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Full Length Research Paper

Monitoring the influenza pandemic of 2009 in Thailand by a community-based survey

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As an international traveling hub of South-East Asia, Thailand was one of the countries hardest and earliest hit by the influenza A (H1N1) 2009 pandemic. In order to understand the epidemic spread in the country, we conducted community-based surveys in metropolitan, urban, and rural areas using questionnaire interviews. We also determined sero-positive rates from randomly selected samples within the surveyed population. Recalled incidences of fever and acute respiratory symptoms in the survey correlated well with systematic reports of 2009 pandemic influenza cases from hospitals in the same areas, giving a ratio of total cases extrapolated from the surveyed data for persons who sought medical attention reported in the hospital-based surveillance system at 275:1. Conducting a large scale survey of the influenza outbreak is time consuming and also can be difficult to complete in a short time. Therefore, we used the survey for monitoring the outbreak of respiratory disease in the early pandemic phase. The seroprevalence rate was 8 to 10%, with higher rate for younger age groups, and suggests that sufficient herd immunity may have been reached in Thailand, especially in urban areas, while others may still be vulnerable to the second wave of the pandemic.

Key words: Pandemic, influenza, survey, Thailand.

INTRODUCTION

Influenza epidemics can be unpredictable and vary enormously in severity (Bramley et al., 2009). Novel influenza A (H1N1) 2009 was first reported in the USA and Mexico in April 2009 (CDC, 2009). The World Health Organization announced Phase 6 of the influenza pandemic on June 11, 2009. The virus is new to humans, so there are uncertainties about transmission efficiency and disease severity as pandemic influenza continues to evolve rapidly (AlMazroa et al., 2009).

As an international traveling hub of South-East Asia, Thailand was one of the countries hardest and earliest hit by the influenza A (H1N1) 2009 pandemic. The initial cases of laboratory-confirmed influenza A (H1N1) 2009 in Thailand were among travelers and students returning from epidemic area of the American Continent, The Thai Ministry of Public Health (MOPH) reported. In Thailand, locally acquired epidemics of pandemic influenza were first detected in June 2009. In the early epidemic, wide and rapid spreads of influenza transmission primarily occurred in schools in Bangkok metropolitan and major tourist cities (Apisarnthanarak, 2009; Suchada, 2009; Jongcherdchootrakul, 2010).

Having accurate and timely information on the extent of spread of outbreaks is crucial to informed decisions, and to deployment of proper interventions and mitigating measures. Thailand has a well-developed public health infrastructure, however, getting accurate information on numbers of cases and their distribution is often difficult in a widespread outbreak situation (Fraser et al., 2009).

Most public health authorities have relied on systematic reporting of laboratory-confirmed cases, influenza-like illnesses in out-patient settings, hospitalized patients with severe cases, and deaths (Kitler et al., 2002; Rao, 2003;

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Flahault, 2006). Having estimated the total cases based on available data, one has to assume fixed proportions of total cases to the surveyed population, which can be acquired from studies at the beginning of the outbreak when the number of total cases is small enough to be tracked.

In addition, the proportion of patients who seek medical attention can be strongly affected by perception about disease severity, for instance, information gathering through the media, however, this perception can vary with time depending on several factors. The proportion of admitted cases and deaths can also vary as the severity of the virus changes while it evolves. Community-based active survey seems reasonable alternative (Ghosh, 2008; Levy-Bruhl, 2009). However, it is difficult to get accurate data from a large-scale clinical-based survey, because influenza symptoms are mostly non-specific, and other infectious and non-infectious illnesses can confound the findings. To overcome this problem, we designed a community-based survey to monitor the situation of influenza-like illnesses and tested its reliability to detect pandemic influenza infections by serological testing.

METHODS

Questionnaire and survey

The tool we used in the survey was a structured questionnaire, which was tested for feasibility and practicality in a district of the Bangkok Metropolitan Administration (BMA) called Dusit. The set of questions on demographic information of respondents and their families included gender, age, education level, religion, number of household members, and type of property resided in. Also, questions about the trend of the influenza outbreak comprised the number of household members who had influenza-like symptoms in the immediately preceding 2 weeks and in the 3 months prior to the survey; details of influenza-like illness and chronic diseases of each household member; behavior of sick individuals to prevent transmission to others; behavior to prevent oneself from getting influenza; and willingness to receive vaccination. For quality control of the interview process, a group of interviewers were trained, educated, and observed by an investigator as they moved step-by-step to follow the guideline for the survey.

Population and sampling method

The initial outbreaks of influenza A (H1N1) 2009 occurred at Thailand at different times. Areas of the country can be categorized into four groups on the basic of timing of outbreaks as May, June, July and August. We selected the province that first reported an outbreak of pandemic influenza in each of the four time-based groups for our survey including Bangkok Metropolitan Administration, NakhonRatchasima, ChiangMai, and Nakhonsrithammarat, respectively. The household survey by poll method (Poll-1), cluster sampling technique, in urban and suburban areas of BMA was conducted by a poll conducted by experienced interviewers in October 2009. For the other three provinces, their cities were purposively chosen, and rural areas were simply randomized to be surveyed with similar periods and techniques.

We also conducted a serologic survey based on a proportional-cluster sampling technique classified by number of households in sub-districts of ChiangMai and Nakhonsrithammarat. Since the prevalence of Influenza A (H1N1) 2009 in the Thai population was estimated at 20% with 5% as worst acceptable value, a total of 246 respondents should be tested. Face-to-face interviews and blood sample collection was done by an investigator and a team of health professionals. Exclusion criteria for recruitment for blood sample collection was a person who was <5 years old, and when fewer than 75% of family members in the household agreed to provide blood samples.

Case definition

An acute respiratory illness (ARI) case was defined as one affecting a person who had history of at least two of the following symptoms; fever, cough, sore throat, and running noses within recent three months prior to the survey.

Serologic testing

Hemagglutination-inhibition assay (HI assay) was performed as previously described (Iconic et al., 2009). The protocol called for A/Thailand/104/2009(H1N1) live virus as the test antigen; and 0.5% goose erythrocytes were used as the detector. The test sera were rid of non-specific inhibitors by pre-treatment with receptor destroying enzyme (Denka Seiken, Tokyo, Japan), at 37°C overnight, followed by heat inactivation at 56°C for 30 min; nonspecific agglutinator was removed by addition of 50% goose erythrocytes and incubated at 4°C for 1 h. Two-fold serial dilutions of test sera were prepared in duplicate, followed by incubation with the test antigen at a working concentration of 4 HA-units, the highest dilution of antigen that gives complete haemagglutination of cells, for 30 min at room temperature. Erythrocyte suspension was added to the reaction plates, and further incubation at 4°C for 30 min was performed before the result was read. HI antibody titer was defined as the reciprocal of the highest serum dilution that completely inhibited hemagglutination.

A microneutralization assay (MicroNT) was performed as previously described (Kitphati et al., 2009). The assay was based on a reduction in the amount of nucleoprotein produced in the virus-infected Madin-Darby Canine Kidney Cells (MDCK) monolayer as infectivity of the test virus is neutralized by specific antibody. A/Thailand/104/2009 pandemic strain was used as the test virus. The test sera were two-fold serially diluted, added with the test virus at a final concentration of 100TCID50 for 2 h at 37°C. The serum-virus mixture was then added onto the MDCK cell monolayer and further incubated for 24 h at 37°C. The reaction plate was fixed and tested by enzyme-linked immunosorbent assay (ELISA) for presence of the viral nucleoprotein using mouse monoclonal antibody (Chemicon, Temecula, CA) as the primary antibody and goat anti-mouse Igs (Southern Biotech, Birmingham, AL) as the secondary antibody. Antibody titer was defined as reciprocal of the highest serum dilution that could reduce ≥50% of the amount of nucleoprotein when compared with the virus control.

RESULTS

The surveyed incidence of ARI correlated with systematically reported ARI

We initially conducted a small exploratory survey to test tools in a district of Bangkok in August 2009. A total of 90 households were recruited with data for each household member obtained in an interview with a family member

Table 1. Demographic characteristics and practices of prevention measures in community based survey (Poll-1) in 8 areas, Thailand, July - September 2009

Characteristics	Bangkok		ChiangMai		NakornRatchasima		Nakornsrithammarat	
	City	Suburb	City	Rural	City	Rural	City	Rural
Population* (mid-year 2009)	99,994	150,166	238,460	21,405	329,531	83,639	254,261	30,816
Interviewed family (n = 826)	100	115	101	106	103	98	101	102
Family members (n = 3,351)	479	450	352	373	429	434	408	426
- Median of member in family [range]	4 [2-25]	3 [1-20]	3 [1-15]	3 [1-8]	4 [1-10]	4 [1-12]	4 [2-7]	4 [2-8]
Any member developed ARI between July and September 2009 (n = 460) (%)	48 (10)	62 (14)	36 (10)	29(8)	101 (24)	60 (14)	66 (16)	58 (14)
- Male (%)	16 (33)	34 (55)	13 (36)	15 (52)	42 (42)	32 (53)	28 (42)	30 (52)
- Median of age (year) [range]	21 [1-67]	25 [1-55]	24 [2-65]	23 [1-71]	18 [1-76]	7 [1-62]	16 [1-70]	12 [1-52]
Interviewee developed ARI (n = 128) (%)	18 (18.0)	22 (19.1)	14 (13.9)	6 (5.7)	35 (34.0)	6 (6.1)	18 (17.8)	9 (8.8)
- Mask use by interviewee (%)	38.9	63.6	71.4	83.3	31.4	16.7	11.1	22.2
- Mask use by family members (%)	5.6	22.7	0.0	50.0	11.4	33.3	5.6	0.0
- Take a sick leave (%)	33.3	59.1	57.1	50.0	60.0	50.0	83.3	55.6
- Hand covers mouth when cough (%)	77.8	63.6	100.0	83.3	82.9	66.7	94.4	88.9
- Not sharing bedroom (%)	22.2	31.8	21.4	50.0	25.7	0.0	33.3	44.4
- Frequent hand washing (%)	83.3	59.1	92.9	100.0	77.1	50.0	100.0	88.9

* Mid-year 2009 Thai population from Department of Provincial Administration, Ministry of Interior, Thailand.

whose age was higher than 15 years. Report for those households indicated that 62.2% had experienced symptoms of acute respiratory infections, example, fever and/or respiratory tract symptoms during the previous 3-month period. The highest frequency of illness was reported for children and young adults (ages 0 to 19 years).

We subsequently conducted surveys (Poll-1) in two areas each of ChiangMai, Nakhonsrithammarat, Nakhon-ratchasima, and Bangkok Metropolitan Administration (BMA), simultaneously in October 2009. These eight areas represented different levels of impact by pandemic influenza according to the national surveillance system for ARI. A total of 826 families were recruited in the eight survey sites. Among those, a member of each family was randomly selected for interview. Table 1 shows popu-lation density and characteristics of the surveyed areas, as well as demographic data of the surveyed subjects. Of all 460 persons who reported having ARI in the surveyed households, 128 (28%), one from each household, were interviewed for their practice of personal hygiene and their family members' preventive measures, example, a face mask wearing, home rest, personal hygiene practice, bedroom separation, and frequent hand washing (Table 1). The survey showed a cumulative incidence of ARI since the beginning of the epidemic to be just under 14%. Concurrent illnesses of two or more members in the same household were reported in 39% of all reported illnesses. Younger age groups had higher incidence of ARI, with cumulative incidence of ARI up to 43% for children <5 years old (Figure 1). The surveyed ARI incidence was extrapolated to the total population in the eight survey areas. Total numbers of ARI by age group correlate well with numbers of reported pandemic influenza A (H1N1) 2009 by age group in the MOPH system (r^2 = 0.733, p<0.005) (Figure 2) giving a ratio of total community-based ARI cases to the reported pandemic H1N1 cases seeking medical attention in the MOPH system was estimated to be 275:1.

We repeated the survey in ChiangMai city and

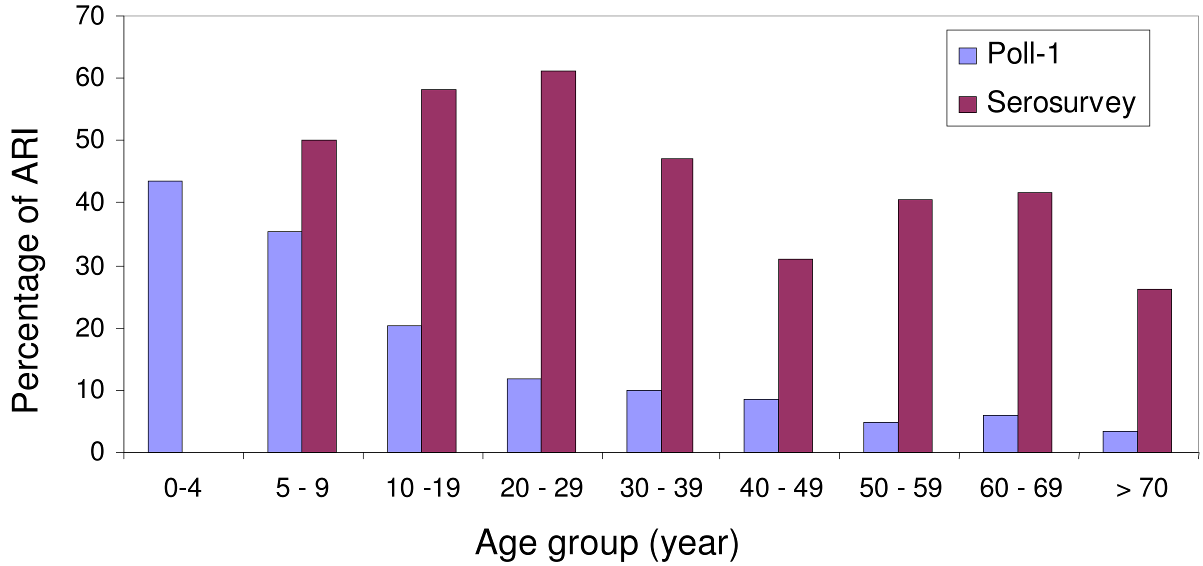

Figure 1. Prevalence of acute respiratory illnesses (ARI) by age-group, community based survey. (Poll-1) in October 2009 (n=3,351) and sero-survey in December 2009 (n=222).

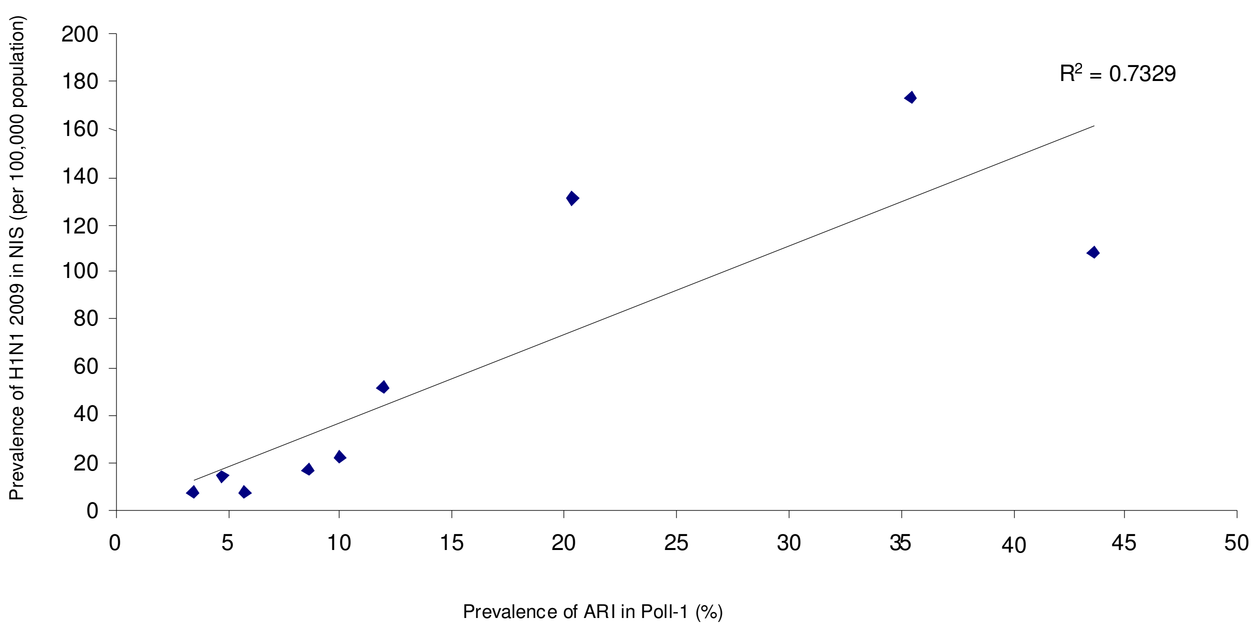

Figure 2. Correlation between prevalence of ARI and prevalence of reported pandemic H1N1 2009 infections by age group in 4 provinces of Thailand, July - September 2009.

Nakhonsrithammarat city in December 2009. The cumulative incidence of ARI increased to 43%, but the incidence of ARI during the 2-week period prior to the survey was 16%.

Antibody detection

In the second survey, we performed serologic tests for antibody to the influenza A (H1N1) 2009 targeted to 246 subjects. Only 222 (90%) subjects from 40 households in ChiangMai city and 44 households in Nakhonsrithammarat city had blood samples taken. Among the 222 subjects, a total of 20 serologically positive cases were detected with HI and MicroNT assays, and the proportion of serologically positive subjects was 9% (95%CI: 5.6 to 13.6%). Among the 96 ARI cases, the proportion of serologically positive cases was 14.6% (95%CI: 8.2 to

Table 2. Characteristics of respondents in sero-survey for a novel influenza H1N1, Thailand, December 2009

Factor	All respondents (n=222)	ILI cases (n=49)	ARI cases (n=96)	Seropositive cases (n=20)
Study area				
- ChiangMai city (%)	110 (50)	23 (47)	52 (54)	11 (55)
- Nakornsrithammarat city (%)	112 (50%)	26 (53%)	44 (46%)	9 (45%)
Median of age (years)	45 (range 5 - 89)	35 (range 7 - 70)	39 (range 7 - 89)	12 (range 7 - 57)
Male : Female	0.57 : 1	0.75 : 1	0.58 : 1	0.43 : 1
Education				
- Primary school (%)	86 (39)	17 (35)	32 (33)	12 (60)
- Secondary school (%)	73 (33)	16 (33)	29 (30)	4 (20)
Had history of any chronic illness (%)	85 (38)	16 (33)	40 (42)	4 (20)
Type of house				
- Separated house (%)	154 (69)	32 (65)	64 (67)	13 (65)
- Dormitory / Apartment (%)	64 (29)	17 (35)	32 (33)	7 (35)
Got seasonal influenza vaccine within 6 months (%)	11 (5)	0	3 (3%)	1 (5)

23.3%). Among 126 non-ARI cases, the proportion of serologically positive cases was 4.8% (95%CI: 1.8 to 10.1%).

The demographic findings for all respondents with reported ARI cases, and with sero-positive cases are shown in Table 2. None of the 222 individuals had a history of pneumonia, hospitalization, and Oseltamivir administration. Only 10 (4.5%) individuals had a history of seasonal influenza A vaccination in the previous year, and 162 (73%) of them stated that they would like to get a novel influenza A (H1N1) vaccination. Age-distribution of the sero-positive cases indicates that children and young adults were most affected by the virus. Subjects living in households with more family members had higher of seropositivity rates, indicating the importance of intra-household transmission. Among 20 case families, 11 families had 1 case each, 3 families had 2 cases each, and 1 family had 3 cases. Among the 11 families with 1 case each, the mean attack rate was 30%. Among the other 4 families, the mean attack rate was 58%. The difference in seropositivity rates among the areas is consistent with the survey data (Table 2). Using serologic data as a gold standard, our survey showed that ARI has a sensitivity of 70%, a specificity of 59%, and a positive predictive value of 15%.

DISCUSSIONS AND CONCLUSIONS

In Thailand, the first wave of the 2009 H1N1 influenza pandemic peaked in mid-July, and the low level of transmission activities lasted until October 2009. During late December 2009 to early January 2010, the rising trend in the influenza pandemic signaled an upcoming second wave of the pandemic in Thailand (Figure 3). In all the studied areas, the outbreaks subsided to an insignificant level by the time blood samples were obtained. So the seroprevalence rate reflected the cumulative incidence of the novel H1N1 influenza infections toward the end of the first wave.

It is not clear why the epidemic declined with herd immunity levels of as low as <10%. Behavioral changes caused by increased awareness and public campaigns may have contributed to this pattern (Neumann et al., 2009; Wiwanitkit, 2009). Weather changes, including reduced rainfall, which usually synchronizes with reduction in seasonal influenza outbreaks, may have also played a role. Another possibility is that the H1N1 pandemic may not have been able to sustain itself in the general population but required continuous sources with higher transmission rates and reproduction numbers of greater than one within the subpopulations. With enough herd immunity within these subpopulations, outbreaks might have been interrupted. Our data that show higher sero-positive rates among children support this hypothesis. However, pharmaceutical and non-pharmaceutical preventions and controls were implemented countrywide during the first pandemic wave. The different levels of saturated infection and immunity in different areas may reflect different timing of deployment of interventions, as areas with delayed onset of outbreak could start interventions earlier in the outbreak, or they may reflect different contact rates and transmission rates, thereby required different levels of herd immunity to stop

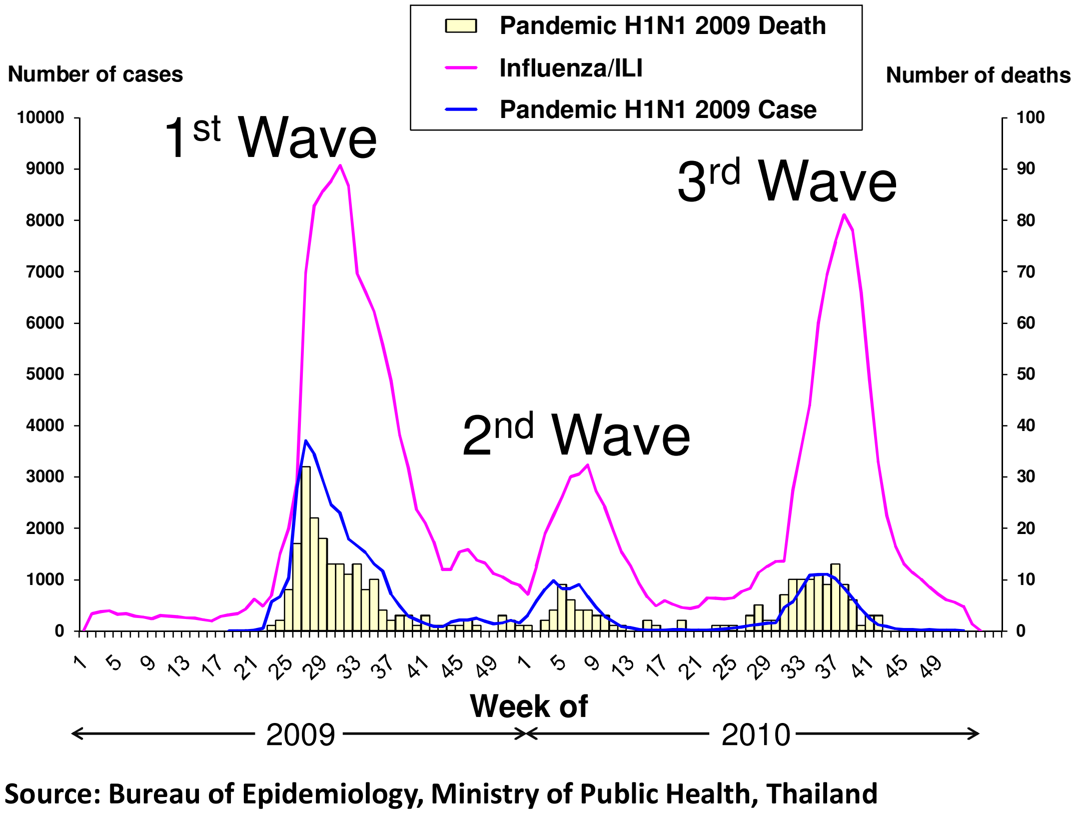

Figure 3. Three waves of the 2009 H1N1 influenza pandemic in Thailand. Source: Bureau of Epidemiology, Ministry of Public Health. Thailand.

an outbreak.

Our study showed that the rural areas had smaller proportion of ARI cases than cities did. This finding seems to support the pattern of a spread of pandemic influenza from cities to rural area in the early phase of pandemic, and similar patterns have been observed in many countries (Hien et al., 2009; Lopez-Cervantes et al., 2009; Yasuda and Suzuki, 2009). However, the different perceivable to ARI symptoms between city and rural people and the survey without serologic confirmation in countrywide were not allowed to conclude the spread of pandemic influenza A (H1N1) 2009 from cities to rural area. In addition, we found inadequacy of non-pharmaceutical prevention measures in the families that reported at least one ARI case. Therefore household transmission of influenza virus was inevitable, and sick children served as effective spreaders in families (Appuhamy et al., 2009; Health Protection Agency, 2009).

The limited sensitivity of a questionnaire to detect pandemic influenza infection was likely due to asymptomatic infections or mild infections, which may have been discountenanced. In an outbreak in a military camp in Thailand, in which all subjects were tested for H1N1 pandemic influenza-specific antibody response, we found an asymptomatic infection rate to be just under 30% of all reported infections (Wattanasak, 2010). Nevertheless, survey results are predictive enough to show good correlation with data in official reporting system. This gave a rather constant ratio of extrapolated total cases to reported cases of 275:1. This ratio is very useful for estimation of total cases and of the impact of the outbreak from the existing reporting system all over the country. This information is important for policy makers and for strategic implementation of outbreak control measures.

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