionary relationships and effective reproduction number (Re(t)) of HK-wave3 (B.1.1.63) and HK-wave4A (B.1.36.27) estimated using tree heights and sequenced incidence data. Node shapes indicate posterior probability >0.5. Histogram shows the number of genomes by collection date. Control-measure stringency applied in Hong Kong is based on the Oxford COVID-19 Government Response Tracker17. Black line shows the instantaneous effective reproduction number (Rt), estimated based on infection dates inferred from reported symptom onset or confirmation dates for asymptomatic cases.

Air travel-related outbreak of multiple SARS-CoV-2 variants [Funding: US National Institute of Allergy and Infectious Diseases (NIAID) (HHSN272201400006C)]

A large cluster of 59 cases were linked to a single flight with 146 passengers from New Delhi to Hong Kong in April 2021. This outbreak coincided with early reports of exponential pandemic growth in New Delhi, which reached a peak of > 400 000 newly confirmed cases on 7 May 2021. Epidemiological information including date of symptom onset, date of positive-sample detection and travel and contact history for individual cases from this flight were collected. Whole genome sequencing was performed, and sequences were classified based on the dynamic Pango nomenclature system. Maximum-likelihood phylogenetic analysis compared sequences from this flight alongside other cases imported from India to Hong Kong on 26 flights between June 2020 and April 2021, as well as sequences from India or associated with India-related travel from February to April 2021 and 1217 reference sequences. Sequence analysis identified six lineages of SARS-CoV-2 belonging to two variants of concern (Alpha and Delta) and one variant of public health interest (Kappa) involved in this outbreak (**Figure 11**). Phylogenetic analysis confirmed at least three independent sub-lineages of Alpha with limited onward transmission, a superspreading event comprising 37 cases of Kappa and transmission of Delta to only one passenger (**Figure 12**). Additional analysis of another 26 flights from India to Hong Kong confirmed widespread circulation of all three variants in India since early March 2021. The broad spectrum of disease severity and long incubation period of SARS-CoV-2 pose a challenge for surveillance and control. As illustrated by this particular outbreak, opportunistic infections of SARS-CoV-2 can occur irrespective of variant lineage, and requiring a nucleic acid test within 72 hours of departure may be insufficient to prevent importation or in-flight transmission.

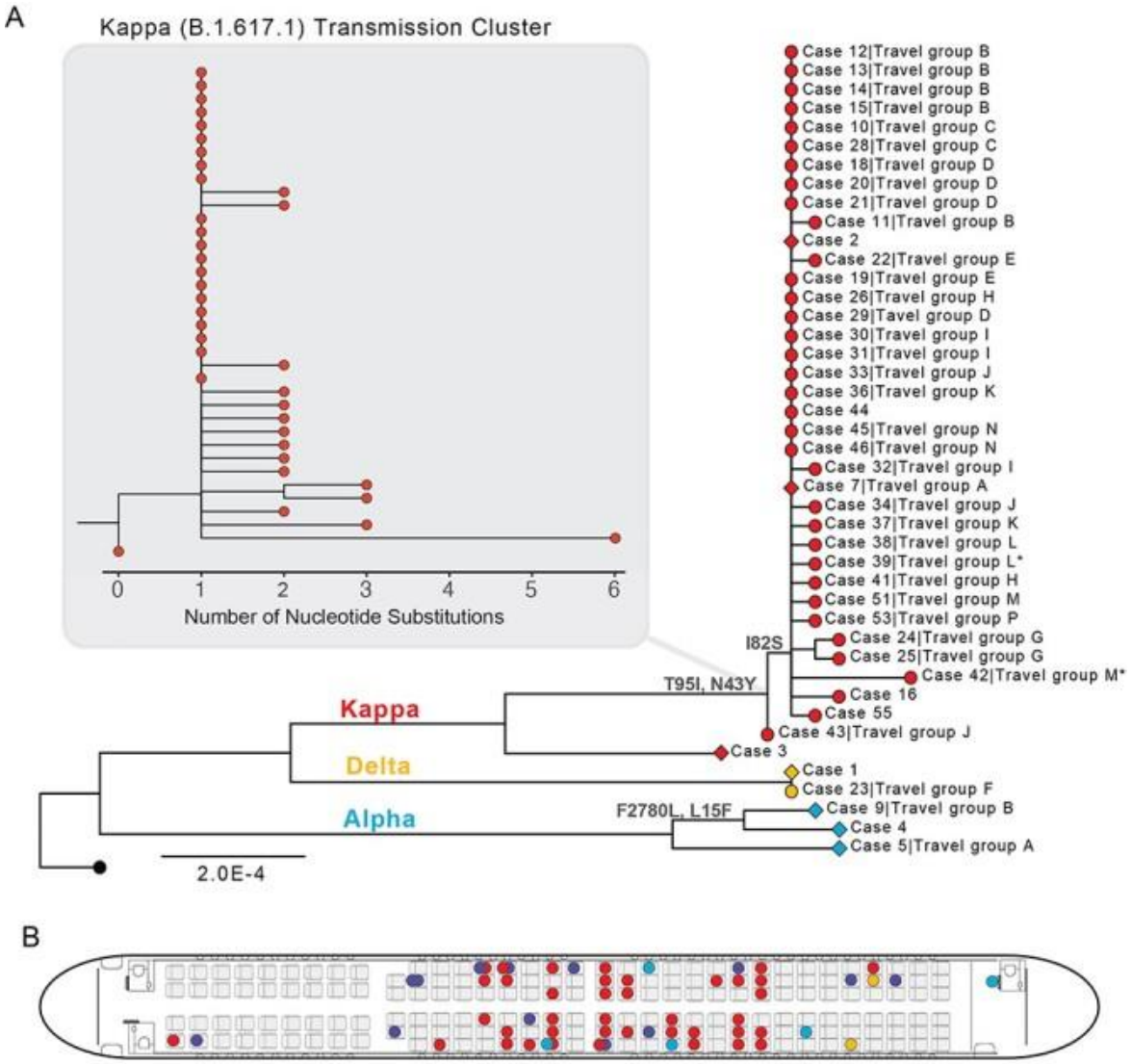

Figure 11. SARS-CoV-2 transmission associated with a flight from New Delhi to Hong Kong in April 2021. Colours represent WHO VOC/VOI and Pango lineage designations. **A**, ML phylogeny of 43 genomes sequenced from 59 cases. Wuhan-Hu-1 (MN908947.3, black tip) was used to root the tree. Diamond tip shapes indicate index cases and circles represent secondary-transmission cases; tips are labelled with case numbers and travel groups. Asterisk denotes sequences with lower coverage (~73%). Nodes are labelled with observed amino acid mutations. The Kappa (B.1.617.1) transmission cluster is magnified to show nucleotide substitutions among samples. **B**, Seat assignments of passengers testing positive. Seats with two positive cases indicate adults seated with children under the age of two.

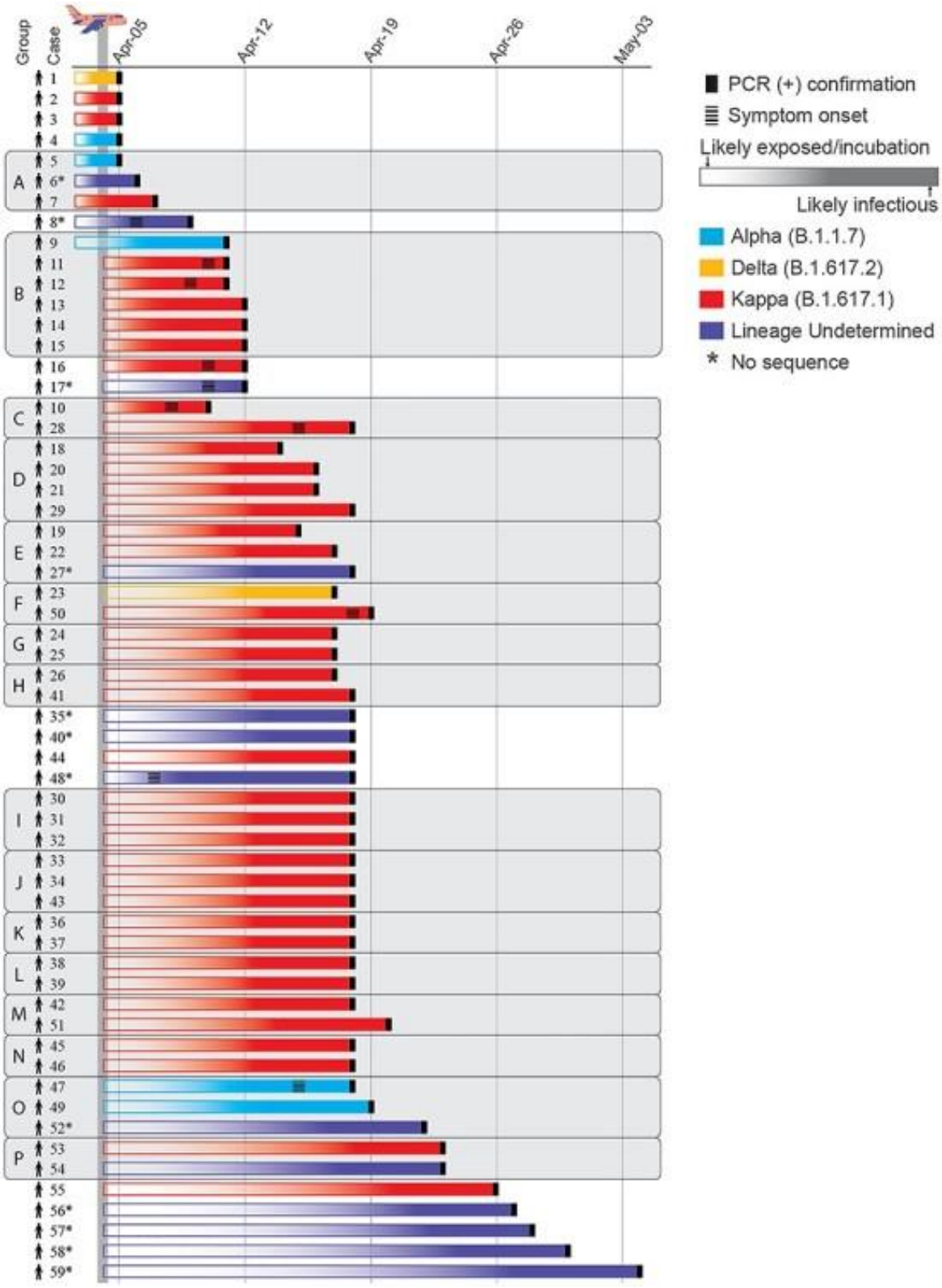

Figure 12. Timeline from exposure to detection for each of 59 SARS-CoV-2 cases associated with a flight from New Delhi to Hong Kong in April 2021. Colours represent WHO VOC/VOI and Pango lineage designations. Individuals travelling together are grouped and shaded in grey. Asterisk indicates cases from which samples were unavailable or unable to be sequenced.

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Publications

1) Dhanasekaran V, Sullivan S, Edwards KE, Xie R, Valkenburg SA, Cowling BJ, Barr IG (2022) Human seasonal influenza under COVID-19 and the potential consequences of influenza lineage elimination. *Nat Commun*, **13**:1721.

2) Gu H, Xie R, Adam DC, Tsui JL, Chu DK, Chang LDJ, Cheuk SSY, Gurung S, K.Ng DYM, Liu GYZ, Wan CKC, Cheng SSM, Edwards KM, Leung KSM, Wu JT, Tsang DNC, Leung GM, Cowling BJ, Peiris M, Lam TTY, Dhanasekaran V, Poon LLM† (2022) Genomic epidemiology of SARS-CoV-2 under an elimination strategy in Hong Kong. *Nat Commun*, **13**:736.

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6) Gu H, Chu DKW, Chang LDJ, Cheuk SSY, Gurung S, Krishnan P, Ng DYM, Liu GYZ, Wan CKC, Xie R, Cheng SSM, Cowling BJ, Tsang DNC, Peiris M, Dhanasekaran V, Poon LLM (2021) Genetic Diversity of SARS-CoV-2 in Travelers Arriving in Hong Kong. *Emerg Dis Infect* **27**:2666-2668.

7) Vieira MC, Donato CM, Arevalo P, Rimmelzwaan GF, Wood T, Lopez L, Huang QS, Dhanasekaran V, Koelle K, Cobey S (2021) Lineage-specific protection and immune imprinting shape the age distributions of influenza B cases. *Nat Commun* **12**:4313.

8) Sanjuán R, Illingworth CJR, Geoghegan JL, Iranzo J, Zwart MP, Ciota AT, Moratorio G, Gago-Zachert S, Duffy S, Dhanasekaran V (2021) Five challenges in the field of viral diversity and evolution. *Front Virol* **1**:684949.

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10) Hoye BJ, Donato CM, Lisovski S, Deng Y-M, Warner S, Hurt AC, Klaassen M, Dhanasekaran V (2021) Reassortment and persistence of influenza A viruses from diverse geographic origins within Australian wild birds: evidence from a small, isolated population of Ruddy turnstones. *J Virol* **95**:e02193-20.

Teaching

1) Vijay Dhanasekaran (2021) Introduction to the Art and Science of Medicine – Problem Based Learning (MBBS Year 1), The University of Hong Kong, Hong Kong SAR (*Tutor*).
2) Vijay Dhanasekaran (2021) Head Neck and Nervous Sytem – Problem Based Learning (MBBS Year 2), The University of Hong Kong, Hong Kong SAR (*Tutor*).
3) Vijay Dhanasekaran (2021) Biological basis of disease – Sequencing revolution: the Genetic Basis of Disease (MPH Year 1), The University of Hong Kong, Hong Kong SAR (*Lecturer*).

Collaborations

1) **Ian Barr** (WHO Collaborating Centre for Reference and Research on Influenza , Peter Doherty Institute, Australia): Genomic epidemiology of influenza and RSV.
2) **Ben Cowling** (School of Public Health, The University of Hong Kong): Genomic epidemiology of respiratory viruses.
3) **Edward Holmes** (Sydney University, Australia): Evolutionary dynamics of RNA viruses.
4) **Trevor Lithgow** (Monash University, Australia): Antimicrobial resistance of *Klebsiella*.
5) **Gavin Smith** (Duke-NUS Medical School, Singapore): Ecology and evolution of influenza and other respiratory viruses.
6) **Kanta Subbarao** (WHO Collaborating Centre for Reference and Research on Influenza , Peter Doherty Institute, Australia):Genomics of avian influenza viruses.
7) **Richard Webby** (St Jude Children's Research Hospital, USA): Evolutionary studies of influenza and SARS-CoV-2 viruses.

Funding

1) Characterising the changing epidemiology of respiratory viruses through public health genomics. (**Principal Investigator**; HKU Seed Fund – Ends: 05/2023).
2) Control of Influenza: Individual and Population Immunity (**Sub PI** ; Research Grants Council of the Hong Kong Special Administrative Region, China – Ends 12/2022).
3) Evolutionary Studies. Centers for Influenza Research and Surveillance (**Principal Investigator**; NIH NIAID – Ends: 03/2021).
4) UHK Identity presence/absence of genetic markers of virulence. Centers for Influenza Research and Response (**Principal Investigator**; NIH UHK – Ends: 03/2028).

Personnel

Name	Position
Vijaykrishna DHANASEKARAN	Associate Professor
Kimberly E. EDWARDS	Project Manager
Xiaoman WEI	Postdoctoral Fellow
Ruopeng Xie	M.Phil student
Shreya GURUNG	M.Phi student
Sonia YOUNAS	Ph.D. student
Zhou YANG	Research Assistant

3.2 Leo POON Lab

Main Objectives and Strategy

Our lab joined HKU-Pasteur in 2020, following the appointment of myself as Co-Director. The main focus is on studying viruses found at the animal and human interface. In particular, our research interests are primarily related to influenza virus and coronaviruses. The overarching goal is to generate experimental evidences to develop evidence-based control measures. Our research projects can be mainly classified into 3 major areas:

1. Emerging Infectious Diseases
2. Basic Virology and vaccinology
3. Molecular diagnosis

Achievements and Ongoing Research

Our research on COVID-19 resulted in 26 publications in this reporting period. The major scientific achievements of these studies can be summarized as follows:

1. Monitor SARS-CoV-2 variant introductions and their transmission dynamic in HK, leading to revising governmental policy against COVID-19 (e.g. setting up quarantine hotels for incoming travelers and banning importation of Syrian hamsters for commercial purposes) (Nat Comm 2022, Lancet 2022).
2. Conduct a serological surveillance study to investigate vaccine-induced antibody response against Omicron variant in HK , leading to the governmental recommendation of using Comirnaty as a booster (Nat Immun 2021).
3. Develop several new materials for developing "smart surfaces" to inactive infectious SARS-CoV-2 (Chem Eng J).
4. Study and understand SARS-CoV-2 pathogenesis using human ex vivo and mouse models (Nature 2022 and Stem Cell Reports 2022).

In addition, our lab has the following impacts:

1. I have been serving as an expert in different working groups under various international healthcare organizations (e.g. WHO and OIE). I have been involved in developing guidelines for these organizations to control COVID-19 (e.g. Guidelines for molecular surveillance of COVID-19 for WHO).
2. My studies on the molecular epidemiology of SARS-CoV-2 in Hong Kong have provided critical scientific information for policy making. Because of our contributions on the COVID-19 control, my team has received an award (Outstanding Project Team on COVID-19 Research Award) from Food and Health Bureau, Hong Kong Government.
3. Because of our cutting-edge work on COVID-19, we have successfully secured a highly competitive and prestigious program grant to research on COVID-19 (Theme-based Research Scheme, 5 years). This program has 10 principal investigators, with me as the project coordinator and two other HKU-Pasteur investigators as co-principal investigators.

Currently, my lab is still heavily invovled in following COVID-19 studies:

1. Molecular epidemiology of SARS-CoV-2
2. Seroepidemiology of SARS-CoV-2
3. Development of antiviral materials
4. SARS-CoV-2 vaccinology and pathogenesis

Publications

1. Yen HL, Sit THC, Brackman CJ, Chuk SSY, Gu H, Tam KWS, Law PYT, Leung GM, Peiris M, Poon LLM (2022) Transmission of SARS-CoV-2 delta variant (AY.127) from pet hamsters to humans, leading to onward human-to-human transmission: a case study. *Lancet* **399**:1070-1078.
2. Gu H, Xie R, Adam DC, Tsui JL, Chu DK, Chang LDJ, Cheuk SSY, Gurung S, Krishnan P, Ng DYM, Liu GYZ, Wan CKC, Cheng SSM, Edwards KM, Leung KSM, Wu JT, Tsang DNC, Leung GM, Cowling BJ, Peiris M, Lam TTY, Dhanasekaran V, Poon LLM (2022) Genomic epidemiology of SARS-CoV-2 under an elimination strategy in Hong Kong. *Nat Commun* **13**:736.
3. Valkenburg SA, Poon LLM (2022) Exploring the landscape of immune responses to influenza infection and vaccination. *Nat Med* **28**:239-240.
4. Gu H, Krishnan P, Ng DYM, Chang LDJ, Liu GYZ, Cheng SSM, Hui MMY, Fan MCY, Wan JHL, Lau LHK, Cowling BJ, Peiris M, Poon LLM (2022) Probable Transmission of SARS-CoV-2 Omicron Variant in Quarantine Hotel, Hong Kong, China, November 2021. *Emerg Infect Dis* **28**:460-462.
5. Hosseini M, Chin AWH, Williams MD, Behzadinasab S, Falkinham JO, 3rd, Poon LLM, Ducker WA. Transparent Anti-SARS-CoV-2 and Antibacterial Silver Oxide Coatings (2022) *ACS Appl Mater Interfaces* **14**:8718-8727.
6. Miot EF, Worthington BM, Ng KH, de Lataillade LG, Pierce MP, Liao Y, Ko R, Shum MH, Cheung WY, Holmes EC, Leung KS, Zhu H, Poon LL, Peiris MJ, Guan Y, Leung GM, Wu JT, Lam TT (2022) Surveillance of Rodent Pests for SARS-CoV-2 and Other Coronaviruses, Hong Kong. *Emerg Infect Dis* 2022 **28**:467-470.
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11. Hui KPY, Ho JCW, Cheung MC, Ng KC, Ching RHH, Lai KL, Kam TT, Gu H, Sit KY, Hsin MKY, Au TWK, Poon LLM, Peiris M, Nicholls JM, Chan MCW (2022) SARS-CoV-2 Omicron variant replication in human bronchus and lung ex vivo. *Nature* 603:715-720.Deng Y, Xu X, Zheng X, Ding J, Li S, Chui HK, Wong TK, , Zhang T (2022) Use of sewage surveillance for COVID-19 to guide public health resp Poon LLM onse: A case study in Hong Kong. *Sci Total Environ* **821**:153250.

12. Ma Z, Li X, Fan RLY, Yang KY, Ng CSH, Lau RWH, Wong RHL, Ng KK, Wang CC, Ye P, Fu Z, Chin AWH, Lai MYA, Huang Y, Tian XY, Poon LLM, Lui KO (2022) A human pluripotent stem cell-based model of SARS-CoV-2 infection reveals an ACE2-independent inflammatory activation of vascular endothelial cells through TLR4. *Stem Cell Reports* **17**: 538-555.

13. Liu LT, Chin AWH, Yu P, Poon LLM, Huang MX (2022) Anti-pathogen stainless steel combating COVID-19. *Chem Eng J* **433**:133783.

14. Poon LLM (2021) A Push for Real Normal: Mass Screening for COVID-19. *Clin Chem* **68**:4-6.

15. Dhanasekaran V, Edwards KM, Xie R, Gu H, Adam DC, Chang LDJ, Cheuk SSY, Gurung S, Krishnan P, Ng DYM, Liu GYZ, Wan CKC, Cheng SSM, Tsang DNC, Cowling BJ, Peiris M, Poon LLM (2021) Air travel-related outbreak of multiple SARS-CoV-2 variants. *J Travel Med* **28**:taab149.

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17. Gu H, Cheng SSM, Krishnan P, Ng DYM, Chang LDJ, Liu GYZ, Cheuk SSY, Hui MMY, Fan MCY, Wan JHL, Lau LHK, Chu DKW, Dhanasekaran V, Peiris M, Poon LLM (2021) Monitoring International Travelers Arriving in Hong Kong for Genomic Surveillance of SARS-CoV-2. *Emerg Infect Dis* 2022 **28**: 247-250.

18. Gu H, Chu DKW, Chang LDJ, Cheuk SSY, Gurung S, Krishnan P, Ng DYM, Liu GYZ, Wan CKC, Xie R, Cheng SSM, Cowling BJ, Tsang DNC, Peiris M, Dhanasekaran V, Poon LLM (2021) Genetic Diversity of SARS-CoV-2 among Travelers Arriving in Hong Kong. *Emerg Infect Dis* **27**: 2666-2668.

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20. Behzadinasab S, Chin AWH, Hosseini M, Poon LLM, Ducker WA.(2021) SARS-CoV-2 virus transfers to skin through contact with contaminated solids. ***Sci Rep*** **11**:22868.

21. Behzadinasab S, Williams MD, Hosseini M, Poon LLM, Chin AWH, Falkinham JO, 3rd, Ducker WA (2021) Transparent and Sprayable Surface Coatings that Kill Drug-Resistant Bacteria Within Minutes and Inactivate SARS-CoV-2 Virus. *ACS Appl Mater Interfaces* **13**:54706-5714.

22. Lau EH, Hui DS, Tsang OT, Chan WH, Kwan MY, Chiu SS, Cheng SM, Ko RL, Li JK, Chaothai S, Tsang CH, Poon LL, Peiris M (2021) Long-term persistence of SARS-CoV-2 neutralizing antibody responses after infection and estimates of the duration of protection. *EClinicalMedicine* **41**:101174.

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24. Xu X, Zhang L, Chu JTS, Wang Y, Chin AWH, Chong TH, Dai Z, Poon LLM, Cheung PP, Huang X (2021) A novel mechanism of enhanced transcription activity and fidelity for influenza A viral RNA-dependent RNA polymerase. *Nucleic Acids Res* **49**:8796-8810.

25. Cohen CA, Li APY, Hachim A, Hui DSC, Kwan MYW, Tsang OTY, Chiu SS, Chan WH, Yau YS, Kavian N, Ma FNL, Lau EHY, Cheng SMS, Poon LLM, Peiris M, Valkenburg SA (2021) SARS-CoV-2 specific T cell responses are lower in children and increase with age and time after infection. *Nat Commun*; **12**:4678.

Seminars and Invited Presentations

1. Leo Poon (2021) "SARS-CoV-2 and its transmission", organized by NIH, USA (*webinar*).
2. Leo Poon (2021) "SARS-CoV-2 and its infection", organized by Centre for Oncology and Immunology, InnoHK (*seminar*).
3. Leo Poon (2021) "SARS-CoV-2 and its infection: from science to public health" in World Science Culture Forum (*webinar*).
4. Leo Poon (2021) "Molecular epidemiological study of COVID-19 cases in Hong Kong" organized by Food and Health Bureau, Hong Kong Government. (*seminar*).
5. Leo Poon (2022) "Molecular epidemiological study of COVID-19 cases in Hong Kong" organized by Pasteur, Paris (*webinar*)

Knowledge Exchange activities

1. Leo Poon (2021) "COVID-19 vaccine", School of Public Health, The University of Hong Kong (webinar).
2. Leo Poon (2021) "Influenza and Coronavirus Infections in Hong Kong", HKU-Space (webinar).

Teaching

1. Leo Poon (2021) CMED6105 – Infectious Diseases in Public Health (Master of Public Health), The University of Hong Kong, Hong Kong SAR (*Course director and lecturer*).
2. Leo Poon (2021) Life Science- (BNur Year 2 and BCMed Year 3), The University of Hong Kong, Hong Kong SAR (*Lecturer*).
3. Leo Poon (2021) Outbreak – Problem Based Learning (MBBS Year 4), The University of Hong Kong, Hong Kong SAR (*Tutor*).
4. Leo Poon (2021) Musculoskeletal System Block– Problem Based Learning (MBBS Year 2), The University of Hong Kong, Hong Kong SAR (*Tutor*).
5. Leo Poon (2021) Introduction to the Art and Science of Medicine (MBBS Year 1), The University of Hong Kong, Hong Kong SAR (*Lecturer*).

Funding

1. Virological, immunological and epidemiological characterization of COVID-19 (**Principal coordinator;** University Grants Committee/Theme-based Research Scheme – Ends 2026)
2. Stability and transmissibility of 2019-nCoV (**Principal Investigator**; Health and Medical Research Fund – Ends 2021).
3. Molecular epidemiological study of COVID-19 cases in Hong Kong (**Principal Investigator**; Health and Medical Research Fund – Ends 2023).
4. Deciphering the role of RNA-RNA interactions between influenza viral segments on reassortment (**Principal Investigator**; Research Grants Council/General Research Fund – Ends 2023).
5. Control of influenza: individual and population immunity (**Co-Principal Investigator**; Research Grants Council/Theme based Research Scheme – Ends 2024).
6. Grid monitoring of SARS-CoV-2 in sewage for an early-warning sign of community outbreak (**Co-Investigator**; Health and Medical Research Fund – Ends 2021).
7. Control of emerging, epidemic and endemic infectious diseases (**Co-Investigator**; HMRF - Commissioned Research on Control of Infectious Diseases – Ends 2025).
8. Replication-defective SARS-CoV-2 mutant vaccines with abnormal codon usages (**Principal Convenor**; University Grants Committee/Collaborative Research Fund – Ends 2024).
9. Airborne virus harvesting, detection and diagnostics inspired by origin of life (**Co-Principal Investigator**; University Grants Committee /Collaborative Research Fund – Ends 2024).

Personnel

Name	Position
Leo Lit Man POON	Professor
Alex Wing Hong CHIN	Research Assistant Professor
Alan Haogao GU	Postdoctoral Fellow
Julie Tung Sem CHU	Postdoctoral Fellow
Nigeer TE	Postdoctoral Fellow
Sylvia Pui Ngan LAU	Senoir Research Assistant
Alison Man Yuk LAI	Postgraduate Student
Sammi Sum Yee CHEUK	Postgraduate Student
Yi CAO	Postgraduate Student
Lydia CHANG	Postgraduate Student
Winnie SUN	Postgraduate Student
Xiaofing XU	Research Assistant
Tansy YAU	Research Assistant
Mani HUI	Research Assistant

3.3 Hein Min TUN LAB

Main Objectives and Strategy

Our group uses conventional microbiology and molecular biology techniques, cutting-edge sequencing technologies, coupled with bioinformatics, statistical and epidemiological approaches to study: 1) the composition, function, and dynamics of human and animal microbiomes in health and disease; and 2) to monitor antimicrobial resistance (AMR) bacteria and resistome in humans, animals, and the environment using a holistic One Health approach. Our goal is to contribute to improving scientific understanding on the impact of microbiome and AMR in public health.

Unwanted Souvenirs for Hong Kong Travelers: A prospective epidemiological approach to study the emergence and dissemination of antimicrobial resistance

Antimicrobial resistance (AMR) constitutes an increasing health hazard worldwide. Recently, the World Health Organization produced a global map of AMR, warning that a 'post- antibiotic' world could soon become reality. Drugs that were once lifesavers are now useless and treatment of many infectious diseases now relies on just one or two drugs. Furthermore, AMR jeopardizes the achievements of modern medicine, since the success of interventions such as organ transplantation, chemotherapy and major surgery depends on effective antimicrobial agents for prevention and treatment of infections. With a dearth of novel antibiotics in the pipeline, the conservation of existing ones is imperative. Adequate global surveillance of AMR is a prerequisite for informing global strategies, monitoring the effectiveness of interventions and detecting new trends and threats. Despite successful regional surveillance programs in Europe, North America, and Australia, global surveillance of AMR generally is neither coordinated nor harmonized comprising the ability to assess and monitor the situation. Moreover, many low and middle-income countries in Africa and Asia, often with a high AMR prevalence, lack a surveillance programs entirely. Last but not least, current national and regional surveillance programs are often based on samples taken from patients with severe (nosocomial) infections neglecting the AMR problem in the community. Therefore, we should aim for alternative ways to monitor AMR around the world in its full breadth in a standardized and flexible fashion.

The extensive overuse and misuse of antibiotics in both humans and animals is fueling AMR, especially in, but not limited to, South (East) Asia. The exponential increase in international travel to these regions may substantially contribute to the emergence and spread of AMR, since it allows resistant bacteria or bacterial mobile genetic elements (MGEs) carrying resistance genes (e.g. plasmids) to be rapidly transported between regions. The intercontinental spread of novel resistance genes via travelers has been demonstrated by the reported transfer of Enterobacteriaceae with New Delhi metallo-beta-lactamase-1 from the Indian subcontinent to Europe. Additionally, case reports of infections with KPC-, VIM-, and OXA-48-producing Enterobacteriaceae in developed countries have been associated with visiting and being hospitalized in endemic areas such as the USA, Greece and Israel for KPCs, Greece for VIMs and the Middle East for OXA-48. The extent to which foreign travel poses a risk for the acquisition of AMR remains largely

unknown, as the presence of resistant bacteria in the normal human microbiota following travel remain undetected unless they cause manifest infections.

The gastrointestinal tract is an open system, which every day encounters myriad of bacterial acquisitions originating from the environment (e.g. from food, water, soil, and other humans or animals). These incoming bacteria acquired in countries with a high prevalence of antimicrobial resistance often harbor AMR genes. In case of opportunistic pathogens of environmental or food-borne origin, such AMR bacteria can pose a direct threat to the host. Alternatively, these incoming microbes might transfer their resistance elements through horizontal gene transfer to the indigenous microbial communities. Next to this lateral gene transfer, AMR microorganisms can also spread from the traveler to other family members and beyond through the fecal-oral route. Travelers can therefore be viewed as interactive biological units who pick up, process, carry and drop off microbial genetic material. Consequently local emergence of AMR can rapidly become a worldwide health problem.

To date, only two studies have been published analyzing geographical differences in the human resistome by means of sequence-based metagenomics. Both cross-sectional studies showed strong correlations between antibiotic consumption and the abundance and type of resistance genes on a population level, clearly proving the strength of resistome analyses. In particular, it remains unknown to what extent traveling to geographic areas with high rates of AMR affects one's resistome. Moreover, information on AMR in countries with a lack of surveillance, like many countries on the African and Asian continent, is currently missing. The proposed project provides a unique opportunity to prospectively study the shifts in the resistome of Hong Kong travelers following travel to different countries. At the beginning of 2018 December, we launched the "Hong Kong Traveler Cohort" by sending the recruitment information to HKU students and staffs via email or posting flyers around the campus. After sending out the first recruitment email, a total of 113 travelers enrolled in the study. rom our pilot cohort, we have generated a manuscript based on AMR and Microbiota data of 90 travelers. In our study, we identified travel associated risk factors for the acquisition of ESBL bacteria and other AMR genes, which are also influenced by the diversity of gut microbiota. This led to develop a model to predict both acquisition and decolonization of AMR bacteria and genes using baseline microbiota and traveler associated risk factors.

Early-life gut microbiota predicts the neurodevelopment of children

Dysbiosis of gut microbiota has been retrospectively linked to autism spectrum disorders but the temporal association between gut microbiota and early neurodevelopment in healthy infants is largely unknown. We undertook this study to determine associations between gut microbiota at two critical periods during infancy and neurodevelopment in a general population birth cohort. We analyzed data from 405 infants (199 females) from the CHILD (Canadian Healthy Infant Longitudinal Development) Cohort Study. Neurodevelopmental outcomes were objectively assessed using the Bayley Scale of Infant Development (BSID-III) at 1 and 2 years of age. Microbiota profiling with 16S rRNA gene sequencing was conducted on fecal samples obtained at a mean age of 4 and 12 months.

Using clustering methods, we identified three groups of infants based on relative abundance of gut microbiota at 12 months: Proteobacteria-dominant cluster (22.4% higher abundance at 12 months), Firmicutes-dominant cluster (46.0% higher abundance

at 12 months) and Bacteroidetes-dominant cluster (31.6% higher abundance at 12 months). Relative to the Proteobacteria-dominant cluster, the Bacteroidetes-dominant cluster was associated with higher scores for cognitive (4.8 points; FDRp = .02), language (4.2 points; FDRp≤0.001), and motor (3.1 points; FDRp = .03) development at age 2 in models adjusted for covariates. When stratified by sex, only male infants with a Bacteroidetes-dominant microbiota had more favorable cognitive (5.9 points, FDRp = .06) and language (7.9 points; FDRp≤0.001) development.
Genus Bacteroides abundance in gut microbiota was positively correlated with cognitive and language scores at age 2. Fully adjusted linear mixed model analysis revealed a positive association between Bacteroidetes-dominant cluster and change in cognitive and language performance from 1 to 2 years, predominantly among males. No associations were evident between 4-month microbiota clusters and BSID-II scores. Noteworthy is that enhanced sphingolipid synthesis and metabolism, and antagonism or competition between Bacteroides and Streptococcus were characteristic of a Bacteroidetes-dominant gut microbiota. This study found strong evidence of positive associations between Bacteroidetes gut microbiota in late infancy and subsequent neurodevelopment, most prominently among males but not females.

Gut microbiome and resistome changes during the first wave of the COVID-19 pandemic in comparison with pre-pandemic travel-related changes

Our gut microbes are sensitive to environmental changes and exposures. Travel-associated alterations to gut microbial composition and antibiotic resistance gene (ARG) profiles have been reported. Despite cross-border travel restrictions are in place during the COVID-19 global pandemic, various pandemic control measures including physical distancing, extensive hygiene and greater disinfectant and antibiotic usage all have the potential to disrupt normal composition of gut microbiota. However, we do not yet fully understand the extent to which gut microbiota and resistance genes against antibiotics and biocides will change in response to the current pandemic control measures.

We performed shotgun metagenomic sequencing on fecal samples collected before (BT) and after (AT) pre-pandemic travels and during the first wave of the COVID-19 pandemic (FW) of 32 Hong Kong adults in a prospective cohort. change in microbial beta diversity between AT and FW was significantly greater than that between BT and AT, and these changes were not correlated with the length of the intervals between sampling time points (P > 0.05). We also observed significantly lower Actinobacteria richness and higher Bacteroidetes richness in the gut microbiome of the participants during the pandemic; neither the first travel episode nor additional travel changed microbial alpha diversity. Normal inhabitants of the oral cavity as well, members of the phylum Actinobacteria become more diverse in human gut with greater exposure to the natural environment. It is conceivable then that lowered species richness within the Actinobacteria during the COVID-19 pandemic reflects a reduction in outdoor recreational activities subsequent to pandemic control measures. At the species level, higher abundance of Lactococcus lactis (of the Firmicutes phylum) and Corynebacterium durum (of the Actinobacteria) was identified following episodes of international travel; the opposite trend was noted during the first wave of the pandemic. By contrast, higher Bacteroides thetaiotaomicron and Parabacteroides distasonis, both members of the Bacteroidetes

phylum, and lower Actinomyces oris (of the Actinobacteria) were observed during the first wave of the pandemic. B. thetaiotaomicron and P. distasonis, mostly resistant to beta-lactam antibiotics, became more plentiful during the pandemic. Beta-lactam resistance genes harbored by these Bacteroidetes species could be transferred to other microbes in the gut. Among the identified ARGs, the abundance of rifamycin resistance genes declined, whereas that of the beta-lactam resistance genes rose during the COVID-19 pandemic. Besides, we found statistical enrichment of polystyrenes and phthalate resistance genes in the gut microbiome during the pandemic, findings attributed to two membrane transporter genes, ttgA and ttgB. Polystyrenes are components of face masks, whereas phthalates are used as plasticizers in food packaging and building materials. Persistent use of masks in Hong Kong, enhanced exposure to food packaging due to pandemic-related in-dining restrictions at restaurants, and prolonged indoor stays due to physical distancing measures might have led to these changes.

Ethnicity Associations With Food Sensitization Are Mediated by Gut Microbiota Development in the First Year of Life

Increasing evidence supports the role of early-life gut microbiota in developing atopic diseases, but ecological changes to gut microbiota during infancy in relation to food sensitization remain unclear. We aimed to characterize and associate these changes with the development of food sensitization in children. In this observational study, using 16S rRNA amplicon sequencing, we characterized the composition of 2844 fecal microbiota in 1422 Canadian full-term infants. Atopic sensitization outcomes were measured by skin prick tests at age 1 year and 3 years. The association between gut microbiota trajectories, based on longitudinal shifts in community clusters, and atopic sensitization outcomes at age 1 and 3 years were determined. Ethnicity and early-life exposures influencing microbiota trajectories were initially examined, and post-hoc analyses were conducted.

Four identified developmental trajectories of gut microbiota were shaped by birth mode and varied by ethnicity. The trajectory with persistently low *Bacteroides* abundance and high Enterobacteriaceae/Bacteroidaceae ratio throughout infancy increased the risk of sensitization to food allergens, particularly to peanuts at age 3 years by 3-fold (adjusted odds ratio [OR] 2.82, 95% confidence interval [CI] 1.13–7.01). A much higher likelihood for peanut sensitization was found if infants with this trajectory were born to Asian mothers (adjusted OR 7.87, 95% CI 2.75–22.55). It was characterized by a deficiency in sphingolipid metabolism and persistent *Clostridioides difficile*colonization. Importantly, this trajectory of depleted *Bacteroides*abundance mediated the association between Asian ethnicity and food sensitization. This study documented an association between persistently low gut *Bacteroides* abundance throughout infancy and sensitization to peanuts in childhood. It is the first to show a mediation role for infant gut microbiota in ethnicity-associated development of food sensitization.

Wastewater-based epidemiology of COVID-19 in Hong Kong

Early detection and surveillance of severe acute respiratory syndrome-coronavirus 2 (SARS-CoV-2) virus are key pre-requisites for the effective control of coronavirus disease (COVID-19). So far, sewage testing has been increasingly employed as an alternative surveillance tool for this disease. However, sampling site characteristics impact the testing results and should be addressed in the early use stage of this emerging tool. In

this study, we implemented the sewage testing for SARS-CoV-2 virus across sampling sites with different sewage system characteristics. We first validated a testing method using "positive" samples from a hospital treating COVID-19 patients. This method was used to test 107 sewage samples collected during the third wave of the COVID-19 outbreak in Hong Kong (from June 8 to September 29, 2020), covering sampling sites associated with a COVID-19 hospital, public housing estates, and conventional sewage treatment facilities. The highest viral titer of 1975 copy/mL in sewage was observed in a sample collected from the isolation ward of the COVID-19 hospital. Sewage sampling at individual buildings detected the virus 2 days before the first cases were identified. Sequencing of the detected viral fragment confirmed an identical nucleotide sequence to that of the SARS-CoV-2 isolated from human samples. The virus was also detected in sewage treatment facilities, which serve populations of approximately 40,000 to more than one million people. This wastewater-based epidemiology of COVID-19 has been used by Hong Kong government as an effective tool to trace asymptomatic cases and active infections during the fourth wave and recent fifth wave of COVID-19 in Hong Kong.

Achievements and Ongoing Research

In, the group published 19 peer-reviewed articles,most of them as orginal research articles. Of note, the Gastroenterology paper (doi:10.1053/j.gastro.2021.03.2016) led by Hein Min Tun and his PhD student, Peng Ye, was selected by editors from Journal of Allergy and Clinical Immunology to feature in their journal. In addition, we won two significant team awards: the "Gold Medal" at the 2021 Inventions Geneva Evaluation Days and the "Outstanding Project Team on COVID-19 Research Award" by the Food and Health Bureau of Hong Kong, to recognize our contribution in wastewater-based epidemiology of COVID-19 in Hong Kong. In addition, the Tun lab was visited by the Chief Executive, Mrs. Carrie Lam, accompanied by legistlative council members as well as officials from Food and Health Bureau and Environmental Protection Department. Hein Min Tun is also involved as a main inventor in 3 US provisional patents. One of our PhD students, Peng Ye, also received the HKU-Pasteur Research Pole Followship and HKUMed's postgraduate student exchange scholarship to perform research at the Insititut Pasteur, Paris for 3 months. The Tun lab had a successful bid in the URC Postdoctoral Scheme and received a 50% salary support for a3-year postdocatral fellow. The lab also secured two new extramural fundings from the FHB's HMRF Commissioned Research on the Novel Coronavirus Disease (COVID-19) and HKU's Enhanced New Staff Start-up Research Grant.

Antimicrobial Resistance Comprehensive Etiology Study [Funding: Wellcome Trust and RGC/Research Impact Fund]

ACES is an international, multisite scientific project whose primary goal is to provide new scientific information on the how much health, agriculture, food and the environment each contribute to the development of antimicrobial resistance affecting humans. This information is needed to stimulate further the development and implementation of effective AMR policies that are synergistic across these sectors. ACES is explicitly intended to position such information – through international organizations, and in written publications and comments – to influence policy. The project was initiated by researchers at the University of Hong Kong (HKU), School of Public Health (SPH). This group has been in discussion with the AMR program at the Wellcome Trust, and with numerous scientific

researchers across the world, to develop the concepts and methods for ACES. Hein Min Tun plays a significant leadership role for ACES projects in Mainland China and Thailand. Moreover, the Tun lab standardized all the testingmethodologies and is responsible to test samples collected from ACES Hong Kong project.

ACES proposes characterizing and clarifying the etiology and epidemiology of AMR by applying a coordinated, systematic and prospective approach to collection of both epidemiological and laboratory data from human, food, food-producing animals and environment settings within different study sites. ACES has 2 main aims:

Aim 1: Determine what proportion of human AMR bacterial infections are attributable to antibiotic related practices, conditions or outcomes in human, plants, food-producing animals, food and the environment;

Aim 2: Determine the main pathways and mechanisms by which such practices, conditions or outcomes result in human AMR bacterial infections.

The basic methodological approach will be to use standardized methods to collect information across multiple sites on AMR patterns, AMR pathogens, antimicrobial resistant genes and antibiotic use. Current, study sites are Hong Kong, Khon Kaen (Thailand) and Guangxi Province (PR China). In the future, funds permitting, additional sites may be added in either North America and/or Europe. In each site, ACES will collect data from two, geographically separate communities that differ in terms of exposures, particularly to food-producing animals. Cross-sectional data collection will be done repeatedly over a 2-years period. Analyses will focus initially on looking for similarities and differences in correlations and patterns. The central study hub will be located at the HKU SPH. Results will be disseminated in ways intended to influence AMR prevention and management policies and strategies, including through international organizations such as WHO, FAO, OIE, UNEP as well as local community stakeholders and civil society groups.

The association of baseline gut microbiota composition with SARS-CoV-2 vaccine immunogenicity and adverse events [Funding: HKU's Enhanced New Staff Start-up Research Grant]

Vaccination elicits protective immune responses against SARS-CoV-2 and provides hope for containing the COVID-19 pandemic. As of 17 January 2022, more than 9.3 billion doses of vaccine have been administrated worldwide1 with substantial efficacy. Recent observational studies reported a steady decline of antibody levels among vaccinated individuals which implied a growing risk of breakthrough infection over time but factors influencing immunogenicity and durability of vaccine remains poorly understood. Evidence from clinical or animal studies suggested that the composition and functions of the gut microbiota are crucial in modulating immune responses of vaccination. Mucosal or systemic microbiota exposure shapes T and B cell repertoires that have an important implication for regulating responses to vaccination. Whether host microbiota composition influences responses of COVID-19 vaccines in humans has not been determined. We conducted a prospective observational study of adults who have received either the inactivated vaccine (CoronaVac; Sinovac) or the mRNA vaccine (BNT162b2; BioNTech; Comirnaty) to examine gut microbiota determinants of vaccine immune responses and vaccine-related adverse events. We performed shotgun metagenomic sequencing in stool samples of 138 COVID-19 vaccinees (37 CoronaVac and

101 BNT162b2 vaccinees) collected at baseline and 1 month after second dose of vaccination. Immune markers were measured by SARS-CoV-2 surrogate virus neutralisation test and spike receptor-binding domain IgG ELISA.

Epilepsy-associated gut microbiome and metabolites in children with cerebral palsy [Funding: HKU's seed fun]

Cerebral palsy (CP) refers to a group of motor function disorders due to non-progressive insult, lesion or abnormality of the developing brain. Dysbiosis of gut microbiota has been implicated in acute phases of traumatic brain injury, and amongst children with cerebral palsy. This cross-sectional study used shotgun metagenomic sequencing coupled with untargeted metabolomics analysis of gut microbiome in children with CP with or without epilepsy.

Functional role and therapeutic potential of Lactococcus lactis in non-alcoholic fatty liver disease

Non-alcoholic fatty liver disease (NAFLD) is the world's most common chronic liver disease, with a population-based prevalence of 24-46% in Western countries and 7.9-54% in Asia, affecting both lean and obese individuals, but lacking in viable therapeutic options. More recently, the gut microbiota has emerged as an important player in the pathogenesis of NAFLD. In general, NAFLD patients showed altered microbial composition, manifested as a decreased proportion of Oscillibacter, an increased proportion of Roseburia, Robinsoniella, Dorea and Lactobacillus via 16s rRNA sequencing. Microbial dysbiosis leads to the distinct shifts in bacterial products, including ethanol, lipopolysaccharides, short-chain fatty acids, conjugated bile acids, and trimethylamine N-oxide. These metabolites translocate into the portal vein through an impaired intestinal barrier ("leaky" gut), bind to specific toll-like receptors in the liver, and activate the proinflammatory pathways. While there is numerous evidence of an altered gut microbiota being associated with NAFLD, any potential causality has yet to be established. In this study, we performed FMT in C57BL/6J mice on a high-fat diet using human donor feces. We aimed to investigate for potential causality between the gut microbiota and NAFLD, and to identify the altered bacterial species that may play a role in the onset and initiation of NAFLD.

Publications

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2. Zheng X, Deng Y, Xu X, Li S, Zhang Y, Ding J, On HY, Lai JCC, Yau CI, Chin AWH, Poon LLM, Tun HM, Zhang T (2022). Comparison of virus concentration methods for SARS-CoV-2 sewage surveillance. *Sci Total Environ* **824**:153687.
3. Zhang S, Tun HM, Zhang D, Chou H, Huang F, Kwok H, Wong DK, Mak L, Yuen M, Seto WK. (2022). Alleviation of Hepatic Steatosis: Dithizone-related gut microbiome restoration during Paneth Cell Dysfunction. Front Microbiol **13**:813783.
4. Peng Y, Zhang D, Chen T, Xia Y, Wu P, Seto WK, Kozyrskyj AL, Cowling BJ, Zhao J, Tun HM (2021).Gut microbiome and resistome changes during the first wave of the COVID-19 pandemic in comparison with pre-pandemic travel-related changes. *J Travel Med* **28**:taab067.
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7. Tamana SK, Tun HM, Konya T, Chari R, Field CJ, Guttman DS, Becker AB, Moraes TJ, Turvey SE, Subbarao P, Sears MR, Pei J, Scott JA, Mandhane PJ, Kozyrskyj AL (2021). Bacteroides-dominant gut microbiome of late infancy is associated with enhanced neurodevelopment. *Gut Microbes* **13**:1-17.
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9. Peng Y, Zhao J, Tun HM (2021) The new foe and old friends: are we ready for microbiota-based therapeutics in treating COVID-19 patients? *Gastroenterology* **160**:192-2193.
10. Vu K, Lou W, Tun HM, Konya TB, Morales-Lizcano N, Chari RS, Field CJ, Guttman DS, Mandal R, Wishart DS, Azad MB, Becker AB, Mandhane PJ, Moraes TJ, Lefebvre DL, Sears MR, Turvey SE, Subbarao P, Scott JA, Kozyrskyj AL (2020) From birth to overweight and atopic disease: multiple and common pathways of the infant gut microbiome. *Gastroenterology* **160**:128-144.e10.

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15. Chan OSK, Naing T, Tun HM (2021) Upholding veterinary services as a pillar of one health in Myanmar. *One Health* **13**:100329.

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19. Orsso CE, Peng Y, Deehan EC, Tan Q, Field CJ, Madsen KL, Walter J, Prado CM, Tun HM, Haqq AM (2021) Composition and Functions of the Gut Microbiome in Pediatric Obesity: Relationships with Markers of Insulin Resistance. *Microorganisms* **9**:1490.

20. Tan Q, Orsso CE, Deehan EC, Kung JY, Tun HM, Wine E, Madsen KL, Zwaigenbaum L, Haqq AM (2021) Probiotics, prebiotics, synbiotics, and fecal microbiota transplantation in the treatment of behavioral symptoms of autism spectrum disorder: A systematic review. *Autism Res* **14**:1820-1836.

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22. Deehan EC, Colin-Ramirez E, Triador L, Madsen KL, Prado CM, Field CJ, Ball G, Tan Q, Orsso C, Dinu I, Pakseresht M, Rubin D, Sharma AM, Tun H, Walter J, Newgard CB, Freemark m, Wine E, Haqq AM (2021) Efficacy of metformin and fermentable fiber combination therapy in adolescents with severe obesity and insulin resistance: study protocol for a double-blind randomized controlled trial. *Trials* 22:148.

Seminars and Invited Presentations

1. Hein Min Tun (2021) International Conference on Current Problems of Biological Safety in the Modern Conditions. Kazakhstan (*webinar*).
2. Hein Min Tun (2021) 26th Medical Research Conference, Department of Medicine, University of Hong Kong (*seminar*).

Teaching

1. Hein Min Tun (2021) CMED6227 – Biological Basis of Disease (Master of Public Health), The University of Hong Kong, Hong Kong SAR (*Course Director and Lecturer*).
2. Hein Min Tun (2021) EBDM- Evidence-Based Decision Making for Patient Care and Public Health MBBS I and II, The University of Hong Kong, Hong Kong SAR (*Tutor*)
3. Hein Min Tun (2021) Fleming Fund Fellowship Program, Laboratory Fellows from Sri Lanka (*Mentor*)
4. Hein Min Tun (2021) BBMS2011/BPHM1121- Research Methods in Medicine and Health Sciences, The University of Hong Kong, Hong Kong SAR (*Course Coordinator*)

Collaborations

1. **Anita Kozyrskyj** (Department of Paediatrics, University of Alberta, Edmonton, Canada): Gut microbiota maturation during infancy.
2. **Andrea Haqq** (Department of Pediatrics, University of Alberta, Edmonton, Canada): Gut microbiota profile in children with Prader-Willi Syndrome.
3. **John Penders** (Department of Medical Microbiology, Maastricht University, Maastricht, The Netherlands): Antimicrobial dissemination in international travellers.
4. **Tanja Sobko** (School of Biological Sciences, The University of Hong Kong, Hong Kong SAR): Impact of nature connectedness on gut microbiome and mental health of children.
5. **Jincun Zhao** (Guangzhou Medical University, Mailand China): Multi-Platform Omics 1 Analysis Reveals Molecular Signature for COVID-19 Pathogenesis, Prognosis and Drug Target Discovery **&** Characterization of respiratory microbial dysbiosis in hospitalized COVID-19 patients.
6. **Wai-Kay Seto** (Department of Medicine, The University of Hong Kong, Hong Kong SAR): Functional role and therapeutic potential of Lactococcus lactis in non-alcoholic fatty liver disease **&** Investigating aerosol generation and transmissibility implications in upper and lower gastrointestinal endoscopy: a multicentre study.
7. **Tong Zhang** (Department of Civil Engineering, The University of Hong Kong, Hong Kong SAR): Grid monitoring of SARS-CoV-2 in sewage for an early-warning sign of community outbreak.
8. **Xudong Zhou** (School of Public Health, Zhejiang University, Mailand China): Antimicrobial resistance comprehensive etiology Study (ACES)-Mailand China.
9. **Rungtip Chuanchuen** (Department of Veterinary Public Health, Chulalongkorn University, Thailand): Antimicrobial resistance comprehensive etiology Study (ACES)-Thailand.

10. **Alongkorn Amonsin** (Department of Veterinary Public Health, Chulalongkorn University, Thailand): Coronavirus seroprevalence among bat exposure villagers in Thailand.
11. **Sophelia Chan** (Department of Paediatrics and Adolescent Medicine, Li Ka Shing Faculty of Medicine, University of Hong Kong): Microbiome in paediatric epilepsy.
12. **Siew C Ng** (Department of Medicine and Therapeutics, Faculty of Medicine, The Chinese University of Hong Kong, Hong Kong): Mommy Cohort and MiVac Cohort.

Funding

1. Cross-sectional study of antimicrobial use pattern, antimicrobial resistant pathogen and bacterial genomic association in urinary tract infection patients.(**Co-Investigator**; HKU – Seed Funding for Basic Research – Ends: 06/2021).
2. Fleming Fellowship Scheme for Sri Lanka (**Co- Investigator**; Fleming Fund – Ends: 06/2022).
3. Deciphering the role of gut microbiota in the development of depression, anxiety and stress (**Principal Investigator**; HKU – Seed Funding for Basic Research – Ends: 07/2021)
4. Antimicrobial resistance comprehensive etiology Study (ACES) (**Co-Principal Investigator**; Wellcome Turst – Ends: 08/2021)
5. Grid monitoring of SARS-CoV-2 in sewage for an early-warning sign of community outbreak (**Co-Investigator**; HMRF Commissioned Research on the Novel Coronavirus Disease (COVID-19) – Ends: 09/2021)
6. Computational imaging of the spatiotemporal distribution of forces in gut tissue: a study of the cross talk between cell mechanics, microbiome and infectious processes (**Principal Investigator**; Institut Pasteur – PTR 232 Grant – Ends: 09/2021).
7. Provision of Laboratory Testing Services for Detecting and Quantifying the Presence of Antimicrobials in Local Livestock Faecal Waste (**Principal Investigator**; Agriculture, Fisheries and Conservation Department – Ends: 10/2022).
8. Acquisition and persistence of antimicrobial resistance following travels to resource-limited countries: a multi-layered metagenomic epidemiological study (**Principal Investigator**; Health and Medical Research Fund – Ends: 11/2022).
9. Investigating aerosol generation and transmissibility implications in upper and lower gastrointestinal endoscopy: a multicentre study (**Co-Investigator**; HMRF Commissioned Research on the Novel Coronavirus Disease (COVID-19) – Ends: 11/2021)
10. Antimicrobial resistance comprehensive etiology Study (ACES) (**Co-Principal Investigator**; Research Grants Council/Research Impact Fund – Ends: 03/2023).
11. Role of the indoor environment in the transmission of antimicrobial resistance to humans: a household-based longitudinal study (**Principal Investigator**; HKU – Internal Research Grant – Ends: 05/2023).

12. Computational imaging of the spatiotemporal distribution of forces in gut tissue: a study of the cross talk between cell mechanics, microbiome and infectious processes (**Principal Investigator**; UGC – Research Matching Scheme – Ends: 06/2023).
13. Household-based longitudinal monitoring on transmission dynamics of antimicrobial resistance bacteria/genes between humans and built environments (**Co-Investigator**; HKU – Research Start-up Award – Ends: 12/2023).
14. Bacterial carriage in the upper respiratory tract among community healthy subjects in Hong Kong and Guangzhou (**Co-Investigator**; HKU – Bau Tsu Zung Kwan Yeu Hing Research Start-up Research Grant – Ends: 12/2023).
15. Computational imaging of the spatiotemporal distribution of forces in gut tissue: a study of the cross talk between cell mechanics, microbiome and infectious processes (**Principal Investigator**; UGC Research Matching Grant – Ends: 12/2023)
16. Antimicrobial Resistance (AMR) Policy Framework in Big Bay Area (Guangdong - Hong Kong - Macau) (**Principal Investigator**; UGC Research Matching Grant – Ends: 12/2023)
17. Fiber supplementation and metformin combination therapy in adolescents with severe obesity and insulin resistance: interactions with the gut microbiome ((**Co-Principal Investigator**; Weston Family Microbiome Initiative Grant – Ends: 12/2023).
18. Donation in support of AMR Research work. The webinar theme is "Antimicrobial Resistance and Antimicrobial Use in Companion Animals" (**Principal Investigator**; UGC – Research Matching Scheme – Ends: 05/2024).
19. Strengthening sewage surveillance for SARS-CoV-2. (**Co-Investigator**; HMRF Commissioned Research on the Novel Coronavirus Disease (COVID-19) – Ends: 08/2024).
20. M2I : Microbiome, Immunity and Infections (**Principal Investigator**; HKU – URC Postdoctoral Fellow (PDF) Scheme – Ends: 10/2024).
21. Control of Influenza: Individual and Population Immunity (**Co-Investigator**; Theme-based Research Scheme – Ends: 12/2024).
22. Assess antibiotic resistome flows from pollution hotspots to environments and explore the control startegies (**Co-Principal Investigator;** Theme-based Research Scheme – Ends: 12/2025).

Personnel

Name	Position
Hein Min TUN	Assistant Professor
Xi ZHANG	Postdoctoral Fellow (since Oct 2021)
Ka Pui NG	IT Technician (until Aug 2021)
Darren Chak Lun CHAN	PhD Student
Suisha LIANG	PhD Student
Ye PENG	PhD Student
Hogan WAI	PhD Student
Xiawan ZHENG	PhD Student – Department of Civil Engineering, HKU
Shuxian LI	PhD Student – Department of Civil Engineering, HKU
Yulin ZHANG	PhD Student – Department of Civil Engineering, HKU
Flice PAK	Research Assistant i (until Dec 2021)
Dengwei ZHANG	Research Assistant I (until May 2021)
Shilin ZHAO	Research Assistant I
Jie ZHU	Research Assistant I (from Aug 2021)
Alex WONG	Research Assistant II (until June 2021)
Gary CHAN	Research Assistant II
Ingrid CHAN	Research Assistant II (from Nov 2021)
Chloe LIU	Research Assistant II (P/T) (from Oct 2021)
Xin LIU	Research Assistant II (from Aug 2021)
Rista SHRESTHA	Research Assistant II
Daniel SIN	Research Assistant II (from Oct 2021)
Hilda ON	Research Assistant II (P/T)
Thomas CHU	Research Assistant II (P/T) (from Mar 2022)

Suet Ying NG	Research Assistant II (P/T) (until Aug 2021)
Anxin PAN	Research Assistant – Department of Civil Engineering, HKU
Xianghui SHI	Research Assistant – Department of Civil Engineering, HKU
Mengying WANG	Research Assistant – Department of Civil Engineering, HKU
Wenxi ZENG	Student Intern
Wing Ki LIU	Student Intern
Wai Hang CHAN	Student Intern
Wai Yee TSANG	Student Intern

3.4 Sophie VALKENBURG Lab

Main Objectives and Strategy

The main objectives of the lab are to define immune correlates of protection for influenza viruses from infection and vaccination. These approaches were adapted to address the COVID-19 pandemic. Our research is centred on the role of protective heterologous T and B cell immunity in mouse and human systems, by investigating novel vaccines and immune correlates of protection for influenza and SARS-CoV-2. Our primary focus is to study adaptive immunity to these viruses, and how this could be harnessed and optimized by vaccination to improve protection from infection. Hemagglutinin (HA) and Spike (S)-specific antibodies can block influenza and SARS-CoV-2 infection respectively, whilst T cells recognize influenza-infected cells and antibodies to non-strcutral proteins expressed during infection can impact viral replication and Fc mediated cellular functions. A vaccine which ultimately combines antibody and T cell-based immunity will provide a full-proof immunological barrier to infection and variants, which our studies will ultimately help develop. Our major research projects, which aim to elucidate how cross-reactive T and B cell responses provide broad immunity, are listed below.

Broadly reactive immune correlates for improved influenza vaccines

A vaccine that is broadly protective against different strains and subtypes of influenza is needed *in lieu* of the current seasonal vaccine that requires yearly update and is not protective against pandemic or outbreak strains.

(a) A T-cell based universal vaccine: In collaboration with Leo LM Poon at HKU, and Liyange Perera at NIH, our group is determining the mechanism of protection of a Vaccinia Wyeth vaccine vector encoding 5 influenza proteins, HA, NA, NP and Matrix 1 and 2 proteins, with a molecular IL-15 adjuvant to enhance vaccine memory responses, termed Wyeth/5Flu/IL-15. The vaccine has been highly effective in mice providing protection against avian, pandemic and seasonal strains of influenza. The vaccine elicits effective influenza-specific T cell memory responses that establish early local T cell responses upon influenza challenge, significantly reducing viral lung titers and thus survival. Importantly, depletion of T cell subsets showed that memory CD4 T cell responses were necessary for vaccine mediated protection, an under-appreciated role of helper subset. The vaccine is being further investigated to determine the role T cell mediated immune pressure may play in viral escape by next generation sequencing to identify mutations at T cell epitopes. Circumventing virus escape is important for the design of next generation influenza vaccines.

(b) Enhanced inactivated influenza vaccines and repeat vaccination for older adults: Since 2016 in collaboration with Prof. Ben Cowling, I have been conducting a longitudinal randomized clinical trial via a co-operative agreement with the US CDC, to assess longitudinal vaccine responses in older adults over 5 years in Hong Kong. Specifically, whether already available enhanced vaccines (recombinant, adjuvant, high dose) were more immunogenic than standard vaccines, and whether an alternating vaccine regime of enhanced vaccines would have further synergy, resulting in an 11-arm clinical trial of nearly 2,000 older adults. Experiments are ongoing.

Immune correlates of SARS-CoV-2 infection

The COVID-19 pandemic has caused devastating morbidty, mortality and impacted to world economy. We have generated a large biobank of clinical samples and probed T cell and antibody responses in patients and vaccine models.

(a) Immune responses to SARS-CoV-2 infection: Recovery from SARS-CoV-2 infection requires the coordinated innate, cellular and antibody response. However there is a breadth of symptoms and majority of cases being asymptomatic, whilst children experience minimal impact may have an immunological basis. In collaboration with Hong Kong clinicians we have sampled longitudinally from infected patients, asymptomatic cases and young children to generate a unique perspective on the immune responses to SARS-CoV-2 infection.

Achievements and Ongoing Research

Over the past year the research output from my team has consolidated, and adapted to the COVID-19 pandemic. Further movement within my research team saw the recruitment of a new postdoctoral fellow (Prathanporn Kaewpreedee) and the deperature of several staff (Luke Wong, Asmaa Hachim and Fionn Ma) and PhD students (Maireid Bull and Athena Li). A 5-year Theme based Research Scheme (TRS) project grant by the Research Grants Council (RGC), and a 3 year Collabortive Research Fund (CRF) grant were awarded to Professor Leo Poon to investigate COVID-19 for both virology and immunology and vaccine, respectively. I am a co-investigator in both and the lab will contribute significantly to over the next coming years. In recognition of my sucessful research program I was promoted to tenure track Assistant Professor in April 2021. However, after 10 years building my research program in Hong Kong, due to various personal circumstances I applied for a 5 year Australian fellowship from the NHMRC, and was successfully awarded a Emerging Leader level 2 fellowship, which facilitated my return to my hometown and a new position as Associate Professor in the Department Microbiology and Immunology, Peter Doherty Institute, at the Univeristy of Melbourne. I remain an honorary Assistant Professor at HKU-Pasteur and maintain a small team for ongoing projects.

Broadly reactive immune correlates for improved influenza vaccines

[Funding: RGC/GRF; CDC; CEIRS]

(a) A T-cell based universal influenza vaccine

The vaccine is being further investigated by Maireid Bull (PhD student) T cell activated vaccines (**Figure 1A**) for immune mediated pressure by full influenza genome Next Generation Sequencing (**Figure 1B-C**) to determine the impact of versus standard vaccination (**Figure 1D**). Virus rescue of newly identified escape variants by reverse genetics was unsuccessful showing these adaptive mutations were not viable. The manuscript was published in Science Advances (April 2022).

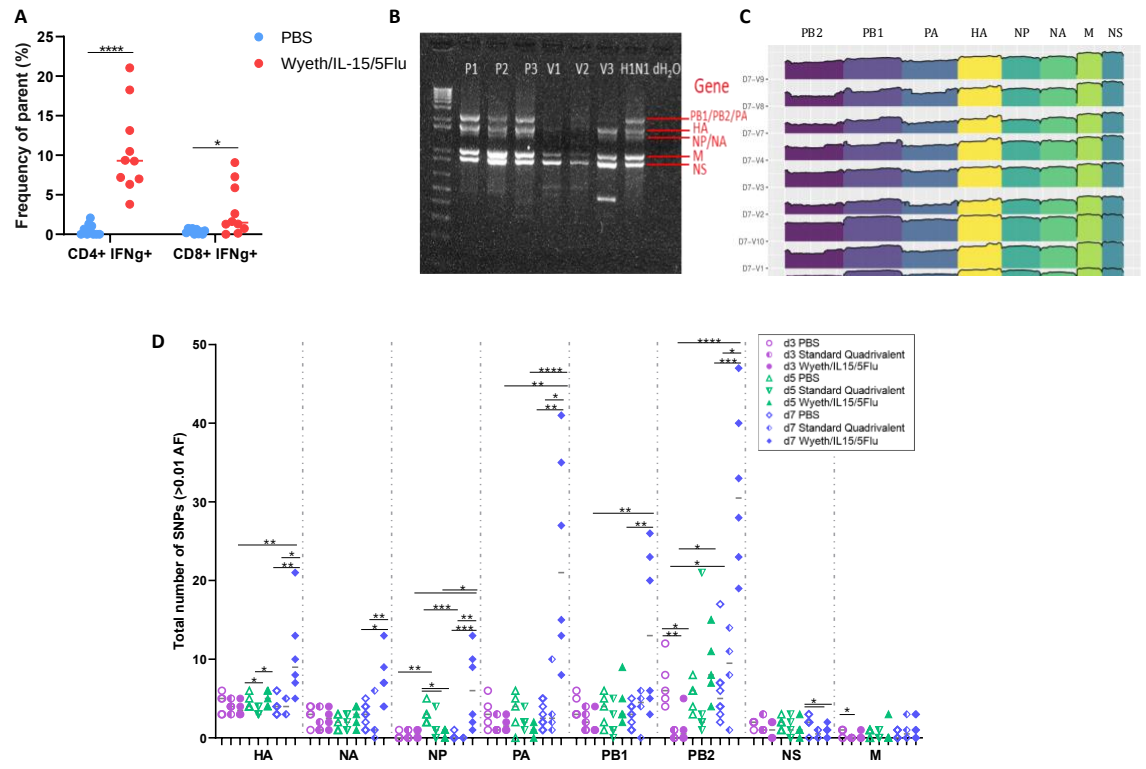

Figure 1: T cell activating influenza vaccines drive genetic diversification of influenza virus during infection. (A) T cell activating vaccine Wyeth/IL-15/5flu and mock PBS vaccinated mice were assessed for H1N1-specific T cells at day 21 after 2 dose vaccination. (B) vRNA was extracted from the lungs of infected mice and amplified by RT-PCR (C) for whole genome next generation sequencing. (D) The cumulative number of variants (from input H1N1 virus) above 1% (allele frequency (AF) 0.01) from day 3, 5 and 7 from PBS, standard quadrivalent inactivated influenza vaccines or Wyeth/IL-15/5flu .

(b) Enhanced inactivated influenza vaccines and repeat vaccination for older adults

In our randomized clinical trial of enhanced vaccines in older adults , enhanced vaccines, particularly adjuvanted vaccines, had increased antibody responses, up to one year later, compared to standard vaccination (**Figure 2A**), and increased cytokine positive T cell responses (**Figure 2B**), leading to polyfunctional responses (**Figure 2C**). (Cowling *et al.*, Clin Infect Dis 2019, Li *et al.*, NPJ Vaccines 2021).

The same enhanced and seasonal inactivated vaccines were assessed in a mouse vaccination and challenge model to determine vaccine memory and survival from lethal influenza challenge. In mice, the adjuvanted vaccines led to earlier IgG class switching of vaccine specific antibodies (**Figure 2D**), which coincided with increased viral clearance (**Figure 2E**), and early viral neutralisation at infection with a distinct H3N2 virus (Figure 2F) (Kavian *et al.*, Clin Transl Immunol 2020). Experiments on further vaccine combinations are underway, and formed the basis of Athena Li's PhD project which was completed in August 2021.

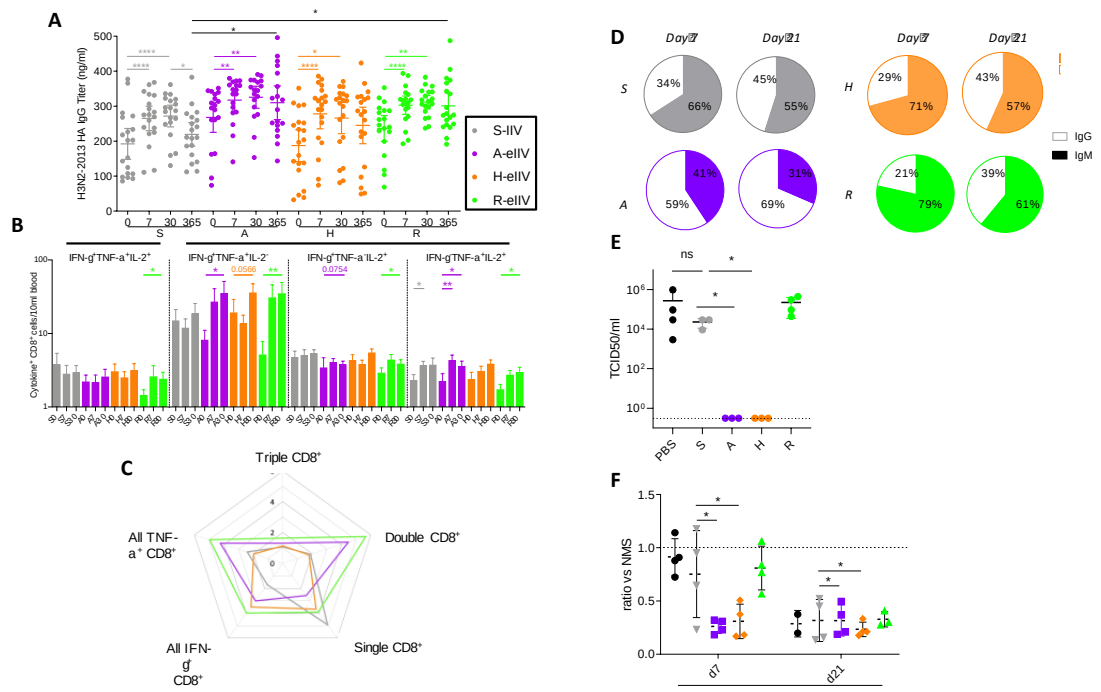

Figure 2: Enhanced IIV in older adults and mice to determine protective correlates. (A) H3-2013 IgG in standard (S), adjuvanted (A), high dose (H) and recombinant (R) influenza vaccination at baseline (day 0), 7, 30 and 365 days post vaccination (n=20 per group). (B) CD8+ T cell responses from vaccinated older adults and (C) cytokine profiles. (D) Antibody switching (based on area under the curve by endpoint titrations of H3-2013 HA IgG and IgM response) from vaccinated mice. (E) Vaccinated mice were challenged with 10LD50 H3N2 HK-1968 and lung viral titers measured at day 7 post infection. (F) The serum response for *in vitro* virus infection of vaccinated mice from day 7 and day21 after H3N2-1968 challenge normalized to naïve normal mouse serum (NMS).

Immune correlates of SARS-CoV-2 infection

(a) Immune responses to SARS-CoV-2 infection

In an unbiased quantitative serological assay, we measured the antibody responses in SARS-CoV-2 infected patients, and found the N-specific antibody response is by far the most immunodominant antigen (**Figure 4A**). Secondly, the ORF8 and ORF3b anitgens that are unique and distinct to SARS-CoV-2 elicited antibody responses. The combined use of N, ORF8 and ORF3b allows the accurate discrimination of SARS-CoV-2 infected cases from pre pandemic controls (**Figure 4B**), with distinct kinetics longitudinally (**Figure 4C**) (Hachim *et al.*, Nat Immunol 2020). Children experience mild COVID-19, and compared to mild/asymptomatic infected adults have a reduced response to structural proteins and increased contribution of antibodies to accessory non-structural proteins to their SARS-CoV-2 antibody landscape (**Figure 4D-E**). This maybe attributable to different viral pathogenesis and prior immunity in children. Further experiments on the cytokine response in relation to the antibody landscape showed unique drivers by prinicipal component analysis on age related differences for SARS-CoV-2 responses. This work was recently accepted at Nature Communications (April 2022).

We have further investigated in collaboration with Chris Mok at the PRP, the T cell (**Figure 5A**) and antibody quality by avidity (**Figure 5B**) and FcR binding (**Figure 5C**) to compare the inactivated whole virion Coronovac and mRNA Spike Pfizer vaccine responses in healthy adults. This work was published in Respirology (December 2021).

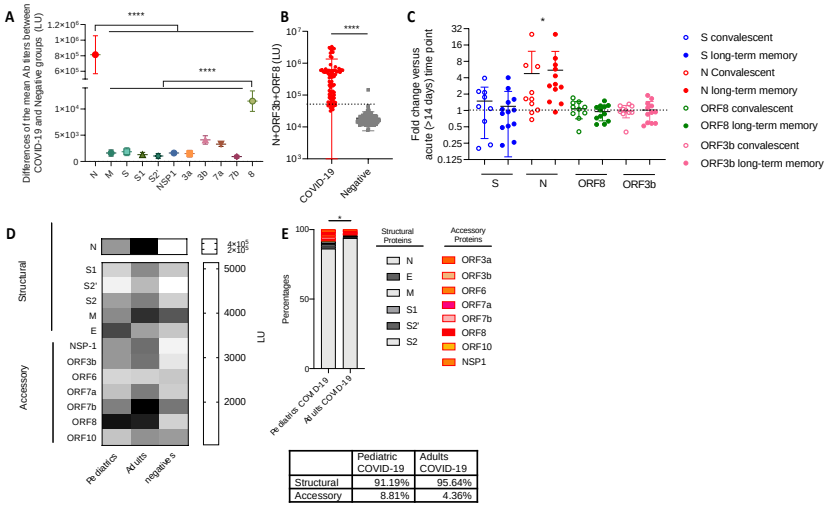

Figure 4: SARS-CoV-2 antibody landscapes for accurate diagnosis and differences in children. (A) 11 antigens of SARS-CoV-2 were screened in a luciferase style pulldown assay for COVID-19 patients and healthy controls to determine responses with the largest difference from controls. (B) Combined antibody responses for N, ORF3b and ORF8 from COVID-19 patients and healthy controls. (C) Longitudinal paired kinetics of SARS-CoV-2 specific antigen responses in COVID-19 patients. (D) The SARS-CoV-2 antibody response of infected children, adults and negative controls, and (E) the proportion of antibody responses for structural and accessory proteins.

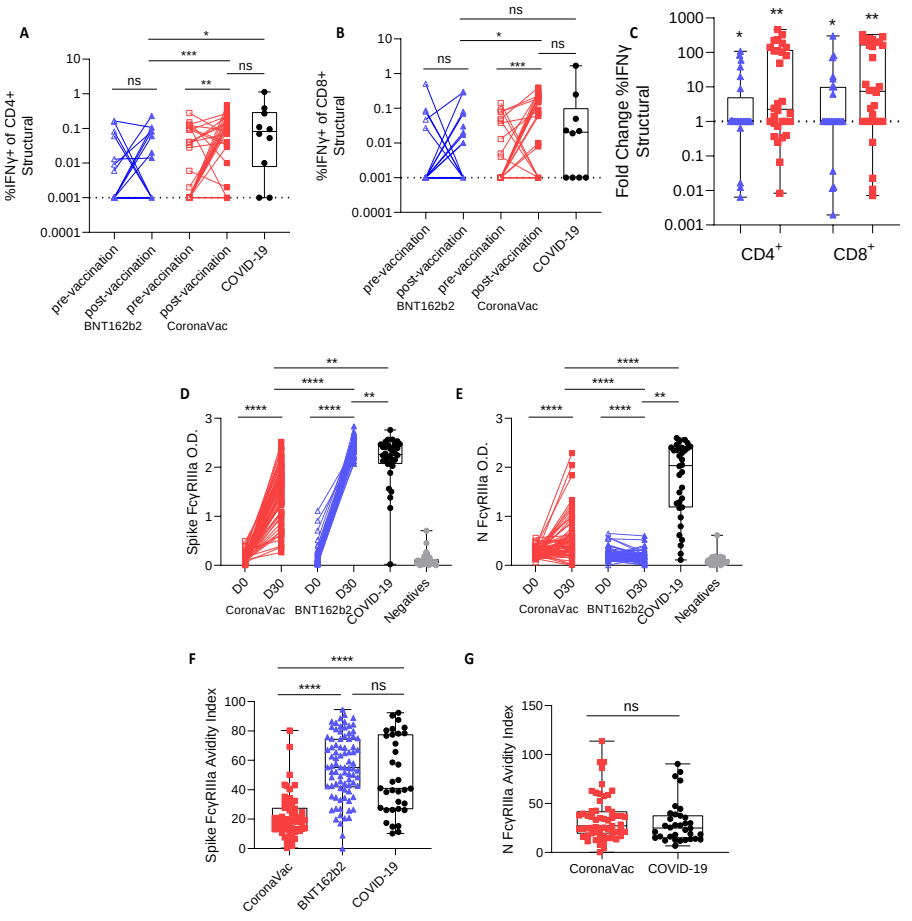

Figure 5: SARS-CoV-2-specific T cell and antibody quality be vaccination. (A) SARS-CoV-2 structural peptides were used to stimulate CD4⁺ (A) and CD8⁺ (B) T cell responses from baseline (D0) and 1 month post 2 dose vaccination (D30) of inactivated whole virion Coronavac and Spike mRNA Pfizer BNT162b2, and the fold change (C) of responses post vaccination. FcgRIIIa binding antibodies for ADCC responses for Spike (D) and Nucleocapsid (E), and for avidity (F,G).

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2. Bull MB, Gu H, Ma FNL, Perera LP, Poon LLM, Valkenburg SA (2022) Next Generation T cell-activating vaccination increases influenza virus mutation prevalence. *Science Advances*, in press.
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5. Dhanasekaran V, Sullivan S, Edwards KM, Xie R, Khvorov A, Valkenburg SA, Cowling BJ, Barr IG (2022) Human seasonal influenza under COVID-19 and the potential consequences of influenza lineage elimination. *Nat Commun* **13**:1721.
6. Mok CKP, Cohen CA, Cheng SMS, Chen C, Kwok KO, Yiu K, Chan TO, Bull MB, Ling KC, Lai HL, Dai MZ, Ng SS, Lui GCY, Amarasinghe GK, Leung D, Wu C, Wong SYS, Valkenburg SA, Peiris JSM, Hui DSC (2022) Comparison of the immunogenicity of BNT162b2 and CoronaVac COVID-19 Vaccines in Hong Kong: An observational cohort study. *Respirology* **27**:301-310.
7. Yen HL, Valkenburg S, Sia SF, Choy KT, Peiris JSM, Wong KHM, Crossland N, Douam F, Nicholls JM (2021) Cellular tropism of SARS-CoV-2 in the respiratory tract of Syrian hamsters and B6.Cg-Tg(K18-ACE2)2Prlmn/J transgenic mice. *Vet Pathol*, in press.
8. Nguyen THO, Cohen CA, Rowntree LC, Bull MB, Hachim A, Kedzierska K, Valkenburg SA (2021) T cells targeting SARS-CoV-2: by infection, vaccination and against future variants. *Front Med Research* **8**:793102.
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10. Hachim A, Kavian N, Valkenburg SA (2021) Antibody landscapes of SARS-CoV-2 can reveal novel vaccine and diagnostic targets. *Curr Opin Virol* **52**:139-146.
11. Valkenburg SA, Cheng SMS, Hachim A, Peiris M, Nicholls J (2021) Postmortem stability of SARS-CoV-2 in mouse lung tissue. *Emerg Infect Dis* **27**:3173-3175.
12. Cohen CA, Li APY, Hachim A, Kwan MYW, Chan WH, Yau YS, Chiu SS, Tsang OTY, Hui DSC, Kavian N, Ma FNL, Lau EHY, Cheng SMS, Poon LLM, Peiris JSM, Valkenburg SA. (2021) SARS-CoV-2 specific T cell responses are lower in children and increase with age and time after infection. *Nat Commun* **12**:4678.
13. Kavian N, Hachim A, Cowling BJC, Valkenburg SA (2021) Repeated influenza vaccination provides cumulative protection from distinct H3N2 viruses. *Clin Transl Immunology* **10**:e1297.

14. Wu C, Qavi AJ, Hachim A, Kavian N, Cole AR, Moyle AB, Wagner ND, Sweeney-Gibbons J, Rohrs HW, Gross ML, Peiris JSM, Basler CF, Farnsworth CW, Valkenburg SA, Amarasinghe GK, Leung DW (2021) Characterization of SARS-CoV-2 N protein reveals multiple functional consequences of the C-terminal domain. *iScience* **24**:102681.

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Seminars and Invited Presentations

1. Sophie Valkenburg (2021) Chinese University of Hong Kong, Hong Kong SAR (webinar)
2. Sophie Valkenburg (2021) Peter Doherty Institute, Melbourne, Australia (webinar).
3. Sophie Valkenburg (2021) *From HKU-Pasteur to C2i: Confronting the challenges of infectious diseases* C2i, HKSTP, Hong Kong SAR.
4. Sophie Valkenburg (2021) Dangerous liaisons: Emerging viruses at the human, animal and environmental interface (webinar).

Presentations at Meetings

1. Sophie Valkenburg (2021) Australasian Society for Immunology annual meeting, December (*Oral*)
2. Sophie Valkenburg (2021) Australian Influenza Symposium (*Oral*)
3. Sophie Valkenburg (2021) ISIRV WHO COVID influenza RSV (*Poster*, *virtual event*)

Teaching

1. Sophie Valkenburg (2021) "Biological Basis of Disease" (Master of Public Health, CMED-6227), School of Public Health, The University of Hong Kong, Hong Kong SAR (*Course Director and Lecturer*).
2. Sophie Valkenburg (2021) Outbreak – Problem Based Learning (MBBS Year 1), The University of Hong Kong, Hong Kong SAR (*Tutor*).
3. Sophie Valkenburg (2021), Health Research Project, The University of Hong Kong, Hong Kong SAR (*Tutor*).
4. Sophie Valkenburg (2021), Problem based learning tutor for MBBS students, The University of Hong Kong, Hong Kong SAR (*Tutor*).

Collaborations

1. **Benjamin Cowling** (School of Public Health, The University of Hong Kong, Hong Kong SAR): Longitudinal impact of repeat influenza and combined COVID-19 vaccines.
2. **Leo LM Poon** (School of Public Health, The University of Hong Kong, Hong Kong SAR): Broadly reactive influenza and COVID-19 vaccines in mouse models.

3. **JS Malik Peiris** (School of Public Health, The University of Hong Kong, Hong Kong SAR): SARS-CoV-2 patient cohorts for immune characterisations.
4. **Gaya Amarasinghe** (Univeristy of Washington, St Louis, USA): SARS-CoV-2 N protein functions.
5. **Liyange Perera** (NIH, USA): next generation vaccines.
6. **Ragahavan Varadarajan** (Indian Institute of Science, Bangalore, India): Characterization of a headless-trimeric pre-fusion conformation HA recombinant protein vaccine in a mouse model to generate broadly reactive HA-stem antibodies.
7. **Amy Chung** (University of Melbourne, Australia): Antibody effector functions after influenza exposure by multiplex assay.
8. **Adam Wheatley** (University of Melbourne, Australia): Recombinant influenza protein expression system.
9. **Mark Hogarth** (Burnet Institute, Australia): Reagents for antibody effector functions.

Funding

1. Assessing novel coronavirus antibodies for specificity and function during clinical infection and community asymptomatic cases (**Principal investigator**, FHB, HMRF commissioned grants, ends 2021).
2. Community based sero-epidemiological study of COVID19 to provide data in real time on age- stratified infection attack rates, disease severity and population-immunity, for guiding intervention policy (**Co-investigator**, FHB, HMRF commissioned grants, ends 2021).
3. Multiplexed antibody function for protection from influenza (**Principal investigator**; HKU – Seed Funding for Basic Research – Ends: 03/2021).
4. Influenza viruses adapt to escape T cell responses (**Principal Investigator**; Research Grants Council/General Research Fund – Ends: 05/2021).
5. Research on the epidemiology, vaccine effectiveness and treatment of influenza and other respiratory viruses in Southeast Asia and the Western Pacific (**Co-Investigator**; US Center for Disease Control – Ends: 07/2021).
6. Influenza ADCC-antibody responses in vaccination and infection of children as a correlate of protection (**Principal Investigator**; Health and Medical Research Fund – Ends: 08/2021).
7. Immunity to viruses by infection and vaccination for correlates of protection (**Principal Investigator**; HKU – Enhanced Research Start-up Award – Ends: 12/2021).
8. COVID-19 severity and cytokine modulation of the antibody landscape (**Co-investigator**, Doherty Sino grant, 2021).
9. The protective role of antibody effector functions for influenza in mice and humans (**Principal investigator**; Research Grants Council/General Research Fund – Ends 06/2022).

10. Replication-defective SARS-CoV-2 mutant vaccines with abnormal codon usages (**Co-investigator**; RGC,CRF grant, C7145-20G, 2021-2023).
11. Control of influenza: individual and population immunity (**Co-investigator**; Research Grants Council/Theme based Research Scheme – Ends 2024).
12. Immunity to SARS-CoV-2 and influenza viruses by vaccination and infection (**Principal investigator**; UGC – Research Matching Grant – Ends 04/2022).
13. Virological, immunological and epidemiological characterization of COVID-19 (**Co-investigator**; RGC, Theme based Research Scheme – Ends 2027).
14. Pasteur Talent Award 2020 (**Awardee**; Institut Pasteur – Ends: 2199).

Personnel

Name	Position
Sophie VALKENBURG	Research Assistant Professor
Prathanporn KAEWPREEDEE	Postdoctoral fellow (from November 2021)
Maireid BULL	PhD Student (until August 2021)
Carolyn COHEN	PhD Student (HKPF awardee)
Athena LI	PhD Student (until August 2021)
Janice JIA	Research Assistant I
Fionn MA	Research Assistant I (until November 2021)
Alan CHEUNG	Research Assistant II
Asmaa HACHIM	Research Assistant II (until January 2022)
Luke WONG	Research Assistant II (until February 2021)
Emma YANG	Student Intern
Hoi Yan SZE	Student Intern
Vanessa TSANG	Student Intern

3.4 Sook-San WONG Lab

Main Objectives and Strategy

The laboratory is focused on understanding the immunological and virological determinants of robust antibody responses after respiratory virus infection and vaccination at a population as well as at the individual level. Our interests also include studying population immunity to zoonotic viruses and how that contributes to our risk assessment process of such pathogens. This research is critical to our understanding of respiratory viruses' vaccine efficacy and pathogenesis and how that affects the population's susceptibility to respiratory viruses. The laboratory research platform is based on immunological assays focusing on humoral immunity, using samples from clinical and epidemiological cohorts as well as from animal models. We are currently developing the following projects under the specified research themes:

The role of IL-10 and vaccine responses

(a) Cytokine markers to predict vaccine responses: Vaccination remains one of the most effective public health strategy to control community transmission as well as to reduce the disease burden of respiratory viruses. However, some vaccinees may fail to mount sufficient protective antibody responses after primary or booster vaccination. We have previously discovered that high levels of IL-10 at time of vaccination were associated with subsequent poor antibody responses upon receipt of the inactivated influenza vaccines in older adults. We hypothesize that IL-10, as a marker of the underlying state of low-grade inflammation (inflammaging) in older adults, can be used to identify potential poor vaccine responders and further probe the mechanisms that leads to vaccine failures in older adults. In collaboration with Ben Cowling and Malik Peiris at HKU, we are developing a project to evaluate this in the 'Immunogenicity of Alternative Annual Influenza Vaccination Strategies in Older Adults in Hong Kong (PIVOT)' study, which was a head-to-head comparison on the immunogenicity of the standard-dose inactivated vaccine, MF59-adjuvanted vaccine, high-dose vaccine and recombinant-hemagglutinin (rHA) vaccine given in an alternating or non-alternating regimen over the course of four-years. Our primary goal for this project would be to determine if these serum biomarkers can be used to identify poor vaccine responders in the elderly. The proposed study using PIVOT samples can contribute towards precision vaccination in older adults, identifying optimal vaccine regimen that is suitable for the immune or health status of the vaccinee using already licensed vaccines.

(b) Mechanisms underlying the IL-10 mediated vaccine failure: Previous study have suggested that dysregulation in the monocytic subpopulations is a potential cause of the IL-10 mediated poor influenza vaccine responses in the older population. To follow up on this hypothesis and as an extension to the above study, we will be collaborating with Sophie Valkenburg, formerly of HKU Pasteur and now at the Peter Doherty Institute at The University of Melbourne, to investigate the immune mechanisms underlying the IL-10 mediated vaccine failure. Once established, we will expand this study within the context of other vaccines, such as COVID-19, and age-groups to examine the generalizability of this phenomenon.

The population dynamics of influenza hemagglutinin (HA) and neuraminidase (NA)-antibodies.

(a) Age-and population specific dynamics

We had previously showed that the immunodominance of the influenza HA and NA antibody responses occur in an age-specific manner after infection (Wong et al., 2020). We hypothesize that the pre-existing pool of memory B-cells will skew the pattern of response to potentially a HA-biased or NA-biased response. Furthermore, it is hypothesized that the high pre-existing antibodies may reduce the likelihood of viral antigens being presented for a robust humoral response, contributing to the "antibody ceiling effect". This research aims to elucidate the immunological principles underlying the age-dependence immunodominance by examining the pattern of HA and NA antibody responses in human sera from naturally-infected and vaccinated cohorts and will be recapitulated using animal models.

(b) Neuraminidase (NA) immunity as correlates of protection against zoonotic influenza

NA-antibodies are more broadly cross-reactive compared to HA-antibodies and may provide better protection against antigenic variants. Furthermore, repeated exposure through infection or vaccination with vaccines containing residual NA can increase the breadth of antibody cross-reactivity. The goal of this program is to identify the role of NA-antibodies in risk-assessments of zoonotic influenza viruses, in particular those posed by avian and swine influenza viruses, and potentially utilize existing influenza vaccines as a safe, low-cost strategy to reduce the risks of zoonotic infections.

Previous achievements and Ongoing Research

My key contribution to science is in the identification of cellular correlates to influenza antibody responses and disease pathogenesis. Using samples from two human cohort studies (SHIVERS and FLU09), I was able to demonstrate the distinct roles of the monocyte subsets and CD4+ and CD8+ T-cells in regulating antibody responses and disease severity. I also contributed to the identification of correlates of vaccine-induced immunity for avian influenza viruses, an important issue that is challenging to address in the field. I led several studies to address these issues in vitro and in the ferret model. We found that the avian influenza vaccines are not inherently poorly immunogenic compared to seasonal influenza vaccines but, crucially, that the structural integrity of the vaccine components may play a role in eliciting neutralizing antibodies. We were able to link the contribution of each immune arm to disease outcome in the H5N1 ferret challenge model, providing the basis for an understanding of avian influenza disease pathogenesis in humans. Finally, I also contributed to epidemiology and clinical studies of SARS-CoV-2 in China. I co-led a study to predict the epidemic curve of SARS-CoV-2 in China under the introduced control measures in March 2020 using a modified SEIR model and a machine learning approach. This study was a significant contribution to the understanding on the transmission of SARS-CoV-2 in China early in the outbreak. I presented these findings to the first joint WHO mission to China at the invitation of Guangdong CDC in February 2020. I also led a cross-sectional serosurvey to investigate the seroprevalence of SARS-CoV-2 antibodies across Guangdong Province after the emergence of SARS-CoV-2 in 2020.

Immune correlate of antibody responsiveness and disease severity after influenza virus infections and vaccination

(a) Activated CD4+ T-cells and CD14++CD16+ monocytes correlate with antibody response following influenza virus infection in humans

The failure to mount an antibody response following viral infection, or seroconversion failure, is a largely underappreciated and poorly understood phenomenon, especially prior to the emergence of COVID-19. Using samples collected from two independent human cohorts, SHIVERS and FLU09, based in Auckland, New Zealand and Memphis, Tennessee, USA, respectively, we identified immunologic markers associated with robust antibody responses after influenza virus infection (Wong et al., 2021). In the SHIVERS cohort, seroconversion significantly associates with (i) hospitalization, (ii) greater numbers of proliferating, activated $CD4^+$ T-cells **(Figure 1A and 1B)**, but not $CD8^+$ T-cells, in the periphery during the acute phase of illness, (iii) more activated $CD16^+$ monocytes **(Figure 1C)** and, (iv) fewer inflammatory monocytes ($CD14^{++}CD16^+$) by convalescence **(Fig. 1D)**. In the FLU09 cohort, individuals that mounted influenza-specific mucosal IgA and IgG-antibodies **(Figure 1E-F)** also had fewer $CD14^{++}CD16^+$ monocytes during early illness in the nasal mucosa **(Figure 1G)**. Our study demonstrates that seroconversion failure after infection is a definable immunological phenomenon, associated with quantifiable cellular markers that can be utilized to improve diagnostics, vaccine efficacy and epidemiologic efforts.

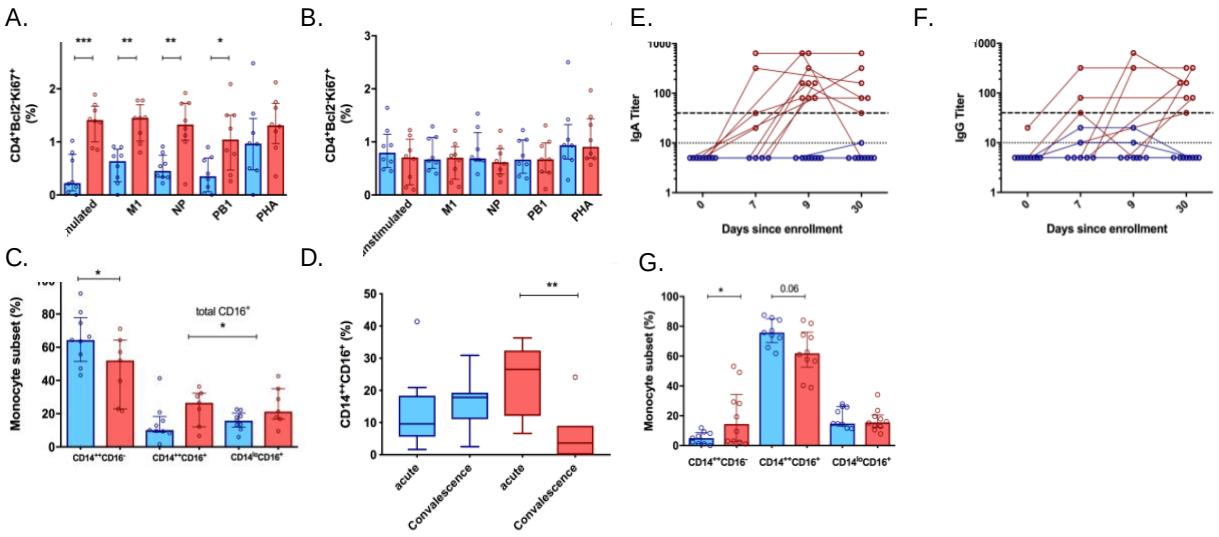

Figure 1. Cellular correlates of antibody responsiveness in the peripheral and nasal airways. (**A**) Frequency and (**B**) number of proliferating $CD3^{+}CD4^{+}$ T-cells ($Bcl2^{-}$ and $Ki67^{+}$) during the acute and convalescence phase, respectively in the seroconverters (SC, red, n=7) and non-seroconverters (Non-SC, blue, n=9) in the SHIVERS study. (**C**) Percentages of each monocyte subpopulations (per total monocytes) during acute phase and (**D**) percentages of $CD14^{++}/CD16^{+}$ monocytes in the acute and convalescent time-points samples in SC and non-SC. Immune responses in the nasal airways of participants in the FLU09 cohort. Influenza-specific (**E**) IgA and (**F**) IgG-titers in the nasal washes collected on day 0, 7, 9 and 28 post enrolment of converters (red, n=10) and non-converters (blue, n=9). Converters were defined as individuals with IgA-titers > 40 at any time points during the study, whereas non-converters were individuals that did not. (**G**) Percentages of each monocyte subpopulations (per total monocytes) in the nasal airways of converters and non-converters collected within a week of enrolment.

(b) Severe influenza results in prolonged immune activation and is associated with less $CD8^{+}$ T-cells activation and increased $CD14^{lo}CD16+$ monocytes

While mild influenza can be subclinical or manifest as fever with unremarkable respiratory symptoms, severe influenza is characterized by respiratory complications such as pneumonia, respiratory distress, and even death. Although risk factors such as extremes of age, preexisting health conditions, and genetics have been identified, a cohesive understanding of the underlying immunological process, particularly within cellular immunity, is still not well-defined. As part of the SHIVERS study, we compared the immunological profiles of patients diagnosed with mild (defined as those with influenza-like illness, ILI) and severe influenza (defined as those with severe acute respiratory infection, SARI) (Wong et al., 2018). We found that most cases of SARI, despite no significant difference in the monocyte subpopulation in the early acute phase, showed increased numbers of $CD16^{+}$ monocytes, and in particular the $CD14^{lo}CD16^{+}$ "patrolling" monocytes which had potent antiviral functions during the convalescent phase of disease **(Figure 2A-B)**. Activation of the homing receptor, $CD192^{+}$ on these monocytes were also lower in SARI during the acute phase **(Figure 2C-D)** but had increased by the later time point **(Figure 2E)**.

Activation of the homing receptor, CD192^{+} on these monocytes were also lower in SARI during the acute phase **(Figure 2C-D)** but had increased by the later time point **(Figure 2E)**. Also by convalescent phase, most SARI cases showed continued immune activation that was characterized by increased numbers of activated CD16^{+}monocytes **(Figure 2F)**, CD8^{+} and CD4^{+} T cells **(Figure 2G-H)** as well as proliferating CD8^{+} T cells **(Figure 2I-J)**. Notably, there were also less influenza virus–specific CD8^{+} T cells in SARI during the acute phase as compared to ILI cases **(Figure 2K-L)**. Taken together, findings from both studies showed that CD8^{+} T-cells and CD14^{lo}CD16^{+} monocytes are important regulating disease severity, while CD4^{+} T cells and CD14^{++}CD16^{+} monocytes are important for eliciting a robust antibody response after infection. This is in agreement with current findings being reported for COVID-19.

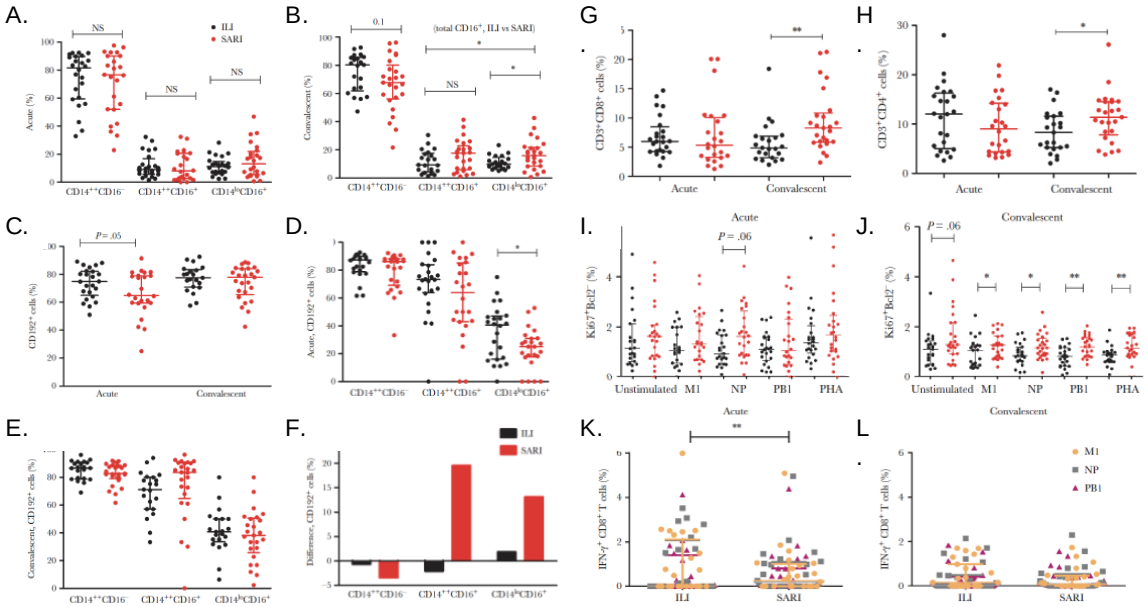

Figure 2. Differences in the monocyte subpopulations and T-cells in influenza-like illness (ILI; black) and severe acute respiratory illness (SARI; red) influenza cases during the acute and convalescent phases of disease. Monocyte populations are identified on the basis of the relative expression level of the markers CD14 and CD16 as CD14^{++}/CD16^{-} (classical),CD14^{++}/CD16^{+} (inflammatory), and CD14^{lo}/CD16^{+} (patrolling) monocytes. (**A**, **B**) Percentages of each monocyte subpopulation (per the total monocyte population) during the acute and convalescent phases. (**C**) Percentages of CD192^{+} cells in the total monocyte population during the acute and convalescent phases. (**D**, **E**) Percentages of CD192^{+} cells per monocyte subpopulation in ILI and SARI during the acute and convalescent phases. Lines represent medians, and error bars represent interquartile ranges. (**F**) Change in percentages of CD192^{+} monocytes in each subpopulation between acute and convalescent samples. Values were obtained by subtracting acute values from the convalescent value. (**G**, **H**) Percentages of CD3^{+}CD8^{+} and CD3^{+}CD4^{+} T cells in ILI and SARI during the acute and convalescent phases. (**I**, **J**) Percentages of proliferating CD3^{+}CD8^{+} T cells (Ki67^{+}Bcl2^{-}) after peptide stimulation (M1, NP, and PB1) in ILI and SARI during the acute and convalescent phases. (**K**, **L**) Total influenza virus–specific IFN-γ–expressing CD8^{+} T-cell responses after peptide (M1, NP, and PB1) stimulation (with the baseline value subtracted) during acute and convalescent phase. Data are expressed as medians, and error bars represent interquartile ranges. *P < .05, **P < .01.

Epidemiology of human coronaviruses (hCoV) and SARS-CoV-2 in China

(a) Modeling the COVID-19 epidemic curve in China under public health control measures

On January 23 2020, China introduced unprecedented nationwide public health control policy in an effort to contain the spread of this newly emerged SARS-CoV-2 from Wuhan, Hubei Province. These policies included large-scale quarantine, strict controls on travel and extensive monitoring of suspected cases. However, it was unknown whether these policies have had an impact on the epidemic.

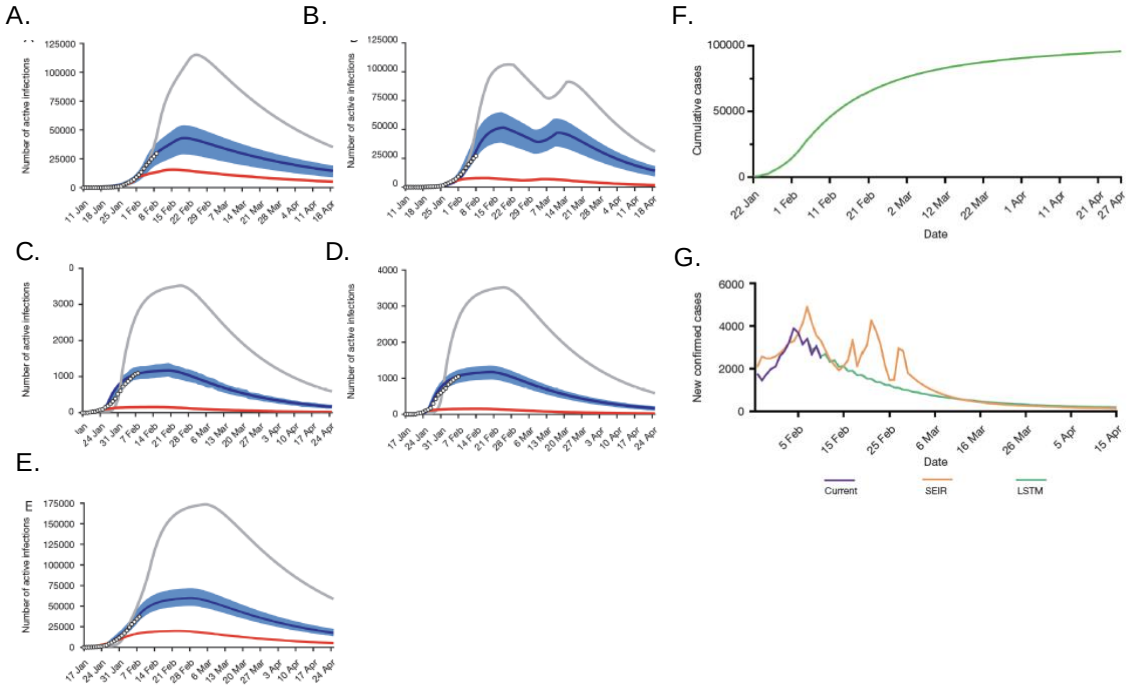

Figure 3. Number of active infections predicted by the modified SEIR model. (**A**) Hubei province under strict quarantine, (**B**) Hubei province under eased quarantine, (**C**) Guangdong province, (**D**) Zhejiang province and (**E**) China when interventions were introduced on January 23 (blue), five days later (grey) and five days earlier (red). Actual data of daily confirmed infections were fitted onto the curve (circles). (**F**) LSTM-predicted cumulative number of COVID-19 cases in China. (**G**) Number of new COVID-19 cases according to actual data (purple), SEIR-model (orange) and LSTM model (green). SEIR, Susceptible-Exposed-Infectious-Removed; LSTM, Long-Short-Term-Memory; COVID-19, coronavirus disease 2019.

Along with colleagues and as part of the COVID-19 response team, we used a modified Susceptible-Exposed-Infectious-Removed (SEIR) model that integrated population migration data before and after January 23 and the most updated COVID-19 epidemiological data to predict the epidemic curve for Hubei under strict quarantine **(Figure 3A)**, Hubei under eased quarantine **(Figure 3B)**, Guangdong **(Figure 3C)**, Zhejiang **(Figure 3D)** and China **(Figure 3E)** (Yang et al., 2020). To complement the classical statistical model, we also used an artificial intelligence (AI) approach, using the Long-Term-Short Memory algorithm and trained on the 2003 SARS data, to predict the epidemic **(Figure 3F)**. Our dynamic SEIR model was accurate in predicting the COVID-19

epidemic peaks and sizes. The implementation of control measures on January 23 2020 was indispensable in reducing the eventual COVID-19 epidemic size. This study was one of ESI's top cited paper for 2020.

(b) ***Seroepidemiology of SARS-CoV-2 in Guangdong Province*** [Funding: GIRH-Harvard COVID-19 Fund GIRHHMS012]

Guangdong province is one of the largest and most populous provinces in China. Due to its status as an economic powerhouse in Southern China, it draws in an estimated 30 million migrant workers annually from neighbouring provinces. Given its importance as a economic hub, we wanted to monitor the SARS-CoV-2 activity in the province. In collaboration with the largest clinical service provided in China, we conducted an age-stratified seroprevalence study across Guangdong province to determine the extent of SARS-CoV-2 spread within the first 6-months after the emergence of the virus.

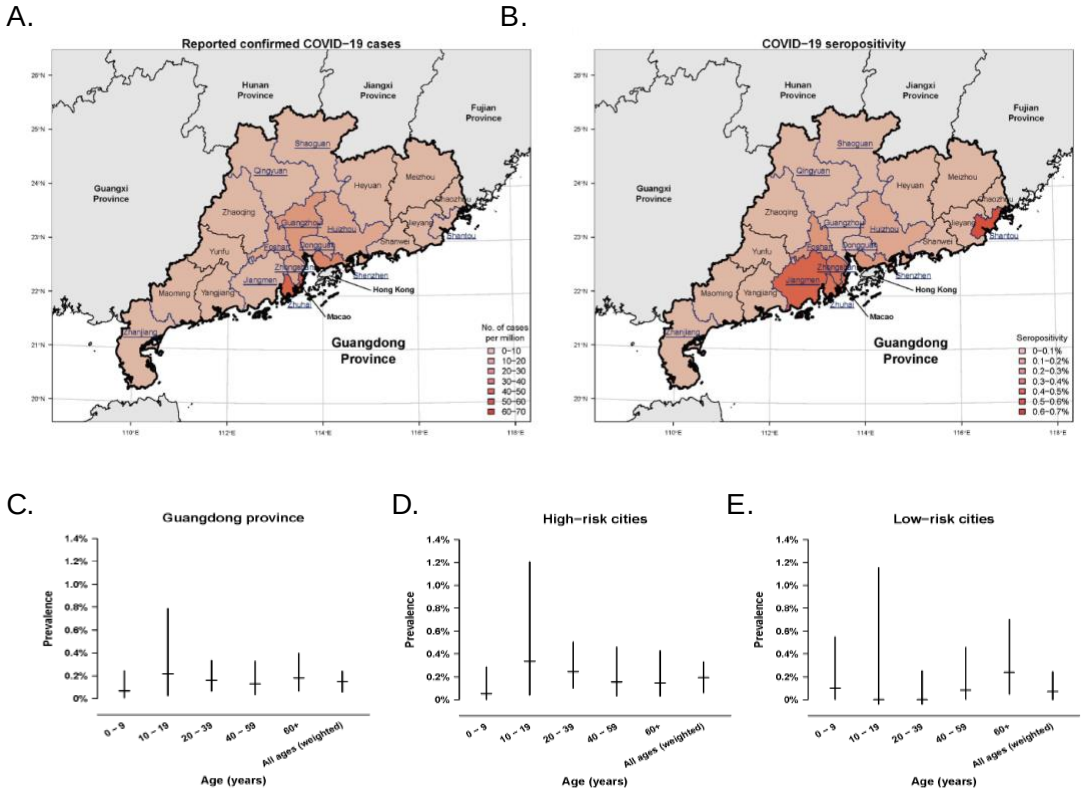

Figure 4. Seroepidemiology of SARS-CoV-2 after the first wave in Guangdond Province. (**A**) Reported confirmed COVID-19 cases, not including asymptomatic cases, based on local official surveillance data in the different prefectural cities in Guangdong province. (**B**) Seropositivity of antibodies to SARS-CoV-2 as identified from the present study, in prefectural cities of the Guangdong province within the first six-months post COVID-19 emergence. Cities that had relatively higher connectivity with Wuhan prior to 23 January 2020, were underlined and highlighted in blue. Estimates of age-specific SARS-CoV-2 seroprevalence in Guangdong province in (**C**) all cities or stratified by (**D**) high or (**E**) low risk of COVID-19 activities according to local officialsurveillance data. Seroprevalences in all ages were age- and sex-weighted according to the

We screened 14,629 sera samples, and found 21 SARS-CoV-2 IgG-positive sera, bringing an age and sex-adjusted seroprevalence estimate in Guangdong province to be 0.15% (95% CI=0.06-0.24%) (Xiao et al., 2021). Overall, there was no hidden epidemics in Guangdong and seropositivity was detected in cities with high mobility index with Wuhan

in the 5 days preceeding the national lockdown on January 23 2020 **(Figure 4A-B)**. No significant age-specific seroprevalence difference was observed due to the low seropositivity detected **(Figure 4C-E)**. This suggests that importation of cases from the outbreak center was likely the cause of seropositivity.

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Human post-infection serological response to the spike and nucleocapsid proteins of SARS-CoV-2. *Influenza Other Respir Viruses* **15**:7-12..

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Collaborations

1. **Benjamin Cowling** (School of Public Health, The University of Hong Kong, Hong Kong SAR): The association of serum cytokine levels with responses to influenza vaccination in older adults receiving standard and enhanced influenza vaccines; Observational study on incidence and epidemiological characteristics of influenza virus infection in adults aged 60 and over - understanding the potential preventive value of twice-yearly influenza vaccination.
2. **JS Malik Peiris** (School of Public Health, The University of Hong Kong, Hong Kong SAR): SARS-CoV-2 patient cohorts for immune characterisations.
3. **Sophie Valkenburg** (Doherty Institute, The University of Melbourne): collaboration on mechanisms of vaccine failures in the PIVOT study
4. **Richard J Webby** (Department of Infectious Diseases, St. Jude Children's Research Hospital): Mechanisms of avian influenza disease
5. **Yang Zifeng, Zhong Nanshan** (State Key Laboratory of Respiratory Disease, Guangzhou Institute of Respiratory Health): Mechanisms of avian influenza disease
6. **Maureen McGargill, Paul Thomas** (Department of Immunology, St. Jude Children's Research Hospital): Mechanisms of avian influenza disease
7. **Mariette Ducatez** (National Institute for Agriculture Research (INRA) and of the National Veterinary School of Toulouse (ENVT): Contribution of virological factors and herd immunity in H9N2 influenza transmission and human disease
8. **Kang Min (Guangdong Centers for Disease Control and Prevention)**: Characterizing the serological response in an influenza vaccination study in older adults in Guangdong Province.

Funding

1. The association of serum cytokine levels with responses to influenza vaccination in older adults receiving standard and enhanced influenza vaccines. (**Principal investigator**, submitted to FHB/HMRF 2022 funding round).
2. Observational study on incidence and epidemiological characteristics of influenza virus infection in adults aged 60 and over - understanding the potential preventive value of twice-yearly influenza vaccination (Guangzhou Institute of Respiratory Health, Subcontract from Prof. Benjamin Cowling, HKU, ends July 2023)

Personnel

Name	Position
Sook-San WONG	Assistant Professor
Lewis SIU	Senior Technician
Cheng XIAO	Research Assitant II (starting June 2022)

3.7 Teaching and Education

HKU-Pasteur Course Series

HKU-PRP has pioneered a unique course series in Hong Kong and in the region that provides state of the art lectures and practical workshops in a "Master class" setting to outstanding postgraduate students and postdoctoral fellows coming from countries with markedly different resources. The alumni network demonstrates that this educational program helps intensify human and scientific links between HKU-PRP, the School of Public Health at HKU, the Institut Pasteur International Network and the international scientific community, while continuing to attract to Hong Kong top scientists and highly motivated students. HKU-Pasteur courses are extremely competitive and comparable in quality to that of established benchmarks, such as EMBO and Cold Spring Harbor courses and, therefore, are solidifying the reputation of HKU-PRP and Hong Kong as the premier regional hub for biomedical education. All HKU-Pasteur Courses have been approved by the Research Postgraduate Committee of HKU for inclusion in the coursework curriculum of MPhil/PhD students, and are supported with external grants from the Institut Pasteur International Network, the French Consulate in Hong Kong and Macau, the Regional Health Cooperation Office of the French Ministry of Foreign Affairs the Pasteur Foundation Asia and other private donations. **All courses were cancelled in 2020 owing to the coronavirus pandemic. They will resume when travel restrictions are lifted**.

The Institut Pasteur and the Pasteur International Network association (PINA) have launched the official Pasteur International Courses "PIC" label for the Institut Pasteur International Network's (IPIN) training programmes, workshops and Massive Open On-Line Courses (MOOCs). This programme, coordinated by the Education Department, the International Department and Pasteur International Network association, will strengthen the visibility and attractiveness of education programmes, digital and onsite, across the Network. It will also promote Institut Pasteur courses/MOOC in local and regional universities, developing partnerships with other stakeholders and contributing to the career development of future scientific and public health leaders. We have submitted applications to obtain the Pasteur International Course (PIC) label for all our programs. The new PIC Labelling Programme will ensure that, in an increasingly competitive environment, the "Institut Pasteur" excellence will be recognised as best-in-class for research and training, attracting the students and researchers at all stages of their career, as well as laboratory technicians and health professionals.

Additional teaching and training

Besides their involvement in the HKU-Pasteur course series, the **Co-Directors and Group Leaders** at HKU-Pasteur are also teaching courses in the undergraduate and postgraduate curriculum and the Problem-Based Learning modules for MBBS students (see cpmplete list at the end of this section). HKU-PRP regularly hosts undergraduate/postgraduate students from overseas institutions for internships. In 2021 we welcomed one international trainee for an internship period. In addition, we resumed o**ur training programs for high school students from the French International School (FIS) in Hong Kong and the Hong Kong Institute of Vocational Education (IVE)**. We hosted two students from FIS and six from IVI.

Complete list of taught and international courses

1. Roberto Bruzzone (2021) Molecular Biology of the Cell Course, Institut Pasteur, Paris, France (*Course Director*).
2. Roberto Bruzzone (2021) CMED6227 – Biological Basis of Disease (Master of Public Health), The University of Hong Kong, Hong Kong SAR (*Course Director and Lecturer*).
3. Roberto Bruzzone (2021) Introduction to the Art and Science of Medicine, Problem Based Learning (MBBS Year 1), The University of Hong Kong, Hong Kong SAR (*Tutor*).
4. Roberto Bruzzone (2021) Endocrine and Reproductive Systems, Problem Based Learning (MBBS Year 2), The University of Hong Kong, Hong Kong SAR (*Tutor*).

4) Vijay Dhanasekaran (2020) Outbreak – Problem Based Learning (MBBS Year 1), The University of Hong Kong, Hong Kong SAR (*Tutor*).

5) Vijay Dhanasekaran (2021) Introduction to the Art and Science of Medicine – Problem Based Learning (MBBS Year 1), The University of Hong Kong, Hong Kong SAR (*Tutor*).

6) Vijay Dhanasekaran (2021) Head Neck and Nervous Sytem – Problem Based Learning (MBBS Year 2), The University of Hong Kong, Hong Kong SAR (*Tutor*).

7) Vijay Dhanasekaran (2021) Biological basis of disease – Sequencing revolution: the Genetic Basis of Disease (MPH Year 1), The University of Hong Kong, Hong Kong SAR (*Lecturer*).

8) Leo Poon (2021) CMED6105 – Infectious Diseases in Public Health (Master of Public Health), The University of Hong Kong, Hong Kong SAR (*Course director and Lecturer*).

9) Leo Poon (2021) Outbreak – Problem Based Learning (MBBS Year 4), The University of Hong Kong, Hong Kong SAR (*Tutor*).

10) Leo Poon (2020) Current topics in Medical Microbiology (RPG), The University of Hong Kong, Hong Kong SAR (*Lecturer*).

11) Leo Poon (2021) Life Science (BNur Year 2 and BCMed Year 3), The University of Hong Kong, Hong Kong SAR (*Lecturer*).

12) Leo Poon (2021) Musculoskeletal System Block– Problem Based Learning (MBBS Year 2), The University of Hong Kong, Hong Kong SAR (*Tutor*).

13) Leo Poon (2021) Introduction to the Art and Science of Medicine, Problem Based Learning (MBBS Year 1), The University of Hong Kong, Hong Kong SAR (*Lecturer*).

14) Hein Min Tun (2021) CMED6227 – Biological Basis of Disease (Master of Public Health), The University of Hong Kong, Hong Kong SAR (*Course Director and Lecturer*).

15) Hein Min Tun (2021) EBDM- Evidence-Based Decision Making for Patient Care and Public Health MBBS I and II, The University of Hong Kong, Hong Kong SAR (*Tutor*)

16) Hein Min Tun (2021) Fleming Fund Fellowship Program, Laboratory Fellows from Sri Lanka (*Mentor*)

17) Hein Min Tun (2021) BBMS2011/BPHM1121- Research Methods in Medicine and Health Sciences, The University of Hong Kong, Hong Kong SAR (*Course Coordinator*).

18) Sophie Valkenburg (2021) "Biological Basis of Disease" (Master of Public Health, CMED-6227), School of Public Health, The University of Hong Kong, Hong Kong SAR (*Course Director and Lecturer*).

19) Sophie Valkenburg (2021) Outbreak – Problem Based Learning (MBBS Year 1), The University of Hong Kong, Hong Kong SAR (*Tutor*).

20) Sophie Valkenburg (2021), Health Research Project, The University of Hong Kong, Hong Kong SAR (*Tutor*).

21) Sophie Valkenburg (2021), Problem based learning tutor for MBBS students, The University of Hong Kong, Hong Kong SAR (*Tutor*).

Complete list of interns

Emma Yang	University of Chicago, USA
Uma Keomany	French International School, Hong Kong SAR
Ethan Tchitchiama	French International School, Hong Kong SAR
Wenxi Zeng	Hong Kong Institute of Vocational Education
Wing Ki Liu	Hong Kong Institute of Vocational Education
Wai Hang Chan	Hong Kong Institute of Vocational Education
Wai Yee Tsang	Hong Kong Institute of Vocational Education
Hoi Yan Sze	Hong Kong Institute of Vocational Education
Vanessa Tsang	Hong Kong Institute of Vocational Education

3.8 Other Major Activities

HKU-PRP exerts a leadership role in a number of research and educational programs of global scope.

International Severe Acute Respiratory and Emerging Infection Consortium (ISARIC)

Roberto Bruzzone was appointed as the Interim Chair in December of 2018 and became the Chair at the beginning of 2020, for a three-year term. ISARIC launched in 2011, is a consortium of over 40 clinical research networks operational in 131 countries (http://isaric.tghn.org/). ISARIC's vision is to change the approach to collaborative, patient-oriented research between and during epidemics of rapidly emerging public health threats, in order to generate new evidence-based knowledge, maximize the availability of clinical information and, thereby, save lives. ISARIC provides a collaborative platform through which global, patient-oriented clinical studies can be developed, executed and shared. In response to the COVID-19 pandemic, ISARIC networks and scientists have been involved in many important clinical studies, including two randomized clinical trials with COVID-19 patients in China. **Thanks to the efforts of many collaborators around the world the ISARIC WHO Clinical Characterisation Protocol for emerging infections is now being used to generate policy-influencing evidence, relying on this common protocol and harmonised data**. A new paper titled "The role of adaptive observational studies in global outbreak research response" is in preparation. In December 2020 **ISARIC**, the research funders group **GloPID-R** (The Global Research Collaboration for Infectious Disease Preparedness) and global group, **Long Covid Support**, **jointly organised a "Long Covid Forum"**, which aimed to gain a better understanding of 'Long Covid' and to define research priorities for funders and researchers to take forward. ISARIC and the Institut Pasteur du Madagascar have received a major grant from the Wellcome Trust to coordinate a clinical study to generate evidence for plague treatment regimens in Madagascar.

Establishment of the Center for Immunology & Infection (C2I)

We have been awarded a major 5-year grant, totaling over 40 million euros, from the Innovation and Technology Commission to establish the Center for Immunology & Infection (C2I) within the framework of InnoHK, a recent collaborative scientific research scheme set up set up by the Government of the Hong Kong Special Administrative Region. This research program, which expands the scope of the partnership between the University of Hong Kong and the Institut Pasteur will work around four major research programs to face public health challenges and making Hong Kong a global center of excellence for precision medicine population strategies. **Malik Peiris is the Managing Director of C2I and Roberto Bruzzone serves as the Co-Director**. Through C2I, we will contribute to Hong Kong's transformation into an international innovation and technology hub of the Greater Bay Area of Guangdong, Hong Kong and Macau. C2I's major focus will be the immune system, which is responsible for maintaining a healthy state and preventing infection in the majority of cases. However, dysfunction of the immune system can result in increased susceptibility to infections, inflammation, autoimmunity or even development of cancer in some individuals. Moreover, individual heterogeneity in the immune response can have an enormous impact on the likelihood to

respond to therapy or the development of side effects secondary to vaccine administration. Thus, knowledge of these parameters in healthy humans, as envisaged in this program, is essential to establish personalized and precision medical care, and disease management. This information will accelerate research to develop new effective vaccines and drug candidates, as well as inform pertinent risk assessment that would facilitate the discovery of solutions for critical issues facing Hong Kong and the world within the next decade, such as pollution, ageing of the population or pandemics. This partnership has the ambition to contribute to Hong Kong's transformation into an international innovation and technology hub of the Guangdong-Hong Kong-Macao Bay Area.

Other key actions

We retain leadership roles in a number of global projects. **Malik Peiris** continues to serve on a number of WHO working groups in relation to both avian and swine origin influenza virus and is the Co-Director of the WHO H5 Reference Laboratory at HKU. **Leo Poon** is a committee memberof the Coronavirus study group, ICTV, IUMS, and an Advisor to the Hong Kong SAR for Food and Environmental Hygiene. He is also an Ad Hoc Expert of the WHO Influenza Molecular Diagnosis Working Group, and of the WHO Expert group for COVID-19 for clinical diagnosis and virus evolution.

3.9 Knowledge Exchange

HKU-Pasteur Research Pole 20th Anniversary

We launched the celebration of its 20th Anniversary in a very challenging public health context that well illustrated the added value of international collaborations during a pandemic. The opening ceremony of the anniversary year was officiated by The Honorable Mrs Carrie Lam, the Chief Executive of the Hong Kong Special Administrative Region, and Mr Alexandre Giorgini, Consul General of France in Hong Kong and Macau. The anniversary year has been punctuated throughout 2021 by a series of events to mark two decades of outstanding partnership between the Institut Pasteur and HKUMed with an outlook to future initiatives.

20th Anniversary Exhibition "20 Years Fighting Emerging Diseases"

We organized a touring exhibition combining historical elements with present activities and future objectives of the laboratory, its parent institutions and the Pasteur Network in the context of the major global health challenges we face today. The exhibition opened on April 12 at the University of Hong Kong Centennial Campus before moving to the Hong Kong Science Park and the French International School. Visitors were guided through the history of parent institutions, the Institut Pasteur in Paris and the University of Hong Kong, before learning about the events that led to this partnership and the establishment of HKU-Pasteur. The exhibition invited visitors visitors to dive into the exciting life of Louis Pasteur, the French scientist at the origin of the Institut Pasteur, as well as the groundbreaking discovery of one of its pupils, Alexandre Yersin, who isolated the bacillus causing bubonic plague in Hong Kong in 1894. The exhibition continued with an in-depth description of ongoing research projects at HKU-Pasteur, currently focused on COVID-19, and the international courses that have made its international reputation a major training centre in biomedical research.

Dangerous liaisons: Emerging viruses at the human, animal and environmental interface

In the context of the COVDI-19 pandemic, we organized a webinar (open online to everyone), featuring researchers from diverse scientific perspectives ranging from epidemiology, virology, and entomology towards microbial ecology and medical anthropology. Gathering local and worldwide experts in the field, this workshop was a great platform to discuss the pressing topic of emerging viruses and their interactions between diverse environments and outline the next steps for research, our knowledge gaps and the ongoing challenges to apply rational scientific approaches to improve health with the successful implementation of innovative research and policy interventions. On the first day, speakers have outlined the state of research on the origin and spread of coronaviruses. The panel included Marion Koopmans (Erasmus University MC), Shi Zhengli (Wuhan Institute of Virology), Malik Peiris (HKU-Pasteur and Cneter for Immunology & Infection), Veasna Duong (Institut Pasteur du Cambodge), and Dr Huiling Yen (HKU School of Public Health) and Dr Nancy Leung (HKU School of Public Health). The second day focused on the tools needed to enhance virus surveillance, with Marco Vignuzzi (Institut Pasteur), Véronique Chevalier (Institut Pasteur du Cambodge & CIRAD)

and Anna-Bella Failloux (Institut Pasteur), whereas Frédéric Keck (French National Research Center, CNRS) and Christos Lynteris (University of St Andrews) provided and anthropological and social perspective to close the forum.

DAY 1 THE TWO SIDES OF VIRUSES: COLLECTING VIRUSES AT THE HUMAN/ANIMAL INTERFACE	
Time	Topic / Speakers
3:30	**Welcome and Presentation of the workshop** Roberto Bruzzone, Co-Director of HKU-Pasteur Research Pole
3:40	**Keynote Lecture** Moderator: Roberto Bruzzone (HKU-Pasteur) Marion Koopmans (Erasmus Medical Center, Rotterdam, The Netherlands)
4:25	**Panel Discussion – The two sides of viruses** Moderators: Vijaykrishna Dhanasekaran (HKU-Pasteur) & Tamara Giles-Vernick Institut Pasteur (Paris, France) **Process of emergence**: - Shi Zhengli Institute of Virology – Wuhan (PR China) - Malik Peiris School of Public Health @ HKUMed **Pandemic transmission** - Hui-Ling Yen School of Public Health @ HKUMed - Nancy Leung School of Public Health @ HKUMed
6:05	**Closing remarks** Vijaykrishna Dhanasekaran (HKU-Pasteur)

DAY 2 ENHANCING VIRUSES SURVEILLANCE	
Time	Topic / Speakers
4:00	**Introduction of Day 2 and Keynote Speaker** Leo Poon Co-Director of HKU-Pasteur Research Pole
4:10	**Keynote Lecture** Marco Vignuzzi Institut Pasteur (Paris, France)
4:55	**Anthropological perspective on surveillance of emerging viruses** Frédéric Keck, Laboratory for Social Anthropology – CNRS (Paris, France)
5:15	**Panel Discussion – Enhancing viruses surveillance** Moderators: Frédéric Keck, CNRS & Chris Mok HKU-Pasteur Research Pole - Véronique Chevalier, Institut Pasteur in Cambodia & CIRAD - Anna-Bella Failloux Institut Pasteur (Paris, France) - Christos Lynteris, University of St Andrews (UK)
6:45	**Closing remarks** Roberto Bruzzone, HKU-Pasteur Research Pole

Joint events with the French International School

We have co-organized with the French International School several events that enabled students to discover the world of biomedical research and the impact of science in driving solutions to save lives and support public health policies. This educational series included a visit of the HKU-Pasteur laboratories and interviews organized by the students of some of our Group Leaders, Sophie Valkenburg and) Vijay Dhanasekaran, and the Pole's Co-

Director, Leo Poon. The students produced 20 videos on diverse topics from the origin and evolution of viruses and the future of Covid-19 pandemic to vaccination and testing explanations. A roundtable featuring Ben Cowling (epidemiologist at HKU) and Sophie Valkenburg, animated by students, focused on the importance of scientific research in the Covid-19 pandemic and the impact on our lives. Finally, during two weeks In June and July, two students from the French International School completed their high school internship at the HKU-Pasteur Research Pole, getting a first-hand taste of the daily life of a research team.

Unseen Enemy: Film Screening and Public Debate at the Asia Society Hong Kong

In the midst of its 20th Anniversary Year Celebrations (July 2, 2021), HKU-Pasteur Research Pole teamed up with the Asia Society Hong Kong to present the screening of a film by award-winning director Janet Tobias: Unseen Enemy. The film focuses on three case studies: Ebola, influenza, and Zika - all local outbreaks that became widespread epidemics, exploring together with some of the world's leading experts in public health how many people's lives are deeply changed by the impact of the epidemic. From the Ebola epidemic in West Africa to the Zika outbreak in Brazil, the film investigates the reason why a virus can spread and cause a pandemic. The screening was followed by a 30-minute Q&A session chaired by Professor Lau Chak Sing, Professor at the HKU LKS Faculty of Medicine, with the participation of Leo Poon, Co-Director of HKU-Pasteur Research Pole and Dr Hui-ling Yen, Associate Professor in the HKU School of Public Health, who discussed the insights and lessons learned from the past and current global pandemics and answered questions from the public.

Joint Symposium in June 2021

From HKU-Pasteur to C2i: Confronting the challenges of infectious diseases

HKU-Pasteur and the Centre for Immunology & Infection (C2i) organized a webinar as part of the 20th Anniversary celebrations, in partnership with the Hong Kong Science and Technology Parks Corporation, to highlight the scientific journey that has led, from the creation of HKU-Pasteur in 2000, to the establishment of C2i. This long-standing collaboration naturally led the way towards a new translational research venture, C2i, which will contribute to Hong Kong's transformation into an international hub of biotechnological innovation in the Guangdong-Hong Kong-Macau Greater Bay Area. In this webinar, principal investigators of C2i and HKU-Pasteur Research Pole discussed their latest research development of new technological platforms for biomarker discovery and of new vaccine and therapeutic strategies.

16:00 – 16:05	**Welcoming Remarks: Oscar Wong** *Head of Business Development, Corporate Development Division, Hong Kong Science & Technology Parks Corporation*
16:05 – 16:10	**Opening Remarks: Isabelle Buckle** *Executive Vice-President for Technology Transfer, and Industrial Partnerships of the Institut Pasteur*
16:10 – 16:30	**Introduction: Roberto Bruzzone** *Co-Director, HKU-Pasteur Research Pole & C2i*

16:30 – 17:00	**From Milieu Interieur to Healthy Human Global Project in Hong Kong: Understanding healthy human diversity: Darragh Duffy** *Head of Translational Immunology, Institut Pasteur, Principal Investigator, C2i*
17:00 – 17:30	**From HKU-Pasteur to C2i: The importance of universal vaccines and pathogenesis studies: Sophie Valkenburg** *Assistant Professor, HKU-Pasteur Research Pole, Co-Investigator, C2i*

20th Anniversary Closing Workshop: Know Thy Microbes

To conclude the celebrations of its 20th Anniversary**,** HKU-Pasteur Research Pole organized a workshop on 21 October 2021. This event gathered local and worldwide public health experts to discuss how our relationship to the microbial world is being influenced by the current pandemic, and outline next steps for researchers and policy makers in the field. The seminar featured researchers from diverse scientific perspectives who addressed the challenges to frame human cohabitation with microbes in the context of the ongoing Covid-19 pandemic, which has led to increased surveillance and travel restrictions.

Time	Topic / Speakers
15:00	**Welcome address by Roberto Bruzzone**, Co-Director of HKU-Pasteur Research Pole
15:15	**Keynote Lecture "You'll Love Your Microbes Like Yourself"** Speaker: Philippe Sansonetti, Institut Pasteur Paris and Center for Microbes in Development and Health, Institut Pasteur of Shanghai – Chinese Academy of Sciences Introduced by: Keiji Fukuda, Director, HKU School of Public Health
15:45	**Panel Discussion** Moderator: Keiji Fukuda, Director, HKU School of Public Health Panelists - Hein Min Tun, HKU-Pasteur Research Pole - Sophie Valkenburg, HKU-Pasteur Research Pole - Tommy Lam, School of Public Health and Centre for Immunology & Infection - Zhiwei Chen, Department of Microbiology, HKU LKS Faculty of Medicine
16:15	***Break***
16:30	**Keynote Lecture – "Traveling with the Unseen"** Speaker: Annelies Wilder-Smith, London School of Hygiene and Tropical Medicine, Nanyang Technological University, Singapore and Heidelberg Institute of Global Health, University of Heidelberg, Germany Introduced by: Leo Poon, Co-Director of HKU-Pasteur Research Pole
17:00	**Panel Discussion** Moderator: Leo Poon, Co-Director of HKU-Pasteur Research Pole Speakers - Hui-ling Yen, HKU School of Public Health - Ben Cowling, HKU School of Public Health - Vijay Dhanasekaran, HKU-Pasteur Research Pole
17:30	**Closing remarks** Leo Poon, HKU-Pasteur Research Pole

The HKU-Pasteur Research Pole Fellowship Program

HKU-Pasteur, together with the Pasteur Foundation Asia and the Consulate of France in Hong Kong and Macau, launched a new initiative providing a unique opportunity for postgraduate students and postdoctoral fellows in Hong Kong and Macau to pursue their research projects in the outstanding scientific environment of the Institut Pasteur in Paris, France. A similar program had been conducted between 2012 and 2015 and proven to be very useful for Hong Kong-France scientific cooperation and the professional path of the laureates. The scholarship includes round-trip air tickets between Hong Kong/Macau and France, housing, monthly living allowance and coverage of Covid-19 pandemic related expenses. For the first edition of the HKU-Pasteur Research Pole, we received several strong applications, despite the uncertainty related to travel restrictions. The three laureates were:

- **Mr Ye Peng**, PhD at HKU-Pasteur, worked in the Biological Image Analysis Laboratory under the supervision of **Dr Elisabeth Labruyère to study gut microbiome using an organ-on-chip approach;**
- **Dr Simon Chi-Chin Shiu**, post-doctoral student in the School of Biomedical Sciences of the LKS Faculty of Medicine of the University of Hong Kong, carried out his internship in the Laboratory of Bio-Organic Chemistry of Nucleic Acids under the supervision of **Dr Marc Hollenstein, to study innovative drug delivery approaches using nanostructures.**
- **Mr Terence Tak Wang Lee**, a PhD student in the School of Biomedical Sciences of the LKS Faculty of Medicine of the University of Hong Kong, carried out research under the supervision of **Dr Thomas Wollert in the Membrane and Transport Biochemistry Laboratory, to characterize new strategies to reduce the severity of Influenza A virus infection**.

Given the resounding success of the first edition, all sponsors pledge to extend their support for 2022, awarding fellowships to four students and allowing them to extend their stay at the Institut Pasteur for up to four months.

Major Published Research News

HKU-Pasteur has proven to be an important actor in facing the SARS-CoV-2 pandemic in Hong Kong. Therefore, multiple discoveries have been highlighted in local and international media such as a multidisciplinary project for detecting SARS-CoV 2 in sewage waters, **involving Dr Hein Min Tun's team, which has led to improvement in the surveillance of outbreaks in local communities and potential clusters**. Sponsored by the Health and Medical Research Fund (HMRF) under the Food and Health Bureau, this project allowed the collection of domestic sewage samples from sewage collection systems in different areas for nucleic acid tests of the new coronavirus SARS-CoV-2. The team, in collaboration with other departments of the University of Hong Kong, has been able to demonstrate that sewage surveillance could provide early warning of COVID-19 outbreaks, reflecting the overall spread of virus in the community. It also helps tracking the development trend of community outbreak. **The significance of these results for public health prompted the Chief Executive Carrie Lam to visit HKU-Pasteur** and praise its work, underlining the importance of multi-disciplinary approaches to mitigate the impact of infectious diseases.

4. Scientific Output

4.1 Publications

1. Au KK, Chen C, Chan YM, Wong WWS, Lv H, Mok CKP, Chow CK (2021) Tracking the Transcription Kinetic of SARS-CoV-2 in Human Cells by Reverse Transcription-Droplet Digital PCR. *Pathogens* **10**:1274.

2. Behzadinasab S, Chin AWH, Hosseini M, Poon LLM, Ducker WA (2021) SARS-CoV-2 virus transfers to skin through contact with contaminated solids. *Sci Rep* 11:22868.

3. Behzadinasab S, Williams MD, Hosseini M, Poon LLM, Chin AWH, Falkinham JO 3rd, Ducker WA (2021) Transparent and Sprayable Surface Coatings that Kill Drug-Resistant Bacteria Within Minutes and Inactivate SARS-CoV-2 Virus. *ACS Appl Mater Interfaces* **13**:54706–54714.

4. Bull MB, Cohen CA, Leung NHL, Valkenburg SA (2021) Universally Immune: How Infection Permissive Next Generation Influenza Vaccines May Affect Population Immunity and Viral Spread. *Viruses* **13**:1779.

5. Burckhardt RM, Dennehy JJ, Poon LLM, Saif LJ, Enquist LW (2021) Are COVID-19 Vaccine Boosters Needed? The Science behind Boosters. *J Virol*, in press.

6. Carson G; Long Covid Forum Group* (2021) Research priorities for Long Covid: refined through an international multi-stakeholder forum. *BMC Med* **19**:84.
*Gail Carson, Louise Sigfrid, Piero Olliaro, Alice Norton, Giuseppe Paparella, Romans Matulevics, Annelies Gillesen, Peter Horby, Claire Hastie, Margaret O'Hara, Jake Suett, Sarah Moore, Richard Vaux, Jean Marie Habarugira, Genevieve Boily-Larouche, Ella Clark, Peter Hart, Josie Golding, Claire Madelaine, Brian Mackenwells, Kristina Ruze, Nina Jamieson, Liliana Resende, Evelyn Depoortere, Anna Kinsey, Daniel Munblit, Fahmy Hanna, Janet Diaz, Annelies Wilder-Smith, Charles Wiysonge, Charu Kaushic, Luis Felipe Reyes, Roberto Bruzzone, Yazdan Yazdanpanah, Tedros A Ghebreyesus, Alex van Blydenstein, Alpha Keita, Andre Siqueira, Angela Cheung, Charitini Stavropoulou, Frances Simpson, Janet Scott, Joseph Breen, Joseph Fokam, Ken Baillie, M Netravathi, Margaret Herridge, Nisreen Alwan, Priscilla Rupali, Ryan Zarychanski, Seong-Ho Choi, Lwazi Mlaba, Moses Badio, Bhasha Mewar, Gina Assaf, Christopher Brightling, Danny Altmann, Fernando Bozza, Ivan Hung, Norio Ohmagari, Sally Singh, Shinichiro Morioka, Simon Hatcher, Simone Piva.

7. Chan OSK, Bradley KCF, Grioni A, Lau SKP, Li WT, Magouras I, Naing T, Padula A, To EMW, Tun HM, Tutt C, Woo PCY, Bloch R, Mauroo NF (2021) Veterinary Experiences can Inform One Health Strategies for Animal Coronaviruses. *Ecohealth* **20**:1-14.

8. Chan OSK, Naing T, Tun HM (2021) Upholding veterinary services as a pillar of one health in Myanmar. *One Health* **13**:100329..

9. Chen T, Qin Y, Chen M, Zhang Y, Wang X, Dong T, Chen G, Sun X, Lu T, White RA 3rd, Ye P, Tun HM, Xia Y (2021) Gestational diabetes mellitus is associated with the neonatal gut microbiota and metabolome. *BMC Med* **19**:120.

10. Chen YY, Zhao X, Moeder W, Tun HM, Simons E, Mandhane PJ, Moraes TJ, Turvey SE, Subbarao P, Scott JA, Kozyrskyj AL (2021) Impact of Maternal Intrapartum Antibiotics, and Caesarean Section with and without Labour on Bifidobacterium and Other Infant Gut Microbiota. *Microorganisms* **9**:1847.

11. Chu DKW, Gu H, Chang LDJ, Cheuk SSY, Gurung S, Krishnan P, Ng DYM, Liu GYZ, Wan CKC, Tsang DNC, Peiris M, Poon LLM (2021) SARS-CoV-2 Superspread in Fitness Center, Hong Kong, China, March 2021. *Emerg Infect Dis* **27**:2230-2232.

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18. Hosseini M, Behzadinasab S, Chin AWH, Poon LLM, Ducker WA (2021) Reduction of Infectivity of SARS-CoV-2 by Zinc Oxide Coatings. *ACS Biomater Sci Eng* **7**:5022-5027.

19. Hosseini M, Chin AWH, Behzadinasab S, Poon LLM, Ducker WA (2021) Cupric Oxide Coating That Rapidly Reduces Infection by SARS-CoV-2 via Solids. *ACS Appl Mater Interfaces* **13**:5919-5928.

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4.2 Presentations at Meetings

1. Sophie Valkenburg (2021) Australasian Society for Immunology annual meeting, December (*Oral*)
2. Sophie Valkenburg (2021) Australian Influenza Symposium (*Oral*)
3. Sophie Valkenburg (2021) ISIRV WHO COVID influenza RSV (*Poster*; *virtual event*)

4.3 Seminars and Invited Presentations

1. Roberto Bruzzone (2021) ISARIC Member Assembly: Clinical Research Networks Addressing COVID-19 & Future Challenges, Fondation Mérieux, Annecy, France.
2. Leo Poon (2021) "SARS-CoV-2 and its transmission" in the annual meeting of Centers of Excellence for Influenza Research and Surveillance organized by NIH (webinar).
3. Leo Poon (2021) "SARS-CoV-2 and its transmission", organized by NIH, USA (*webinar*).
4. Leo Poon (2021) "SARS-CoV-2 and its infection", organized by Centre for Oncology and Immunology, InnoHK (*seminar*).
5. Leo Poon (2021) "SARS-CoV-2 and its infection: from science to public health" in World Science Culture Forum (*webinar*).
6. Leo Poon (2021) "Molecular epidemiological study of COVID-19 cases in Hong Kong" organized by Food and Health Bureau, Hong Kong Government. (*seminar*).
7. Leo Poon (2022) "Molecular epidemiological study of COVID-19 cases in Hong Kong" organized by Pasteur, Paris (*webinar*).
8. Leo Poon (2021) "COVID-19 vaccine", School of Public Health, The University of Hong Kong (webinar).
9. Leo Poon (2021) "Influenza and Coronavirus Infections in Hong Kong", HKU-Space (webinar).
10. Hein Min Tun (2021) International Conference on Current Problems of Biological Safety in the Modern Conditions. Kazakhstan (*webinar*).
11. Hein Min Tun (2021) 26th Medical Research Conference, Department of Medicine, University of Hong Kong (*seminar*).
12. Sophie Valkenburg (2021) Chinese University of Hong Kong, Hong Kong SAR (webinar)
13. Sophie Valkenburg (2021) Peter Doherty Institute, Melbourne, Australia (webinar).
14. Sophie Valkenburg (2021) *From HKU-Pasteur to C2i: Confronting the challenges of infectious diseases* C2i, HKSTP, Hong Kong SAR.
15. Sophie Valkenburg (2021) Dangerous liaisons: Emerging viruses at the human, animal and environmental interface (webinar).

5. Annexes

5.1 List of Staff

Name	Position
Malik Peiris	Honorary Director
Roberto Bruzzone	Co-Director
Leo Poon	Co-Director
Vijaykrishna Dhanasekaran	Associate Professor
Hein Min Tun	Assistant Professor
Sooksan Wong	Assistant Professor
Sophie Valkenburg	Assistant Professor (ended December 2021)
Chris Mok	Research Assistant Professor (ended April 2021)
Anne Li	Administration Manager
Jimmy Lai	Laboratory Manager (ended December 2021)
Wendy Yu	Laboratory Manager
Mathilde Boisserin	Communication Officer (ended Jan-2022)
Karen Chan	Executive Assistant (ended March-2021)
Dilys Li	Executive Assistant
Christine Wong	Executive Assistant
Ka Pui Ng	IT Technician (ended August-2021)
Prathanporn Kaewpreedee	Post-doctoral Fellow
Yun Lan	Postdoctoral Fellow (ended March 2021)
Xiaoman Wei	Post-doctoral Fellow
Xi Zhang	Post-doctoral Fellow
Darren Chan	PhD Student (Non-study leave till 31-July-2022)
Maireid Bull	PhD Student (ended January-2021) Research Associate (ended September-2021)
Carolyn Cohen	PhD Student
Athena Li	PhD Student (ended May-2021)
Suisha Liang	PhD Student
Tomas Lyu	PhD Student
Ye Peng	PhD Student
Ray So	PhD Student (ended August-2021)

Qiwen Teo	PhD Student (ended August-2021)
Hogan Wai	Research Assistant II (ended August-2021) PhD Student
Ho Him Wong	PhD Student (ended August-2021)
Sonia Younas	PhD Student
Xiawan Zheng	PhD Student – Department of Civil Engineering, HKU
Shuxian Li	PhD Student – Department of Civil Engineering, HKU
Yulin Zhang	PhD Student – Department of Civil Engineering, HKU
Shreya Gurung	Mphil Student
Weiwen Liang	Mphil Student
Ruopeng Xie	Research Assistant I (ended August-2021) Mphil student
Lewis Siu	Senior Technician
Kimberly Edwards	Senior Research Assistant
Janice Jia	Research Assistant I
Yihan Lin	MPhil student (ended December-2021) Research Assistant I
Shilin Zhao	Research Assistant I
Yang Zhou	Research Assistant I
Jie Zhu	Research Assistant I
Wilson Ng	Research Assistant I (ended February-2021)
Garrick Yip	Research Assistant I (ended February-2021)
Dengwei Zhang	Research Assistant I (ended June-2021)
Fionn Ma	Research Assistant I (ended November-2021)
Felice Pak	Research Assistant I (ended January-2022)
Gary Chan	Research Assistant II
Ingrid Chan	Research Assistant II
Alan Cheung	Research Assistant II
Xin Liu	Research Assistant II
Rista Shrestha	Research Assistant II
Daniel Sin	Research Assistant II
Zi Xi Dai	Research Assistant II (ended September-2021)

Asmaa Hachim	Research Assistant II (ended December-2021)
Jinlin Wang	Research Assistant II (ended March-2021)
Alex Wong	Research Assistant II (ended September-2021)
Luke Wong	Research Assistant II (ended January-2021)
Cheng Xiao	Research Assistant (starts June-2022)
Thomas Chu	Research Assistant II (P/T)
Chloe Liu	Research Assistant II (P/T)
Hilda On	Research Assistant II (P/T)
Suet Ying Ng	Research Assistant II (P/T) (ended August-2021)
Anxin Pan	Research Assistant – Department of Civil Engineering, HKU
Xianghui Shi	Research Assistant – Department of Civil Engineering, HKU
Mengying Wang	Research Assistant – Department of Civil Engineering, HKU
Jieying Leung	Laboratory Attendant
Emma Yang	Student Intern, University of Chicago, USA
Uma Keomany	Student Intern, French International School, Hong Kong SAR
Ethan Tchitchiama	Student Intern, French International School, Hong Kong SAR
Wenxi Zeng	Student Intern, Hong Kong Institute of Vocational Education
Wing Ki Liu	Student Intern, Hong Kong Institute of Vocational Education
Wai Hang Chan	Student Intern, Hong Kong Institute of Vocational Education
Wai Yee Tsang	Student Intern, Hong Kong Institute of Vocational Education
Hoi Yan Sze	Student Intern, Hong Kong Institute of Vocational Education
Vanessa Tsang	Student Intern, Hong Kong Institute of Vocational Education

Visiting and Honorary Appointments

Name	Position	Apointment End date
James Di Santo	Visiting Research Professor	31-December-2022
Sumana Sanyal	Visiting Associate Professor	31-December-2021
Noël Tordo	Honorary Professor	31-December-2023
Jincun Zhao	Honorary Professor	31-December-2022
Ralf Altmeyer	Honorary Associate Professor	5-November-2022
Chris Mok	Honorary Assistant Professor	30-April-2023
Sophie Valkenburg	Honorary Assistant Professor	10-November-2023
Iris Wai Sum Iris Li	Honorary Clinical Assistant Professor	30-June-2022
Barbara Gayraud-Morel	Honorary Research Associate	31-December-2023
Niloufar Kavian	Honorary Research Associate	31-December-2022
Yun Lan	Honorary Research Associate	30-April-2023
Qiwen Teo	Honorary Research Associate	31-October-2022
Simon Muller	Honorary Tutor	31-December-2023

5.2 Income & Expenses for the year ending June 2021 (Period: 1 July 2020 to 30 June 2021)

INCOME:

Central Fund	$ 3,000,000.00	17.77 %
Faculty in-kind	$ 1,875,000.00	11.11 %
Institut Pasteur	$ 2,962,345.49	17.55 %
Private Donation	$ 0.00	0.00 %
External Grants**	**$ 9,040,302.85**	**53.56 %**
Teaching/Training	$ 0.00	0.0 %
	$ 16,877,648.34	**100.00%**

EXPENSES:

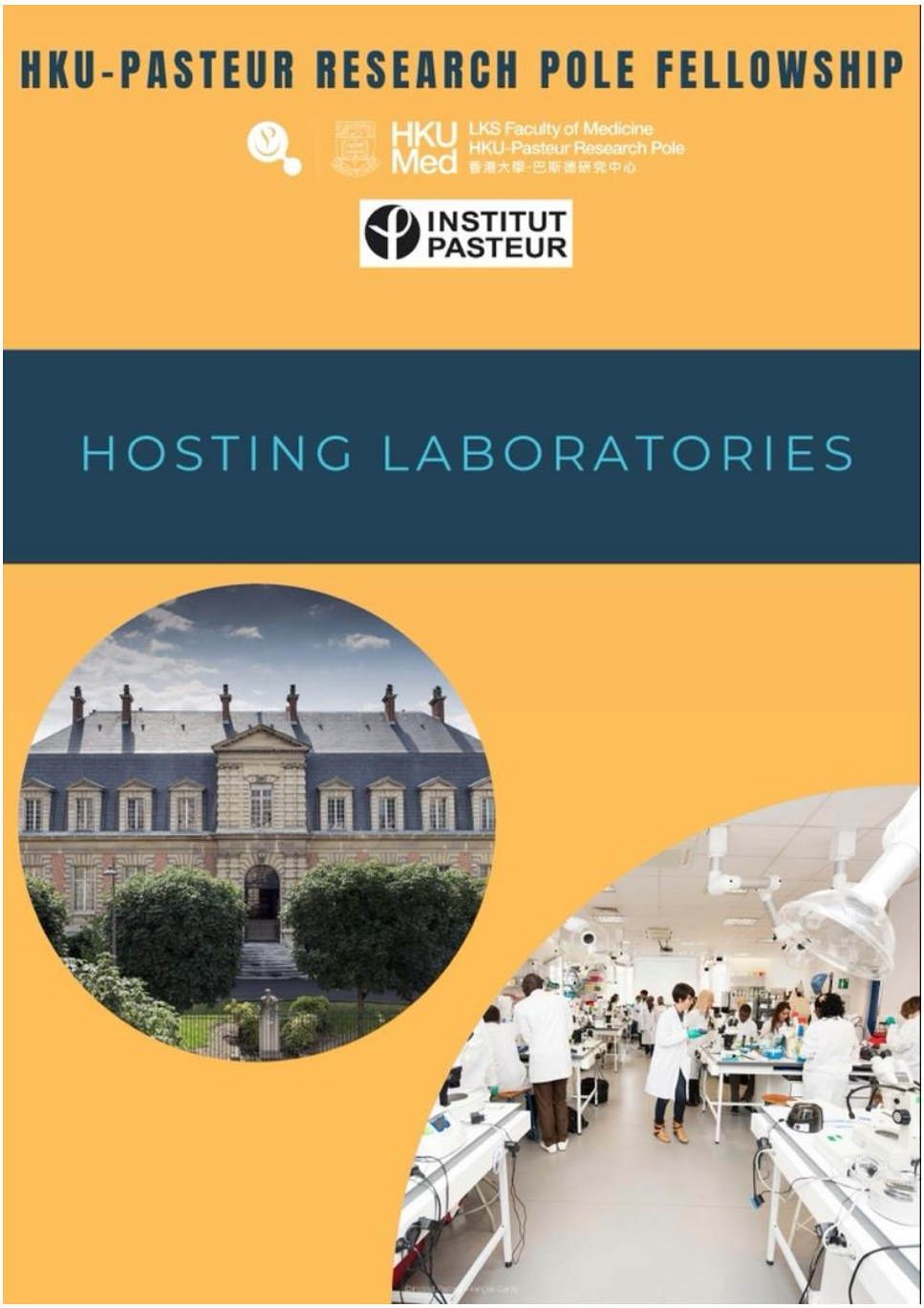
Staff cost	$ 8,025,153.80	47.73 %
Stipend	$ 1,061,439.29	6.31 %
Research / Equipment / Maintenance	$ 7,373,163.55	43.86 %
Conference / Meeting	$83,898.93	0.50 %
Administration	$ 142,137.01	0.85 %
Teaching/Training	$ 126,463.87	0.75 %
TOTAL	**$ 16,812,256.45**	**100.00%**

BALANCE CARRY FORWARD TO 2021/2022 **$ 65,391.89**

5.4 Dangerous Liaisons: Emerging viruses at the human, animal and environmental interface

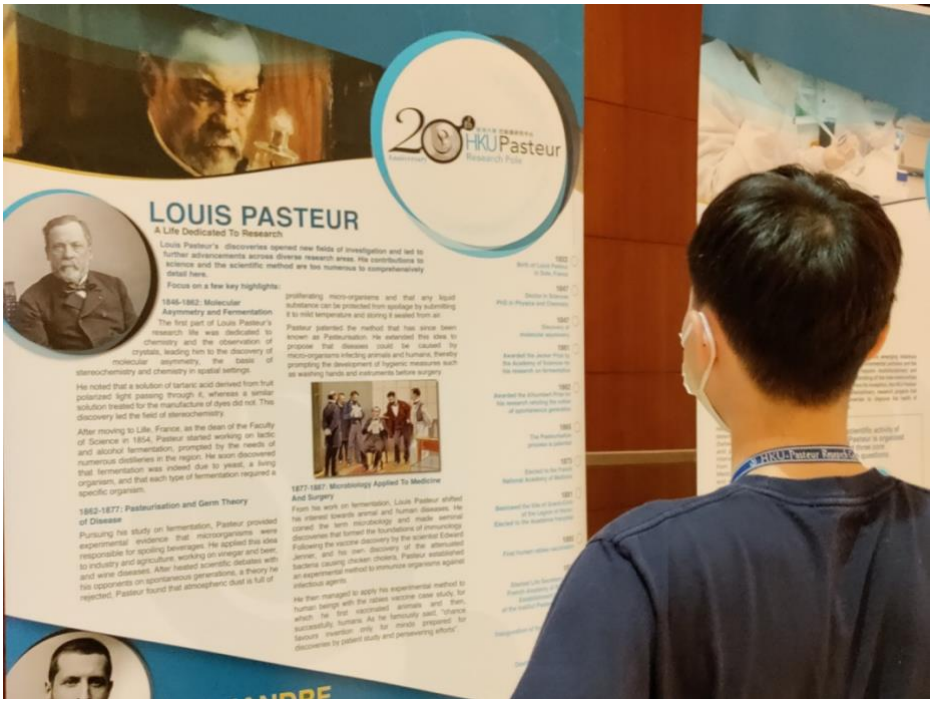
5.5 HKU-Pasteur Research Pole Fellowship

Hosting Laboratories Booklet:
http://www.hkupasteur.hku.hk/news/hkuprp_fellowship_booklet_maj250221.pdf



5.6 HKU-Pasteur 20th Anniversary Exhibition

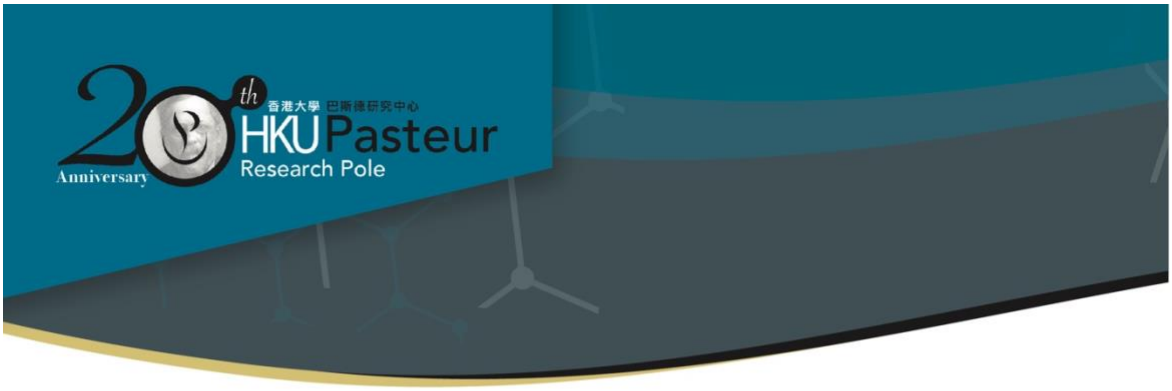
The University of Hong Kong Centennial Campus:



Hong Kong Science Park:

The University of Hong Kong campus of Medicine:

5.7 20th Anniversary Program



20th Anniversary Program

21 Oct 2020	Opening Ceremony
Dec 2020	Book Publication
Jan 2021	HKU-Pasteur Fellowship Program launch
24-25 Feb 2021	Dangerous Liaisons Worksop
23 Mar 2021	French International School in Hong Kong (FIS) visiting the HKU-Pasteur Research Pole
Apr 2021	Publication of a series of videos by FIS and HKU-Pasteur
12-26 Apr 2021	HKU-Pasteur 20th Anniversary Exhibition at HKU Centennial Campus
30 Apr 2021	Event at the French International School in Hong Kong
7-18 Jun 2021	HKU-Pasteur 20th Anniversary Exhibition at the Hong Kong Science Park
11 Jun 2021	Movie screening and Q&A at Asia Society Hong Kong
Sept-Oct 2021	HKU-Pasteur 20th Anniversary Exhibition at HKUMed Campus
Sept-Oct 2021	HKU-Pasteur 20th Anniversary Exhibition at the FIS

5.8 C2i symposium June 2021

5.9 HKU-PRP 20th Anniversary Closing symposium

"KNOW THY MICROBES – Living With the Unseen"

5.10 Inauguration of C2i

5.11 Events organized with the French International School (FIS)

Visit of HKU-Pasteur and interview of Professor Leo Poon by the FIS Students:

Workshop with Doctor Vijaykrishna Dhanasekaran:

Meet Dr. Vijaykrishna DHANASEKARAN - HKU Pasteur Research Pole

Roundtable with the FIS Students and HKU-Pasteur researchers:

5.12 Unseen Enemy Film Screening with Asia Society Hong Kong

LKS Faculty of Medicine
HKU-Pasteur Research Pole
香港大學-巴斯德研究中心

Annual Report 2021

Roberto Bruzzone, Co-Director
Leo Poon, Co-Director
Malik Peiris, Honorary Director

HKU-Pasteur Research Pole
7/F Jockey Club Building for Interdisciplinary Research
5, Sassoon Road, Hong Kong SAR