Investigation and analysis of a cluster event of enterovirus infection in a kindergarten in Hsin-Chu County Hu-Kou Village

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Abstract

On Jan. 23 and 25, 2008, 2 suspected cases of enteroviruses infection with severe complications were reported to the Second Branch of the Department of Health Center for Disease Control. During disease surveillance, the 2 cases were found to live in the same area and had a common suspected infection source, a kindergarten which led to further investigation.

This investigation was to understand the cluster event in the kindergarten in order to avoid an outbreak of secondary cases of enteroviruses infection with severe complications in students or their family members.

A total of 29 symptomatic cases (including the first reported cases) were found in the surveillance. A total of 19 viral throat swabs and 11 stool samples were gathered. The results showed that all of the 19 viral throat swabs were negative and among the 11 stool samples, 7 cases were confirmed to be infected by enterovirus 71. These results show that a cluster infection of enterovirus 71 did

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occur in the kindergarten during the time of surveillance.

The first 2 cases were concluded to have a direct relationship, but each had the infection separately at home. This surveillance showed that the cluster event infection source was the basement; transmission was through student contact which spread to their family members and then back to the school.

As for disease prevention, symptomatic cases were quarantined at home with follow up services done by phone by the Health Bureau. Monitoring continued for 2 weeks after they came back to school. The level of disease prevention was raised to the "community" level to control the disease in order to prevent secondary severe complications from appearing.

Keywords: enterovirus cluster infection, enteroviruses infection with severe complications, herpangina, class suspension measurement

Introduction

Enteroviruses belong to the family *Picornaviridae*, with Enterovirus Type 71 causing the most neurological complications besides poliovirus. The virus was first isolated in California, USA in a 1969 endemic which caused multiple cases of aseptic meningitis and encephalitis [1]. Enteroviruses can cause many diseases while most (about 50-80%) infected people are asymptomatic; with some experiencing a mild fever or flu-like symptoms. In a few cases, symptoms such as hand-foot-mouth disease, herpangina, aseptic meningitis, viral encephalitis, limb paralysis syndrome, acute hemorrhagic conjunctivitis, and myocarditis appear [1,2].

Pandemics have occurred in Taiwan in 1998, 2000, and 2001. The one in 1998 was the most severe, leading to 78 deaths with about 90% of them younger than 4 [1-5], and general panic among parents. In the last 10 years, the academia,

doctors, and health authorities have gain more insight into the transmission, treatment, and prevention of enteroviruses. Previous studies have shown that age is a risk factor of enterovirus type 71 infection, the younger the age the higher the infection rate [3,6]. Infants younger than 1 have the highest incidence of severe complications and a mortality rate between 10.0% and 25.7%. Since young infants can not care for themselves, immature immune system, and no protective antibodies, they are easily infected by Enteroviruses leading to high incidence and mortality rate [7]. The main focus of infectious disease prevention of Enteroviruses is in young children since they are the high risk group of developing severe complications and death.

Transmission of Enterovirus is through the gastrointestinal tract (fecal-oral, water, and food contamination), respiratory tract (aerosol, coughing, or sneezing), contact with a patient's secretion, or liquid from skin blisters [3, 6, 8]. Children are often infected by touching secretions, blister fluid, asymptomatic carriers such as infected siblings or adults, hands of care givers, and ingestion of contaminated food. With toys, another common vector, transmission occurs when children bite the infected object [3, 5].

Enterovirus has a very high transmission rate in households. Previous studies have shown that the transmission rate in a household is 52%, 84% among siblings, 41% from the parents, and 28% from grandparents [3]. Another significant risk factor is contact with symptomatic patients expressing cases of hand-foot-mouth disease and herpangina [5]. Evidence suggests that children with an enterovirus infection should be separated from siblings to avoid secondary infection. Studies have shown that the major areas of transmission among pre-school kids are through households and schools, especially kindergartens [5]. Therefore households and kindergartens are the key locales of health education and disease

prevention.

According to the monitoring results of Department of Health Center for Disease Control, there have been no outbreaks of disease for 2 years. The surveillance by the CDC in Tao-yuan in 2007 showed that no antibodies were found in children younger than 3 years old. Enterovirus activity seemed to increase after October 2007 which caused severe complications in southern cities and counties. Suspecting large scale outbreaks this year, the northern area quickly prepared itself.

Occurrence and report of this cluster event

The second branch of the Department of Health Center for Disease Control received reports concerning 2 suspected cases of enteroviruses infection with severe complications on Jan. 23 and 25, 2008, respectively. After surveillance, we found that the 2 cases were living in Hu-Kou Village, Hsin-Chu County. Although they were not in the same kindergarten, a geographic relationship was suspected because their houses were only 1,048 meters apart. Direct contact transmission was ruled out when preliminary surveillance found that Case 2 never went to school or daycare. We suspect that both cases were infected in their households since there were cases of herpangina in both homes before the onset of enterovirus. We also found that a younger sister of Case 1 and a cousin of Case 2 were in the same kindergarten. Since both cases were confirmed to be enterovirus type 71 infections with severe complications, surveillance and prevention were required to prevent a second wave of cases with severe complications.

Demographic data of the two cases of enterovirus infection are summarized in the following:

Case 1: 5-year-old, male, attended a class for 5 years old in a kindergarten,

symptoms appeared on Jan. 17, reported in on Jan. 23, confirmed to be a case of enterovirus type 71 infection on Jan. 31.

Case 2: 8-month-old, male, never went to school or daycare, symptoms started on Jan. 20, reported in on Jan. 25, confirmed to be a case of enterovirus type 71 infection on Jan. 28.

Goal of the surveillance

In order to establish a connection between the 2 cases in terms of people, timing, and location and assess the risk and potential level of an outbreak, the CDC conducted a surveillance on Feb. 1 with the following goals:

1. To understand the outbreak in the kindergarten.
2. To understand the level, potential routes of transmission and infection sources.
3. To prevent students and their family members from getting a second wave of enterovirus infection with severe complications.

Material and methods

The kindergarten

The kindergarten is a 5-floor spacious building with independent toilets in each classroom located in Hu-Kou Village, Hsin-Chu County. There were a total of 9 classes organized by the age of the child and 168 students; 65 students in 3 classes for 6 year olds, 59 students in 3 classes for 5 year olds, 30 students in 2 classes for 4 year olds, and 14 students in 1 class for 3 year olds and younger. The classes for 5 year olds were on the first floor, the classes for 6 year olds were on the second floor and the classes for the 4 year olds and the 3 and under were in the basement.

Activities of each class were independent from each other. They had their own class schedule, lunch time, nap time in their own classroom, and shuttle

buses.

Subjects of the surveillance

All students in the kindergarten of the case 1

Surveillance period

We tried to find the suspected first case in the kindergarten 10 days (2 incubation periods) before the onset of Case 1 (Jan. 18). Hence, the surveillance period was from Jan.8 to Feb. 1 (the first day of the surveillance).

Definition of symptomatic patients

Those who had absences due to illness, symptoms of flu, or suspected enterovirus infection during the period of surveillance were examined by doctors one by one. Those who confirmed to have suspected symptoms of infection were defined as symptomatic patients in this surveillance.

Surveillance tools and methods

To confirm the cluster event in the kindergarten, doctors performed examinations and gathered samples on the day of surveillance [9]. Students who were symptomatic during the period of surveillance were checked one by one with the help of teachers. We checked symptoms including fever, rash, oral ulcer, cough, sore throat, running nose, malaise, and anorexia along with dates of onset, sick leaves, medical interventions, diagnoses, and infections of family members. Samples gathered included viral throat swabs and/or stool samples. Throat swabs were gathered from symptomatic patients and stool samples were gathered from those who had recovered, or had symptoms for more than a week. The Health Bureau took stool samples from children who were absent due to illness.

Laboratory examination

Viral throat swabs and/or stool samples were gathered from symptomatic cases during the surveillance period in the kindergarten of the case 1 since Feb. 1.

Samples were sent to Kun-Yang Laboratory for RT-PCR and virus isolation. Positive cases were confirmed if either of the tests came out positive [10,11]. Virus types were also identified.

Results

Relationship of cases in the kindergarten

Surveillance found that prior to first case, the younger sister of case 1 started showing symptoms on Jan. 12. She was in the 3 years and under class temporarily before the onset of the symptoms, and left soon afterward. The second case that occurred prior to Case 1 and 2 was in the class of 4 year olds whose symptoms started on Jan. 14 and is cousin of Case 2. Case 1 was the third case, exhibiting symptoms on Jan. 17, and was in the class of 5 year olds. Case 2 had symptoms appear on Jan. 21 but he did not attend school.

Disease surveillance and its scope

168 subjects were involved in this surveillance, 29 cases having identifiable symptoms of an enterovirus infection. Among them, 28 were mild cases and 1 was reported as a case of enterovirus infection with severe complications. Doctors checked and gathered samples from 19 cases while the other 10 cases, who were absent due to illness, were followed by the local Health Bureau. Symptoms of the 19 cases were mainly flu-like symptoms, including running nose (73.7%, 14/19), cough (68.4%, 13/19), fever (31.6%, 6/19), throat infection (31.6%, 6/19), and hand rash (10.53%, 2/19).

Among the 29 cases with symptoms of enterovirus, 6 were in the class of 6 year olds, 10 in the class of 5 year olds, 8 and the class of 4 year olds, and 5 in the 3 year old and under class. The infection rates were 9.23%, 16.95%, 26.67%, 35.71%, respectively, with an overall infection rate of 17.26% (Table 1).

Table 1. Distribution, sample gathering, and infection rates of suspected enterovirus cases in a kindergarten in Hsin-Chu County

Classes		6 year olds			5 year olds			4 year olds		3 year olds and under	Total
		A	B	C	A	B	C	A	B		
Floor		2F			1F			B1		B1	
Students		22	21	22	20	21	18	15	15	14	168
Symptomatic students (Total)		1	2	3	5	4	1	5	3	5	29
		6			10			8		5	
Sample number	Throat swabs	1	1	2	3	4	0	4	1	3	19
	Stool	0	0	1	2	1	1	3	2	1	11
Infection rate		9.23%			16.95%			26.67%		35.71%	17.26%

As to the distribution on floors, 13 (44.83%) of the cases were in the basement, followed by 10 (34.48%) of the cases on the first floor, and 6 (20.69%) of the cases on the second floor. The infection rates were highest in the basement (29.54%) followed by the first and the second floor.

Thirteen (44.83%) of the cases were male, and sixteen (55.17%) were female.

Samples gathering and examination results

A total of 19 viral throat swabs and 11 stool samples were examined. All 19 viral throat swabs processed by RT-PCR were negative. Out of 11 stool samples, 5 were cultured positive for enterovirus type 71 and 5 were found positive by molecular analysis. A total of 7 cases were confirmed to be enterovirus type 71 infections (Table 2).

The examination results confirmed that there was a cluster outbreak of Enterovirus type 71 in the kindergarten.

Table 2. List of cases in the cluster enterovirus infection in a kindergarten in Hsin-Chu County

No	Classes	Gender	Date of onset	Samples	Date of sample gathering	Examination results
1	3 years old and under class Younger sister of No. 3	F	97/01/12	Throat swab	×	×
				Stool	97/02/05	Negative
2	4 year olds class(B) Cousin of Case 2	M	97/01/14	Throat swab	×	×
				Stool	97/02/05	Enterovirus type 71
3	5 year olds class(B) Reported Case 1	M	97/01/17	Throat swab	97/01/23	negative
				Stool	97/01/24	Enterovirus type 71
4	4 year olds class (A) Younger sister of No. 7	F	97/01/21	Throat swab	×	×
				Stool	97/02/07	Enterovirus type 71
5	4 year olds class (A)	M	97/01/23	Throat swab	97/02/04	negative
				Stool	97/02/04	Enterovirus type 71
6	4 year olds class (A)	F	97/01/25	Throat swab	97/02/04	negative
				Stool/molecular biology	97/02/02	Enterovirus type 71
7	6 year olds class(C)	M	97/01/28	Throat swab	×	×
				Stool	97/02/05	Enterovirus type 71
8	4 year olds class (B)	F	97/01/29	Throat swab	×	×
				Stool/ molecular biology	97/02/02	Enterovirus type 71

Note: Case 2 did not enter school, so he was not listed in this table.
×: not gathered.

Disease prevention measures

After local surveillance and disease prevention meetings in the kindergarten, the following preventive measures were undertaken:

1. The Health Bureau continued monitoring the outbreak in the kindergarten for new cases, giving daily reports even through the Chinese New Year Vacation (Feb. 4 ~ 11).
2. The Health Bureau monitored all symptomatic children through telephone

everyday for 14 days (till Feb. 18). Uninfected siblings were monitored as well to prevent family cluster events and cases with severe complications. Results: a total of 32 cases were found to be positive for flu or suspected enteroviruses infection during Jan. 28 ~ Feb. 4. While one was diagnosed with hand-foot-mouth disease, the rest were diagnosed as having a common cold. These children were quarantined at home with samples taken from them with test results being negative. Up until February 25, no new cases of enterovirus infection with severe complications were found.

3. The kindergarten stopped large scale activities including the graduation ceremony. Health monitoring continued 14 days after the Chinese New Year Vacation to prevent a second wave of outbreak. Up until Feb. 25, 6 children were on sick leave and no new cases of enterovirus infection with severe complications were found, showing that the outbreak was limited. The 6 cases recovered and were released from quarantine.
4. Disease prevention measures were raised to the “community” level to monitor the outbreak. The Health Bureau actively surveyed 34 kindergartens in the Ko-Ho Village concerning sick absences because of symptoms of enterovirus infection. A total of 2,212 children were followed with only 1 child diagnosed with enterovirus infection on Feb. 18. Others had no suspected symptoms. Up to late May, there were no new cases of Enterovirus with severe complications found in the Hu-Kou Village, showing that the preventive measures were effective.

Discussion

Suspected transmission route and infection sources

A geographic relationship was suspected since the two reported cases were

living in Hu-Kou Village, Hsin-Chu County with their houses only 1,048 meters apart. Preliminary surveillance found that Case 2 had never been to school or a daycare center, thus transmission through direct contact between the 2 cases was ruled out. However, when the health status of their siblings was monitored, we found that symptoms of both cases started after other family members had herpangina. Therefore we think that the two cases were infected in their house. We also found that a younger sister of Case 1 and a cousin of Case 2 attended the same kindergarten, which led us to suspect that the kindergarten is the common source of the infection. This led us to believe that there might be an ongoing outbreak of enterovirus type 71 in the kindergarten.

However after the CDC surveyed the environment and looked over the different schedules of each class in the kindergarten, we saw that the building was spacious, each class had their own floors and toilets, and meals were eaten separately in their own classroom. We then ruled out direct transmission between Case 1 and the cousin of Case 2, and suspected that there might be other sources of indirect infection. With further investigation in the kindergarten we found that although the 3 years old and under classes and 4 years old classes were not beside each other and had no common activities, they had lunches in the same room. We also found that students were not separated when they had lunches, but crowded together in the dining room. The dining room was small and in a poorly ventilated basement. Since the basement had the most symptomatic cases, it is likely that the dining room was the common infection source. After cross comparison we found that acquaintances of the two reported cases, younger sister of Case 1 in the 3 years old and under class and a cousin of Case 2 who was in the 4 year old class, were the earliest cases in the kindergarten. We suspected that the dining room in the basement was the common infection source, with the

acquaintances of the cases as the first wave of infection followed by Case 1 and Case 2 becoming infected in the households.

The epidemiological curve of this event, following the incubation period, showed three waves of outbreaks occurred in the 3 year old and under classes, followed by the 4 year olds, 5 year olds, and finally the classes of 6 year olds, suggesting the transmission route in the kindergarten (Fig. 1). We think the outbreak started from the basement, and disseminated from the 3 year old and under and 4 year olds class.

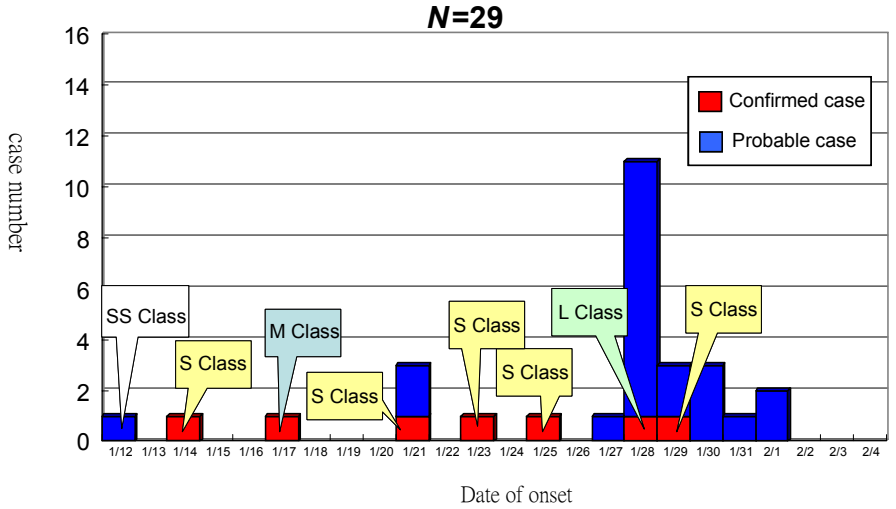

Fig 1. Epidemiological Curve of cases of suspected enterovirus infection in a kindergarten in Hsin-Chu County

This outbreak had a positive sample rate of 63.64% with all the viruses classified as type EV71. The analysis results [8] showed that the kindergarten had an EV71 cluster outbreak, which confirmed our hypothesis. Positive stool sample rates were highest in the basement (45.46%, 5/11), more so than the first floor

(9.09%, 1/11) and the second floor (9.09%, 1/11), showing the basement was the main infection focus.

As we further investigated the infection source for positive cases on the first and second floor, we found that they all had younger family members in the 4 year olds and 3 year olds and younger classes who had the disease first with positive samples., We deciphered that the basement was the infection source transmitting the disease into households and then back to the kindergarten which lead to this enterovirus outbreak. The results reflect in the epidemiological curve, which showed three waves of outbreak in the 3 years and younger classes, followed by the 4 year olds classes, 5 year olds and then 6 year olds classes. The results further showed that friends of the 2 reported cases were the first waves of infection, followed by the second wave in the households, and the third wave back in the kindergarten.

Previous studies have shown that the virus can be isolated 2 weeks after infection from the throats of the patients. The viruses can also pass through the intestinal tract between 6 to 8 weeks and even up to 12 weeks [12]. In this survey, we were not able to find positive throat swab samples even from cases having positive EV71 stool samples. This is contrary to previous studies that had a positive throat swab rate of 90% [1,3,13] which suggested that throat swabs are most effective during outbreaks [9]. The reason for the difference might be that throat swabs taken in this surveillance was taken more than 7 days after onset of the disease. Since many cases had no acute symptoms, the viral levels might be significantly decreased. On the other hand, our positive stool rate was 63.64%, higher than the previous studies' (32%) [1,3,13]. This can also be explained by the fact that the patients released more viruses long after the onset of the disease. Meanwhile, our study showed that stool samples are also important during disease

surveillance.

This study also showed that the timing and type of sampling correlate with the progression of disease; such that correct samples should be taken in order to confirm outbreaks [9].

Since cases of enterovirus infections can continuously shed viruses from the respiratory and intestinal tracts, isolation of infected cases is vitally important for prevention of the disease. Stools and oral/nasal secretions of cases should also be handled with great care. Personal hygiene, including frequent hand washing and reduced hand to mouth/nose contact, should be emphasized to prevent the virus from spreading to other people.

Previous studies have shown that children younger than 3 years old, especially one to two years old, are more likely to be infected by enterovirus due to the disappearance of passive antibodies and a lack of active antibodies. They also have higher chances of getting severe complications. The younger the age, the more serious the disease and the poorer the prognosis [3,5-7]. Previous studies have also shown that the second case in the family is more likely to have severe complications. Similar results were found in the two cases in this study. Hence, children infected by enterovirus should be isolated to prevent younger children in the family from infection.

Limitation

In this study, most cases had only one sample (throat swab or stool samples). Since the throat swabs were taken on average 7 days after the onset of the disease and we didn't have stool samples to help confirm infection, this study might underestimate the infection rate. Besides, asymptomatic or mild symptomatic adults might also be infection sources [3]. Hence, we could not know from this

study whether the original infection source was from adults or asymptomatic children in the school or families.

Conclusion

This outbreak was an event in which two cases of enterovirus infection with severe complications were infected as second cases in families from family members who were infected at the school. This study also confirmed an outbreak of enterovirus occurred in the kindergarten during the surveillance period. The surveillance also showed that the infection started in the basement and spread from the school to households, followed by infection of other family members and then transmitted back to the school to cause this cluster event.

The CDC started surveillance on Feb. 1 in the kindergarten, assisted disease follow-up, suggested disease prevention measures including quarantine of symptomatic patients, follow-up by phone by the Heath Bureau during the Chinese New Year holidays, continuous follow-up for 2 weeks after coming back to the school, and raising the prevention measures to the “community“ level. No new case was reported after these measures, showing the effectiveness of disease prevention.

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