Influenza Updates

The newsletter of the WHO Collaborating Centre for Reference and Research on Influenza in Melbourne

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Thank you and happy holidays

As 2016 draws to a close we would like express our sincere thanks to all the laboratories who sent us influenza samples during the past year. We wish you all the best for the holiday season and look forward to working with you again in 2017.

Introducing our new Director

We are pleased to introduce Dr Kanta Subbarao as the new Director for the WHO Collaborating Centre for Reference and Research on Influenza in Melbourne. Kanta was previously at the National Institutes of Health in Bethesda MD, USA where she was a Senior Investigator and Chief of the Emerging Respiratory Viruses Section, Laboratory of Infectious Diseases, National Institute of Allergy and Infectious Diseases (NIAID). Her research interests focus on the pathogenesis of influenza and influenza vaccinology, especially pandemic influenza, as well as studies of other emerging respiratory viruses. Having worked extensively in both influenza public health and research for many years, Kanta is looking forward to combining her interests as part of her new role at the Centre. We warmly welcome Kanta and hope that many of you will have an opportunity to meet her in the coming year.

We also thank Prof Ian Barr who has been Acting Director for his stewardship of the Centre for most of 2015 and 2016. Ian has resumed his role as Deputy Director of the Centre.

WHO Shipping Fund Project reminder

In anticipation of the WHO Consultation on the Composition of Influenza Vaccines for the Northern Hemisphere 2017-2018, which will be held in February 2017, this is a friendly reminder that the WHO Shipping Fund Project (SFP) is available to assist National Influenza Centres in shipping samples to WHO Collaborating Centres up to three times per year. The recommended timing of the three shipments is: one between the end of December to mid-January and one between the end of June and mid-August, to support the WHO vaccine composition recommendation-making for each hemisphere; the third shipment can be used at your own judgement.

We encourage you to send samples in a timely manner as soon as possible after collection. Please avoid sending your samples in large batches collected over long periods, as up-to-date data for the current season are the most useful for WHO GISRS surveillance and vaccine formulation.

If you have any questions about shipping samples or would like information about accessing the WHO Shipping Fund, please contact us at *whoflu@influenzacentre.org*.

External Quality Assurance Project (EQAP) for Viral Isolation

The Centre has undertaken the development of an external quality assurance project (EQAP) to test proficiency of virus isolation techniques in National Influenza Centres (NICs) in the WHO Western Pacific Region (WPR) and South-East Asia Region (SEAR). The Centre has prepared a panel of 16 test influenza samples which were validated by real-time RT-PCR and isolation both at our Centre and WHO Collaborating Centres for Influenza in Japan and China.

A total of 21 NICs — 14 in WPR and 7 in SEAR — are participating in the EQAP and received the panel between September and October of this year. To date, 14 NICs have completed and returned their results. We hope to receive the remaining 7 before the end of the year and encourage those laboratories who are yet to return their results to do so. We will inform all participating NICs of the provisional results and identity of the 16 samples as soon as all results have been received. This will be followed by more detailed analysis of results and the completed questionnaires in 2017.

We thank all of the laboratories who are participating in this EQAP - the information provided through your results and feedback will help us to assess the capacity of NICs in our region to isolate and accurately identify influenza viruses. This will assist us to organise and provide appropriate training opportunities to best support your surveillance activities, which will in turn strengthen the influenza surveillance network in our region and around the world.

Visiting scientists

We were pleased to welcome Ms Phally Vy (*left*), from the National Public Health Laboratory, Phnom Penh, Cambodia; and Ms Kimlay Chea (*centre*) and Ms Sokhoun Yann (*right*), from the Institut Pasteur, Phnom Penh, Cambodia, who visited the Centre from 21 November to 2 December.

Ms Vy, Ms Chea and Ms Yann undertook training in various techniques related to detection and characterisation of seasonal influenza viruses, including:

- sequencing of influenza genes and phylogenetic analysis
- viral isolation in MDCK cells and serological analysis of isolates
- egg inoculation and harvest for growth of influenza viruses
- analysis and management of surveillance data.

Ms Vy said she found great value in the training as her laboratory is working towards designation as a WHO National Influenza Centre and she was now familiar with a number of new techniques, some of which will ultimately be introduced into her laboratory. Ms Chea commented that she was very pleased to diversify her skill set and felt this background would assist with her goal of rotating between different divisions of her current laboratory.

Visit to the National Influenza Centre in Fiji

The Centre's Educator, Prof Patrick Reading visited the **National Influenza Centre at the Fiji Centre for Communicable Disease Control,** Suva, Fiji in November to facilitate discussions regarding the implementation of cell culture and virus isolation. Plans were developed to modify existing laboratories and obtain appropriate equipment and reagents, with the aim of establishing cell culture and virus isolation techniques in 2017. Practical training in real-time RT-PCR was also provided to new staff members at the National Influenza Centre.

Training workshops at regional laboratories

Patrick Reading has also participated in the organisation, facilitation and delivery of two training activities at regional laboratories in recent months.

The Influenza-like Illness Surveillance Workshop was held at the Institute of Medical Research in Goroka, Papua New Guinea, on 24–26 October. Approximately 20 participants attended the workshop, with representatives from a number of regions in Papua New Guinea to plan implementing enhanced surveillance for human and avian influenza.

Image courtesy of Berry Ropa, National Department of Health, Papua New Guinea

The Introduction to Laboratory Quality Management and Good Laboratory Practice workshop was held at the National Institute of Health Research and Development (NIHRD), in Jakarta, Indonesia on 21–25 November. The workshop was attended by 24 laboratory managers and technicians from laboratories from across different provinces in Indonesia that are involved in laboratory testing for influenza virus. The workshop involved a mixture of lectures and practical exercises to strengthen principles of Laboratory Quality Management and Good Laboratory Practices.

Recent activity at the Centre (1 October — 30 November 2016)

Following is a summary of surveillance activities at the Centre from 1 October to 30 November. Concurrent with the end of the Southern Hemisphere winter and influenza season we have observed a steady reduction in the number of samples received since September.

Samples received

The Centre received 737 influenza samples from the laboratories and institutions listed below during the period 1 October—30 November, 2016.

AUSTRALIA: Canberra Hospital, Royal Darwin Hospital, John Hunter Hospital, Prince of Wales Hospital, Queensland Health Forensic and Scientific Services, SA Pathology, North West Pathology, Royal Hobart Hospital, Alfred Hospital, Monash Medical Centre, Dorevitch Pathology, VIDRL

MALAYSIA: Institute for Medical Research

SOLOMON ISLANDS: National Referral Hospital

Recent activity at the Centre (1 Oct — 30 Nov 2016, continued)

	Antigenic analysis: A total of 362 influenza isolates were analysed by HI assay.				**Neuraminidase inhibitor susceptibility:** A total of 509 influenza isolates were tested by neuraminidase inhibition (NAI) assay for susceptibility to oseltamivir, zanamivir, peramivir and laninamivr.			
Country of submitting laboratory	**No. of viruses analysed by HI assay***				**No. of viruses tested by NAI assay***			
	A(H1N1) pdm09	A(H3N2)	B/ Victoria	B/ Yamagata	A(H1N1) pdm09	A(H3N2)	B/Victoria	B/Yamagata
Australia	62	197		25	68	332	6	22
Malaysia	9	1	47	9	9	3	59	9
New Zealand				4		1		
Singapore		2		5				
Vietnam		1						
Total	**71**	**201**	**47**	**43**	**77**	**336**	**65**	**31**

* *Subtypes and lineages are based on analysis of the HA and in some cases confirmed by genetic analysis of NA.*

Genetic analysis: Sanger sequencing was performed on 55 HA, 56 NA, 37 MP and 15 NS genes from 56 viruses. Next Generation Sequencing (NGS) techniques were used to sequenced the HA, NA and MP genes of an additional 71 viruses and the full genomes of 53 influenza A viruses. In total, 145 sequences from 63 human viruses received for surveillance purposes were deposited with the GISAID EpiFlu™ database (http://www.gisaid.org).

Country of submitting laboratory	**No. of viruses with individual genes (HA/NA/ MP/NS) analysed by Sanger sequencing or NGS techniques**				**No. of viruses with full genomes analysed using NGS techniques**	
	A(H1N1) pdm09	A(H3N2)	B/Vic	B/Yam	A(H1N1)pdm09	A(H3N2)
Australia	10	96	4	7	5	3
Cambodia					38	
Fiji					1	1
Malaysia			2			
New Caledonia					2	
New Zealand		2	1	1		2
Singapore		1				
Sri Lanka					1	
Vietnam		3				
Total	**10**	**102**	**7**	**8**	**47**	**6**

Isolation of viruses in eggs

The Centre undertakes primary isolation of selected viruses in eggs to obtain potential vaccine strains. From 1 October to 30 November 2016, 2 A(H3N2) and 4 B/Yamagata viruses were successfully isolated in eggs at the Centre.

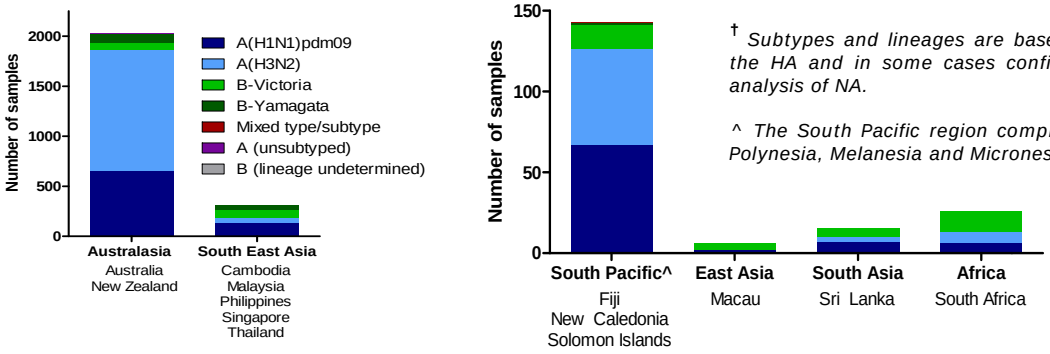
Surveillance update: Virus activity 1 January—30 November 2016

The data below are results for viruses collected between 1 January and 30 November 2016 that have been analysed at the Centre as of 6 December 2016.

Virus types / subtypes†

The type and subtype/lineage of 2525 viruses have been determined. The predominant type/subtype amongst viruses analysed to date was A(H3N2) (52.1%).

Figure: Types and subtypes/lineages of viruses collected 1 Jan—30 Sept 2016, by world region



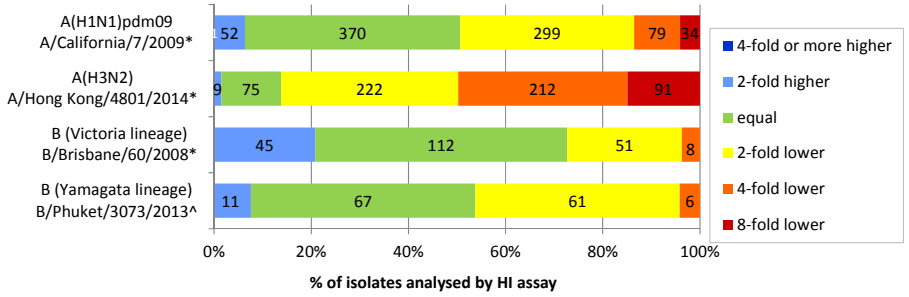
† *Subtypes and lineages are based on analysis of the HA and in some cases confirmed by genetic analysis of NA.*

^ *The South Pacific region comprises countries in Polynesia, Melanesia and Micronesia.*

Antigenic analysis

Haemagglutination inhibition (HI) assays indicate that with the majority of isolates were antigenically similar to the 2016 Southern Hemisphere and 2016-2017 Northern Hemisphere vaccine strains, which are used as reference strains until the end of the year. Viruses were identified as low-reactors if their titre with the reference antiserum was at least 8-fold lower than the titre of the reference virus. Detection of low reactors with specific antisera may be due to several different factors, so further analyses such as genetic sequencing are performed to determine whether antigenic drift has occurred.

Figure: HI assay titres of viruses collected 1 Jan—30 Sept 2016 relative to reference virus



* *indicates reference strains included in the 2016 Southern Hemisphere and 2016-2017 Northern Northern Hemisphere WHO vaccine recommendations.*

^ *Indicates strains included in the WHO quadrivalent vaccine recommendations*

Neuraminidase inhibitor susceptibility

Viral isolates are routinely tested for their susceptibility to the antiviral drugs oseltamivir (Tamiflu), zanamivir (Relenza), peramivir and laninamivir using the neuraminidase inhibition (NAI) assay. Viruses that demonstrate reduced inhibition by antiviral drugs in the NAI assay undergo genetic analysis of the neuraminidase gene to detect known or novel mutations associated with the functional change.

Of 2444 viruses tested, three A(H1N1)pdm09 viruses (from Queensland, New South Wales and Victoria) showed highly reduced inhibition to oseltamivir and peramivir, whilst two B/Victoria viruses from Malaysia showed highly reduced inhibition to all four neuraminidase inhibitors. The relationship between reduced inhibition and the clinical effectiveness of a neuraminidase inhibitor is not well understood. Further studies would be required to determine whether a virus with reduced inhibition in the NAI assay is clinically resistant.

Table: Neuraminidase inhibitor susceptibility of viruses collected 1 Jan—30 Sept 2016

Type/ subtype	A(H1N1) pdm09	A(H3N2)	B/Victoria	B/Yamagata	A (Mixed subtype)
No. viruses tested	846	1239	217	141	1
Number of viruses with highly reduced inhibition					
Oseltamivir	3	0	2	0	0
Peramivir	3	0	2	0	0
Zanamivir	0	0	2	0	0
Laninamivir	0	0	2	0	0

Previous editions of this newsletter can be found at: http://www.influenzacentre.org/centre_reports.htm

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