Risk-Based Food Safety Inspection During Incident Event in Thailand

Submitted by: Thailand
Risk-Based Food Safety Inspection during Incident Event in Thailand

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Content

• Investigation of a Multi-provinces Cholera Outbreak in Thailand which expanded to Laos People Democratic Republic in 2007
  • Investigation and develop risk-based inspection (RBI) along exported Cockles' food supply chain (from Thailand across several check points to Laos PDR)
  • Future RBI during incident event uses Government Information Network (GIN) system
Situation of Cholera in Thailand, 2007

- 988 cases with 7 death (CFR 0.7%)
- spread to 50 provinces of total 76 provinces
- Inaba: most occur in Tak province
- Ogawa: distribute for the whole country
- age group 15-44 years

Number

<table>
<thead>
<tr>
<th>Age group</th>
<th>Number</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;1 month</td>
<td>50</td>
</tr>
<tr>
<td>1-4 months</td>
<td>10</td>
</tr>
<tr>
<td>5-9</td>
<td>20</td>
</tr>
<tr>
<td>10-14</td>
<td>50</td>
</tr>
<tr>
<td>15-24</td>
<td>200</td>
</tr>
<tr>
<td>25-34</td>
<td>250</td>
</tr>
<tr>
<td>35-44</td>
<td>150</td>
</tr>
<tr>
<td>45-54</td>
<td>100</td>
</tr>
<tr>
<td>55-64</td>
<td>50</td>
</tr>
<tr>
<td>&gt;65</td>
<td>10</td>
</tr>
</tbody>
</table>

Cockle farm
### Cholera cases by *V. cholerae* serotype in Thailand, 2007

<table>
<thead>
<tr>
<th>Category</th>
<th>Number (or Percentage)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hospitalized</td>
<td>926</td>
</tr>
<tr>
<td>Community search</td>
<td>433</td>
</tr>
<tr>
<td>Death</td>
<td>7</td>
</tr>
<tr>
<td>Geographical distribution</td>
<td>50 provinces</td>
</tr>
<tr>
<td>Ogawa</td>
<td>1,087</td>
</tr>
<tr>
<td>Inaba</td>
<td>266</td>
</tr>
<tr>
<td>Hikojima</td>
<td>1</td>
</tr>
<tr>
<td>O139</td>
<td>5</td>
</tr>
</tbody>
</table>

- **Ogawa**
  - 1,087 cases
- **Inaba**
  - 266 cases
  - 38th week highlighted

**Graphical Representation**

- **Active cases**
- **Passive cases**
Multi-provinces Cholera outbreak associated with cockles

In 2007: Cholera outbreak

Thailand

10 Oct. 2007 (38th wk), expanded to Laos People Democratic Republic

Most *V. cholerae* strains isolated from patients

“*V. cholerae* O1 serotype Ogawa”

The cockles were assumed to be a source of cholera outbreak (OR 2.24, 95% CI 1.43 – 4.02)
Thai-Laos ‘s Joint investigation on Cholera outbreak, 2007

Organism: RSC positive for *Vibrio cholerae* O1 El Tor Ogawa (10th Oct.)

Investigation: 120 workers in factory (80 living), 10 RSC sample found 1 positive the same organism (case’s 2 months pregnant wife)
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Cockles: *Anadara granosa*

: marine animal which is habitat of marine microorganisms

In Thailand, cockle is usually cooked by boiled or roasted transmitted the pathogens to human via contaminated cockle

**Scientific classification**

- **Kingdom:** Animalia
- **Phylum:** Mollusca
- **Class:** Bivalvia
- **Subclass:** Pteriomorphia
- **Order:** Arcoida
- **Family:** Arcidae
- **Genus:** Anadara
- **Species:** A. granosa
Investigation of cockles’ food supply chain from Thailand to Laos PDR

Pass out at 4 check points (Thailand) and enter at 2 check points (Laos PDR)

Transport to Northeastern Provinces of Thailand

Farm in Surat Thani
Inspection of cockles’ farm

- **Departments of Fisheries**
  Coastal Fisheries Research and Development Surat Thani: care, promote and monitor of water quality affecting cockle farming. The amount of salinity of sea water should be 10-30 ppt.

- **Fisheries Office Surat Thani**:
  1. Promote the coastal fishery and Good Aquaculture Standard (GAP) and registration cockles’ farm.
  2. Research and monitor of water quality and aquatic products from Surat Thani.
  3. Check for contamination and quality of aquatic products.
In 2007, the salinity of sea water was less than 10 ppt. for long and continuous months (Aug. – Oct.).
In 2007, total rainfall in Surat Thani was more than 90.1 millimeter.
Food quality inspection for *V. cholerae*

| Date  | Sample no. | Lab code | Test Results |  
|-------|-------------|----------|--------------|---
|       |             |          |              | **V. cholerae** |
| 9/7/50 | N50400755   | CC/3     | TVC 3800 CFU/ml | ND |
| 6/8/50 | N50400844   | CA/1     | TVC 9600 CFU/ml | ND |
| 6/8/50 | N50400845   | CA/2     | TVC 12000 CFU/ml | ND |
| 6/8/50 | N50400846   | CA/3     | TVC 4700 CFU/ml | ND |
| 6/8/50 | N50400856   | CB/1     | TVC 3300 CFU/ml | ND |
| 6/8/50 | N50400857   | CB/2     | TVC 3600 CFU/ml | ND |
| 6/8/50 | N50400858   | CB/3     | TVC 4300 CFU/ml | ND |
| 6/8/50 | N50400867   | CC/1     | TVC 3800 CFU/ml | ND |
| 6/8/50 | N50400868   | CC/2     | TVC 870 CFU/ml | ND |
| 6/8/50 | N50400869   | CC/3     | TVC 2300 CFU/ml | ND |

**Note:** Some results are marked as "DETECTED."
The outbreak of cholera in Laos – Thailand linked together because the investigation revealed that the suspect food was exported - imported cockles which found *V. cholerae*

Decreasing of sea water’s salinity in 2007 which promoted the high prevalence of *V. cholerae* might be because of heavy rain fall.
• Transportation time of cockles from Surat Thani through Loas PDR was greater than 20 hours may be an important factor in inducing the increasing number of *V. cholerae* until reaching infective dose \((10^9)\).

• To prove the hypothesis, pulse field gel electrophoresis should be used to identified *Vibrio cholerae* strain caused this outbreak was the same or not?

• GAP should be strengthening by regulatory agencies.
RBI for cholera outbreak during 2007 to 2010, Thailand

Cockles samples \(N=161\)

Rectal swab samples \(N=328\)
- Patients, family members, neighboring

Detection of *Vibrio cholerae* from specimens by culture method

Detection of *Vibrio cholerae* serogroups by Uniplex and Duplex PCR, PFGE, Ribotyping, MLVA techniques.

*V. cholerae* O1
*V. cholerae* non-O1

PCR: Polymerase Chain Reaction, PFGE: Pulsed field gel electrophoresis
MLVA: Ribotyping, multiple-locus variable-number tandem-repeat analysis
Risk based inspection for cockles, 2010

Sample collection
1. Khon Kaen province: 30 samples
2. Udon Thani province: 7 samples
3. Nong Khai province: 56 samples
4. Exported cockles: 68 Samples (sample from truck (once/ month from Mar. to Nov. 2010))

Study method
1. Standard culture method
2. PCR

Obj: To define the prevalence of *V. cholerae* in cockles
Detection of *Vibrio* spp. in cockles from 3 sources of collection

- **250 g of cockle**
- **250 ml of PBS**
- **250 g of cockle suspended in 250 ml of PBS**

Add **20 ml** (10 g) of cockle suspension to **80 ml** of APW

Incubated at 37°C for 6 h

**APW**

- **PCR method**
  - **1 ml**
  - **DNA extraction**
  - **PCR amplification**
- **Culture method**
  - **A loopful**
  - **Streak on TCBS agar**
  - **Biochemical test**
The prevalence of *Vibrio* spp. detected by tetraplex PCR was slightly lower than uniplex PCR however, it is higher than culture method.
Prevalence of *Vibrio* spp. in three different sources of cockle samples (uniplex PCR)

- *Vibrio* spp. among three sources were not significantly different, except *V. mimicus* was found very high (47.2%) in Khon Kaen
- *V. parahaemolyticus* was found in 100% in all 3 sources
- *V. vulnificus* was found about 90% in all 3 sources
- *V. cholerae* was found about 80% in all 3 sources
**Mixed Vibrio spp. found in cockle samples in the northeastern Thailand**

<table>
<thead>
<tr>
<th></th>
<th>VP+VC+VV+VM</th>
<th>VP+VC+VV</th>
<th>VP+VC</th>
<th>VP+VV</th>
<th>VP</th>
</tr>
</thead>
<tbody>
<tr>
<td>Exported to Lao PDR (34)</td>
<td>14.7</td>
<td>3.6</td>
<td>18.9</td>
<td>58.8</td>
<td>58.7</td>
</tr>
<tr>
<td>Nong Khai (56)</td>
<td></td>
<td></td>
<td></td>
<td>75</td>
<td>11.8</td>
</tr>
<tr>
<td>Khon Kaen (53)</td>
<td></td>
<td></td>
<td></td>
<td>11.8</td>
<td>14.7</td>
</tr>
<tr>
<td>Total</td>
<td>14.7</td>
<td>3.6</td>
<td>18.9</td>
<td>58.8</td>
<td>58.7</td>
</tr>
</tbody>
</table>

Most of cockle samples were contaminated with combination of various species of *Vibrio*.
Summary of the sensitivity of the tetraplex PCR and uniplex PCR compared with culture method using spiked *Vibrio* spp. in sterile cockle samples

<table>
<thead>
<tr>
<th>Pathogens</th>
<th>Tetraplex PCR</th>
<th>Uniplex PCR</th>
<th>Culture method</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0 h</td>
<td>3 h</td>
<td>6 h</td>
</tr>
<tr>
<td><em>V. cholerae</em></td>
<td>-</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td><em>V. parahaemolyticus</em></td>
<td>-</td>
<td>10^1</td>
<td>1</td>
</tr>
<tr>
<td><em>V. vulnificus</em></td>
<td>-</td>
<td>10^2</td>
<td>1</td>
</tr>
<tr>
<td><em>V. mimicus</em></td>
<td>-</td>
<td>10^2</td>
<td>10^2</td>
</tr>
</tbody>
</table>

After **6 h of enrichment** in APW is sufficient for detection of *Vibrio* spp. in cockle samples because **as few as 1 CFU** can be detected by tetraplex PCR
According to the DMSC food standard, the cockles distributing in the market in Nong Khai and Khon Kaen, and exported to Laos, were not up to standard due to almost of cockle samples were presence of *V. parahaemolyticus* and *V. cholerae* in 10 g of cockle sample whereas the criteria for *V. parahaemolyticus* and *V. cholerae* require has to be absent in 25 g of food sample (frozen and chilled fishery product).
Conclusion RBI on cockles, 2010

- *V. cholerae* was detected only by PCR method.
- Enrichment at least 6 hr was increased the detection.
- The reason of low prevalence of *V. cholerae* by culture method because of it might be in the stage of viable but non-culturable (VBNC)
- The results showed the important of cockles to spread *V. cholerae* and demonstrated PCR should be used for RBI
The cholera outbreak in Thailand during 2007 – 2010 were exclusively caused by the *V. cholerae O1* El Tor variant carrying the classical ctxB and El Tor rstR genes. It probably appeared in Thailand during recent years.

PFGE differentiated Thai El Tor variant isolates into nine pulsotypes that share the similarity of 88%.
Figure 1. Pulsed-field gel electrophoresis (PFGE) patterns among 343 Thai V. cholerae O1 isolates.

http://www.plosone.org/article/info:doi/10.1371/journal.pone.0030863
Conclusion RBI on human samples, 2007 -2010, Thailand (2)

• MLVA typing among isolates during outbreak episodes in different geographical and association of causative *V. cholerae* in cholera outbreak showed the different of MLVA typing.. In 2007, in northeastern region were triggered by the consumption of cockles contaminated with *V. cholerae* MLVA type 2.

• The study can be the data to trace the emergence, year long survival , or disappearance of a particular type (s) of isolate in terms of spatial and temporal association
Figure 3. Distribution of major MLVA types of V. cholerae O1 isolates during the 2007–2010 cholera outbreaks in Thailand.


http://www.plosone.org/article/info:doi/10.1371/journal.pone.0030863
Drug sensitivity of *V. cholerae* Ogawa

- C: 100%
- Nor: 67.9%
- Gm: 50%
- Amp: 85.7%
- E: 81.8%
- Te: 98.2%
- Sxt: 100%

Green: sensitive, light orange = intermediate, thick orange = resistance
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GIN system
Development Strategies

1. Leadership and management,
2. Development of e-Government services,
3. Infrastructure development,
4. Development and improvement of laws, regulations and obligations relating to government servicing process.
Integrated e-government Services Architecture

Access devices:
- Internet
- Call center
- Email
- Mobile phone
- Kiosk

Common Access Layer:
- Customers
  - General Publics
  - Enterprises
- Civil Service
- Commissions
- Organizations
- Departments

eGovernment Portal Domain:
- Authentication/Authorization services
- e-Payment services
- Registration Services
- Directory Services
- Cluster Services
- Information Exchange Services

Gateway Domain:
- Government Gateway

Back-end Ministries/Departments:
- Min-1
- Min-2
- Dept-1
- Dept-2
- Dept-3
Percentage of 6 Major chemical contamination, 2003 - 2012

Beta agonist, Borax, Sodium hydrosulfite, Formalin, anti fungal, pesticide residue
Risk based monitoring is needed?