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Field Epidemiology Training Program, Bureau of Epidemiology Department of Disease Control, Ministry of Public Health, Thailand Tel: +6625901734-5, Fax: +6625918581, Email: osireditor@osirjournal.net, http://www.osirjournal.net

# An Outbreak of Gastrointestinal Illness Associated with Khmer Noodles: A Multipronged Investigative Approach, Kandal Province, Cambodia, June 2014

Panha Kimsean<sup>1,\*</sup>, Kosal Sreng<sup>1</sup>, Phalmony Has<sup>3</sup>, Sowath Ly<sup>1</sup>, Sansam Sim<sup>1</sup>, Sokheng Chhay<sup>1</sup>, Dara Prak<sup>1</sup>, Sreng Bun<sup>2</sup>

- 1 Cambodia Applied Epidemiology Training, Communicable Disease Control Department, Ministry of Health, Cambodia
- 2 Communicable Disease Control Department, Ministry of Health, Cambodia

\*Corresponding author, email address: kimseanpanha@gmail.com

#### Abstract

Food safety is a high priority for the Cambodian Ministry of Health, with standard operational procedures for foodborne diseases: intersectoral outbreak investigations and response finalized in 2015. On 29 Jun 2014, Kandal Provincial Health Department reported 143 patients with acute gastrointestinal illness, and 28 out of them were hospitalized. The number of patients increased to 215 within hours of the first report. A case-control study was conducted as well as laboratory testing, and an environmental survey to investigate the outbreak and institute necessary control measures. Three villages were randomly selected due to the provincial-wide distribution of the cases. A total of 37 cases and 36 controls were identified. Cases predominantly reported vomiting (97.3%) and diarrhea (75.7%). Consumption of Khmer noodles on 29 Jun 2014 was strongly associated with the illness (OR = 71.0, 95% CI = 8.6-582.2). Laboratory testing of food samples revealed *Staphylococcus aureus* and *Bacillus cereus*, which was consistent with the epidemiological and clinical findings. The investigation revealed the source as a sole local noodle producer and distributer. Environmental investigation of the noodle factory demonstrated multiple sources of contamination. The factory was temporarily shut down, and the team recommended destroying the remaining noodles. Local authorities were recommended to conduct regular inspection of the noodle factory.

Keywords: foodborne disease, disease outbreaks, gastroenteritis, Cambodia

#### Introduction

Contaminated food contributes to an estimated 1.9 billion cases of diarrhea cases.<sup>1</sup> The World Health Organization estimated that in 2010, foodborne disease contributed to approximately 600 million illnesses and 420,000 deaths.<sup>2</sup> In Southeast Asia, approximately one million children under five die from diarrheal diseases every year.<sup>3,4</sup>

Nowadays, acute foodborne infections and intoxication pose higher concern to governments and food industries when compared to a few decades ago. Some of the emerging issues include impact of foodborne diseases on vulnerable population such as children, elderly and immunocompromised persons, importing traditional foods of immigrants into the countries of settlement, ease in worldwide shipment of fresh and frozen foods, and development of food industries such as aquaculture.<sup>5</sup> Other issues of consideration for food safety are climate change, new technologies and antimicrobial resistance.

Monitoring of trends and burden of foodborne diseases requires an effective surveillance system at local, national and international levels, and resources to investigate outbreaks. Due to limited resources, many countries rely on passive reporting mechanisms to report foodborne diseases such as event-based surveillance of media or by health care workers.

Although global networks may assist countries with identifying and tracking clusters of foodborne illnesses from imported foods, the primary burden still falls on the countries where the cases occur. Unfortunately, when outbreaks occur, these countries may not have sufficient human or financial resources to identify the source of the outbreak, and effectively implement control and prevention activities. The rapid response team (RRT) in Kandal Province Disease notified the Communicable Control Department under the Cambodian Ministry of Health of 143 people with vomiting and acute diarrhea, and rapidly escalated to 215 people within a few hours on 29 Jun 2014. The ill individuals lived in six communes of Kandal Stueng District in Kandal Province, and received treatment at local health centers and hospital. An investigation team was assembled to investigate the outbreak by describing epidemiology of the cases, and identifying mode of transmission and etiology of the illness.

#### Methods

The team, made up of applied epidemiology training officers and the local and central RRT, conducted an investigation and collected food samples for laboratory testing. The team also performed an environmental investigation with a traceback process to find the source of the outbreak.

#### **Epidemiological Investigation**

The RRT in Kandal Province obtained a list of people who had attended local health centers with acute gastrointestinal illness. As the patients came from six different communes, a case-control study was conducted in one of the affected communes which was randomly selected. Three out of total 10 villages in the selected commune were further randomly selected. Definition of a case was a resident living in the selected villages who had onset of vomiting and/or diarrhea during 29 Jun to 1 Jul 2014. A control was a person who lived in a neighboring household to a case, and did not report vomiting or diarrhea between 29 Jun and 1 Jul 2014.

standardized paper-based questionnaire Α was administered by face-to-face interview. Collected information included demographics, signs and symptoms, onset dates of illness, and types of food and water consumed in previous two days. Data were double-entered to ensure data quality and cleaned. The data were analyzed in Epi Info  $7^6$ , and odds ratios (OR) and 95% confidence interval (CI) were calculated.

#### **Environmental Investigation**

An environmental survey was conducted to investigate food production and processing in the Khmer noodle factory that the RRT hypothesized as the source. The team performed a walk-through examination of the premises, observed noodle production processes, cleanliness of machinery as well as storage conditions of raw ingredients and final products. Based on the noodle distribution list provided by staff at the factory, a traceback investigation was performed to remove the products from stalls across the province.

#### Laboratory Investigation

The investigation team collected noodle samples and noodle ingredients (rice, flour and water) from the implicated factory, the only factory in the area with widespread distribution of fresh noodles to stalls. Samples were stored in a refrigerator and sent to Institute Pasteur du Cambodge for laboratory testing.

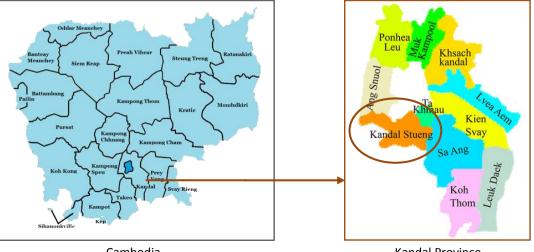
#### **Ethical Issue**

An ethical review was not required as this investigation was considered to be an emergency response to an outbreak.

#### Results

#### Epidemiology

Patients lived in six communes of Kandal Stueng District, Kandal Province, which is located 25 km east of the capital city, Phnom Penh (Figure 1).



Cambodia

Kandal Province

Figure 1. Map of Ampov Prey Commune, Kandal Stueng District, Kandal Province, Cambodia

Total 215 people were treated at local health centers and hospital for gastrointestinal illness. Out of 28 people who were hospitalized, 12 had severe diarrhea and were transferred to Kandal Provincial Hospital. No deaths or ongoing sequelae were reported.

The randomly selected commune was Ampov Prey Commune. Three selected villages in this commune were Ampov Prey 1, Ampov Prey 2 and Tadol 1. A total of 37 cases and 36 controls were recruited in the study. Majority of study participants were females, with 70.3% among cases and 66.7% among controls. Most were older than 16 years, with 70.3% among cases and 86.1% among controls (Table 1).

The most common symptom reported was vomiting (97.3%), followed by diarrhea (75.7%), abdominal pain (65.0%), headache (35.0%) and nausea (30.0%).

Although the cases reported consuming many types of foods, only five items with high incidence were considered for analysis, including Khmer noodles (67.1%), fish Khmer soup (42.5%), water (41.1%), rice (28.8%) and curry (15.1%). We found that those who consumed Khmer noodles had the highest OR (Table 2). The incubation period between consuming the suspected noodles and becoming sick ranged from 3-6.5 hours, with median of 4.5 hours (Figure 2).

Table 1. Sex and age composition of cases and controls from
a gastrointestinal outbreak in Kandal Province, Cambodia,
29 Jun - 1 Jul 2014

Variable	Case (	n=37)	Control (n=36)			
Vallable	Number	Percent	Number	Percent		
Gender	-	-	-			
Male	11	29.7	12	33.3		
Female	26	70.3	24	66.7		
Age group (year)						
0-5	5	13.5	1	2.8		
6-15	6	16.2	4	11.1		
≥16	26	70.3	31	86.1		

#### Environmental

A local noodle factory was implicated in the outbreak as it was the main source of raw noodles for all the food stalls involved. In the noodle factory, seven employees produced an average of 26 pots (15 kg each) of noodles per day. Khmer noodles were usually made in two batches: 5 pm (first day) and 4 am (second day).

# Table 2. Analytic findings on selected food products consumed from a gastrointestinal outbreak in Kandal Province, Cambodia, 29 Jun - 1 Jul 2014

E d in	Cas	Cases (n=37)		rols (n=36)	Odds ratio	05% 01
Food item	Ate	Did not eat	Ate	Did not eat	(OR)	95% CI
Khmer noodles	36	1	10	26	71.0	8.6-582.2
Fish Khmer soup	21	16	10	26	3.1	1.2-8.2
Curry	9	28	2	34	5.1	1.0-25.6
Rice	1	36	12	24	0.04	0.01-0.2
Water	3	34	12	24	1.9	0.4-8.8

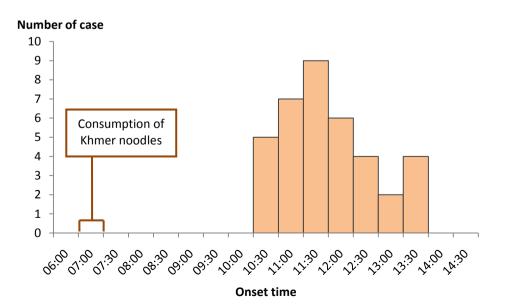


Figure 2. Distribution of cases by onset time of illness from a gastrointestinal outbreak in Kandal Province, Cambodia, 29 June 2014

First, rice was ground into flour, this flour was then kept under a stone for 2-3 hours and then boiled. The whole process took 5-6 hours. The noodles were then cut and stored at room temperature. Finally, batches of noodles were distributed by motorbikes around 6 am on the next day to noodle sellers in six communes. The transportation time from factory to seller took approximately 30 minutes. Noodle stalls cook the noodles in many different ways. The most common method for breakfast is boiling for soups.

The investigation team performed an environmental survey of the noodle factory and found several issues of concern. These findings included 11 potentially contaminated noodle pots (one pot with cut noodles and 10 pots with noodles that were not yet cut); employees with gastrointestinal symptoms in the food processing vicinity; a muddy stream running close by noodle pots on the ground; and potentially



(a) Pots used for noodle preparation on the ground and left uncovered



(c) Contaminated drainage system at the noodle factory



(e) Inappropriate storage of uncut noodles at the factory

contaminated multiple points along the line of noodle production such as broken and dirty water supply system, pots on the ground by the muddy stream, and animals in the factory (Figure 3). One factory employee reported that neighbors sprayed pesticides a week before the outbreak. The neighbors also complained that the noodle factory smelled bad, especially after the rain.

#### Laboratory

Laboratory results revealed high level of *Staphylococcus aureus* and *Bacillus cereus* in multiple samples of old (first day) and new (second day) Khmer noodles and flour. These results implied potentially injurious contamination levels and were unfit for human consumption according to British Health Protection Agency guidelines for assessing the microbiological safety<sup>7</sup> (Table 3).



(b) Inappropriate storage of rice packs to be used for noodle preparation



(d) Broken water supply at the noodle factory



(f) Uncovered Khmer noodle pots accessed by animals

Figure 3. Noodle production from the implicated factory in Kandal Province, Cambodia, 29 Jun - 1 Jul 2014

Type of sample	Bacteria found	Amount detected	Interpretation <sup>7</sup>
Rice	Staphylococcus aureus	<1 CFU/g	Low
	Bacillus cereus	4.0x10 <sup>2</sup> CFU/g	Low
Khmer noodles (first day)	Staphylococcus aureus	<sup>8</sup> 2.6x10 CFU/g	High
	Bacillus cereus	<sup>5</sup> 1.5x10 CFU/g	High
Khmer noodles (second day)	Staphylococcus aureus	9 1.7x10 <sup>°</sup> CFU/g	High
	Bacillus cereus	<1 CFU/g	Low
Flour	Staphylococcus aureus	9 1.5x10 CFU/g	High
	Bacillus cereus	6 8.0x10 <sup>6</sup> CFU/g	High

#### Discussion

The clinical, laboratory and epidemiological findings revealed that this outbreak in Kandal Stueng District most likely caused by consumption was of contaminated Khmer noodles which were originated from the same factory though they were sold in different noodle stalls. Predominant upper gastrointestinal clinical signs and symptoms of the cases, short incubation period of 3-6.5 hours, identification of S. aureus and B. cereus in food samples, and results of  $\mathbf{the}$ environmental investigation supported that S. aureus and B. cereus were probably the cause of the outbreak.

The findings from the case-control study implicated the Khmer noodles as the potential source. However, other food items also resulted a significant risk. The environmental findings, high bacterial load found in the Khmer noodles and flour as well as high OR for this food item suggested that the source of the outbreak was most likely to be Khmer noodles. Other food items with high OR might be due to cocontamination or eating together with the noodles.

*B. cereus* spores have been found in many types of foods such as milk, corn starch, rice, dried legumes and spices. Vegetative cells of some strains can start to multiply at 4°C. As the emetic toxin produced by *B. cereus* is heat stable, reheating of left-over foods may not destroy the bacteria and toxins produced. Storing food at 5°C for less than seven days may prevent *B. cereus* growth.<sup>8</sup>

*S. aureus* is commonly found in raw foods, which can be destroyed by an effective pasteurization process. The presence of this pathogen indicates post-process contamination due to poor personal hygiene and hand washing, and cross-contamination from other raw animal products. As it does not produce toxin at temperature below 10°C, refrigeration of foods should be done at less than 5°C.  $^{\rm 8}$ 

Both *B. cereus* and *S. aureus* are characterized by short incubation periods and short duration of illness, which fit the clinical picture of illness seen in this outbreak. Other studies also detected enterotoxigenic *B. cereus* in rice noodles, wet wheat noodles, dry wheat noodles and other foods.<sup>9</sup> In addition, another study showed that common ready-to-serve starchy foods such as noodles could serve as a food poisoning vehicle if temperature control was not correct.<sup>10</sup>

#### Limitations

The investigation team was unable to retrieve or test any clinical samples from the sick factory workers or cases; therefore, unable to directly link organisms in humans and foods to establish causality. The study was also constrained by small sample size of randomly selecting three villages in one commune. Due to limited investigation time, cases and controls were not matched and only a two-day food history was taken. Potential for recall bias existed as media and local authorities had already pointed to the noodles as the source of illness prior to the investigation. Moreover, selection bias could have played a role as the investigation team could not interview more controls. The investigation team also relied upon the local village chief to identify possible cases; this might have led only to people known to have eaten the noodles and who were sick to be interviewed. The investigation team did not review the possibility of contamination during transportation.

#### Recommendations

Several control measures were recommended for the noodle processing factory to reduce further contamination and thereby, preventing future outbreaks from consumption of the noodles. Immediate recommendations included discontinuing delivery of Khmer noodles produced in the processing area, destroying left-over Khmer noodles, cleaning up food processing equipment and premises, restricting sick workers from the food production area, improving water supply system in the factory, protecting the production area by using netting or plastic, keeping all materials used for food processing above the ground, and utilizing safe and treated water in all noodle preparation processes. In addition, local government authorities were recommended to conduct regular inspection of noodle-making facilities and provide health education in villages about hygienic food preparation practices.

### Conclusion

In summary, the findings of this study identified consumption of Khmer noodles as the likely source of the outbreak in Kandal Stueng District in Kandal Province. Noodles were contaminated by *B. cereus* and S. aureus, which were detected in the food items reported to be consumed by the cases and came from the Khmer noodle factory. Clinical signs and symptoms of the cases also fit with this finding. The RRT detected and responded to this event in a timely manner and ensured noodles were removed from further distribution. Our multipronged approach of highlighted the advantages integrating epidemiologic, laboratory and environmental tools for addressing disease outbreaks. The concordant findings helped to strengthen evidence needed to implement timely control measures. The introduction of food safety regulations as well as policing of these regulations in Cambodia would assist in detecting and preventing future outbreaks.

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