Occurrence and distribution of *Vibrio cholerae* in the coastal environment of Peru


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**Vibrio cholerae**

- Causative agent of cholera
- Gram negative curved rod
- Ø-Proteobacteria
- Two serogroups that cause the vast majority of cholera - El Tor (O1) and O139
  - O1 detected worldwide, 0139 only detected in Asia at present
**Vibrio cholerae**

- Reservoirs include humans and aquatic sources
  - Occurs naturally in fresh, brackish and salt water, attached primarily to copepods (zooplankton) and other crustaceans

- VBNC stage

- Sequenced genome
  - Two chromosomes
  - Chromosome 1 harbors the virulence factors
V. *Cholerae* chromosomes

*Vibrio cholerae* O1 biovar eltor str. N16961 chromosome I, co...

Accession: NC_002505
Length: 2,961,149 bp; Genes: 2,860

*Vibrio cholerae* O1 biovar eltor str. N16961 chromosome II, c...

Accession: NC_002506
Length: 1,072,315 bp; Genes: 1,097
Virulence Factors

- Toxin co-regulated pilus (TCP)
  - Essential colonization factor
- Cholera toxin (CT)
  - Enterotoxin - cause of diarrhea
- Regulatory protein (ToxR)

ADP ribosylation

Nicotinamide adenine dinucleotide (NAD)
Cholera - A history

- Ancient disease (references date back to Hippocrates and Sanskrit writings)
  - Originally confined to India (Ganges delta)
  - Spread to other areas in the 19th century (to date 7 [8?] global pandemics have taken place)

The global spread of cholera during the seventh pandemic, 1961–1971. (CDC)
The Cholera Pandemics

- 1st - Bengal, India (1816-1826)
- 2nd - Reaches Europe & America (1829-1851)
- 3rd - Mainly Russia, Europe & America (1852-1860)
- 4th - Europe and Africa (1863-1875)
- 5th - Germany (1881-1896)
- 6th - Russia again (1899-1923)
- 7th - Indonesia, Bangladesh, India and Russia (1961-1970)
1991 New Strain Identified in Bangladesh, 8th pandemic?
Cholera cont’d

- John Snow recognized the epidemic spread of cholera in 1854

- While *V. cholerae* was first described & termed by Fillippo Pacini (1854)

Original map by Dr. John Snow showing the clusters of cholera cases in the London epidemic of 1854
Cholera cont’d

- Properly characterized as a waterborne disease by Robert Koch (1884)

- Devastating disease
  - Claims thousands of lives each year (WHO Report 1999)
  - 236,896 cases were notified from 52 countries in 2006 (WHO)
Drawing of Death bringing the cholera, in Le Petit Journal (Second pandemic)

1889 Cartoon (Fifth pandemic)
Cholera cont’d

• Infection is due to consumption of contaminated water or food and eating seafood
Cholera

- Need LARGE \(10^{11}\) infectious dose

- Disease characterized by a sudden onset of acute watery diarrhea (rice water stool) - death by severe dehydration and kidney failure
A Cholera Bed

A Chinese hygiene pamphlet
Occurrence and distribution of *Vibrio cholerae* in the coastal environment of Peru


Why Peru?

• In 1991, cholera reappeared in Latin America (initially Peru) after being absent for >100 years

• Between 1991-2000 cholera cases occur in a seasonal pattern
Objectives

• Determine the occurrence and distribution of this bug in sea water and plankton in Peruvian coastal waters

• To monitor occurrence of cholera cases in nearby cities
  – Identify a start of an epidemic and relate it to climate and environmental conditions
Table 2. Environmental data collected at the study sampling sites with minimum, maximum (upper line) and mean ± standard deviation (lower line) provided.

<table>
<thead>
<tr>
<th>Sampling sites</th>
<th>n</th>
<th>Air temperature (°C)</th>
<th>Water temperature (°C)</th>
<th>Salinity (p.p.t.)</th>
<th>Chlorophyll a (mg m⁻³)</th>
<th>Total bacterial counts (10⁵ cell ml⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Trujillo</td>
<td>36</td>
<td>16.0–22.0</td>
<td>12.0–29.0</td>
<td>35.0–37.0</td>
<td>1.5–150.9</td>
<td>0.02–11.6</td>
</tr>
<tr>
<td></td>
<td></td>
<td>19.6 ± 2.3 a</td>
<td>17.5 ± 4.0 a</td>
<td>35.6 ± 0.7 a</td>
<td>35.8 ± 48.8 a</td>
<td>1.9 ± 2.5 a</td>
</tr>
<tr>
<td></td>
<td>36</td>
<td>22.0–26.0</td>
<td>14.1–24.3</td>
<td>34.0–36.0</td>
<td>0.0–122.6</td>
<td>0.4–18.0</td>
</tr>
<tr>
<td></td>
<td></td>
<td>24.4 ± 1.7 b</td>
<td>17.8 ± 2.7 a</td>
<td>35.0 ± 0.4 b</td>
<td>18.6 ± 32.3 b</td>
<td>4.1 ± 5.3 a</td>
</tr>
<tr>
<td>Callao</td>
<td>36</td>
<td>16.0–30.3</td>
<td>15.0–25.1</td>
<td>34.0–37.0</td>
<td>0.0–136.2</td>
<td>0.01–14.1</td>
</tr>
<tr>
<td></td>
<td></td>
<td>22.8 ± 3.6 b</td>
<td>18.4 ± 2.5 a</td>
<td>35.1 ± 0.6 b</td>
<td>13.2 ± 28.6 b</td>
<td>2.5 ± 3.1 a</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>13.2 ± 28.6 b</td>
<td></td>
</tr>
<tr>
<td>Lima</td>
<td>68</td>
<td>15.0–30.0</td>
<td>12.0–23.5</td>
<td>30.0–37.0</td>
<td>0.0–16.6</td>
<td>0.003–10.8</td>
</tr>
<tr>
<td>Arequipa</td>
<td>39</td>
<td>15.0–30.0</td>
<td>12.0–23.5</td>
<td>30.0–37.0</td>
<td>0.0–16.6</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>23.8 ± 3.7 b</td>
<td>16.7 ± 2.9 a</td>
<td>35.1 ± 1.2 b</td>
<td>2.3 ± 3.2 c</td>
<td>1.9 ± 2.5 a</td>
</tr>
</tbody>
</table>

For each column, means that are statistically different are indicated by different letters (a, b or c); P < 0.05.

n = number of sampling events per site.
Table 1. Presence of *V. cholerae* O1 at sampling sites included in this study, three fractions combined.

<table>
<thead>
<tr>
<th>Sampling sites</th>
<th>% positive (no of positive samples/total samples analysed)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Culturable <em>V. cholerae</em> O1</td>
</tr>
<tr>
<td>Trujillo</td>
<td>0% (0/35)</td>
</tr>
<tr>
<td>Callao</td>
<td>0% (0/36)</td>
</tr>
<tr>
<td><strong>Lima</strong></td>
<td><strong>14.7% (10/68)</strong></td>
</tr>
<tr>
<td>Arequipa</td>
<td>0% (0/39)</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>5.6 % (10/178)</strong></td>
</tr>
</tbody>
</table>

DFA, direct fluorescent antibody assay.
Fig. 2. Percentage of samples positive for *V. cholerae* O1.
N – total samples analysed; W – sea water; P64 – plankton ≥ 64  m but < 202  m
P202 – plankton ≥ 202  m in size.
DFA, direct fluorescent antibody assay.
Q statistic (Fleiss, 1981) was used to compare the proportion of *V. cholerae* in each fraction; values statistically different from one another are indicated by a different letter (a, b or c); P < 0.05.
El Niño / La Niña

- Are important temperature fluctuations in surface waters of the tropical Eastern Pacific Ocean

- El Niño - Spanish for “the little boy”
  - characterized by unusually warm ocean temperatures in the Equatorial Pacific

- La Niña - Spanish for “the little girl”
  - characterized by cooler than normal waters in the Equatorial Pacific
Normal Conditions

El Niño Conditions
Fig. 3. Seasonal data of cholera incidence rate and sea surface temperature (SST) in Peru during the time of this study. (TR, Trujillo; LI, Lima; CA, Callao; AQP, Arequipa).
Conclusions

- *V. cholerae* O1 is present in sea water and plankton in the Peruvian coast

- *V. cholerae* O1 is culturable in polluted sites, although difficult in isolating them in unpolluted areas
  - Fluorescent monoclonal antibody allows for detection

- Seasonal changes in water temperature is a factor to predict cholera outbreaks
Global Warming

• Average increase in the temperature of the atmosphere near the Earth's surface and in the troposphere
  – Can contribute to changes in global climate patterns

• Burning fossil fuels (coal, oil) and deforestation cause heat trapping green house gasses to increase
  – Green house gases help keep the planet's surface warmer (prevents heat from escaping to space)
  – Too much will cause earth’s temperature to climb

• Affects agriculture, forests, energy production, polar ice caps melting…etc

• But…..
Global Warming

- Global warming might create a favorable environment for *V. cholerae* and increase the incidence of the disease in vulnerable areas
  - More epidemics

- Other diseases that could have an impact include*
  - Yellow fever
  - Dengue
  - Malaria
  - Lyme disease
  - Tick borne encephalitis