Case Presentation

• 18 year old native from Thailand (moved to US at 10 years of age) presents in August with 2 days of illness including fever, headache, bone pain, and nausea 3 days after returning from a vacation in Puerto Rico. He vacationed in San Juan, stayed in a hotel, ate and drank local foods and beverages from the hotel and restaurants, and swam in the hotel pool and ocean. He does not recall mosquito exposure and did not reliably use DEET. He had no animal exposures. He has no past medical history except for a single STI. He takes no medications to include those acquired OTC. He does not abuse ETOH nor use illicit drugs.

• The LEAST likely diagnosis would be:
  1. chikungunya
  2. dengue
  3. leptospirosis
  4. acute HIV
Lecture Objectives

1. Attendees will understand the **global distribution** of dengue virus circulation and disease.
2. Attendees will understand the spectrum of **dengue clinical phenotypes** and the clinical and laboratory findings and parameters which distinguish mild from severe forms of the disease.
3. Attendees will understand the nuances of **treating dengue** and best management practices.
4. Attendees will become familiar with **countermeasure development** efforts.
Lecture Outline

• Basics
• Epidemiology
• Clinical Phenotypes
• Pathophysiology
• Diagnostics
• Treatment
• Vaccine Development

Kuhn, R., Purdue University
Dengue

• Basics
  – Family Flaviviridae, Genus Flavivirus, Species Dengue
    • Same family as WNV, YF, JE, Zika
  – RNA virus, 3 structural and 7 non-structural genes
    • Different functions during infection process
    • Different targets for drugs/vaccines

  4 dengue virus types: DENV-1-4
  • Multiple genotypes within each dengue virus type
• Transmission
  – Feeding vector
  – Laboratory acquired
  – Blood supply?
  – Organ donation?

• Vector
  – Aedes aegypti
  – Aedes albopictus

Is dengue a threat to the blood supply?

<table>
<thead>
<tr>
<th>Table 3. Dengue and donor deferral</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Country</strong></td>
</tr>
<tr>
<td>Singapore*</td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td>Hong Kong*</td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td>Sri Lanka*</td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td>Australia†</td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td>New Zealand‡</td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td>UK‡</td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td>United States‡</td>
</tr>
<tr>
<td></td>
</tr>
</tbody>
</table>

*Endemic for dengue.
†Non-endemic parts of Northern Australia.
‡Non-endemic.
Dengue Epidemiology

Dengue Ward: QSNICH, Bangkok, Thailand (Photo: Christopher Brown, IHT)
Areas supporting dengue virus transmission.
Dengue Burden
Under-estimated and under-reported

Table 1 | Estimated burden of dengue in 2010, by continent

<table>
<thead>
<tr>
<th>Type</th>
<th>Apparent Millions (credible interval)</th>
<th>Inapparent Millions (credible interval)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Africa</td>
<td>15.7 (10.5–22.5)</td>
<td>48.4 (34.3–65.2)</td>
</tr>
<tr>
<td>Asia</td>
<td>66.8 (47.0–94.4)</td>
<td>204.4 (151.8–273.0)</td>
</tr>
<tr>
<td>Americas</td>
<td>13.3 (9.5–18.5)</td>
<td>40.5 (30.5–53.3)</td>
</tr>
<tr>
<td>Oceania</td>
<td>0.18 (0.11–0.28)</td>
<td>0.55 (0.35–0.82)</td>
</tr>
<tr>
<td>Global</td>
<td>96 (67.1–135.6)</td>
<td>293.9 (217.0–392.3)</td>
</tr>
</tbody>
</table>

389.9M infections/ year

Figure 3. Average number of dengue cases in 30 most highly endemic countries/territories as reported to WHO, 2004–2010
Average dengue incidence per 100,000 by country, Region of the Americas, 1980–2007.
## Table 1: Confirmed cases and deaths from 2006 to 2011 in the affected areas of Pakistan.

<table>
<thead>
<tr>
<th>Year</th>
<th>Khyber Pakhtunkhwa</th>
<th>Sindh</th>
<th>Karachi</th>
<th>Punjab</th>
<th>Lahore</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Cases</td>
<td>Deaths</td>
<td>All parts</td>
<td>Cases</td>
<td>Deaths</td>
</tr>
<tr>
<td>2006</td>
<td>31</td>
<td>1</td>
<td>1500 50</td>
<td>1500 50</td>
<td>800 1</td>
</tr>
<tr>
<td>2007</td>
<td>0</td>
<td>0</td>
<td>950 22</td>
<td>950 20</td>
<td>258 0</td>
</tr>
<tr>
<td>2008</td>
<td>30</td>
<td>4</td>
<td>585 6</td>
<td>585 6</td>
<td>1450 20</td>
</tr>
<tr>
<td>2009</td>
<td>100</td>
<td>7</td>
<td>550 7</td>
<td>550 7</td>
<td>300 2</td>
</tr>
<tr>
<td>2010</td>
<td>0</td>
<td>0</td>
<td>5000 35</td>
<td>4500 16</td>
<td>4000 3</td>
</tr>
<tr>
<td>2011</td>
<td>296</td>
<td>8</td>
<td>952 18</td>
<td>755 15</td>
<td>21,314 337</td>
</tr>
</tbody>
</table>

*a Data collected from National Institute of Health Islamabad.*

*b Data collected from provincial health departments.*
Africa

Emerging Infectious Diseases • www.cdc.gov/eid • Vol. 17, No. 8, August 2011

Figure. Dengue and *Aedes aegypti* mosquitoes in Africa. Brown indicates 34 countries in which dengue has been reported, including dengue reported only in travelers, and *Ae. aegypti* mosquitoes. Light brown indicates 13 countries (Mauritania, The Gambia, Guinea-Bissau, Guinea, Sierra Leone, Liberia, Niger, Chad, Central African Republic, Republic of the Congo, Malawi, Zimbabwe, and Botswana) in which dengue has not been reported but that have *Ae. aegypti* mosquitoes. White indicates 5 countries (Western Sahara, Morocco, Algeria, Tunisia, and Libya) for which data for dengue and *Ae. aegypti* mosquitoes are not available.

Brown – dengue reported

Light Brown – dengue not reported but vector exists

White – data not available
CDC Dengue Map – 1 JUL – 10 SEP 2014

Reporting sources – WHO, MOHs, ProMed, GeoSentinel, EuroSurveillance, World Org
High financial and human cost

Economic Impact of Dengue Illness in the Americas

Donald S. Shepard,* Laurent Coudeville, Yara A. Halasa, Betzana Zambrano, and Gustavo H. Dayan
Brandeis University, Waltham, Massachusetts; Sanofi Pasteur, Lyon, France; Sanofi Pasteur, Swiftwater, Pennsylvania

**Figure 1.** Number of dengue reported cases in the Americas from 2000 to 2007.

**Figure 3.** Annual economic burden in the Americas from 2000 to 2007 (in 2010 US$).

Economic and Disease Burden of Dengue in Southeast Asia

Figure 3. Aggregate values of dengue episodes and economic burden by year for 12 countries in SEA (2001–2010).

United States

Dengue Fever, Hawaii, 2001–2002

Emerging Infectious Diseases • www.cdc.gov/eid • Vol. 11, No. 5, May 2005

Paul V. Effler,* Lorrin Pang,* Paul Kitsutani,† Vance Vorndam,† Michele Nakata,* Tracy Ayers,* Joe Elm,* Tammy Tom,* Paul Reiter,† José G. Rigau-Perez,† John M. Hayes,† Kristin Mills,* Mike Napier,‡ Gary G. Clark,‡ and Duane J. Gubler* for the Hawaii Dengue Outbreak Investigation Team

First suspect dengue case reported 9/12

Legend

Kauai
Oahu
Maui

Weekly interval beginning

Infections (n)

20 3 17 1 15 12 9 6 3 0 17 1 15 12 9 6 3 0

May Jun Jul Aug Sep Oct Nov Dec Jan Feb

Legend

Maui Cases

Count

1 2 5 28 52

Kaanapali 7379
Wailea 35388
Waikoloa 6542
Waipahu 11334

Laie/Keahamau 35388
Waimanalo 10324
Kaneohe 40000
Kailua 25203

Oahu Cases

Count

1 3 9 11

Sunset Beach 5035
Lualualei 25203

Honolulu Area

Community

Island
United States

Identification of Dengue Fever Cases in Houston, Texas, with Evidence of Autochthonous Transmission Between 2003 and 2005


DENGUE SURVEILLANCE IN TEXAS, 1995

JULIE A. RAWLINGS, KATHERINE A. HENDRICKS, CHRISTINE R. BURGESS, RICHARD M. CAMPMAN, GARY G. CLARK, LAURA J. TABONY, AND MARY ANN PATTERSON

Infectious Disease Epidemiology and Surveillance Division and Bureau of Laboratories, Texas Department of Health, Austin, Texas; Dengue Branch, Centers for Disease Control and Prevention, San Juan, Puerto Rico

Dengue Hemorrhagic Fever --- U.S.-Mexico Border, 2005


Epidemic Dengue and Dengue Hemorrhagic Fever at the Texas–Mexico Border: Results of a Household-based Seroepidemiologic Survey, December 2005

UNCLASSIFIED
Locally Acquired Dengue — Key West, Florida, 2009–2010

FIGURE. Number of locally acquired dengue cases (N = 28), by week of illness onset and method of identification — Key West, Florida, 2009–2010

TABLE. Characteristics of patients (N = 28) with locally acquired dengue — Key West, Florida, 2009–2010

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>No.</th>
<th>(%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>19</td>
<td>(68)</td>
</tr>
<tr>
<td>Female</td>
<td>9</td>
<td>(32)</td>
</tr>
<tr>
<td>Age group (yrs)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt;20</td>
<td>1</td>
<td>(4)</td>
</tr>
<tr>
<td>21–40</td>
<td>11</td>
<td>(39)</td>
</tr>
<tr>
<td>41–60</td>
<td>11</td>
<td>(39)</td>
</tr>
<tr>
<td>&gt;60</td>
<td>5</td>
<td>(18)</td>
</tr>
<tr>
<td>Race</td>
<td></td>
<td></td>
</tr>
<tr>
<td>White</td>
<td>24</td>
<td>(86)</td>
</tr>
<tr>
<td>Black</td>
<td>3</td>
<td>(11)</td>
</tr>
<tr>
<td>Asian/Pacific Islander</td>
<td>1</td>
<td>(4)</td>
</tr>
<tr>
<td>Ethnicity</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Non-Hispanic</td>
<td>25</td>
<td>(89)</td>
</tr>
<tr>
<td>Hispanic</td>
<td>3</td>
<td>(11)</td>
</tr>
<tr>
<td>Symptoms</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fever</td>
<td>28</td>
<td>(100)</td>
</tr>
<tr>
<td>Headache</td>
<td>22</td>
<td>(79)</td>
</tr>
<tr>
<td>Myalgia</td>
<td>23</td>
<td>(82)</td>
</tr>
<tr>
<td>Arthralgia</td>
<td>18</td>
<td>(64)</td>
</tr>
<tr>
<td>Eye pain</td>
<td>14</td>
<td>(50)</td>
</tr>
<tr>
<td>Rash</td>
<td>15</td>
<td>(54)</td>
</tr>
<tr>
<td>Bleeding</td>
<td>6</td>
<td>(21)</td>
</tr>
</tbody>
</table>

* Percentages might not add to 100% because of rounding.
Florida 2013

- Locally Acquired Dengue as of SEP 2013:
  - 22 cases
  - 20 residents, 2 out of state
  - Martin (21) and Miami-Dade (1)
- Imported (traveler) Dengue 2013:
  - 88 cases imported into Florida
- 69 / 110 cases serotyped by PCR

<table>
<thead>
<tr>
<th>Serotype</th>
<th># of cases</th>
</tr>
</thead>
<tbody>
<tr>
<td>DENV-1</td>
<td>50</td>
</tr>
<tr>
<td>DENV-2</td>
<td>1</td>
</tr>
<tr>
<td>DENV-3</td>
<td>3</td>
</tr>
<tr>
<td>DENV-4</td>
<td>16</td>
</tr>
<tr>
<td>2013 total</td>
<td>69</td>
</tr>
</tbody>
</table>

References: 5) Florida Dept. of Health Website  
6) Florida Arbovirus Surveillance: Week 36 September 2013
Puerto Rico

Suspected cases reported compared to the historical average

- Totals through 29 JUL 2014: 2,468 suspected, 381 confirmed, 2 severe (DHF), 0 deaths
- Totals through 31 DEC 2013: 18,164 suspected, 9,032 confirmed, 50 severe (DHF), 12 deaths

Total viral identifications in the last 12 months

Serotype Distribution

References: 1) CDC Website  4) Dengue Surveillance Weekly Report, CDC, December 2013
“With respect to specific diagnoses, malaria was one of the three most frequent causes of systemic febrile illness among travelers from every region, although travelers from every region except sub-Saharan Africa and Central America had confirmed or probable dengue more frequently than malaria.”
Dengue and US Military Operations
from the Spanish–American War
through Today

Robert V. Gibbons, Matthew Streitz, Tatyana Babina, and Jessica R. Fried
Emerging Infectious Diseases • www.cdc.gov/eid • Vol. 18, No. 4, April 2012

Figure 1. Captain Percy Ashburn.
Figure 2. First Lieutenant Charles Craig.
Figure 3. Lieutenant Commander J.P. Siler.
Figure 4. Major Albert Sabin.
Figure 6. Airplane spraying of DDT over Manila, the Philippines, 1945.
Dengue Risk / Threat to DoD

• Prevalence and Risk to Soldiers (2003-2012)
  – **Total Cases**: 631
    • Active Duty: 177; Reserve: 35; MHS Beneficiaries: 419
    • No record of attributable deaths
  – Dengue Mission Impact Projections
    • Not severe: hospitalized ~5-7 days, low functioning ~14-28 days
    • Severe: evacuation to MTF, ICU care?, death?, LDD >1 month
  – Deployment
    • DODSR: 500 samples, deployed between 2006-2008
      – 11.2% seroprevalence of dengue antibody
      – 2.4% with monovalent profile (high risk with next infection)

References: *Dengue Tetravalent Vaccine CDD; *DMSS
Seroprevalence of DENV Exposure in Deployed Personnel

- DODSR, 1000 samples, first time deployers, 2008-2011
- 250 samples selected per COCOM
- Tested for presence of neutralizing antibody by microneut assay
- Overall 7.6% seroprevalence rate of past dengue exposure
- 1.5% seroconversion rate during deployment (first infection)
- Increased self report of fever during deployment in those with antibodies

<table>
<thead>
<tr>
<th>Region</th>
<th>Percent</th>
</tr>
</thead>
<tbody>
<tr>
<td>Central America</td>
<td>4.8%</td>
</tr>
<tr>
<td>South America</td>
<td>12.4%</td>
</tr>
<tr>
<td>Asia</td>
<td>7.2%</td>
</tr>
<tr>
<td>Africa</td>
<td>6.0%</td>
</tr>
<tr>
<td>Total</td>
<td>7.6%</td>
</tr>
</tbody>
</table>
Seroprevalence of DENV Exposure in USASOC Personnel

- USASOC and WRAIR viral disease threat characterization
- Pre- and post-deployment sample collection in deploying SOC personnel
- Tested for presence of neutralizing antibody by microneut assay

- NOV 2013: 411 pre-deployment and 7 post-deployment samples tested
  - N = 56 pre-deployment positive (13.6%)
  - N = 8 pre-deployment monovalent profiles (2.0%)
  - N = 2/7 post-deployment seroconversions (qualitative [neg to pos])

- Summary: USASOC personnel are highly primed to dengue, a proportion are in high risk category for severe disease with secondary infection, clinical impact will likely not be documented, is this knowledge changing approach to febrile patient during deployment?
<table>
<thead>
<tr>
<th>Disease</th>
<th>2010 COCOM panel rank</th>
<th>ID-IDEAL Rank</th>
</tr>
</thead>
<tbody>
<tr>
<td>Malaria</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td><strong>Dengue</strong></td>
<td><strong>2</strong></td>
<td><strong>3</strong></td>
</tr>
<tr>
<td>Diarrhea, bacterial</td>
<td>3</td>
<td>1</td>
</tr>
<tr>
<td>MDR wound pathogens</td>
<td>4</td>
<td>NA</td>
</tr>
<tr>
<td>Leishmaniasis</td>
<td>5</td>
<td>19</td>
</tr>
<tr>
<td>Q fever (Coxiella burnetti)</td>
<td>6</td>
<td>26</td>
</tr>
<tr>
<td>Norovirus / viral diarrhea</td>
<td>7</td>
<td>NA</td>
</tr>
<tr>
<td>Influenza</td>
<td>8</td>
<td>NA</td>
</tr>
<tr>
<td>Leptospirosis</td>
<td>10</td>
<td>7</td>
</tr>
<tr>
<td>Diarrhea, protozoal</td>
<td>11</td>
<td>11</td>
</tr>
<tr>
<td>TB</td>
<td>12</td>
<td>NA</td>
</tr>
<tr>
<td>CCHF</td>
<td>13</td>
<td>10</td>
</tr>
<tr>
<td>HIV</td>
<td>14</td>
<td>8</td>
</tr>
<tr>
<td>HFRS</td>
<td>15</td>
<td>17</td>
</tr>
<tr>
<td>Chikungunya</td>
<td>16</td>
<td>4</td>
</tr>
<tr>
<td>Meningococcal meningitis</td>
<td>17</td>
<td>20</td>
</tr>
<tr>
<td>Plague</td>
<td>18</td>
<td>27</td>
</tr>
<tr>
<td>Rickettsioses</td>
<td>19</td>
<td>18</td>
</tr>
<tr>
<td>Viral encephalitides</td>
<td>20</td>
<td>NA</td>
</tr>
</tbody>
</table>
Factors Driving Transmission

- DENVs
  - Travel in hosts
  - Viral evolution
- Naïve hosts
  - Population growth
  - Increased urbanization
- Vector
  - Ecologic changes
  - Evolution
Factors Driving Disease

• There is a significantly increased risk of severe dengue disease (dengue hemorrhagic fever) when infected a second time with a different DENV type than what you were infected with during your first infection (i.e. DENV-4 during first infection, DENV-2 during second).

• Co-circulation of numerous DENV types in similar time and space increases risk of experiencing multiple infections with different DENV types.
DENV Type Distribution - 1970

D. Gubler
Global Air Travel Flight Patterns

http://upload.wikimedia.org/wikipedia/commons/a/ac/World-airline-routemap-2009.png
DENV Type Distribution - 2004

D. Gubler
Case Presentation

• 18 year old native from Thailand (moved to US at 10 years of age) presents in August with 2 days of illness including fever, headache, bone pain, and nausea 3 days after returning from a vacation in Puerto Rico. You suspect he has a dengue infection. He is tolerating PO intake without vomiting and is urinating. Vital signs except for temperature (102.5F) are in the range of normal. Mucous membranes are moist, skin turgor is normal, abdominal exam is normal, and lungs are clear. A CBC reveals a low WBC (3.5k) but otherwise is within normal limits. Electrolytes are normal.

• What is the most reasonable initial management strategy?
  
  – 1. treat as outpt, provide NSAIDS, encourage PO fluids
  – 2. treat as inpt, provide 1L NS bolus, monitor in ICU setting
  – 3. treat as outpt, provide acetaminophen, encourage po fluids, F/U
  – 4. treat as inpt, encourage PO fluids, perform q6 hr HCT evaluations
Clinical Phenotype

AN ACCOUNT OF THE

Bilious Remitting Fever,

AS IT APPEARED IN PHILADELPHIA, IN THE SUMMER AND AUTUMN OF THE YEAR 1780.

The pains which accompanied this fever were exquisitely severe in the head, back, and limbs. The pains in the head were sometimes in the back parts of it, and at other times they occupied only the eyeballs. In some people, the pains were so acute in their backs and hips, that they could not lie in bed. In others, the pains affected the neck and arms, so as to produce in one instance a difficulty of moving the fingers of the right hand. They all complained more or less of a soreness in the seats of these pains, particularly when they occupied the head and eyeballs. A few complained of their flesh being sore to the touch, in every part of the body. From these circumstances, the disease was sometimes believed to be a rheumatism. But its more general name among all classes of people was, the Break-bone fever.
Dengue Infection
Clinical Phenotypes

Dengue virus infection

- Asymptomatic
- Symptomatic
  - Undifferentiated fever (viral syndrome)
  - Dengue fever syndrome
    - Without haemorrhage
    - With unusual haemorrhage
  - Dengue haemorrhagic fever (plasma leakage)
    - No shock
    - Dengue shock syndrome

Dengue fever

Dengue haemorrhagic fever


WHO 95629
**1997 WHO dengue fever case definition**

- **Probable dengue infection**
  - Acute febrile illness and at least 2 of the following:
    - Headache, retro-orbital pain, myalgia, arthralgia, rash, hemorrhagic manifestations, and leukopenia

- **Confirmed dengue infection**
  - Above + lab confirmation (at least one method below)
    - DENV isolation (blood, autopsy samples)
    - 4 fold rise in IgM or IgG to any of the four DENV antigens in paired blood samples
    - Demonstration of DENV antigen (tissue, CSF, serum) by ELISA, Immunohistochemistry, immunofluorescence
  - PCR +
### 1997 WHO case definition for DHF/DSS

**Table 1**

1997 World Health Organization (WHO) case definition for dengue hemorrhagic fever and dengue shock syndrome*

<table>
<thead>
<tr>
<th>DHF, the following must all be present:</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fever, or history of acute fever, lasting 2–7 days, occasionally biphasic</td>
</tr>
<tr>
<td>Hemorrhagic tendencies, evidenced by at least one of the following:</td>
</tr>
<tr>
<td>A positive tourniquet test</td>
</tr>
<tr>
<td>Petechiae, ecchymoses, or purpura</td>
</tr>
<tr>
<td>Bleeding from the mucosa, gastrointestinal tract, injection sites, or other locations</td>
</tr>
<tr>
<td>Hematemesis or melaena</td>
</tr>
<tr>
<td>Thrombocytopenia (100,000 cells/mm³ or less)</td>
</tr>
<tr>
<td>Evidence of plasma leakage caused by increased vascular permeability, manifested by at least one of the following:</td>
</tr>
<tr>
<td>A rise in the hematocrit equal to or &gt; 20% above average for age, sex, and population</td>
</tr>
<tr>
<td>A drop in the hematocrit following volume replacement treatment equal to or &gt; 20% of baseline</td>
</tr>
<tr>
<td>Signs of plasma leakage such as pleural effusion, ascites, and hypoproteinemia</td>
</tr>
<tr>
<td>Case definition for dengue shock syndrome:</td>
</tr>
<tr>
<td>All of the above four criteria for DHF must be present, plus evidence of circulatory failure manifested by:</td>
</tr>
<tr>
<td>Rapid and weak pulse, and</td>
</tr>
<tr>
<td>Narrow pulse pressure (&lt; 20 mm Hg)</td>
</tr>
<tr>
<td>or manifested by:</td>
</tr>
<tr>
<td>Hypotension for age, and</td>
</tr>
<tr>
<td>Cold, clammy skin and restlessness.</td>
</tr>
</tbody>
</table>

*HF = dengue hemorrhagic fever.
2009 WHO dengue case definitions

TABLE 2

<table>
<thead>
<tr>
<th>2009 World Health Organization (WHO) dengue case definitions*14</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Probable dengue</strong></td>
</tr>
<tr>
<td>Live in or travel to dengue endemic area, fever and two of</td>
</tr>
<tr>
<td>the following:</td>
</tr>
<tr>
<td>Nausea, vomiting</td>
</tr>
<tr>
<td>Rash</td>
</tr>
<tr>
<td>Aches and pains</td>
</tr>
<tr>
<td>Tourniquet test positive</td>
</tr>
<tr>
<td>Leucopenia</td>
</tr>
<tr>
<td>Any “Warning Sign”</td>
</tr>
<tr>
<td><strong>Dengue with Warning Signs</strong></td>
</tr>
<tr>
<td>Abdominal pain or tenderness</td>
</tr>
<tr>
<td>Persistent vomiting</td>
</tr>
<tr>
<td>Clinical fluid accumulation</td>
</tr>
<tr>
<td>Mucosal bleed</td>
</tr>
<tr>
<td>Lethargy, restlessness</td>
</tr>
<tr>
<td>Liver enlargement &gt; 2 cm</td>
</tr>
<tr>
<td>Laboratory increase in HCT concurrent with rapid decrease in</td>
</tr>
<tr>
<td>platelet count</td>
</tr>
</tbody>
</table>

Severe dengue (short form)

- Severe plasma leakage
  - Shock (DSS)
    - Fluid accumulation with respiratory distress
  - Severe bleeding (as evaluated by clinician)
  - Severe organ involvement
    - Liver AST or ALT >= 1,000
    - CNS impaired consciousness
    - Heart and other organs

Severe dengue (long form)

- There is evidence of plasma leakage, such as:
  - High or progressively rising hematocrit;
  - Pleural effusions or ascites;
  - Circulatory compromise or shock (tachycardia, cold and clammy extremities, capillary refill time greater than 3 seconds, weak or undetectable pulse, narrow pulse pressure or, in late shock, unrecordable blood pressure).
- There is significant bleeding
- There is an altered level of consciousness (lethargy or restlessness, coma, convulsions).
- There is severe gastrointestinal involvement (persistent vomiting, increasing or intense abdominal pain, jaundice).
- There is severe organ impairment (acute liver failure, acute renal failure, encephalopathy or encephalitis, or other unusual manifestations, cardiomyopathy) or other unusual manifestations.

---

*HCT = hematocrit; DSS = dengue shock syndrome; AST = aspartate aminotransferase; ALT = alanine aminotransferase; CNS = central nervous system.
Figure 1.4 Suggested dengue case classification and levels of severity

**DENGUE ± WARNING SIGNS**

- **With warning signs**
- **Without**

**SEVERE DENGUE**

1. Severe plasma leakage
2. Severe haemorrhage
3. Severe organ impairment

**CRITERIA FOR DENGUE ± WARNING SIGNS**

- **Probable dengue**
  - Live in / travel to dengue endemic area.
  - Fever and 2 of the following criteria:
    - Nausea, vomiting
    - Rash
    - Aches and pains
    - Tourniquet test positive
    - Leukopenia
    - Any warning sign

- **Laboratory-confirmed dengue**
  - Important when no sign of plasma leakage

**Warning signs***

- Abdominal pain or tenderness
- Persistent vomiting
- Clinical fluid accumulation
- Mucosal bleed
- Lethargy, restlessness
- Liver enlargement >2 cm
- Laboratory: increase in HCT concurrent with rapid decrease in platelet count

*(requiring strict observation and medical intervention)*

**CRITERIA FOR SEVERE DENGUE**

- **Severe plasma leakage**
  - Leading to:
    - Shock (DSS)
    - Fluid accumulation with respiratory distress

- **Severe bleeding**
  - As evaluated by clinician

- **Severe organ involvement**
  - Liver: AST or ALT >= 1000
  - CNS: Impaired consciousness
  - Heart and other organs
Dengue Clinical and Lab Parameters

24 hr period around defervescence = danger period
Dengue Fever

6 year old male with acute primary den-1, DF

Negative tourniquet test  Pleural effusion index 0.0

Fever (°C)

Pulse pressure (mm Hg)

White blood cell count

Liver

Hematocrit (%)

Platelet count

Clinical illness day

Fever day

AST

ALT

Albumin

Plt

WBC

500x10^3
400x10^3
300x10^3
200x10^3
100x10^3
Dengue Hemorrhagic Fever

7 year old male with acute secondary den-1, grade III DHF

- Fever (°C)
- Pulse pressure (mm Hg)
- White blood cell count
- Liver

Positive tourniquet test
Pleural effusion index 25.5

Hematocrit (%)
Platelet count
Clinical illness day
Fever day
R LAT decubitus X-ray showing a large pleural effusion, DHF the day after defervescence. Degree of plasma leakage may be quantified by means of the pleural effusion index. The pleural effusion index is calculated as 100 times the maximum width of the R pleural effusion, divided by the maximal width of the R hemithorax.
Hemoconcentration
Diagnosing Dengue

- Maintain high degree of suspicion
  - Geographic location
  - Clustering of cases
- History and physical
  - Clinical presentation
  - Vital signs (HR, BP, Temp)
  - Dengue tourniquet test (TT)
- Clinical lab assessment
  - CBC (WBC, HCT, PLT), AST/ALT
- Dengue tests in US
  - IgM capture ELISA
  - CDC RT-PCR
- Dengue area, +Clinical, +TT, WBC<5k = High PPV (~70%)
Evaluation of diagnostic tests: dengue

Rosanna W. Peeling, Harvey Artsob, Jose Luis Pelegrino, Philippe Buchy, Mary J. Cardosa, Shamala Devi, Delia A. Enria, Jeremy Farrar, Duane J. Gubler, Maria G. Guzman, Scott B. Halstead, Elizabeth Hunsperger, Susie Kliks, Harold S. Margolis, Carl M. Nathanson, Vinh Chau Nguyen, Nidia Rizzo, Susana Vázquez and Sutee Yoksan

Figure 1 | Comparative merits of direct and indirect laboratory methods for the diagnosis of dengue infections. Opportunity refers to the fact that antibody testing is usually the most practical diagnostic option available.
Dengue Tourniquet Test

- Measure BP
- SBP + DBP / 2 = target insufflation pressure for test
- Inspect area near antecubital fossa
  - You will assess delta before / after
- Inflate to target pressure
- Hold for 5 minutes
- Remove cuff
- Reassess antecubital fossa
- Count # of petechiae in 2.5 cm² area
- ≥10 new petechiae is positive

• TT measures capillary fragility, severe disease predictor?
Case Presentation

• 18 year old native from Thailand (moved to US at 10 years of age) presents in August with 2 days of illness including fever, headache, bone pain, and nausea 3 days after returning from a vacation in Puerto Rico. You decide to manage him as an outpatient. He fails to follow up as requested but does return day 6 of his illness afebrile with resolution of some symptoms but he now has abdominal pain, slight shortness of breath, and lethargy.

• What is the most reasonable management strategy at this point?
  – 1. Continue close follow up as outpatient, encourage PO fluid intake, this is the natural history of a resolving dengue infection
  – 2. Admit to the hospital, approach as a critically ill patient, this is a severe dengue virus infection
  – 3. Admit to the hospital, evaluate for another infectious disease process, these new symptoms represent a new medical problem
  – 4. Prescribe doxycycline, he probably has leptospirosis
Dengue Treatment

**Dengue Without Warning Signs**

**Group A**
(May be sent home)

- **Group criteria**
  - Patients who do not have warning signs
  AND
  - who are able:
    - to tolerate adequate volumes of oral fluids
    - to pass urine at least once every 6 hours

- **Laboratory tests**
  - full blood count (FBC)
  - haematocrit (HCT)

**Treatment**
Advice for:
- adequate bed rest
- adequate fluid intake
- Paracetamol, 4 gram maximum per day in adults and accordingly in children.

Patients with stable HCT can be sent home.

**Monitoring**
Daily review for disease progression:
- decreasing white blood cell count
- defervescence
- warning signs (until out of critical period).
Advice for immediate return to hospital if development of any warning signs, and
- written advice for management (e.g. home care card for dengue).
**DENGUE WITH WARNING SIGNS**

**Group B**
(Referred for in-hospital care)

**Group criteria**
Patients with any of the following features:
- co-existing conditions such as pregnancy, infancy, old age, diabetes mellitus, renal failure
- social circumstances such as living alone, living far from hospital

**Monitoring**
Monitor:
- temperature pattern
- volume of fluid intake and losses
- urine output (volume and frequency)
- warning signs
- HCT, white blood cell and platelet counts.

**Laboratory tests**
- full blood count (FBC)
- haematocrit (HCT)

**Treatment**
- Encouragement for oral fluids. If not tolerated, start intravenous fluid therapy 0.9% saline or Ringer’s Lactate at maintenance rate.
Figure 2.2 Algorithm for fluid management in compensated shock

- **Assess**
- **Intervene**
- **Re-assess**

**Compensated shock (systolic pressure maintained but has signs of reduced perfusion)**
Fluid resuscitation with isotonic crystalloid:
5–10 ml/kg/hr over 1 hour

**Flowchart**

- **Improvement**
  - YES
  - IV crystalloid 5–7 ml/kg/hr for 1–2 hours, then:
    - reduce to 3–5 ml/kg/hr for 2–4 hours;
    - reduce to 2–3 ml/kg/hr for 2–4 hours.
    - If patient continues to improve, fluid can be further reduced.
    - Monitor HCT 6–8 hourly.
    - If the patient is not stable, act according to HCT levels:
      - if HCT increases, consider bolus fluid administration or increase fluid administration;
      - if HCT decreases, consider transfusion with fresh whole blood.
    - Stop at 48 hours.
  
  - NO
    - **Check HCT**
    - HCT \(\uparrow\) or high
      - Administer 2nd bolus of fluid
        - 10–20 ml/kg/hr for 1 hour
        - **Improvement**
          - YES
            - If patient improves, reduce to 7–10 ml/kg/hr for 1–2 hours
              - Then reduce further
          - NO
    - HCT \(\downarrow\)
      - Consider significant occult/overt bleed
        - Initiate transfusion with fresh whole blood

UNCLASSIFIED Slide 52
Figure 2.3 Algorithm for fluid management in hypotensive shock

**Hypotensive shock**
Fluid resuscitation with 20 ml/kg isotonic crystalloid or colloid over 15 minutes
Try to obtain a HCT level before fluid resuscitation

- **Improvement**
  - **YES**
  - Crystalloid/colloid 10 ml/kg/hr for 1 hour, then continue with:
    - IV crystalloid 5-7 ml/kg/hr for 1-2 hours, reduce to 3-5 ml/kg/hr for 2-4 hours; reduce to 2-3 ml/kg/hr for 2-4 hours.
    - If patient continues to improve, fluid can be further reduced.
    - Monitor HCT 6-hourly.
    - If the patient is not stable, act according to HCT levels:
      - if HCT increases, consider bolus fluid administration or increase fluid administration;
      - if HCT decreases, consider transfusion with fresh whole blood.
    - Stop at 48 hours.

  - **NO**

- **Review 1st HCT**
  - HCT up or high
    - Administer 2nd bolus fluid (colloid) 10-20 ml/kg over ½ to 1 hour
  - HCT down
    - Consider significant occult/overt bleed
      - Initiate transfusion with fresh whole blood

- **Improvement**
  - **YES**
  - **NO**
Consider occult / overt bleed

- Assess
- Intervene
- Re-assess
### Textbox A. Good clinical practice and bad clinical practice

<table>
<thead>
<tr>
<th>Good practice</th>
<th>Bad practice</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Assessment and follow-up of patients with non-severe dengue and careful instruction of warning signs to watch out for</td>
<td>Sending patients with non-severe dengue home with no follow-up and inadequate instructions</td>
</tr>
<tr>
<td>2. Administration of paracetamol for high fever if the patient is uncomfortable</td>
<td>Administration of acetylsalicylic acid (aspirin) or ibuprofen</td>
</tr>
<tr>
<td>3. Obtaining a haematocrit level before and after fluid boluses</td>
<td>Not knowing when haematocrit levels are taken with respect to fluid therapy</td>
</tr>
<tr>
<td>4. Clinical assessment of the haemodynamic status before and after each fluid bolus</td>
<td>No clinical assessment of patient with respect to fluid therapy</td>
</tr>
<tr>
<td>5. Interpretation of haematocrit levels in the context of fluid administered and haemodynamic assessment</td>
<td>Interpretation of haematocrit levels independent of clinical status</td>
</tr>
<tr>
<td>6. Administration of intravenous fluids for repeated vomiting or a high or rapidly rising haematocrit</td>
<td>Administration of intravenous fluids to any patient with non-severe dengue</td>
</tr>
<tr>
<td>7. Use of isotonic intravenous fluids for severe dengue</td>
<td>Use of hypotonic intravenous fluids for severe dengue</td>
</tr>
<tr>
<td>8. Giving intravenous fluid volume just sufficient to maintain effective circulation during the period of plasma leakage for severe dengue</td>
<td>Excessive or prolonged intravenous fluid administration for severe dengue</td>
</tr>
<tr>
<td>9. Avoiding intramuscular injections in dengue patients</td>
<td>Giving intramuscular injections to dengue patients</td>
</tr>
<tr>
<td>10. Intravenous fluid rate and frequency of monitoring and haematocrit measurement adjusted according to the patient’s condition</td>
<td>Fixed intravenous fluid rate and unchanged frequency of monitoring and haematocrit measurement during entire hospitalization for severe dengue</td>
</tr>
<tr>
<td>11. Close monitoring of blood glucose, i.e. tight glycaemic control</td>
<td>Not monitoring blood glucose, unaware of the hyperglycaemic effect on osmotic diuresis and confounding hypovolaemia</td>
</tr>
<tr>
<td>12. Discontinuation or reducing fluid therapy once haemodynamic status stabilizes</td>
<td>Continuation and no review of intravenous fluid therapy once haemodynamic status stabilizes</td>
</tr>
</tbody>
</table>
Pathophysiology

Dengue Ward: QSNICH, Bangkok, Thailand (Photo: Christopher Brown, IHT)
Exposure and Infection Outcome Determinants

- **Host**
- **Ecology**
- **Virus**
- **Vector**

Space & Time
Exposure Determinants – Infection Risk

**Virus**
- Tropism for Aedes
- Tropism for man
- Replicative kinetics
  - Human / Aedes
- “Immune avoidance”
  - Human / Aedes
- Evolutionary capacity

**Vector**
- Response to ecology
  - Temperature
  - Rain
- Infection resistance
  - Co-infection
- Evolutionary capacity

**Host**
- Immune profile (dengue, other flavivirus)
- Vector exposure dynamics (duration, concentration)
  - “Neighbors” infection status
- Activities of daily living (who, what, where, when)
Infection Outcome Determinants – Disease Risk

- **Virus**
  - DENV type
  - DENV genotype
  - Replicative kinetics
  - Immune evasion
  - Target cell tropism
  - Evolutionary capacity
  - Sequence of infection

- **Vector**
  - Salivary proteins

- **Host**
  - Immune profile (first infection, humoral, cellular)
  - Demographic (age, race, genetic background [HLA])
    - Co-morbidities
    - Medical system sophistication treating dengue
Figure 4 | A balance between favourable and unfavourable factors determines the clinical outcome of dengue virus infection. Virus inoculation by *Aedes aegypti* mosquitoes results in viral dissemination, which in turn results in disease manifestations; high virus titres are necessary but not sufficient to cause severe disease. Viral and host factors affect early viral replication and influence the level of viraemia. Other host factors affect the levels of production of various cytokines (such as interferon-γ (IFNγ) and tumour necrosis factor (TNF)) and determine the severity of illness for any given level of viraemia. APL, altered peptide ligand.
Endothelial dysfunction

- Endothelial cells: infection and replication in selective endotheliocytes
- Liver: replication in hepatocytes and kupffer cells
- Tissue macrophages
- Bone marrow: replication in stromal cells

- Apoptosis
- Necrosis and/or apoptosis in liver and breach of its function
- Apoptosis
- Suppression of haemopoiesis

- Release of toxic products into the blood
- Increase of coagulation
- Activation of fibrinolytic system
- Consumption of platelets

- Soluble mediators: TNF-α, IFN-γ, IL-1, IL-2, IL-6, IL-8, IL-10, IL-13, IL-18, TGF-β, C3a, C4b, C5a, MCP-1, CCL2, VEGF, NO

- Imbalance profile of cytokine and other mediators
Dengue Vaccine Development
Dengue Vaccine Pipeline 2013

Pre-clinical | Phase 1 | Phase 2 | Phase 2b | Phase 3
---|---|---|---|---
SP- Chimerivax | | | | 
WRAIR/GSK - LAV | | | Development halted | 
NIH/JHU - Δ30 mut | | Development halted | | 
NMRC – Tetra DNA + Vaxf. | | Tetravalent product to endemic regions | | 
WRAIR/GSK – PIV + AS | US and Puerto Rico studies - PoConcept | | | 
Inviragen – PDK53 | Ongoing trial in PR, other endemic | | | 
Merck / HBI – r80E + ISCO | Tetravalent study ongoing | | | 
GenPhar – Cad-Vax | Exploring future | | | 
NMRC/Genvec – Adv5_DNA | | NHP studies completed | | 
Mahidol - LAV | | DENV-3 component derailed initial effort, reformulating | | 
FDA – mutF | | NHP study completed, no further development | | 
Carolina – alphavirus vector | | NHP study completed | | 
VaxInnate – flagellin E | | NHP study underway | | 
Altravax – flagellin E | | NHP study underway | | 
Arbovax - mutant | | NHP study underway | | 

Green = human testing; Red = pre-clinical
NMRC & others: DNA (preM+E) + adjuvant

Merck: 80% E recombinant
Expressed in Drosophila cells

NIH: Directed mutagenesis, deletions, point mutations
Stand alone and chimeras

Sanofi P: YF 17D backbone
Dengue prM and E
Monovalents formulated as tetra-

WRAIR/GSK: PIV + adjuvant system
Full genome, inactivated, formalin

Inviragen: DENV-2 PDK backbone,
Directed mutagenesis, DENV-2/-1, -2/-3, -2/-4
Development Challenges

• Each DENV type may cause severe disease/death
  – Viable vaccine requires efficacy against multiple types
  – Immune interference may prevent balanced response

• Incomplete understanding of protection / pathology
  – Will a poor dengue vaccine increase / worsen disease?
  – Extrapolating wild type infection data to immunization?

• No validated immune correlate of protection
  – No metrics or benchmarks for vaccine developers
  – Increases need for larger scale clinical trials
Development Challenges

• No validated animal model of disease
  – NHPs develop viremia and Nab but not disease

• No validated human infection model
  – Advancing vaccines based on Nab and NHP data
  – Efficacy trials may not capture efficacy vs. all DENVs

• Biologic assays used for endpoint determinations
  – Inter-assay variability notorious
  – Neutralizing antibody’s ability to predict efficacy?

• Numerous indications with unique challenges
  – Needs vary at the time, space, and population level
Conclusions

• The world needs a dengue vaccine!

• Global dengue burden is increasing
• Maintain a high index of suspicion in febrile traveler
• High financial and societal cost associated with disease
• Numerous factors continue to drive transmission
• Numerous vaccine development challenges exist
• Dengue vaccine pipeline robust
• Numerous areas for expanded study exist
Questions?