

Preliminary summary on oseltamivir-resistant H1N1 virus: Japan

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During the 2007/2008 Northern Hemisphere influenza season in Japan, a total of 1,360 H1N1 viruses have been tested for oseltamivir resistance (H275Y) and 22 (1.6%) found resistant.

Table 1. Frequency of resistant viruses isolated in Japan during 2007/2008 influenza season.

Month of sampling	09-2007	10-2007	11-2007	12-2007	1-2008	2-2008	3-2008	total
No. of H1N1 isolates tested	2	39	100	136	625	413	45	1360
No. (%) of resistants to Oseltamivir			1		11	7	3	22 (1.6%)

See Table 2 for details of cases with oseltamivir-resistant virus infection.

Specimen collection site: 17 out of 22 resistant viruses were collected at outpatient clinics (77%) and 5 (23%) of them were collected on sites (three specimens from one elementary school, one each from a kindergarten and a nursery school) where the local outbreaks occurred.

Demography: Although specimens have been collected from both adults and children, 19 out of 22 resistant viruses were isolated from children under 14 years old. There are no significant gender differences among patients with resistant viruses.

Oseltamivir use: 2 out of 22 (9%) resistant viruses were collected from patients after 5 days of oseltamivir treatment

Vaccination history: Vaccination history was available in 7 of 22 patients with oseltamivir-resistant virus infection. Four had vaccination history while three were not vaccinated. Vaccination history in patients with oseltamivir-sensitive viruses is not available. .

Geographical distribution: The resistant viruses were scattered in several prefectures of the main island of Japan and more than one resistant viruses were isolated from some cities and prefectures including Yokohama City, Tottori Prefecture, Tochigi Prefecture and Gifu City (Figure 1). According to the preliminary report from a local health laboratory, oseltamivir resistance prevalence have been dramatically increasing in Tottori prefecture located at west of the main island of Japan (30% of total isolates in the prefecture, analysis in detail is ongoing).

Clinical features: All of patients with resistant viruses fully recovered without complications.

Special underlying conditions: All reported cases were previously healthy children/adults.

Other findings:

1. Epidemiological background of oseltamivir-resistant viruses.
 - A/Yokohama/22/2008, A/Yokohama/34/2008, A/Yokohama/35/2008 were collected from one elementary school and A/Yokohama/30/2008, A/Yokohama/31/2008 were isolated from patients visited one outpatient clinic, which is closely located from the elementary school. Consequently, these 5 resistant viruses were considered as a part of the local outbreak.
 - A/Yokohama/77/2008, A/Yokohama/78/2008 and A/Yokohama/79/2008 were detected in a family cluster.
 - These resistant viruses isolated in Yokohama were recovered from the patients without oseltamivir treatment, although some of them were treated by zanamivir prior to sampling.
 - A/Tottori/21/2008, A/Tottori/23/2008, A/Tottori/28/2008 and A/Tottori/29/2008 were isolated from the same outpatient clinic, although the oseltamivir use in these patients is unknown.
 - All oseltamivir resistant viruses were sensitive to zanamivir.

2. Genetic analysis of oseltamivir resistant viruses.
 - Figure 2 shows phylogenetic tree of the NA gene of oseltamivir resistance viruses.
 - In 2007/08 influenza season, most H1N1 viruses isolated in Japan fell into both subclade 2B (representative reference virus; A/Hong Kong/2652/2006) and 2C (representative reference virus; A/Brisbane/59/2007), although they were antigenically indistinguishable from each other.
 - One resistant virus, A/Yokohama/91/2007, was found in subclade 2C.
 - The rest of resistance viruses (21 viruses) belonged to subclade 2B as well as the resistance viruses isolated in EU and USA.
 - In subclade 2B, viruses possessing 354G amino acid marker (so called 'Northern EU-like group') were distinguished from the viruses without the marker (so called 'Hawaii-like group').
 - Majority of Japanese resistant viruses belonged to Hawaii-like group, while three resistance viruses isolated from a family cluster in Yokohama belonged to Northern EU-like group.
3. Antigenic analysis of resistance viruses.
 - Table 3 shows antigenic analysis of representative resistance viruses by hemagglutination inhibition (HI) tests using post-infection ferret antisera against recent H1N1 reference viruses.
 - The resistance viruses were well covered with antiserum to A/Brisbane/59/2007 virus, but not with antisera to A/New Caledonia/20/99 and A/Solomon Islands/3/2006 viruses.
 - The HI data indicate that most of resistance viruses isolated in Japan were antigenically similar to A/Brisbane/59/2007 strain, but not to previous vaccine strains A/New Caledonia/20/99 and A/Solomon Islands/3/2006.

WHO Collaborating Centre for Reference and Research on Influenza at National Institute of Infectious Diseases, Tokyo Japan.
Genome Analysis Center, Department of Biotechnology, National Institute of Technology and Evaluation, Tokyo Japan.
Prefectural Public Health Laboratories in Japan.

Phylogenetic analysis of influenza A/H1N1 NA genes

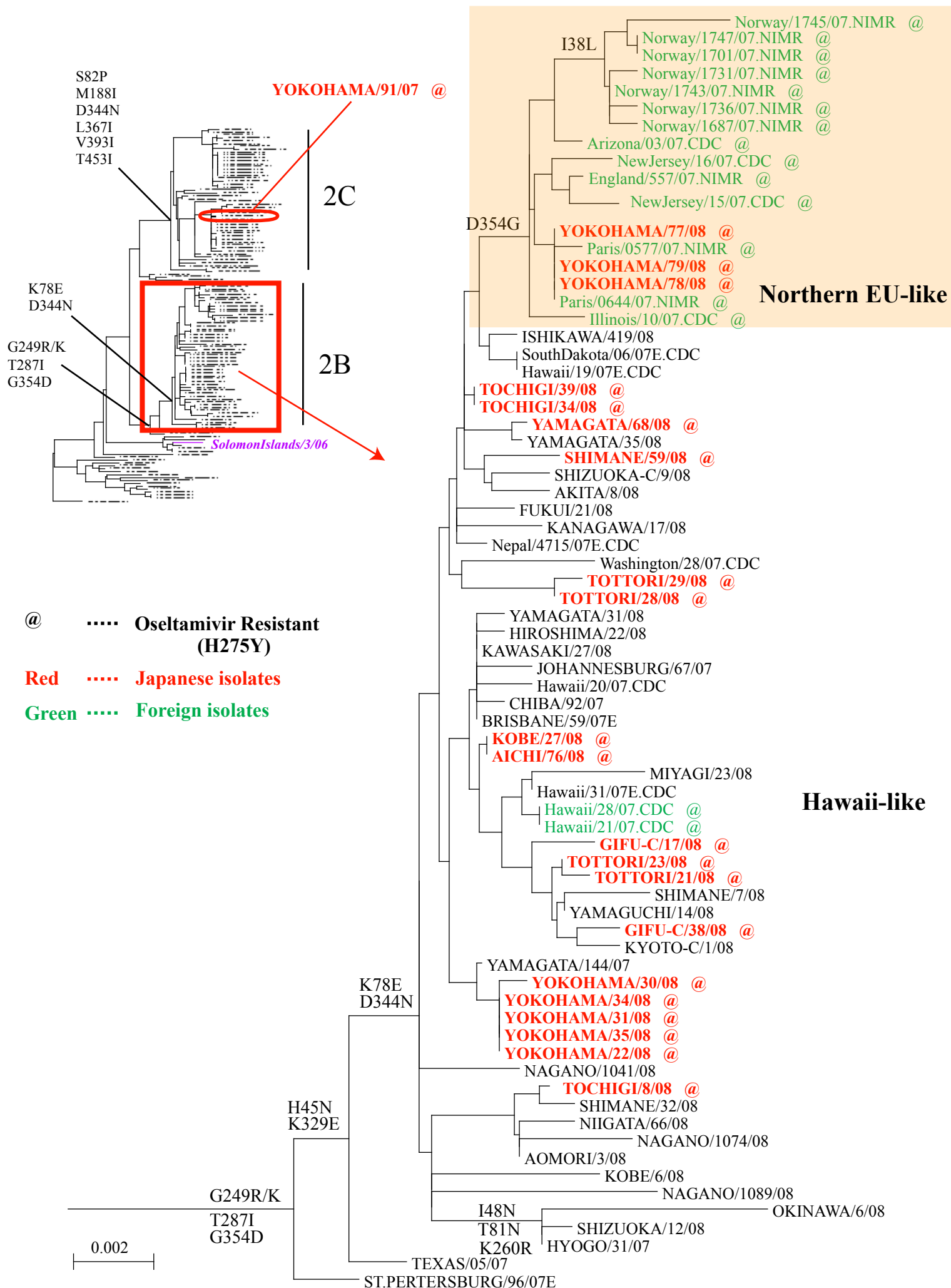


Table 2 Oseltamivir-resistant viruses isolated in Japan, 2007/08 influenza season (as of May 9, 2008)

Virus	Year	Sampling date	H274Y	Status of NAIs treatment	Age	Sex	Specimen collection site	Vaccination history
A/YOKOHAMA/91/2007	2007	2007-11-30	Y	Oseltamivir (after 5days)	5	M	Kindergarten	unkonwn
A/TOCHIGI/8/2008	2008	2008-01-15	HorY	Oseltamivir (after 5days)	4	M	Nursery school	Vaccination
A/GIFU-C/17/2008	2008	2008-01-25	Y	None	4	F	Outpatient clinic	None
A/TOTTORI/29/2008	2008	2008-01-25	Y	Unknown	5	F	Outpatient clinic	unkonwn
A/TOTTORI/28/2008	2008	2008-01-26	Y	Unknown	5	M	Outpatient clinic	unkonwn
A/YOKOHAMA/22/2008	2008	2008-1-28	Y	Zanamivir (after ? days)	10	M	Elementary school	None
A/YOKOHAMA/30/2008	2008	2008-01-28	Y	None	12	M	Outpatient clinic	unkonwn
A/YOKOHAMA/31/2008	2008	2008-01-28	Y	None	13	M	Outpatient clinic	unkonwn
A/YOKOHAMA/34/2008	2008	2008-1-28	Y	Zanamivir (after ? days)	8	F	Elementary school	Vaccination
A/YOKOHAMA/35/2008	2008	2008-1-28	Y	Zanamivir (after ? days)	9	M	Elementary school	Vaccination
A/TOTTORI/23/2008	2008	2008-01-29	Y	Unknown	4	M	Outpatient clinic	unkonwn
A/TOTTORI/21/2008	2008	2008-01-30	Y	Unknown	6	M	Outpatient clinic	unkonwn
A/TOCHIGI/34/2008	2008	2008-02-18	Y	None	1	F	Outpatient clinic	unkonwn
A/TOCHIGI/39/2008	2008	2008-02-18	Y	None	4	F	Outpatient clinic	unkonwn
A/AICHI/76/2008	2008	2008-02-19	Y	Zanamivir (after 5 days)	8	F	Outpatient clinic	unkonwn
A/SHIMANE/59/2008	2008	2008-02-26	Y	None	14	F	Outpatient clinic	Vaccination
A/YOKOHAMA/77/2008	2008	2008-02-26	Y	None	23	M	Outpatient clinic	unkonwn
A/YOKOHAMA/78/2008	2008	2008-02-28	Y	None	50	F	Outpatient clinic	unkonwn
A/YOKOHAMA/79/2008	2008	2008-02-28	Y	None	52	M	Outpatient clinic	unkonwn
A/GIFU-C/38/2008	2008	2008-03-07	Y	None	4	F	Outpatient clinic	Vaccination
A/KOBE/27/2008	2008	2008-03-11	Y	None	3	F	Outpatient clinic	unkonwn
A/YAMAGATA/68/2008	2008	2008-03-11	Y	None	8	M	Outpatient clinic	unkonwn

Table 3

Antigenic characterizations of NIA resistance viruses by HI tests

HI test date:2008/4/18

Strains	Passage History	Sample date	NewCal Lot.05	St. Petersburg 0806 No.2	K140E	K140E	K140E	K140E	K140E
					Solomon Islands 0306 No.2	Fukushima 97 No.1	Fukushima 141 No.1	Brisbane59 07 Egg No.2	Brisbane59 07 No.2
REF.Viruses									
A/New Caledonia/20/99	E3/E2/E1		640	320	160	40	320	80	80
A/St. Petersburg/08/2006	E 1/E2+1		320	160	80	40	320	80	40
A/Solomon Islands/3/2006	E2 +2	2006/08/2	160	640	640	640	1280	1280	640
A/FUKUSHIMA/97/2006	MDCK 2 +3	2006/01/2	80	640	640	320	2560	640	160
A/FUKUSHIMA/141/2006	E3 +1	2006/02/0	320	640	640	320	2560	1280	320
A/Brisbane/59/2007	E2 +1	2007/07/0	80	320	320	160	1280	640	320
A/Brisbane/59/2007	MDCKX/2 +2	2007/07/0	40	80	80	160	160	320	1280
TEST.Viruses									
A/YOKOHAMA/31/2008	MDCK2+1	(2008/2/6)	80	160	80	160	160	320	640
A/TOCHIGI/8/2008	MDCK2+1	2008/1/15	40	80	80	40	160	640	320
A/GIFU-C/38/2008	MDCK1+1	2008/3/3	80	320	320	320	160	640	640
A/YOKOHAMA/77/2008	MDCK2+1	2008/2/26	80	160	160	80	320	640	640
A/TOTTORI/28/2008	MDCK1+1	2008/1/26	80	160	320	80	640	640	320