ISSN 1021-366X

Epidemiological Investigation of a Food Poisoning Outbreak in a Tainan Hotel, Taiwan

1. Introduction

A family in Tainan invited guests of 30 tables to a wedding party at a hotel on 19 May 1996. The party started at 1:00 pm and ended at 2:30. Some 60 guests began to develop symptoms of vomiting and diarrhea in that evening and were treated at the local health station, the Chimei Hospital and other hospitals. Upon report of the health station, the Tainan City Health Bureau took immediate action for investigation on 20 May. As the number affected was too large, request for support was made by the Health Bureau to FETP through the Bureau of Food Sanitation of the Department of Health (DOH). A team from FETP was despatched for investigation together with the Tainan City Health Bureau on 21 May. The team visited homes of victims and the hotel for epidemiological investigation to understand the causes of the incident, the food items contaminated, and the probable pathogenic microbes of the outbreak.

2. Materials and Method

Although there were guests for 30 some tables, through the name list supplied by the host, telephones and addresses of only 132 guests could be traced. They were investigated by the"case-control method".

1) the Questionnaire

A structure questionnaire containing items such as: personal information (age and sex), whether being to the wedding party on 19 May, time of eating, food items eaten, any uncomfortable feeling after party, any symptoms, time of onset of symptoms, medication and/or hospital care, time of recovery, taking some food home for family to share, did family members have any symptoms after eating food, etc. Each was interviewed either face-to-face or through telephone. A total of 116 was interviewed,

giving a coverage rate of 88% (116/132).

2) Inspection of the Kitchen

By the time the team visited the hotel, the kitchen was already cleaned. The sanitary conditions of the environment and the staff were satisfactory. Separate places, facilities and workers were used for the treatment of cooked and cold food. Fishery products were frozen, though the temperatures of the refrigerator and the freezer of 10℃ and －15℃ were not low enough.

3) Laboratory Testings

of human specimens:

10 rectal swabs were collected from patients on 20 May, and 9 from the cooks of the hotel on 21 May for testings by the Southern Branch Laboratory of the National Institute of Preventive Medicine of the Department for: *Vibrio parahoemolyticus, Bacillus cereus, Pathogenic Escherichia coli, Staphylococcus aureus, Salmonella, Vibrio cholerae, Salmonella typhi* and *paralyphi, Bacillus dysenteriae.*

of food specimens:

Three food specimens from leftovers of the day (crab rice, assorted ribs and shrimp balls) were collected on 21 May for testings by the Souther Branch Laboratory of the National Laboratories for Food and Drugs of the Department for: *Vibrio parahaemolyticus, Salmonella, Bacillus cereus, Pathogenic Escherichia coli, Staphylococcus aureus* and *enterotoxin.*

of cooking utensils:

Two tap water specimens, one swab of the kitchen scissors, two swabs of the kitchen knives, and two swabs each of the chop boards used for cooked and cold food were collected on 21 May for testings by the Southern Branch Laboratory of the National Laboratories for Food and Drugs.

4) Data Processing and Analysis

Information collected from questionnaires was keyed in by dBase III Plus and then verified with SAS of the 486 DX66 personal computer. Data were then tabulated and analyzed.

A case was defined according to the frequency of self-reported symptoms. Attack

rate, exposure odds and odds ratio were calculated. χ^2 test and Fisher exact test were used in the univariate analysis.

3. Results

1) Questionnaire Findings

107 copies of the questionnaire out of 116 persons interviewed were valid (42 males and 65 females). Of them, 104 had developed symptoms, and 66 treated. Their symptoms were: diarrhea (90.4%, 94/104), abdominal pain (69.2%, 72/104), vomiting (51.0%, 53/104), nausea (41.3%, 43/104), dizziness (39.4%, 41/104), chill (20.2%, 21/104), fever (18.3%, 191104) and blood stool (2.9%, 3/104). By the distribution of symptoms, a case was defined as one who had eaten at the wedding party at noon of 19 May 1996 in this particular hotel and had had diarrhea more than twice daily and also one of the following symptoms: abdominal pain, vomiting, nausea, fever, or dizziness. Of the 107, 82 (29 males and 53 females) met the definition, giving an attack rate of 76.6% (82/107).

The incubation periods, from the intake of food to the onset of symptoms, ranged from 4 to 43 hours. They are shown in the epidemiological curve in Figure 1. The median was 15 hours and the mode, 11 hours.
Each food item was epidemiologically investigated of its association with the outbreak. Questionnaire findings showed that none of the guests were vegetarian, and none had taken food home for family to share. They said they ate practically all food items, the attack rate hence was unusually high (76.6%). Table 1 gives by each food item the number of person eaten, exposure odds, odds ratio and statistical testings. By food item, the “lobster in garlic” and the “sea slug with pork tendon” were found to be significantly related ($p<0.05$) to the incident. Others were not related (p value larger than 0.05).

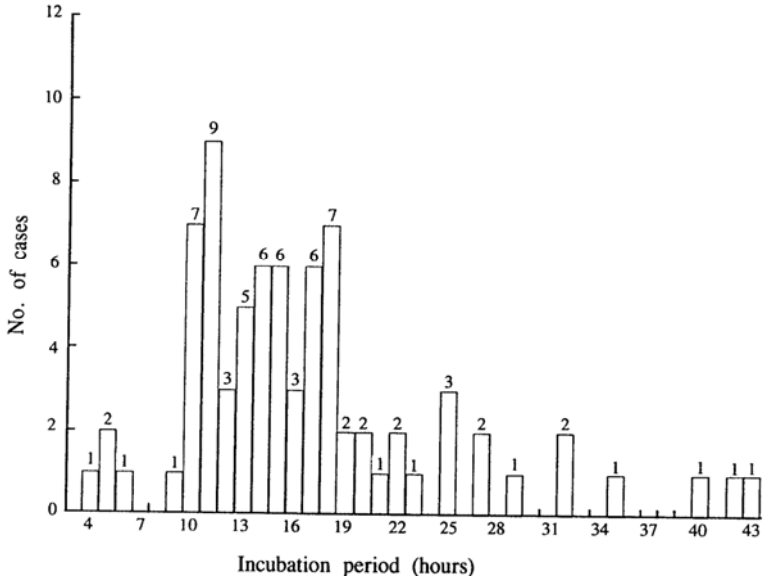
2) Laboratory Testings

K6 serotype of *Vibrio parahaemolyticus* was isolated from all ten rectal swabs of the patients. From the three food specimens, K6 *Vibrio parahaemolyticus* and *Bacillus cereus* were isolated only from the crab rice and the bacterial colonies were larger than 1.1 x 105 MPN/g. The nine rectal swabs of the cooks and the nine specimens of cooking utensils and water were all negative.

4. Discussion and Conclusion

Since Japan isolated *Vibrio parahaemolyticus* from fecal specimens of food poisoning victims in 1950, many reports of food poisoning incidents caused by *Vibrio*

Figure 1. Distribution of Incubation Periods



parahaemolyticus in south-east Asia, Australia, UK and USA have appeared [(1)]. In the past, it was generally believed that *Vibrio parahaemolyticus* survived on the surface or in the internal organs of fish and shellfish in sea water of 17℃ and above. However, Hornstrup et al.[(2)] pointed out that even in cold places such as Sweden, enteritis and extra-intestinal otitis media induced by *Vibrio parahaemolyticus* also occurred.

Desenclos [(3)] et al further indicated that some of those who had eaten food contaminated by *Vibrio parahaemolyticus* could also develop bacteriemia. In particular, for those who had liver disease, diabetes and removal of stomach, their chances of extra-intestinal infections were much higher than normal persons. Kelly and Dan Stroh [(4)], periodically collected, at ten selected sites on the seashore of British Columbia, sea water specimens for laboratory testings for *Vibrio parahaemolyticus*. They found that no *Vibrio parahaemolyticus* could be isolated between December and May each year when the temperature of the sea water was below 14℃ and the salt concentration larger than 13 per 1,000. However, in July when the temperature of the sea water was around 20℃ and the salt concentration lower than 6.5 per 1,000, *Vibrio parahaemolyticus* bred rapidly and in 7% of the sea water specimens, *Vibrio parahaemolyticus* could be cultured. They, therefore, concluded that more gastro-intestinal infections by these microorganisms certainly could be found in summertime.

In Taiwan, SW Fang [(5)] Ct al collected specimens of fishery products from retail

Table 1. Exposure Odds, Odds Ratios and Statistical Testings by Food Item

Food item	Ill			Not ill			Odds ratio (3)/(6)
	Eaten (1)	Not eaten (2)	Exposure odds (3)=(1)/(2)	Eaten (4)	Not eaten (5)	Exposure odds (6)=(5)/(4)	
Assorted plate	78	2	39 0	25	0	—	—
Crab with shark fin	78	2	39 0	23	2	11.5	3.39*
Lobster in garlic	79	1	79.0	22	3	7 3	10.82*#
Sea slug with pork tendon	74	6	12.3	19	6	3 2	3.84*#
Crab rice	74	6	12 3	22	3	7 3	1.68*
Fruits	77	3	25.7	23	2	11 5	2.23*
Assorted ribs	70	9	7 8	23	2	11.5	0.68*
Shrimp balls	68	11	6.2	17	8	2 1	2.95*
Fish	73	7	10.4	21	4	5 3	1.96*
Meat stew in pot	74	6	12 3	21	4	5 3	2.32*
Cake	61	17	3 6	19	6	3.2	1.13**
Dessert	56	22	2.5	19	6	3.2	0.78**

Exposure odds are ratios of those who had eaten a certain food item against those who had not eaten. If the exposure odds to a food item of those who became ill are higher than those who did not become ill, the particular food item could be suspected of being responsible for the incident.

* Fisher exact test

** χ^2 test

statistically significant at $p < 0.05$.

markets in eight coastal counties and cities for the screenings of *Vibrio parahaemolyticus*. They found that 45.7% of the fishery products: 40.0% of fish, 22.3% of raw fish, 44.4% of shrimps and 47.8% of crabs, had been contaminated by *Vibrio parahaemolyticus*. In the Tainan area (Tainan city and county), when the temperature was around 28¢XC or between April and November, the amount of *Vibrio parahaemolyticus* in sea water was the largest. The present food poisoning incident at a hotel in Tainan City was in mid-May and the food contaminated was crab.

Some researchers [6], however, maintained that whether *Vibrio parahaemolyticus* could induce infection depended on the presence of hemolysin. Evidences were that *Vibrio parahaemolyticus* that could induce gastro-intestinal infections in man, almost 95.0% of them would show hemolysis in Wagatsuma culture. In the ordinary environment, only 1.0% of *Vibrio parahaemolyticus* would cause hemolysis [6].

Substances that can cause hemolysis, thus far, are four. They contain phospholipase A, lysophospholipase and glycerophosphoryicholine diesterase [7]. In animal experiments, these substances destroy upper dermis of the small intestine to cause diarrhea [6]. Many reports [1, 6-8] indicated that 4 to 96 hours after *Vibrio parahaemolyticus* infection, on average 15 to 24 hours, men developed diarrhea. The time of onset depended on the amount of microorganisms consumed, the nature of food and the concentration of stomach acid. The incubation period of the present incident ranged from 1 to 66 hours, 95% within the range of 4 to 43 hours; and some victims had blood stool.

Blood stool could be related to the superficial ulcer of the membrane of sigmoid [9]. The mechanism of ulcer of the membrane of sigmoid is yet not clear. Animal experiments [10-11] show that *Vibrio parahaemolyticus* can secret thermostable direct hemolysin (TDH). TDH will induce edema of lamina propria, destroy endoplasmic reticulum, degenerate microvilli, and enlarge mitochondria. In addition, vacuoles will be found in the cytoplasm and the nuclei of some epithelial cell will be degenerated. Fatality of *Vibrio parahaemolyticus* infection is low, around 0.03%, according to Zen-Yoji et al [12]. In the present study, 2.9% of the victims developed blood stool; none died.

The lack of the basic knowledge of *Vibrio parahaemolyticus* on the part of cooks is often the main reason of food poisoning incidents such as this. They must understand that *Vibrio parahaemolyticus* breeds more in summertime on fishery products or shellfish; that it breeds faster than any other bacteria under room temperature; and that it can survive even under 80℃. Zen-Yoji et al [12] maintained that those who processed or cooked sea food had more chances of consuming sea food than any others, they could become inapparent carriers. Zen-Yoji et al examined 200 sushi cooks without symptoms of enteritis, and found *Vibrio parahaemolyticus* in 14 (7%) of them. This was nine times higher than what one would find in the ordinary people. This finding suggested that *Vibrio parahaemolyticus* could be transmitted (secondary infection) by cooks to customers. Peffers et al [7] found that even after 15 minutes of boiling, the temperature of the central part of crab was only 63℃.

Epidemiological investigation of food items showed that “lobster in garlic” and “sea slug with pork tendon” were statistically related to the food poisoning. Specimens of these two food items were not available, no laboratory testings could be made to support the reasoning. Though K6 *Vibrio parahaemolyticus* was isolated from the crab rice, the relationship between crab rice and the incident could not be statistically established. The reason could be that the sample size of interviewing was not large enough, only 116 persons out of guests for 30 tables and only 107 copies of the questionnaire were valid. However, K6 *Vibrio parahaemolyticus* was isolated from rectal swabs, and the distribution of the incubation periods on the epidemiological curve corresponds to the specific features of *Vibrio parahaemolyticus*.

By the findings of Barker [1] and Peffers et al [7], it could be concluded that the present incident was due to the consumption of the *Vibrio parahaemolyticus* contaminated crab rice, or “lobster in garlic”, or “sea slug with pork tendon” (no specimens of the latter two for laboratory testings). Crabs, lobsters and sea slugs are fishery products, chances of their being contaminated by *Vibrio parahaemolyticus* are

high probably because of: 1) *Vibrio parahaenwlyticus* breeding in large quantity on crabs in the process of transportation for poor refrigeration or freezing facilities or process; 2) temperature of the freezing facilities of the hotel was not low enough (only － 15℃, higher than the standard －18℃), or too much food was stored and the temperature of the fishery products in the inner part of the freezer could not be maintained at － 18℃ and lower; 3) in the process of cooking, the temperature was not high enough to kill *Vibrio parahaemolyticus* [1,7,8]

5. Recommendations

To prevent food poisoning outbreaks of *Vibrio parahaemolyticus*, it is recommended that:

1) sea food should be frozen at － 18¢XC and below to prevent the growth of *Vibrio parahaemolyticus*;

2) *Vibrio parahaemolyticus* can survive under 60℃ for 15 minutes and under 80℃ for several minutes; in cooking, the temperature of the central part of food should be kept at 70℃ and above for more than 15 minutes [8]

3) food should be consumed soon after cooking; if not, they should be frozen immediately at 4℃ and below; raw food and cooked food should be stored separately;

4) cooks and other related workers should be physically examined regularly to ensure that they are free of *Vibrio parahaemolyticus* to avoid secondary infection;

5) in the case of food poisoning, in addition to penalty to the restaurant concerned according to law, cooks and other staff of the restaurant should be given training in sanitary control measures to upgrade their professional knowledge and skills and to safeguard the safety of consumers.

Acknowledgement

The authors wish to thank the Tainan City Health Bureau, the Bureau of Food Sanitation of the Department of Health, the Southern Branch Laboratory of the National Institute of Preventive Medicine of the Department, and the Southern Branch Laboratory of the National Laboratories for Food and Drugs of the Department for their assistance in the investigation.

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