

## Molecular Epidemiological Investigation of Childhood

### Rotavirus Infection in Taiwan between 2004 and 2006

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**Abstract:** from Chinese version, pp,448-460

from Chinese version, pp,448-460 Rotavirus is the major cause of severe diarrhea in childhood. It mainly infects children. Once infected, children will experience vomiting and severe watery diarrhea (>10 times/day), which can easily resulted in dehydration, electrolyte imbalance, acidosis, seizures or even death. This study analyzed hospitalized children with acute gastroenteritis referred from outpatient departments or emergency rooms. Out of the 820, 801, and 1946 stool specimens collected from April 2004 to 2006, Rotavirus was identified in 14.1%, 18.73% and 23.7% respectively. Most cases occurred between Novembers to May in the following year, with peak season in March. RT-PCR showed the prevalence of serotypes of rotavirus group A VP7 G (2004/2005/2006): G1 (11.9%, 23.4%, 34.4%), G2 (35.9%, 17.6%, 6.3%), G3 (26.1%, 19.1%, 34.4%), G4 (0, 0.68%, 0), G9 (26.1%, 21.7%, 24.6%). A new strain, G12, was identified this year. Severe diarrhea or vomiting was often observed clinically with up to about 90% of patients requiring hospitalization. Keywords: rotavirus, acute gastroenteritis, molecular epidemiological analysis, G genotyping.

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causing the collective outbreak of diarrhea and vomiting among the residents of this severely handicapped care center. Since 22 out of the entire 48 residents exhibited symptoms that met the case definition, the attack rate was estimated at 45.8%. Besides, only one employee was detected having norovirus, i.e. an attack rate of 4.0%. After ruling out some point-source exposure possibilities, such as sharing drinking water or food, transmission from one particular person as a common source, and through a common household utensil, we have to conclude that the transmission was likely to have occurred in the person-to-person mode. However, our effort to identify the infection source through tracking down the personal, temporal, and geographical connections between the individual patients has been in vain. As to the countermeasures implemented in this case, the care center started on September 28 to quarantine the sick residents, restrict the movement of the its workers while performing their routine duties, and conduct a blanket sterilization program of the living quarters as well as all facilities on the premises. The outcome appeared to be a success as no more individuals have fallen sick since October 1. Four days later, on October 5, the epidemic control measures were lifted.

Key words : Norovirus, outbreak, person-to-person transmission.

### Introduction

Norovirus infection is also known as stomach flu or winter gastroenteritis, which suggests that it is very active during the winter season [1]. Outbreaks often occur in various densely populated places or institutions such as nursing homes [2-3], hospitals [4-5], mental institutions [6], homes for the disabled and handicapped [7], and long-term care centers [8]. Norovirus infection usually leads to some mild symptoms, such as diarrhea, stomachache, nausea, vomiting, fever, etc., and most patients will recover by themselves without the need of any medical help [9]. The spreading route of norovirus may either follow a person-to-person mode [10-11], or through some sort of point-source exposure such as sharing drinking water [12-13] or food [14-15]. This article is essentially an investigation report by a joint task force of the local health authorities and Taiwan CDC on a cluster outbreak of norovirus infection among the residents and staff of a severely handicapped care center located in Taipei City.

This particular severely handicapped care center was founded in July 1997 and its business management has been entrusted to Taipei Mental Rehabilitation Association. Anyone wishing to become a resident of this care center needs to meet the criteria of having been a resident of Taipei City for at least 6 months, being at least 15 years old, having a Registration Card for People with Mild to Severe Disabilities, having been in stable condition and referred by a physician, and having not contracted any infectious diseases according to the applicant's latest physical examination report. The maximum number of residents the center can accommodate is 50.

It was the morning of September 26, 2006, when the severely handicapped care center notified the local health authorities of a few residents simultaneously exhibiting

the symptoms of diarrhea, vomiting, and slight fever. According to the epidemiological correlation status of the people involved and the timing and locality of the event, it was decided that the care center situation met the criteria of a cluster infection case, and it was necessary for the health authorities to conduct a full scale epidemiological investigation right away. The objectives of such investigation include estimating the epidemic scale of the incident, finding out the transmission route, identifying the pathogen responsible for the outbreak, searching for the source of infection, providing recommendations on appropriate control measures and finally assessing the effectiveness of the implemented measures afterwards.

#### **Case definition**

Any staff member or resident of the severely handicapped care center who, during the period from September 20 to 30, 2006, showed diarrheal symptoms at least three times a day, or experienced vomiting or diarrhea at least twice a day in addition to suffering from stomachache or slight fever, was defined as a case of this outbreak of diarrhea and vomiting.

#### **Scale of the outbreak**

At the time the outbreak took place, this particular severely handicapped care center had 25 employees (including 1 director, 1 general manager, 1 social worker, 2 stationed teachers, 2 Substitute Services draftees, 5 nursing staff, 11 service personnel, and 2 cleaners) and 48 residents. All these 73 individuals became the subjects of our investigation after the diarrhea and vomiting outbreak was reported. Among them, 22 individuals who exhibited symptoms that met the above-mentioned case definition were all residents, and they represented 45.8% of the total resident population. The most common symptoms among the patients were, in descending order, diarrhea 95.5% (21/22), fever or slight fever 77.3% (17/22), and vomiting 18.2% (4/21). Besides, there was one staff member who, in spite of not being responsible for providing direct care to the residents and not showing any symptom, had norovirus detected in his stool sample. The attack rate among the 25 staffs was then estimated at 4.0% (1/25).

#### **Determination of the pathogen**

The investigation team collected 17 stool specimens from the symptomatic residents and had these specimens sent to Laboratory Research and Development Center of Taiwan CDC for laboratory analyses. Those samples went through a standard RT-PCR assay and norovirus was detected in 15 samples. Based on the symptoms these 15 patients displayed, we were confident that norovirus was the pathogen of this collective diarrhea and vomiting outbreak among the residents of this severely handicapped care center. Aside from that, the investigation team also collected rectal swabs and stool specimens from 12 asymptomatic employees of the center and had the specimens sent over to the Laboratory Research and Development

Center. Norovirus was detected in one of the stool specimens, and this sample was gathered from one administrative employee who normally had no direct physical contact with any of the sick residents.

### **Transmission route**

When the dates of symptom onset were plotted into an epidemiological curve, (see Figure 1), we noticed the resulting curve had one peak only, which suggested single point-source exposure as a likely transmission route for this outbreak.

However, after we investigated several common factors that are normally in connection with point-source exposure (such as air, drinking water, food, common person, and common utensil shared by all involved), we realized that such a single-peak prevalence curve could not be interpreted as a definite consequence of point-source exposure.

The rationale is that the symptoms surfaced were mainly gastrointestinal ones, but the attack rates of the residents and staff were very different, 45.8% and 4.0% respectively. Such a huge gap between the two can convincingly rule out the possibility of air being the medium of the point-source exposure. Secondly, the drinking water consumed at this particular care center is just common tap water, and no special alteration or treatment had been applied to the drinking water system at the place before the outbreak. Therefore, we can also safely rule out the connection between the drinking water and the point-source exposure. On the other hand, if the possible point-source exposure was through some unclean food item(s), then the number of individual cases showing up on the same or the next day after the tainted meal was served would not have been just one (we know the incubation period of norovirus happens to be 1~2 days [9]). It is shown in Figure 1 that 6 and 7 persons fell sick on the third and fourth day of the outbreak respectively, which effectively ruled out a possible connection with any food stuff shared by people at the care center.

Another strange phenomenon we noticed in this case is that although the residents' bedrooms were assigned to and taken care of by different service workers, those having fallen sick came from all bedrooms (see Figure 2) while none of the care takers of these rooms got sick, nor was norovirus detected in their stool specimens when assayed. Therefore, we can safely cross out the possibility of a certain staff being responsible for spreading the germ and started the outbreak. Although no residents at this particular care center had their own tableware and drinking utensils, given that none of the staff responsible for distributing or cleaning tableware and water tumblers showed any symptoms of the sickness, the possibility of certain meal utensils or drinking water containers being the common source of infection can also be ruled out.

After we had tried but failed in proving the above mentioned factors as having led to point-source exposure, we believed it was just another case of group infection as demonstrated by several previous incidents that showed similar single-peak

prevalence curves but later proved to be a case of person-to-person transmission [5-7]. This phenomenon stems from a combination of two facts: i.e. the incubation period of norovirus is very short, only 1-2 days, and the ability of the virus to transmit from one host to another is so strong that as few as 100 virus particles can effect an infection in a person. Figure 2 shows the floor plan of the care center along with the distribution of the sick residents and the onset date of each individual involved. All these facts made us more certain that this group outbreak of diarrhea and vomiting indeed followed a person-to-person mode as we suspected.

#### **Deduction of the source of infection**

September 23, 2006 happened to be a parent's day at the care center and the day of symptom onset for the first individual case among the residents became sick with diarrhea and vomiting. On that day, this sick resident's mother and older sister came to the care center to pay him a visit. And within a couple of days after they returned home, they also started to have diarrhetic and vomiting symptoms. In view of the sequential order of these events, it's fair to say that they are likely to have caught the germs when briefly visiting the care center rather than they being the source of this infection outbreak.

The investigation team then traced backward and looked into what had happened since four days (twice the length of the incubation period of norovirus) before the onset date of the first symptomatic victim (i.e. September 19, 2006). They checked the use of health insurance cards by all employees of the care center within those four days and found that altogether there were 11 persons who went to see a doctor in that particular period, but only one of them, a female, did consult a physician because of stomach trouble on September 20. However, her trouble as she recollected was about constipation, which was far from diarrhea and vomiting symptoms.

Among the stool specimens collected from 12 asymptomatic employees of the care center, norovirus was detected in only one specimen. We have no way of telling whether this person was the original virus carrier responsible for setting off the incident or just another infected victim of this outbreak. The investigation team also paid visits to the families of the sick residents, but none of them had shown the symptoms of diarrhea and vomiting. Furthermore, although the care center kept a logbook that recorded the date and time of any home visit by the residents as well as any site visit by the residents' family, unfortunately there was no written information about whether the visitors had diarrhea and vomiting symptoms. Another point is norovirus infection usually causes no big harm, and the infected person will recover even without treatment. That is why the investigation team did not collect samples from everyone. Any asymptomatic norovirus carrier is capable of passing on the virus and causing infection in other people [5]. Therefore, it is quite difficult to find out the source of infection for this wave of group outbreak of diarrhea and vomiting.

**Recommendations on control measures**

Effective measures for norovirus infection control and prevention can be divided into three major categories: isolation of symptomatic patients, restraints on the movement of service personnel, and thorough sterilization of the living environment and facilities on the premises.

Since the entire care center is located in a one-storey building, and the six bedrooms in the center were almost fully occupied by 48 multiple handicapped residents at the time the outbreak struck, no extra space was available for quarantine purpose. The best they could do was to designate Bedrooms 1, 6, and 7 as contaminated zones and moved all symptomatic residents into these bedrooms. Then they used desks and chairs to block the entrance to these areas so that asymptomatic residents could be kept off the areas. At the same time, they designated the remaining Bedrooms 2, 3, 5 as clean zones and moved residents without symptoms to these areas. The residents were asked to stay inside these bedrooms and try their best not to mingle with their friends in the social lounge. Also the management of the care center requested their contract food supplier to deliver cooked food in the form of boxed meals, and all residents (whether sick or well) must have their meals inside their own bedrooms to cut down their chances for catching norovirus while eating.

In terms of restraining the movement of the staff members, the center suspended all regular group activities, curriculum arrangements, reception of outside guests and services by voluntary workers. They stopped allowing anyone who was not a current resident or employee to enter the severely handicapped care center and tried not to permit any residents to visit home. For those residents who insisted on going home because they were worried about being infected, the staff must educate their family about norovirus prevention and control and request the residents to contact the care center if symptoms appeared. After a resident went home, the care center would actively contact the family of the resident on a daily basis to keep track of the resident's health situation. All work personnel were encouraged to wear masks, gloves, and disposable clothing at work, and to mark the time in writing whenever they entered or left the contaminated and clean zones, so that the infection source could be traced if further individual cases surfaced. All work personnel were told to monitor their own health condition to see if diarrhetic or vomiting symptoms appeared. Upon experiencing any symptom, the staff must go to see a doctor right away and stop reporting to work at the care center till one week after the symptoms have subsided. Moreover, training on hand washing, hand hygiene, and the accurate way of wearing masks was strengthened at the care center.

With regard to the sterilization of living environment and facilities, the care center placed at its entrance a spray container of 75% alcohol. Anyone entering or leaving the premises was required to sterilize his or her hands with the alcohol spray

to decrease the odds of becoming infected. Since most of the residents were multiple handicapped, the seemingly simple spray was no easy feat for them. Therefore, it was recommended that the work personnel sprayed both hands of every resident with 75% alcohol in the morning and afternoon daily. But there was a catch to watch out for: since norovirus does not have a lipid envelope, alcohol is able to deactivate the virus only with limited efficiency. Therefore, the importance of hand washing cannot be totally replaced by alcohol spray in this case. Aside from that, the center also placed a bucket of bleach water solution (0.4~0.5 ppm) next to the washbasin in the bathroom. Under the staff's supervision, residents must immerse both hands in the bleach solution for at least 30 seconds every time after using the toilet and washing their hands before being allowed to return to their bedrooms. The rationale of this measure is that such bleach solution has a rather strong odor and feels kind of sticky on skin, so it can effectively deter the handicapped residents from putting their hands into the mouth or touching their mouth or nose. Furthermore, in case the residents did not wash their hands thoroughly enough after using the toilet, this tactic of immersion in bleach water would add one more safeguard to diminishing the opportunity of spreading norovirus. One important thing to bear in mind when preparing the bleach was that the solution should not be of higher concentration than the level specified above since more concentrated bleach might cause skin inflammation. After all, bleach is usually used in sterilization of nonbiological equipment. Furthermore, the service personnel of the center were asked to sterilize the premises with diluted bleach water twice a day, i.e. once in the morning and once in the afternoon. They also had to regularly and thoroughly wipe target items including the floor, bathroom, social lounge, doors and windows of bedroom, bed handrails, cabinets, armrests of chairs, railings, tabletops, computers, etc., with disinfectant. It is generally recognized that failure in the sterilization of objects contaminated by patients' excretions or vomits and incomplete environmental disinfection are two major causes of norovirus spreading [16]. According to Taiwan CDC's newly released document "Recommendations on Norovirus Infection Control Measures: A Draft," the suggested concentration of hypochlorite in the bleach water used as disinfectant is 0.1%. Thus, a rather straightforward and appropriate way to prepare such a solution from commercially available bleach, which normally has a hypochlorite concentration of 5.25%, is to dilute the bleach with water to 50 folds of its original volume [17]. It is twice the concentration of the one traditionally used for environmental disinfection, which contains 0.05% or 500 ppm hypochlorite. Since the traditional concentration cannot effectively wipe out norovirus in the environment, people have learnt to double the concentration level to solve the problem. Some people have even tried a bleach solution of 5,000 ppm hypochlorite for this purpose. Finally, the care center also handled and washed the dirty clothing and beddings of the symptomatic residents separately from those of the asymptomatic ones and made sure that the soiled clothing was not placed in the same room where boxed meals were

received and distributed.

After the care center implemented the above-stated control measures, the investigation team reminded the care center to closely monitor the health condition of every resident and staff member every day. If any further case of diarrhea or vomiting surfaced, in addition to sending the case to see a doctor, the care center had to promptly notify the section chief of the Newly Emerging Infectious Disease Section, Joint Office for Disease Control, Taipei City Hospital. This was considered essential because only through monitoring individual cases and reporting new cases, the local health authorities could keep track of the development of the event and understand if the measures taken were effective and whether this collective diarrhea and vomiting outbreak was brought under complete control. The good news is, after these comprehensive countermeasures were carried out, no more individual cases of diarrhea and vomiting showed up at the care center starting from October 1, 2006, and four days later (i.e. twice the length of the incubation period for norovirus), on October 5, 2006, the outbreak control was finally lifted.

### **Conclusions and discussion**

In the course of this collective outbreak of diarrhea and vomiting at a certain severely handicapped care center, the investigation revealed a total of 15 confirmed individual cases of norovirus infection, 7 suspected cases, and 1 asymptomatic positive case. The overall attack rate at the care center was 31.5% (23/73), while the attack rate for residents alone was 45.8% (22/48) and for employees alone was 4.0% (1/25). Since the objective of collecting specimens was to look for the responsible pathogen(s) causing this group outbreak of diarrhea and vomiting sickness, not to identify which resident was a confirmed or asymptomatic case, and given that the cost of detecting norovirus infection was not cheap and legally it was not a notifiable disease yet, the investigation team decided that it was not necessary to collect specimens from everybody at the outbreak site. Of course, a down side of this decision was that it would inevitably lead to an underestimation of the attack rates because they did not include all asymptomatic cases. In this particular norovirus attack, we noticed that among the 22 symptomatic individual cases, in addition to a very high proportion (95.5%) of diarrhea, fever or slight fever also had a rather high percentage (77.3%). This made us wonder if the norovirus causing this particular outbreak of diarrhea and vomiting was not of the same genotype as the virus that had previously led to outbreaks, since a much lower percentage of feverish symptoms was detected in patients in the past [18-19].

Since the arrival of fall 2006, there have been a series of group infection incidents with gastrointestinal symptoms caused by norovirus in Taiwan. Our neighbors such as Japan and Hong Kong reportedly also showed similar trends. Although most symptoms caused by norovirus infection are not that serious, they are still very scary, especially to organizations that has never encountered this disease before. From our



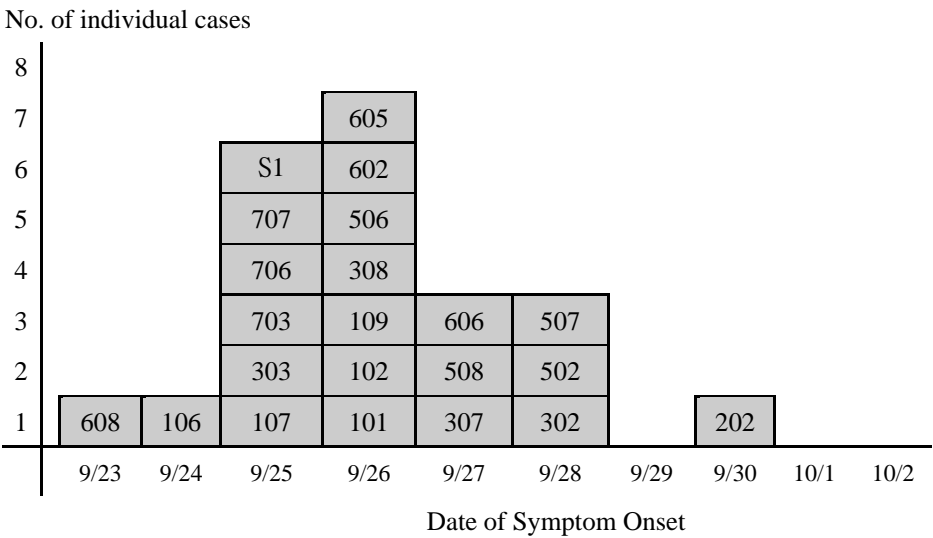
past experiences, most group norovirus infection outbreaks were verified as having been transmitted through a person-to-person mode. Only in a few outbreaks where many individual cases occurred simultaneously at multiple spots but within the same organization would food items, kitchen workers and food delivery personnel come under suspicion as the possible source for the outbreaks. For this very outbreak, we also collected stool specimens from relevant kitchen workers and those who delivered cooked meals to the care center, and we did detect norovirus in some of these specimens. However, since the Bureau of Food and Drug Analysis does not conduct norovirus analysis for samples of leftover food or foodstuffs, we have no way of proving or ruling out the possibility that this norovirus outbreak was caused by consuming certain food items. In other countries, there have been many reports on the detection of norovirus in food items and drinking water [12-13, 15, 20-21], suggesting that we need to strengthen our diagnostic efforts in this respect.

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**Figure 1. Distribution of symptom onset dates of sick persons during the outbreak of diarrhea and vomiting at the care center of the severely handicapped (The number in each box is the bed number of the sick person.)**



**Figure 2. The floor plan of the care center with bed number, onset date, and lab result marked if applicable**

				206	205	106 9/24	105				
				207	203	107 9/24	103				
				208	202	108	102 9/26				
					201	109 9/26	101 9/26				
305	303 9/25	302 9/28	301				S1 9/25	709	708	707 9/25	706 9/25
							S2				
306	307 9/27	308 9/26	309	Social Lounge				701	702	703 9/25	705
				501	509	601	609				
				502 9/28	508 9/27	602 9/26	608 9/23				
				503	507 9/28	603	607				
				505	506 9/26	605 9/26	606 9/27				

Remarks: The top numeral is the bed number. For instance, 102 denotes the 2<sup>nd</sup> bed in Bedroom 1.

The lower numeral is the onset date of symptoms. For example, 9/23 means September 23, 2006.

The gray background means norovirus was detected in the person's stool sample.