Reinfection with Influenza A (H2N2, H3N2, and H1N1) Viruses in Soldiers and Students in Japan

Tadao Sonoguchi, Mitsuo Sakoh, Nobuharu Kunita, Kiyoaki Satsuta, Hideo Noriki, and Hideo Fukumi

Reinfection with influenza A virus was studied by measuring hemagglutination-inhibiting antibody responses to infection in paired sera taken from groups of soldiers and students. Among 62 soldiers severely infected during the first wave of the A/Asian/57 (H2N2) pandemic in 1957, 17 were asymptomatically reinfected with the same virus within six months. In the 1962 epidemic the rate increased to 41%. Among reinfected soldiers studied, 68% had an asymptomatic infection; only 10% were severely symptomatic, and they were found to be infected with a virus closely related to A/Asian/57. For H3N2 epidemics, the rate of reinfection was 17% among students studied in 1970 who were reinfected with a virus closely related to the prototype A/Hong Kong/68 (H3N2). Reinfection with an extremely drifted variant of H3N2 was found to be 32% and 69% in two groups of students studied in 1972. Reinfection with a related virus was 32% in another group studied in 1983. Among the students studied who were reinfected with H3N2 viruses, the rates of asymptomatic infection were similar to those of symptomatic infection. The reinfection rates with a virus related to A/USSR/77 (H1N1) were 9.3% and 20% in two groups studied in 1980.

The high degrees of infectivity and transmissibility of influenza A viruses among susceptible populations are well-known properties of these agents. Less well known is the degree to which reinfection occurs. This aspect of influenza epidemiology was discussed infrequently before 1972 [1, 2]. Since then more reports have become available [3-9].

To examine the immunologic and epidemiologic factors associated with reinfection, we undertook a review of the experience of nine groups of subjects during the A/Asian/57 (H2N2), A/Hong Kong/68 (H3N2), and A/USSR/77 (H1N1) pandemic and interpandemic periods. Though the present study concentrated on juveniles and young adults in closed populations, an improved understanding of reinfection and its virological and clinical consequences will help clarify the basis of immunity to influenza A infections.

This study was greatly facilitated by the fact that since the A/Asian pandemic, surveillance of influenza virus in Japan has been nationwide in scope. Every year the antigens of viruses isolated in the 47 prefectures in Japan are analyzed for antigenic variation. In addition, every year since 1960 field studies on inactivated influenza vaccines have been performed by the Influenza Vaccine Research Group.

Subjects and Methods

Subjects. A description of the populations evaluated in this study is shown in table 1. Nine populations were observed for extended periods beginning in 1957 and ending in 1983. The nine groups studied were all young adult populations; three groups consisted of military recruits and six groups, students. These individuals were evaluated serologically and clinically.

Group 1 was comprised of soldiers from one camp who had severe symptomatic infections with influenza A (H2N2) during the first wave of A/Asian/57 (H2N2), which appeared in Japan beginning 10 May 1957 [10] and in this camp, 6 June 1957 [11, 12]; subjects in this group volunteered for
**Table 1.** Characteristics of subjects who were infected and reinfection with influenza A (H2N2, N3N2, and H1N1) viruses in epidemics that occurred from 1957 through 1983.

<table>
<thead>
<tr>
<th>Subjects</th>
<th>Epidemics</th>
<th>Serological detection of infection and reinfection</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group 1</td>
<td>June-Dec 1957</td>
<td>A/Adachi/57 Same</td>
</tr>
<tr>
<td>1, 70 soldiers (18-35)</td>
<td>A/Adachi/57</td>
<td>(27)</td>
</tr>
<tr>
<td>Group 2</td>
<td>Feb-Mar 1962</td>
<td>A/Tokyo/62 Closely related</td>
</tr>
<tr>
<td>2, 464 soldiers (20-35)</td>
<td>A/Adachi/57</td>
<td>220 (47) 192/78 (41)</td>
</tr>
<tr>
<td>Group 3</td>
<td>Feb-Mar 1962</td>
<td>A/Tokyo/62 Closely related</td>
</tr>
<tr>
<td>3, 97 soldiers (20-35)</td>
<td>A/Adachi/57</td>
<td>38 (39) 50/18 (36)</td>
</tr>
<tr>
<td>Group 4</td>
<td>Jan-Mar 1970</td>
<td>A/Fukuoka/70 Closely related</td>
</tr>
<tr>
<td>4, 858†</td>
<td>Jan-Feb 1972</td>
<td>A/Kumamoto/72 Variant</td>
</tr>
<tr>
<td>5, 858 students† (13-18)</td>
<td>A/Kumamoto/72 Variant</td>
<td>348 (40) 358 (42)</td>
</tr>
<tr>
<td>Group 5</td>
<td>Jan-Feb 1972</td>
<td>A/Kumamoto/72 Variant</td>
</tr>
<tr>
<td>6, 85 students (16-18)</td>
<td>A/Kumamoto/72 Variant</td>
<td>62 (73) 63 (83)</td>
</tr>
<tr>
<td>Group 6</td>
<td>Nov 1979</td>
<td>A/Kumamoto/79 Related</td>
</tr>
<tr>
<td>7A, 56 students‡ (18-20)</td>
<td>A/Kumamoto/79 Related</td>
<td>27 (48) 32/3 (9.3)**</td>
</tr>
<tr>
<td>Group 7</td>
<td>Mar 1980</td>
<td>A/Wakayama/83 Related</td>
</tr>
<tr>
<td>7B, 39 students‡‡ (18-20)</td>
<td>A/Wakayama/83 Related</td>
<td>25 (48) 31/10 (32)</td>
</tr>
<tr>
<td>Group 8</td>
<td>Jan-Feb 1983</td>
<td>A/Wakayama/83 Related</td>
</tr>
<tr>
<td>8, 52 students (16-17)</td>
<td>A/Wakayama/83 Related</td>
<td>27 (48)</td>
</tr>
</tbody>
</table>

* Asympt = asymptomatic; slight = students who continued to attend school despite slight symptoms.
† Most subjects lived in Nagasaki prefecture.
‡ Corrected number.
§ \( P < .001 \) by \( \chi^2 \) test.
¶ All subjects lived in Nagasaki prefecture.
# Only 76 sera were tested because of a shortage of serum volume.
** No statistical significance.
†† All subjects lived in Osaka.
‡‡ All subjects lived in Fukuoka.

The study in the middle of June. During the period of observation a second wave of A/Asian/57 occurred in this camp between October and December 1957. Serum samples were collected from this group of soldiers seven times.

Group 2 was also comprised of soldiers from a single camp. An epidemic attributed to a virus closely related to the prototype A/Asian/57 occurred in this camp (in the center of Tokyo) between February and March 1962 [13]. In early January, serum samples had been taken from all 713 regiment soldiers for testing for syphilis. From this group, 464 soldiers agreed to be followed up and were tested after the epidemic to determine their titers of HAI antibody. During the epidemic, soldiers who were sick were evaluated for clinical manifestations of influenza. After the epidemic, clinical data were related to serological evidence of infection with the A/Asian/57 virus.

Group 3 was comprised of 97 healthy soldiers at
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a camp in the suburb of Tokyo whose pre- and postepidemic serum samples were collected by random sampling (the medical staff did not send the sera of seriously infected persons to us because of a communication gap) [14].

Groups 4 through 7B were comprised of subjects who had all received monovalent vaccine for influenza B as a control for those who received a vaccine for influenza A.

Group 8 was comprised of students who had not received any influenza vaccine for more than three years because of side effects or for other reasons. Pre- and postepidemic serum samples, however, were collected from all members of this group, along with the vaccinees, to examine for HAI antibodies.

Method of surveillance. Groups 1, 2, and 3 were not vaccinated. The subjects in these populations were examined for illness on a regular basis by the military medical officers.

Since the autumn of 1962, ~80%~90% of Japanese students have received commercial, killed influenza vaccine annually in accordance with the recommendation of the Japanese government. In addition, the Welfare Ministry recommends that prefectoral governments conduct surveys for serological evidence of infectious diseases, including influenza, rubella, measles, and other agents. For influenza, the surveillance consists of the measurements of HAI antibody in sera taken from infants, pupils, students, and adult volunteers during the interepidemic periods. During influenza epidemics, acute- and convalescent-phase sera are also taken routinely from affected individuals in every prefecture and their HAI titers measured.

In Japan, collection of three serum specimens in field trials is routinely carried out. The following collection schedule was used for groups 4 through 7B: just before vaccination (end of October or November), four to five weeks after vaccination (December or early January; preepidemic), and four to five weeks after the epidemic (end of March or April; postepidemic). Because epidemics due to H3N2 or H1N1 virus occurred during these periods, it was possible to observe the efficacy of influenza A vaccines and also the rate of reinfection with influenza A virus among recipients of the vaccine for influenza B.

Self-administered questionnaires were given to all subjects in groups 1, 2, 6, and 8, whenever influenza-like illness occurred during the epidemics. Those individuals more seriously affected visited doctors and responded to the questionnaires along with the doctor's diagnosis. Slightly ill subjects did not always respond to the questionnaire. As a result, differences in the number of influenza-like illnesses and infections were significant.

In groups 4, 5, 7A, and 7B, questionnaires were not used. Therefore, in these groups only serological evidence of reinfection was studied.

The clinical definition of influenza used throughout the study was a febrile (37.5°C) respiratory illness lasting more than one day and associated with at least one of the following symptoms: chills, sore throat, headache, or muscle aches. Those individuals with a body temperature ≥38.5°C were defined as seriously infected with influenza, and those who did not manifest any symptoms were considered asymptomatic. In addition, those with just slight symptoms and who attended schools or worked in camps as usual were considered to be well.

Serological methods and procedures for the isolation of virus. Sera were incubated with four volumes of receptor-destroying enzyme (Vibrio cholerae) in tubes or on plastic plates at 37°C for 18 hr and then heated at 56°C for 1 hr. Serial dilutions of 0.25 ml of treated serum were incubated with equal volumes of influenza virus containing four units of HA. After incubation at room temperature for 1 hr, 0.5 ml of fowl red blood cells was added to each tube or well, and the results were noted after red cell sedimentation. The HAI titer was defined as the highest initial serum dilution that completely inhibited HA. (Because of the use of the highest final serum dilution for expression of the titer in Japan, a titer of 1:64 in this report is equivalent to a titer of 1:16 in articles from the United States or European countries.)

The pre- and postepidemic serum samples from each individual were tested simultaneously (as a pair) for HAI antibodies. The following antigens were used: A/Adachi/2/57 (H2N2) [A/Singapore/1/57], A/Aichi/2/68 (H3N2), A/Fukuoka/1/70 (H3N2); figure 1, A/Kumamoto/1/72 (H3N2) [A/Hong Kong/107/71], A/NIigata/102/81 (H3N2; figure 1), A/USSR/91/77 (H1N1), and A/Kumamoto/37/79 (H1N1) [A/Brazil/11/78]. Reference strains are shown in brackets.

The A/Kumamoto/1/72 strain exhibited considerable antigenic drift from the prototype A/Hong Kong (A/Aichi) virus (figure 1), but it was not widespread and caused only local outbreaks in Asia [15]. In Japan particularly, 85% (195 of 229) of isolated viruses were the same as the Kumamoto strain, and
### Figure 1. Antigenic drift between strains of virus.

Strains were tested by the cross-HAI test with postinfection ferret serum. The data represent the reciprocal HAI titer. * = significant antigenic drift was detected by using monoclonal antibodies [25].

<table>
<thead>
<tr>
<th>Strain</th>
<th>A/Adachi/1/57</th>
<th>A/Tokyo/1/62</th>
<th>A/Kumamoto/1/72</th>
<th>A/Bangkok/1/79</th>
<th>A/Tokyo/1/72</th>
<th>A/Kumamoto/1/79</th>
<th>A/Bangkok/1/72</th>
</tr>
</thead>
<tbody>
<tr>
<td>H1N1</td>
<td>512</td>
<td>256*</td>
<td>64</td>
<td>128</td>
<td>64</td>
<td>256*</td>
<td>512</td>
</tr>
<tr>
<td>H3N2</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>A/Adachi/1-1/68</td>
<td>1,024</td>
<td>1,024</td>
<td>64</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>A/Tokyo/1-1/70</td>
<td>256</td>
<td>1,024</td>
<td>64</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>A/Kumamoto/1-1/72</td>
<td>€32</td>
<td>€32</td>
<td>1,024</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>A/Bangkok/1-1/79</td>
<td>2,048</td>
<td>512</td>
<td>128</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>A/Philippines/11/81</td>
<td>512</td>
<td>1,024</td>
<td>512</td>
<td>128</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>A/Tokyo/1-1/62</td>
<td>256</td>
<td>512</td>
<td>1,024</td>
<td>1,024</td>
<td>512</td>
<td>256</td>
<td>512</td>
</tr>
<tr>
<td>A/Kumamoto/1-1/79</td>
<td>€32</td>
<td>€32</td>
<td>€32</td>
<td>128</td>
<td>256</td>
<td>512</td>
<td>256</td>
</tr>
</tbody>
</table>

**VIRUS STRAIN TESTED**

**ANTISERUM TO**

iZcrr, H1N1 A/USSR/92/77 1,024 128

A/Kumamoto/32/79 64 256

H2N2 A/Adachi/2/57 512 256*

A/Tokyo/1/62 512 512

H3N2 A/Adachi/1/68 1,024 1,024 64

A/Tokyo/1/70 256 1,024 64

A/Kumamoto/1/72 €32 €32 1,024

A/Bangkok/1/79 2,048 512 128

A/Tokyo/1/72 512 1,024

A/Kumamoto/1/79 1,024 128

A/Bangkok/1/79 512 1,024

A/Tokyo/1/72 256

15% were similar to the A/Tokyo/1-1/72 strain [A/England/42/72 (H3N2)]. Interestingly, A/Kumamoto/1-1/72 disappeared in 1973, and since then A/Tokyo/72 has become predominant. A fourfold or greater increase in the titer of HAI antibody between pre- and postepidemic serum specimens was regarded as evidence of infection.

Embryonated eggs were used for isolation of virus. Determination of previous primary infection and of reinfection with the prevalent virus. Earlier studies suggested that individuals <70 years of age did not have any H2N2 HAI antibody before the first wave of Asian influenza [16]. The subjects in groups 1, 2, and 3 were all 18–35 years of age. Since these soldiers did not receive an H2N2 vaccine (because it was not available or because they did not have histories of exposure), the 1957 wave of H2N2 viruses represented their initial exposure to this virus. Although groups 2 and 3 were investigated five years after the initial infection with the H2N2 virus, they had the same experience as group 1. The reasons are as follows. Following the first and second waves of the A/Asian/57 pandemic, sera from >7,000 soldiers from all military camps throughout Japan were selected by random sampling, and HAI titers were determined [11]. Thus, it was clearly demonstrated that all the camps were involved in the pandemic twice. These facts strongly suggest that some recruits in groups 2 and 3 have had two infections (figure 2). In addition, from 1958 through 1961 just a few sporadic cases were observed in several districts and in only three to five camps far from Tokyo [17–19]. Therefore, the recruits in groups 2 and 3 should not have had any additional infection before the 1962 epidemic. However, their HAI titers might have been reduced because of the time lapse.

Retrospective serological studies have suggested that before the emergence of H3N2 viruses in 1968, there was a small number of people with existing HAI titers $\geq 1:16$ to these viruses [20]. Our earlier reports indicated that very few individuals in Japan had antibody to H3N2 in inverse proportion to HAI titers before the epidemic [21, 22]. For our studies, we omitted from further analysis those individuals with preepidemic HAI titers $\geq 1:16$ in groups 4, 5, 6, and 8. After subtracting this number, the remainder (corrected number) consisted of those who had a primary infection with influenza A (H3N2) during that epidemic.

Observations of the H1N1 pandemic in 1977–1978 indicated that individuals born after 1957 did not have any HAI antibody to H1N1 virus before that pandemic [2]. Group 7 included only persons born after 1957. Therefore, the 1977–1978 epidemic of H1N1 viruses represented the initial exposure to this virus for these subjects.

A fourfold or greater increase in HAI titer following the second wave of A/Asian/57 in persons in group 1 and following the 1962 epidemic among those in groups 2 and 3 with HAI titers $\geq 1:32$ before the epidemic clearly demonstrated reinfection with the H2N2 virus.

Some individuals in groups 4–8 might have received influenza vaccine $\geq 1.5$ years before each epidemic. However, our previous observations have demonstrated that vaccine-induced antibodies among those who had no HAI antibody before vaccination became undetectable within one year [23,
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Figure 2. The change in frequency distribution of HAI titers of 70 soldiers seriously infected with H2N2 virus and individual prior rises and falls in the HAI titer of 17 reinfection individuals. The antigen used was A/Adachi/2/57. The numbers 1-17 represent reinfect individuals.

Reinfection with the same H2N2 virus shortly after primary infection. Figure 2 illustrates the increase and decrease in the titer of HAI antibody to A/Asian/57 (H2N2) virus in 70 soldiers who had their serious primary infection with the H2N2 virus in the middle of June during the first wave of the A/Asian/57 epidemic, as well as reinfection with the same virus during the second wave (1957). Three to four weeks after the onset of illness this group achieved a geometric mean titer of 6.5 (log₂). The geometric mean titer remained almost unchanged until eight weeks after onset and dropped to a titer of 5.1 (log₂) after 18 weeks (at the beginning of the second wave). Only 62 subjects were present in the military camp and available for study at that time. A fourfold or greater rise in the preepidemic HAI titers was observed in 13 of these subjects 18-23 weeks after the primary infection. Finding it difficult to accept the possibility of reinfection with the same virus in such a short period, we tested paired sera three times and obtained the same results. Similarly, 23-34 weeks after the initial epidemic, fourfold rises in HAI titers were detected in four more soldiers. These individuals probably could have been reinfected at the end of the second wave, although their titers were measured two months later. However, these seventeen (27%) did not show any symptoms of sickness and worked as usual. The HAI titers induced by the same virus during the second wave remained unchanged for rather a long period. These results clearly indicate that reinfection with type A (H2N2) influenza can occur even after a short interval, as HAI titers drop, and that although infection may recur, sickness does not necessarily follow. (Neutralization and CF tests were not carried out, and neuraminidase inhibition and ELISA tests were not available at that time.)

Characteristics of reinfection with a virus closely related to the prototype H2N2 virus five years after primary infection. Number and rates of total infection and reinfection with a virus closely related to A/Asian/57 (H2N2), A/Toyko/1/62 (H2N2) [A/England/12/62] [25], as well as clinical information of reinfection in group 2 in 1962, are shown in table 1. The rate of reinfection with severe illness was far lower than the rate of asymptomatic reinfection (10% vs. 68%). These results demonstrate that immunity acquired by the previous infection with H2N2 virus could mostly prevent the occurrence of symptoms even if reinfection with a closely related virus did occur five years later. The rates of asymptomatic illness became higher as the HAI titers increased.

The results obtained from recruits in group 3 also indicated a high rate of asymptomatic reinfection. Reinfection with a closely related H3N2 virus one year after primary infection. Group 4 was involved in the A/Hong Kong/68 (H3N2) influenza epidemic for the first time from December 1968 through March 1969 and exposed to a closely related virus about one year later (table 1, figure 1). The fairly high rate of total infection (37%) is clearly explained by the fact that the first A/Hong Kong influenza
epidemic was far smaller than that of the second epidemic in 1970 on Kyushu Island, in contrast to the other districts. This fact was distinctly demonstrated by the summary of the Report of Weekly Report of the Welfare Ministry on influenza-like illness, 1968–1969 and 1969–1970. The report showed that the total number of pupils in public schools who were absent during the first epidemic in each prefecture in Kyushu was about half of those in the second epidemic, including group 4. This result coincided with our previous reports [21, 22]. Therefore, a large number of susceptibles remained after the first epidemic and were infected during the second epidemic. That the reinfection rate (17%) was low despite the rather large epidemic may be due to the short lapse of time after the first exposure and also to a small antigenic drift.

Characteristics of infection with a major variant virus within a short interval among those previously infected with the prototype and closely related H3N2 viruses. Groups 5 and 6 were exposed to two epidemics that were due to the prototype H3N2 virus (1968–1969) and to one that was due to A/Fukuoka/1/70 (1970). In these two populations, epidemics caused by a virus with a major antigenic drift from a previous strain (A/Kumamoto/1/72) occurred, and the variant viruses were isolated between January and February 1972. Thus, serum HAI antibody responses were measured in pre- and postepidemic sera by using as antigen the prevalent A/Kumamoto/1/72 and previous A/Fukuoka/70 strains. Antigenic drifts are shown in figure 1.

Before the A/Kumamoto epidemic, the proportion of subjects with preepidemic HAI titers <1:16 against A/Fukuoka was only 5.3% of 858 subjects in group 5, whereas against A/Kumamoto it was 54%. This fact indicates that the antigenic drift of the A/Kumamoto strain from the prototype was significant. After the A/Kumamoto/72 epidemic, the overall rate of serological infection in group 5 was almost the same as that after the A/Fukuoka/70 epidemic in group 4. However, the rate of reinfection in group 4 in 1970 was far less than the rate of fourfold or greater rises in HAI titer against A/Kumamoto during the epidemic among those with preepidemic HAI titers ≥1:32 was 69% (22 of 32). In addition, nine students were proved to have had preepidemic HAI titers ≥1:32 against A/Fukuoka and <1:16 against A/Kumamoto, and their titers rose fourfold or more against A/Kumamoto during the epidemic. Thus, the rate of infection became 97% (31 of 32).

These high rates of serological infection should be attributed to the major antigenic drift of the prevailing virus as well as to living conditions in this dormitory, which might have facilitated the spread of the virus.

In the epidemic, 17 students were seriously ill and had to miss classes. Eight of these had preepidemic HAI titers (against A/Fukuoka) ≤1:16, whereas the other nine (29%) students who had titers ≥1:32 were infected with A/Kumamoto virus. Of 31 students previously infected with preceding strains, 22 (71%) did not miss classes during the epidemic. These facts suggested that less than one-third of previously infected students experienced severe infection with the variant, and the remaining two-thirds were only very slightly or asymptptomatically reinfected.

Reinfection with a closely related H1N1 virus. Groups 7A and 7B were involved in the epidemic that was due to a closely related H1N1 virus, A/Kumamoto/37/79 [A/Brazil/1/11/78], for the second time 20–24 months after the first outbreak of A/USSR/77. (There was no epidemic due to influenza A virus in Japan in 1979.) The rate of infection for the entire group and that for those with preepidemic HAI titers ≥1:32 are shown in table 1. The rate of the latter in group 7A was 9.3% and in group 7B, 20%.

Characteristics of infection with a variant H3N2 virus among those exposed several times to various H3N2 viruses. Group 8 probably was exposed to many H3N2 variants, e.g., A/Victoria/75, A/Texas/77, and A/Bangkok/79, because large epidemics due to these viruses occurred in 1976, 1978, and 1980, respectively, in most schools in Japan. Since sporadic cases occurred in 1981 and an epidemic due only to B virus occurred in 1982, this group had no exposure to H3N2 virus for three years. Both the total rate of infection with and the rate of fourfold or greater
rises in HAI titer against the prevalent (closely related to A/Philippines/82) virus among those with the preepidemic HAI titers $\geq 1:32$ to previous H3N2 virus were higher. The results should be considered to be due to antigenic drift and to the rather long lapse of time from the last exposure to an H3N2 virus.

Of 10 reinfected individuals, three were judged to be seriously affected (temperature $\geq 38^\circ C$), two others mildly affected, and five did not manifest any symptoms of illness.

Discussion

Studies on reinfection with type A influenza (H3N2 and H1N1) virus have been increasing recently [3-9]. The present study attempted to extend these observations by reviewing six epidemics due to three subtypes of influenza A virus (H2N2, H3N2, and H1N1) through measurement of HAI titers of paired sera taken before and after the epidemics from juveniles and young adults in closed populations in Japan.

Two notable features were represented by the observations of reinfection with the same H2N2 virus. First, reinfection with the virus occurred at a comparatively high rate within just six months. Second, none of those who were reinfected had symptoms. Our previous report indicated reinfections with the same virus in the second wave in three populations [10]. These facts clearly demonstrate that reinfection can occur within a very short period.

An interesting fact was observed in the H3N2 subtype era. The extremely high rate of infection with a variant virus during the 1972 epidemic was obtained from those subjects who experienced one exposure to the prototype A/Aichi/68 and one more exposure to a related virus just one or two years before the appearance of the variant, which represented an antigenically significant difference between the two previous viruses.

Comparing one set of observations of reinfection with another is difficult because of the variability in epidemiological factors and in methods used. However, almost all the rates of reinfection with each subtype virus obtained in this study were higher than were found in previous reports [1, 2, 26, 27], except for those found by the Influenza Research Unit in the United Kingdom and by groups in Seattle and Houston [3, 4, 8, 9].

These high rates of reinfection appear to be due to the following factors. First, symptomatic and asymptomatic cases were investigated in this study by the titration of paired sera of all individuals; second, the subjects were mostly young people [3, 4, 10, 15]; third, the subjects were characterized by living in rather or extremely crowded populations such as schools, military camps, and dormitories where there are high rates of exposure; fourth, there was a large degree of antigenic drift in groups 5 and 6; and fifth, the interval between exposures, i.e., the fall in titer of antibody in the period before reinfection in groups 2, 3, and 8.

These very high rates of reinfection are far different than those observed in Seattle and Houston, where variable rates were presented. The reason for the difference between their rates and ours should be attributed to the variability of age, density of population, and antigenic drift to sequential virus. However, the extremely high rates of second infection mentioned above are clearly due to extremely high rates of exposure as well as to major antigenic drift.

Davies et al. [8] also reported extremely high rates of reinfection in boarding schools. This must have come from the high rate of exposure, as happened in our group 6. However, the rate of reinfection with a closely related virus was not as high as in our study [2, 26-28].

Earlier studies by one of the authors (T. S.) indicated that the rate of primary infection with symptoms vs. that without symptoms during the first outbreaks due to new subtypes was 10:1 or 20:1 [29, 30]. However, the rate was reversed in the reinfections in this study. Similar results were obtained by Davies et al. [8].

In this report the rate of asymptomatic cases among reinfeected individuals was high (100% of 17 and 68% of 78) in two epidemics due to the same or to closely related H2N2 viruses. Those rates in two epidemics caused by a variant H3N2 virus were also rather high (61% of 31, and 50% of 10).

In this connection, it should be taken into consideration that Japanese pupils do not attach much importance to upper-respiratory-tract diseases. They do not visit doctors but continue to attend school if the symptoms are slight. However, Davies et al. [8] reported that only nine of 23 reinfeected individuals had symptoms. These results in the United Kingdom and in Japan suggest that the rate of asymptomatic reinfection may possibly be higher in some populations than the rate reported in other studies.

In this study, we define reinfection as fourfold rises in HAI titers during each epidemic among only those...
with preepidemic HAI titers \(\geq 1:32\); those with titers of 1:16 were excluded. Nevertheless, among the latter there may be some individuals whose HAI titers had increased after a previous infection but then fallen to <1:16 before the epidemic because of the time lapse (figure 2). The number of these persons might have increased as time went by. If they are added to our results, and if other methods such as CF, neuraminidase inhibition, ELISA, or isolation of virus were used, the rates of reinfection would be even higher than those shown in the present study.

In order to determine the real rate of reinfection, however, we must undertake large-scale longitudinal observations of various populations, such as the three groups mentioned above [6, 9, 28], that include analysis of host, virus, and environmental aspects [9], as well as cross-sectional studies of many populations.

References