A Waterborne Shigellosis Outbreak in a Primary School, Tai-Chung City, November 2007

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Abstract

Shigellosis is mainly transmitted from person-to-person. Contaminated water or foods can also cause large outbreaks. This study was to evaluate the transmission route and to find the possible common source associated with a waterborne outbreak caused by *Shigella sonnei* in a primary school in Tai-Chung City, Taiwan, in November 2007.

We conducted a case-control study of students in the school on November 22nd, 2007. Cases were defined as illness on students who had diarrhea or at least two of the following symptoms: fever, vomiting, tenesmus, or abdominal pain. We used questionnaire included information about demographic data, hand hygiene, type of lunch, drinking water, and school cleaning assignments. Rectal swabs were collected from suspect cases and their contacts. We also inspected the school environment. Stool and water samples were sent for cultures and

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pulsed-field gel electrophoresis (PFGE).

There were 271 suspect cases occurred during the 4 weeks before investigation. A total of 57 confirmed cases were found in the end of outbreak. Poor hand hygiene after toilet was associated with illness (OR: 1.64; 95% CL: 1.07-2.52). Environmental investigation revealed that groundwater for lavatory-use in four of the six school buildings was contaminated by a damaged sewer connection. Students whose classrooms located in the four buildings had higher attack rate in the study. (16.7% vs. 8.2%, $p < 0.001$). The water sample yielded *Shigella sonnei* which was identical to human isolates in PFGE pattern.

Conclusion. It was a shigellosis waterborne outbreak and lasted for another two weeks after carrying out the control measures. It did not spread to the community. Tai-Chung City government then prohibited any groundwater use in schools and increased the budget for them to access piped water system. Water quality examination should also be done in each semester.

Keywords: *Shigella sonnei*, waterborne outbreaks, groundwater

Background

Shigellosis is usually transmitted from person to person in households [1]. However, outbreaks due to contaminated food or water are also seen and it poses public health problems in both developing and developed countries [2-4]. In Taiwan, the incidence of shigellosis decreased in recent 5 years. Most of cases located in mountainous area [5]. In November 2007, Centers for Disease Control, Taiwan (Taiwan CDC) was notified that a primary school in Tai-Chung city had increasing confirmed cases of shigellosis despite of the use of hygiene, case isolation, and antimicrobial therapy. The city has about one million of population and is the largest urban area in central Taiwan. We thus conducted this study to

evaluate the transmission of shigellosis in this outbreak, especially to find the possible common source other than person-to-person transmission.

Method

1. **Case-control study**

   This case-control study was done on November 22nd, 2007. A student with diarrhea or two of the following symptoms (fever, vomiting, abdominal pain, or tenesmus) during the past one month was defined as a case. A control who had none of the above symptoms was selected from the same class of a case. We used questionnaire regarding demographic data, hand hygiene, type of lunch, drinking water at school, and assigned cleansing work at school. The association between the above indexes with shigellosis infection was represented by Odds Ratio (OR) respectively. 95% Confidence Limits (CL) of OR including 1.0 was defined as no statistical significance. Chi-square test was conduct to compare proportions' difference and a significant level of 0.05 was chosen.

2. **Environmental investigation**

   On November 21st, 2007, we inspected sewage system, water system, hand hygiene practice and the kitchen in the school. Most of the students ate lunch prepared by the school kitchen. Water samples were cultured for enteropathogens and tested for residual chloride content.

3. **Microbiology study**

   We collected rectal swabs from all the cases. Rectal swabs and water samples were delivered in Cary-Blair transport medium at low temperature to the Central Taiwan Laboratory of the CDC Laboratory Center. They were processed on the following media: Hekton Enteric and Shigella-Salmonella media for

enrichment, Triple Sugar Iron, Lysine Iron and Sulfite-Indol-Motility media for biochemical identification. Shigella strains were typed serologically by shigella antiserum and biochemically by API20E. (bioMéreux, France).

4. Drug sensitivity test

Antibiotics susceptibility was determined by disk diffusion method. Eighteen antibiotics tested including amikacin (30 μg), ampicillin (10 μg), cefazolin (30 μg), cefixime (5 μg), cefotaxime (30 μg), cephalothin (30 μg), chloramphenicol (30 μg), ciprofloxacin (5 μg), gentamicin (10 μg), kanamycin (30 μg), nalidixic acid (30 μg), norfloxacin (10 μg), ofloxacin (5 μg), penicillin (10 U), streptomycin (10 μg), Trimethoprin/Sulfamethoxazol(1.25 ug/23.75 ugl), teteracycline (30 μg) and tobramycin (10 μg).

5. Pulsed-field gel electrophoresis

Standardized PulseNet method from the US CDC was used for bacteria embedding, lysis and gel washing. Chromosomal DNA was digested by NotI and XbaI and analyzed by CHEF MAPPER (Bio-Rad Laboratories, Hercules, CA, USA). After ethidium bromide staining, gel images were taken by Kodak Electrophoresis Documentation and Analysis System 290 (Kodak; Rochester, NY) and saved as TIFF files. *Salmonella enterica* serotype Braenderup H9812 was used as molecular size marker. PFGE maps were then analyzed by BioNumerics (Applied Maths, Kortrijk, Belgium), saved into the Shigella DNA fingerprint database of the CDC, and compared with other saved maps to identify the possible origin of the strains.

Results

1. case-control study

There were 1,973 students in the school and 1,783 of them finished the

questionnaire (response rate 90.3%). A total of 271 cases were found. The attack rate was 15.2% (271/1,783) in the time window of our study. Most cases presented with diarrhea (88.9%), fever (52.4%), or abdominal pain (51.7%). Only 9 cases had bloody diarrhea (3.3%) and 35 cases had tenesmus (12.9%). Results of risk factor analysis were shown in Table 1. Incorrect hand hygiene habit after toilet was a risk factor (OR: 1.64, 95% CL: 1.07-2.52). Epidemic curve through the end of this outbreak was shown in Figure 1.

Table 1. Results of risk factor analysis in the case-control study

Risk Factor	Case (n=271)	Control (n=271)	Odds Ratio	95% Confidence Limit
Male	141	123	1.31	0.92-1.86
Poor hand hygiene before meals	134/222	141/225	1.10	0.74-1.64
Poor hand hygiene after toilet *	81/231	54/218	1.64	1.07-2.52
Eat lunch provided by school	256/271	263/271	1.93	0.75-5.05
Assigned lavatory cleaning works	27/244	27/244	1.00	0.55-1.82

* statistically significant, 95% CL not including 1.0

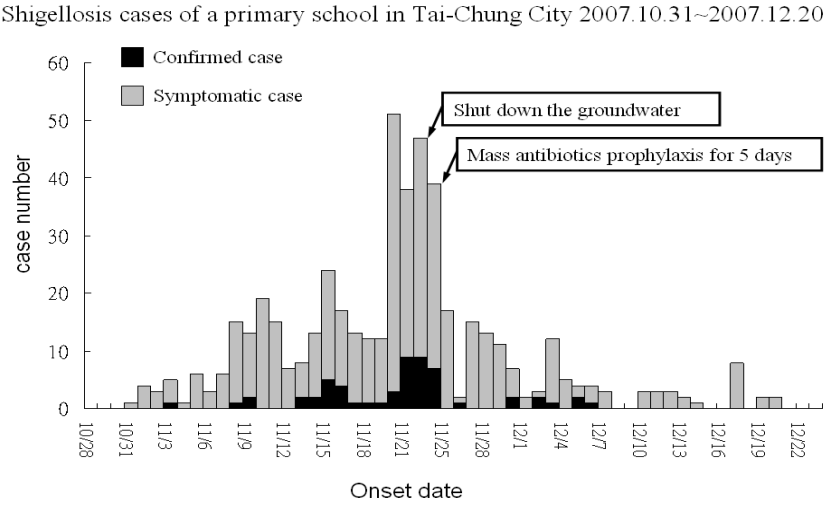

Figure 1. Epidemic curve of shigellosis outbreak. Investigation was started on November 21st, the contaminated well was shut down on November 22nd, and mass antibiotics prophylaxis for 5 days was started on November 23rd.

2. Environmental investigation

The kitchen was closed on Nov. 20th, two days before the investigation. One cook had diarrhea on Nov. 13th and shigellosis was confirmed on Nov. 19th. But the cook was in charge of administrative works, and she denied handling foods. No food samples left for inspection. The cooks reported that they washed hands before handling foods and wore gloves and masks during works. Water sample from the kitchen had no coliform bacteriae.

The school used two kinds of water source. One was piped water used in the kitchen, the drinking fountains, and the bath sinks before classrooms. The other was groundwater used in lavatories. A water sample from lavatories yielded coliform bacteriae and Shigella sonnei. Two wells provided the groundwater. Dye testing showed that water from one of the wells was contaminated by sewage system. Inspection of the sewer discovered a damaged connection near a pump of that well (Figure 2). The contaminated groundwater was for lavatory-use in four of the six school buildings. Students whose classrooms located in the four buildings had higher attack rate in the study. (16.7% vs. 8.2%, $p < 0.001$)

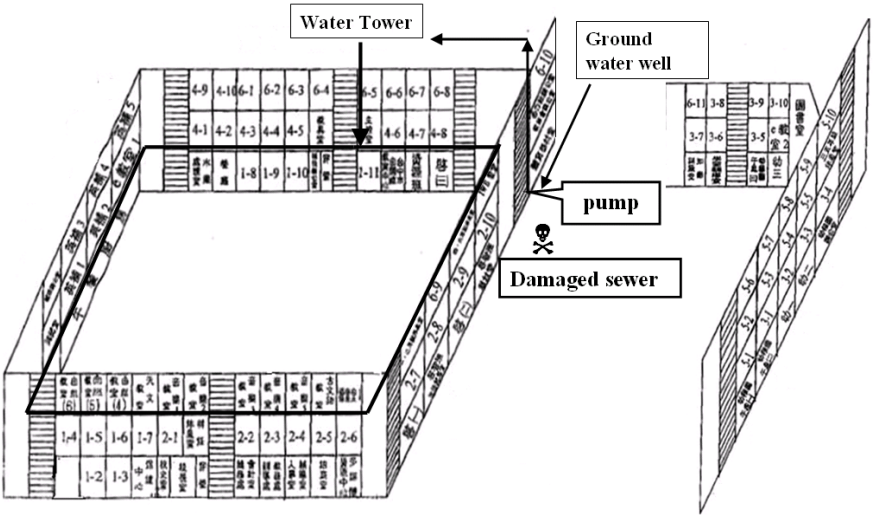

Figure 2. The groundwater was pumped up to a water tower in the top floor. The damaged sewer pipe was located nearby the pump. Leaked fecal material thus contaminated the water tower that provided lavatory-use water in the four connected buildings.

3. Microbiology results

Stool cultures from 25 cases yield *Shigella sonnei* and others had negative results in our case-control study (positive rate 25/271=9.5%). All isolates had identical antibiotic susceptibility which was resistant to nalidixic acid and trimethoprin/sulfamethoxazol and sensitive to Ampicillin, fluroquinolones and third-generation cephalosporins. PFGE was performed on all *Shigella* isolates from cases and the water sample. Human and environmental isolates were identical (as shown in Figure 3). While comparing with maps in the *Shigella* DNA fingerprint database, the strains isolated had close relationship with a strain circulated in the neighboring county (Nan-Tou County) during previous three months (as shown in Figure 4).

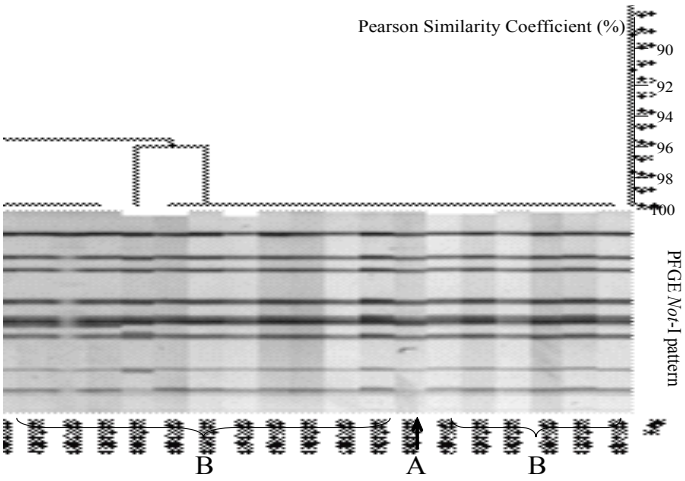

Figure 3. PFGE map of isolates in this outbreak. (A) indicates the isolate from contaminated water sample which was identical to other human cases. (B) indicates human isolates from confirmed cases in this outbreak.

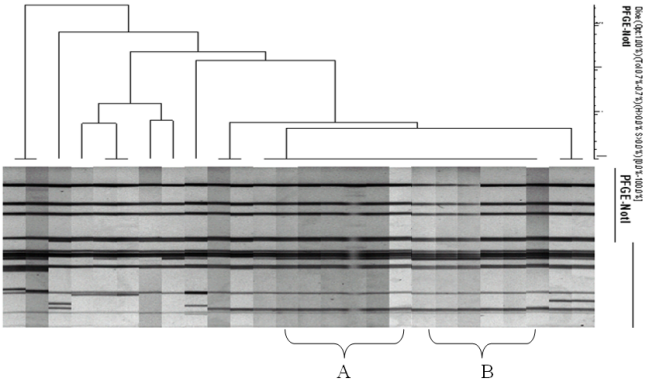

Figure 4. The Shigella DNA fingerprint database of Taiwan CDC. (A) indicates the strains of this outbreak. (B) indicates the strain circulated in the neighboring county (Nan-Tou County) during previous three months. Others indicate the strains circulated in other parts of Taiwan in the same time.

4. Control measures

Wells in the school were soon shut down on November 22nd and lavatories used piped water instead. The school isolated symptomatic students (defined as having diarrhea, fever or abdominal pain) at home until their stool culture results were available. Both contacts and symptomatic students had to send rectal swabs to the CDC lab. Whoever had positive stool culture with *Shigella sonnei* should received antibiotics from physicians and sent rectal swabs to the CDC lab again 24 hours after completed his treatment. Post-treatment cultures were repeated daily for two days. Cases could go back to school after getting two consecutive negative culture results.

For students remained at school, antibiotic prophylaxis with 5 days of Ampicillin were prescribed on November 23rd. Frequent and correct hand wash was emphasized. There was no confirmed case after December 6th. The community was also notified of the outbreak via the school and the press. Intensive monitoring of diarrhea diseases in the community and schools continued for one month after our investigation initiated. There were 13 community cases in the end. Ten of them were relatives of student cases. No shigellosis outbreak was noted in other schools or child-care settings. After this outbreak, Tai-Chung City government prohibited any groundwater use in schools and increased the budget for them to access piped water system. Water quality examination should be done in each semester.

Discussion

This was a school outbreak of shigellosis. All Shigella sonnei isolates belonged to the same strain in PFGE map. The strain was circulated in the rural area of central Taiwan at that time and there were only sporadic cases or family

outbreaks. In this large outbreak, we suspected that the explosive presentation of cases shown in epidemic curve resulted from a common source. Water sample which yield the outbreak strain of Shigella sonnei and the damaged sewer pipe located nearby the groundwater pump were the evidences supporting the hypothesis, especially that shigella spp. are difficult to culture from water samples and isolation from water samples is unusual [6]. The contaminated water source provided lavatory-use in four of the six school buildings. Higher attack rate among students in these buildings was also indicated that groundwater was the common source.

In review, groundwater, the main water system in the community of developing countries, was the common contaminated source of waterborne outbreaks [7, 8]. Most reports clearly demonstrated that the contaminated water had dose-dependent consumption effect or epidemiologic link to cases. In our report, we did not demonstrate that the contaminated water had significant consumption effect. The common use of groundwater in lavatories was for hand washing, cleansing and flushing. Students assigned to clean up lavatories had no significant risk to get disease. But other types of specific exposure to water in lavatories, such as washing mouth or even drinking, were not investigated. Control measures implemented can also evaluate the potential common source of an outbreak [9]. However, shutting down the groundwater and mass antibiotics prophylaxis were done in consecutive two days during the peak of this outbreak (Figure 1). Although it was stopped two weeks later, we were not able to evaluate the effect of water treatment. However, since the absence of any other common factor, groundwater was the most possible explanation of the magnitude of this outbreak.

Five days of oral antibiotic prescription to the non-cases was one of the

control measures in management of this outbreak. Mass antibiotic prophylaxis is not recommended by World Health Organization to control shigellosis [10]. It is concerned that chemoprophylaxis can hasten the emergence of resistant strains, and diverts attention and resources from other, most effective, control measures. After this outbreak, the agency therefore changed the national guideline of shigellosis control. Chemoprophylaxis according to drug sensitivity should be used with caution only in unusual outbreaks (defined by the responsible health authority). Maintaining of other control measures and disease monitoring should also be emphasized until the end of outbreaks [11].

There are few weaknesses of this study that we can improve in the future. (A) In studies of waterborne disease, utilization or consumption of the water should be clearly defined. (B) Some students were not included in our case-control study because they were sick and absence at school for days. To avoid bias of school outbreak investigations, students absent in school should also be included in the data.

Despite lacking of some evidences, it was highly suspected that groundwater played an important role in transmission of shigellosis in the school other than person-to-person mode. Tai-Chung City now is the first city government in Taiwan which prohibited groundwater use in schools . Water safety in schools should be emphasized again to avoid such outbreaks.

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