Case Presentation

• 18 year old native from Thailand (moved to US at 10 years of age) presents on day 2 of illness with fever, headache, eye pain, bone pain, and nausea 3 days after returning from a vacation in Puerto Rico. Dengue is suspected and he is treated symptomatically as an outpatient with NSAIDS and po fluid intake is encouraged. He returns 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 best next step and rationale for the same?
  A. Continue close follow up as outpatient, encourage po fluid intake, this is the natural history of a resolving dengue infection
  B. Admit to the hospital, approach as a critically ill patient, this is a severe dengue virus infection
  C. Admit to the hospital, evaluate for another infectious disease process, these new symptoms represent a new medical problem
  D. Prescribe doxycycline, he probably has leptospirosis
Lecture Objectives

1. Attendees will understand the **global distribution** of dengue virus circulation and disease.
2. Attendees will understand the spectrum of **dengue illness** 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.
Dengue Virus

- Flavivirus (single-stranded RNA virus)
- Spherical, 40-50 nm (dia.) viral particle
- 4 clinically important serotypes, DENV 1-4
  - 5 known serotypes (5th is rare-sylvatic)
  - Multiple genotypes per serotype
- Common progenitor 1,000 years ago
- Serotypes have further divergence
  - 62 to 67% homology based on amino acid sequence
- Varying pathogenicity based on serotype
Basics

• Dengue Viruses
  – 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
History of Dengue

• Clinical descriptions date as far back as 992 AD in China
• David Bylon (Batvia) in 1779
  – “knokkelkoorts” --- joint fever
• Benjamin Rush
  – Termed “breakbone fever”
  – Comes from Swahili “ka dinga pepo” meaning a sudden cramp like seizure and plague
Dengue Viruses (DENVs)

• Transmitted by *Aedes* mosquitoes
  – *Ae aegypti*, *Ae albopictus*
  – Day-biting
  – “Cosmopolitan:” close association with human habitation

• Infection with one serotype induces lifelong immunity to the same serotype

• No cross-protective immunity between 4 serotypes
  – Subsequent infection with a different serotype is the key risk for Dengue Hemorrhagic Fever (DHF)
Areas supporting dengue virus transmission.
Dengue Epidemiology

Dengue Ward: QSNICH, Bangkok, Thailand (Photo: Christopher Brown, IHT)
Dengue disease burden

- Global Impact
  - Endemic in > 100 countries \(^1,2\)
  - 2.5 billion at risk (40% of global pop.) \(^1,2\)
  - ~50-100 million infected annually \(^1,2\)
  - >2 million hospitalized with DHF annually \(^3\)
  - >22,000 deaths \(^1,2\)
  - Emerging disease
  - 30 fold increase in last 50 years \(^1,2\)
  - Epidemics increasing in size and frequency \(^1\)

1) CDC  2) WHO  3) Beatty et al, Nov. 2010
DENV Type Distribution - 1970
Global Air Travel Flight Patterns

http://upload.wikimedia.org/wikipedia/commons/a/ac/World-airline-routemap-2009.png
Factors Driving Transmission

- **DENVs**
  - Rapid travel
  - Viral evolution
- **Naïve hosts**
  - Population growth
  - Increased urbanization
- **Vector**
  - Ecologic changes
  - Evolution
Average dengue incidence per 100,000 by country, Region of the Americas, 1980–2007.
Dengue Burden
Under-estimated and under-reported

389.9M infections/ year

GLOBAL STRATEGY FOR DENGUE PREVENTION AND CONTROL

Figure 3. Average number of dengue cases in 30 most highly endemic countries/territories as reported to WHO, 2004–2010

© World Health Organization 2012
Reports from the last 3 months.
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**
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


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**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).
### Blood supply

**Blood supply?**

**Organ donation?**

---

#### Table 3. Dengue and donor deferral

<table>
<thead>
<tr>
<th>Country</th>
<th>Donor deferral measures for dengue</th>
</tr>
</thead>
<tbody>
<tr>
<td>Singapore*</td>
<td>6 months deferral for history of dengue infection</td>
</tr>
<tr>
<td></td>
<td>3 weeks deferral for history of fever</td>
</tr>
<tr>
<td></td>
<td>No travel-related deferral for dengue</td>
</tr>
<tr>
<td>Hong Kong*</td>
<td>6 months deferral for history of dengue infection</td>
</tr>
<tr>
<td></td>
<td>2 weeks deferral for history of fever</td>
</tr>
<tr>
<td></td>
<td>No travel-related deferral for dengue</td>
</tr>
<tr>
<td>Sri Lanka*</td>
<td>No specific deferral for history of dengue infection</td>
</tr>
<tr>
<td></td>
<td>2 weeks deferral for history of fever</td>
</tr>
<tr>
<td></td>
<td>No travel-related deferral for dengue</td>
</tr>
<tr>
<td>Australia†</td>
<td>4 weeks deferral for history of dengue infection</td>
</tr>
<tr>
<td></td>
<td>No travel-related deferral for dengue</td>
</tr>
<tr>
<td>New Zealand‡</td>
<td>4 weeks deferral for history of dengue infection</td>
</tr>
<tr>
<td></td>
<td>No travel-related deferral for dengue</td>
</tr>
<tr>
<td>UK‡</td>
<td>2 weeks deferral for history of dengue infection</td>
</tr>
<tr>
<td></td>
<td>No travel-related deferral for dengue</td>
</tr>
<tr>
<td>United States‡</td>
<td>4 weeks deferral for history of dengue infection</td>
</tr>
<tr>
<td></td>
<td>No travel-related deferral for dengue</td>
</tr>
</tbody>
</table>

*Endemic for dengue.
†Non-endemic except parts of Northern Australia.
‡Non-endemic.
Dengue in the USA

• Some historical dengue outbreaks in the USA
  – 1780: Philadelphia, PA
  – 1826-8: Savannah, GA
  – 1850-1: Charleston, SC, Savannah, GA, New Orleans, LA, Mobile, AL, Galveston, TX, Augusta, GA
  – 1922: Texas, Savannah, GA
  – 1934: Florida
  – 1945: New Orleans

http://www.topnews.in/number-dengue-cases-delhi-reaches-913-2238269
United States

Dengue Fever, Hawaii, 2001–2002

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

Paul V. Effer,* 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
- Oahu Cases
  - 1
  - 3
  - 9
  - 11
- Maui Cases
  - 1
  - 2
  - 5
  - 8
  - 52

Weekly interval beginning
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
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>

CDC Dengue Map, 15 OCT 2013

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

Suspected cases reported compared to the historical average

Total viral identifications in the 12 month period

Serotype Distribution

Totals through 31 DEC 2013: 18,164 suspected, 9,032 confirmed, 50 severe (DHF), 12 deaths

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. F. Siler.

Figure 4. Major Albert Sabin.

Figure 5. Airplane spraying of DDT over Manila, the Philippines, 1945.
WWII

• South Pacific:
  – 80,000 hospitalized
  – 1942: 1600 hospitalizations (Changi, Singapore); “Almost all troops” (Tulagi)
  – 1943: Espiritu Santos, New Hebrides (5,000 affected (25%); Hawaii (Waikiki beach, resulting in travel restrictions)
  – 1944: Saipan (7 epidemics, 3500/1000/yr), controlled vector by spraying (148th General Hospital, 176th Station Hospital)
  – 1945: Okinawa (275/1000/yr)
  – 1944-1945: New Guinea/Philippines (27,000 cases)
  – China-Burma-India: 8,000 hospitalized: 40/48 US troops developed DF within 5-10 days of arrival.

• Australia:
  – 1942: 80% of service members were affected.
  – 1943: 463 cases
Vietnam War

- 1964: Ubol, Thailand 58/294 confirmed DF
- 1966: 31/110 DF among FUO cases
- 1966-1967: 8th Field Hospital 10/94 (11%)
- 1967: Mekong River Delta 3%
- 1965-1966: average monthly incidence of DF (3.5/1000)
- 3-28% of FUO cases were dengue
Post Vietnam War

• Somalia
  – Operation Restore Hope (1993, 30,000 troops)
  – 129/289 (45%) did not have an immediately identified cause of illness; 59/96 (61%) had laboratory-confirmed DF
  – 69/289 (24%) suspected DF cases

• Haiti
  – Operation Uphold Democracy (1994, 20,000 troops) 30/103 (29%) hospitalized febrile troops had DF
  – Follow-up UN mission: 79/249 (32%) had DF (86th CSH)
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

Seroprevalence Based on 1,000 Post-Deployment Samples in First Time Deployers

<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?
# DOD Infectious Disease Threats

<table>
<thead>
<tr>
<th>Disease</th>
<th>2010 COCOM panel rank</th>
<th>ID-I DEAL 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>
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.
**Clinical disease: Dengue fever**

- **Acute febrile illness**
  - “Breakbone fever”
    - Myalgias, arthralgias
    - Headache, retro-orbital pain
    - Nausea, vomiting, abdominal pain
    - Rash
    - Occasional mild hemorrhagic manifestations

- **Self-limited course**
  - 5-7 days’ duration, correlates with viremia
  - “Post-viral asthenia,” potentially for weeks
  - No long-term sequelae
    - Except: “sensitization” for DHF in future

Classic dengue rash: “islands of white in a sea of red.”
Patient with acute dengue fever, Puerto Limon, Costa Rica, 2010
The risk of developing dengue hemorrhagic fever or dengue shock syndrome is increased among travellers who have a history of:

A. Being bitten frequently by mosquitoes of different species
B. Allergy to bee stings
C. Past dengue infection
D. No prior travel to dengue risk areas
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.
Dengue Immunology: double-edged sword

• Homologous (same serotype) immunity probably lifelong
• Heterologous (different serotype) immunity short-lived (3-6 months)
• Secondary infections occur in regions with hyperendemicity
  – Secondary infections increase risk for DHF
Dengue haemorrhagic fever
Diagnosis, treatment, prevention and control

SECOND EDITION

World Health Organization
Geneva
1997

Dengue Infection
Clinical Phenotypes

Dengue virus infection

Asymptomatic

Symptomatic

Undifferentiated fever (viral syndrome)

Dengue fever syndrome

Dengue haemorrhagic fever (plasma leakage)

Without haemorrhage

With unusual haemorrhage

No shock

Dengue shock syndrome

_ WHO 95829 _

Dengue clinical dx/classification: WHO recs 2009

• Probable dengue, +/- warning signs
  – Exposure + Fever + 2:
  – Nausea, vomiting
  – Rash
  – Myalgias / arthralgias
  – Tourniquet test +
  – Any warning sign

• Warning signs:
  • Abd pain / tenderness
  • Persistent vomiting
  • Clinical fluid accumulation
  • Mucosal bleeding
  • Lethargy / restlessness
  • Hepatomegaly > 2 cm
  • Lab: increased Hct concurrent with rapid decrease in platelets

• Criteria for severe dengue
  – Severe plasma leakage resulting in
    • Shock (DSS)
    • Fluid accumulation with respiratory distress
  – Severe bleeding
    • As evaluated by clinician
  – Severe organ dysfunction
    • AST or ALT > 1000
    • Impaired LOC
    • Heart and other organs

Lab-confirmed dengue

## Dengue Hemorrhagic Fever

### Table 1. WHO Classification of DHF

<table>
<thead>
<tr>
<th>Grades</th>
<th>Increase in Hematocrit*</th>
<th>Hemorrhage†</th>
<th>Shock‡</th>
<th>Profound shock§</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>+</td>
<td>−</td>
<td>−</td>
<td>−</td>
</tr>
<tr>
<td>2</td>
<td>+</td>
<td>+</td>
<td>−</td>
<td>−</td>
</tr>
<tr>
<td>3</td>
<td>+</td>
<td>±</td>
<td>+</td>
<td>−</td>
</tr>
<tr>
<td>4</td>
<td>+</td>
<td>±</td>
<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>

*Hematocrit increased by at least 20%.
†Spontaneous bleeding in skin and/or other sites.
‡Hypotension and/or narrowing of pulse pressure to 20 mmHg or less, with cold clammy skin and restlessness.
§Undetectable blood pressure or pulse.
2009 WHO dengue case definitions

**TABLE 2**

<table>
<thead>
<tr>
<th>2009 World Health Organization (WHO) dengue case definitions</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 \( \geq 1,000 \)
  - 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.*
Which of the following is NOT a “warning sign” of severe dengue?

A. Severe abdominal pain or persistent vomiting
B. Petechiae or ecchymoses
C. Respiratory distress
D. Vomiting blood
E. Fever

20%  20%  20%  20%  20%
Figure 1.4 Suggested dengue case classification and levels of severity

**DENGUE ± WARNING SIGNS**

- **with 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)
- **without warning signs**

**SEVERE DENGUE**

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

**CRITERIA FOR DENGUE ± WARNING SIGNS**

- 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
Pathophysiology

Dengue Ward: QSNICH, Bangkok, Thailand (Photo: Christopher Brown, IHT)
Dengue: Clinical Manifestations

asymptomatic

Undifferentiated Fever

symptomatic

Dengue Fever

No hemorrhage

+ hemorrhage

+ Capillary leak

DHF

Gr I-II

+ shock
Dengue infection outcomes

• Majority of cases > 90% are asymptomatic, undifferentiated fever or non-hospitalized DF
  - Inapparent: apparent infection ratio
    • Depends on age, other host factors, serotype
    • Primary determinant is prior serotype predominance

• < 5% are DHF
  - Secondary infection greater risk
  - Putative mechanisms:
    • Immunopathogenesis
      - ADE
      - Heterologous T cell effects
    • Viral determinants
    • Multifactorial
Clinical disease: Dengue fever

• Acute febrile illness
  – “Breakbone fever”
    • Myalgias, arthralgias
    • Headache, retro-orbital pain
    • Nausea, vomiting, abdominal pain
    • Rash
    • Occasional mild hemorrhagic manifestations

• Self-limited course
  – 5-7 days’ duration, correlates with viremia
  – “Post-viral asthenia,” potentially for weeks
  – No long-term sequelae
    • Except: “sensitization” for DHF in future

Pathogenesis of DHF

• Acute increase in vascular permeability
  – Reversible (unless death supervenes)

• Hemostatic changes/hemorrhagic diathesis
  – Vascular changes
  – Thrombocytopenia
  – Coagulopathy
    • Most patients with DSS have DIC
    • Prolonged PTT, decreased fibrinogen, increased fibrinogen split products
    • 1/3 of patients with DSS have severe bleeding
    • Majority of fatal cases have GI hemorrhage
Putative Immunopathogenic Mechanisms

Antibody Dependent Enhancement (ADE)

- Epidemiological Observations
  - Secondary infection with a heterotypic dengue virus is strongly associated with an increased risk of DHF

- Hypothesis
  - Pre-existing antibodies to one dengue virus serotype complex with the virus and increase efficiency of FcR-mediated virus uptake, resulting in increased infection
  - FcR-bearing cells (monocytic lineage) can produce cytokines mediating pathogenesis
Putative Immunopathogenic Mechanisms

T-Cell Mediated Pathogenesis

- Clinical Observations
  - Marked T-cell activation and levels of several T cell cytokines correlate with disease severity. Rothman, 1999; Green 2002

- Hypothesis
  - Original antigenic sin: cross-reacting memory T cells from primary infection proliferate in second infection
  - Lower affinity T cells contribute to immunopathology but with comparatively lower anti-viral effect
Course of illness: fever, viremia and anti-dengue IgM relative to day of defervescence

Adapted from Figure 1 in Vaughn et al., J Infect Dis, 1997; 176:322-30.
DF (Febrile Phase)

- Lasts 2-7 days
- Abrupt onset high fever (≥38.5°C)
  - 5-7 days fever (biphasic)
- Rash
  - Early flushlike rash may be replaced by a macular/morbilliform rash. Late petechial
- Arthralgias, myalgias
- Severe headache
- Eye, Retro-orbital pain
- Lumbosacral pain
DF (Febrile Phase II)

- Anorexia, nausea, vomiting
- Sore throat, injected pharynx, conjunctiva
- Respiratory symptoms
- Epistaxis, gum bleeding, petechiae
  - Classic DF with some hemorrhage is NOT DHF
  - Massive (GU, GI) rare.
- PE:
  - Fever
  - Generalized rash (may be replaced by macular/morbilliform later on).
    Petechial late
  - Relative bradycardia
  - Generalized lymphadenopathy
  - Hepatomegaly
  - Positive tourniquet test
- Lab
  - Progressive decrease in WBC
Critical Phase I

• Occurs at time of defervescence
  – Around days 3-7 of illness
  – Temperature drops to 37.5-38.0°C or below
  – Lasts 24-48 hours
• Systemic vascular leak syndrome
  – Increasing hematocrit
  – Hypoproteininemia
  – Pleural effusions
  – Ascites
Critical Phase II

• Progressive leukopenia
• Rapid thrombocytopenia
• Degree of plasma leakage varies
  – Increase in Hct a good barometer
• Shock
  – If is to occur, preceded by warning signs
• Follow fluid management closely
  – CXR, US
Recovery Phase

• After critical phase, 48-72 hours of reabsorption of extravascular fluid
• Well-being, appetite improves
• Bradycardia common
• Hemodynamic status improves
• GI symptoms abate
• Blood counts normalize (RBC>WBC>Plt)
• Diuresis occurs
• Prolonged convalescence
Severe Dengue I

• Severe plasma leakage
  – Shock (DSS)
  – Serosal fluid accumulation with respiratory distress
• Severe bleeding
  – Clinically evident
• Multi-organ involvement
  – Liver: AST/ALT >1000
  – CNS: Impaired consciousness, seizures, encephalopathy
  – CV and other
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.

\[ PEI = \frac{A}{B} \times 100 \]
Severe Dengue II

- Clinical signs
  - Tachycardia, peripheral vasoconstriction
  - DBP rises towards SBP (narrowed pulse pressure, ≤ 20 mmHg)
  - Patient may be conscious and lucid
  - Poor perfusion
    - Cold, clammy extremities
    - Rapid, weak pulse
    - Mental status changes
  - Serosal fluid accumulation with respiratory distress
Severe Dengue III

• Severe bleeding
  – Clinically evident

• Multi-organ involvement
  – Liver: AST/ALT >1000
  – CNS: Impaired consciousness, seizures, encephalopathy
  – CV (cardiomyopathy) and other
  – Coagulopathy
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.
Diagnosis and Therapy
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 area, +Clinical, +TT, WBC<5k = High PPV (~70%)
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
Rash
Hemoconcentration
Laboratory Diagnosis

• Virus detection
  – RNA by RT-PCR
  – Virus isolation
  – NS1 antigen detection by ELISA, dipstick

• Antibody detection
  – IgM by ELISA, dipstick
  – Confirmation by convalescent IgG ELISA, virus-neutralizing antibody

• Optimal diagnostic strategy:
  – Combination of NS1 and IgM detection
    • Markedly increased sensitivity and predictive value

• Supporting findings from routine labs:
  – Thrombocytopenia, leukopenia with atypical lymphocytes
### Tests Used for the Lab Diagnosis of Primary Dengue Infection (2)

<table>
<thead>
<tr>
<th>Test</th>
<th>Diagnostic Window</th>
<th>Sample Required</th>
<th>Sample Storage</th>
<th>Turnaround Time</th>
<th>Facilities/Cost</th>
</tr>
</thead>
<tbody>
<tr>
<td>Antigen Detection</td>
<td>1-6 days</td>
<td>Serum, tissues</td>
<td>Refrigerate or frozen</td>
<td>1 day or more</td>
<td>ELISA facilities ($$), Histology facilities ($$$$)</td>
</tr>
<tr>
<td>IgM ELISA</td>
<td>Day 5 to –Day 90 post infection</td>
<td>Serum, plasma, blood</td>
<td>Frozen or refrigerated</td>
<td>1-2 days</td>
<td>ELISA facilities, $</td>
</tr>
<tr>
<td>IgM Rapid Test</td>
<td>Day 5 to –Day 90 post infection</td>
<td>Serum, plasma, blood</td>
<td>Frozen or refrigerated</td>
<td>30 mins</td>
<td>No additional supplies, $</td>
</tr>
<tr>
<td>IgG (paired sera) by ELISA, HI or neutralization</td>
<td>Acute sera: 1-5 days; Convalescent: after 15 days</td>
<td>Serum, plasma, blood</td>
<td>Frozen or refrigerated</td>
<td>7 days or more</td>
<td>ELISA facilities, BSL-2 for neutralization, $</td>
</tr>
</tbody>
</table>
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.
Product Introduction

**#1: IgG/IgM Dengue Duo Cassette**

10μL of serum, plasma, or whole blood

15 minute (time to result)

Wu et. al. CDLI 2000, pp 106-109

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Comparison of Two Rapid Diagnostic Assays for Detection of Immunoglobulin M Antibodies to Dengue Virus

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Viral and Rickettsial Diseases Department, Naval Medical Research Center, Bethesda, Maryland 20889-5000

Integrated Diagnostics Inc., Baltimore, Maryland 21217; Diagnostic Systems Division, U. S. Army Medical Research Institute of Infectious Diseases, Fort Detrick, Frederick, Maryland 21702-5011; and Departments of Virus Diseases, Walter Reed Army Institute of Research, Washington, D.C. 20317-5000

Received 25 June 1999/Returned for modification 4 August 1999/Accepted 10 October 1999

Two easy-to-use commercial diagnostic assays, a dipstick enzyme-linked immunosorbent assay (ELISA) (PanBio, Brisbane, Australia) and an immunochromatographic card assay (PanBio, Brisbane, Australia), were evaluated for detection of immunoglobulin M (IgM) antibody to dengue virus with an in-house IgM antibody capture microplate ELISA as a reference assay. The dipstick ELISA was based on the microplate ELISA format using dengue 2 virus as the only antigen and enzyme-labeled goat anti-human IgM antibody as the detector. The total assay time was 75 min. The immunochromatographic card assay was based on an antibody capture format and separately measured both anti-dengue virus IgM and IgG in the same sample. Using gold-labeled anti-dengue virus monoclonal antibody bound with dengue virus 1 to 4 antigen control as the detector, and anti-human IgM and IgG were the capture antibodies. The total assay time was <1 h. Sera from 164 individuals classified as either anti-dengue virus IgM positive (94) or anti-dengue virus negative (70) in the reference microplate ELISA with a dengue virus 1 to 4 antigen cocktail were tested in two commercial assays. The dipstick ELISA missed 7 of 84 positive samples, for a sensitivity of 92.6%, the immunochromatographic card assay missed two positive samples, for a sensitivity of 97.5%. Of 70 negative samples, four were false positive by the dipstick ELISA and two were false positive in the immunochromatographic card assay, resulting in specificities of 94.3 and 97.1%, respectively. Both commercial assays provide sensitive and specific detection of anti-dengue virus IgM antibody and could prove useful in situations where the microplate ELISA is impractical.
#2: NS-1/IgG/IgM Dengue Duo Cassette

120μL of serum or plasma
15 minute (time to result)


Comparison of the diagnostic accuracy of commercial NS1-based diagnostic tests for early dengue infection

Lyda Osorio¹, Meleny Ramirez¹, Aniliza Bonelo², Luis A Villar³, Beatriz Parra²

Abstract

Background: We compared the diagnostic accuracy and reproducibility of commercially available NS1-based dengue tests and explored factors influencing their sensitivities.

Methods: Paired analysis of 310 samples previously characterized as positive (n = 218) and negative (n = 92) for viral isolation and/or RT-PCR and/or IgM seroconversion. Masked samples were tested by two observers with Platelia™ Dengue NS1 Ag, second generation Pan-E™ Dengue Early ELISA, SD Dengue NS1 Ag ELISA, Dengue NS1 Ag STRIP™, and SD BIOLINE™ Dengue Duo (NS1/IgM/IgG).

Results: SD BIOLINE™ NS1/IgM/IgG had the highest sensitivity (80.7% 95%CI 75-85.7) with likelihood ratios of 7.4 (95%CI 4.1-13.8) and 0.21 (95%CI 0.16-0.28). The ELISA-format tests showed comparable sensitivities; all below 75%. STRIP™ and SD NS1 had even lower sensitivities (<65%). The sensitivities significantly decreased in samples taken after 3 days of fever onset, in secondary infections, viral serotypes 2 and 4, and severe dengue. Adding IgM or IgG to SD NS1 increased its sensitivity in all these situations.
Standard Diagnostics Dengue Duo (NS-1) RDT

**NS1 Ag**

3 drops (110 μl) of plasma or serum for early acute phase samples (day 1 ~5)

**IgG/IgM Ab**

10 μl of plasma or serum for early convalescence phase samples (after day 5 ~ 14)

Slide courtesy of Dr. Subhamoy Pal
Interpretation

- **Negative**
- **Primary**
- **Secondary**
Indonesian elephants
Taman Safari, Bogor, West Java, Indonesia
Management: Diagnosis and Therapy

• Most often diagnosed on clinical grounds
  – Rapid and point-of-care tests in development
• No specific antiviral therapy
  – Conservative management, observation
    • Seek and treat alternative etiologies in DDx
  – For DF
    • Antipyretics, analgesics
    • AVOID aspirin, NSAIDS due to bleeding risk
    • Observe for signs of DHF
  – For DHF
    • Fluid management
    • Supportive care
Therapy of DF and DHF

• Treatment of DF
  – Observation, evaluation for DFH
  – R/O treatable etiologies
  – Avoid agents with bleeding risk
    • ASA, NSAIDs

• Treatment of Suspected DHF
  – When to hospitalize
    • Before shock develops
    • DSS frequently fatal, but rapidly reversible with fluid resuscitation
    • Prognosis dependent upon early recognition of shock
  – Warning signs
    • Decreasing platelet count, increasing Hct
    • Clinical signs of hypoperfusion
      – Lethargy/restlessness, cold extremities, severe abdominal pain, refractory vomiting, oliguria

• NB Differential Dx:
  • Influenza
  • Measles
  • Rubella
  • Malaria
  • Typhoid fever
  • Leptospirosis
  • Meningococcemia
  • Rickettsial infections
  • Bacterial sepsis
  • Other VHFs
<table>
<thead>
<tr>
<th>Intervention</th>
<th>Conclusion</th>
</tr>
</thead>
<tbody>
<tr>
<td>Choice of intravenous fluid</td>
<td>RCTs show no benefit of colloids over crystalloids; trend towards benefit with colloids over crystalloids in severe cases</td>
</tr>
<tr>
<td>Rate of fluid infusion</td>
<td>No evidence available, regimens based on experience of centers treating large numbers of cases</td>
</tr>
<tr>
<td>Transfusion of blood products</td>
<td>Small RCT suggests fresh frozen plasma may increase platelet counts</td>
</tr>
<tr>
<td>Nasal CPAP</td>
<td>RCT shows improvement of hypoxemia and reduction of need for ventilation in dengue with acute respiratory failure</td>
</tr>
<tr>
<td>Carbazochrome sodium sulfonate</td>
<td>RCT shows no evidence of benefit, but underpowered</td>
</tr>
<tr>
<td>Corticosteroids</td>
<td>Systematic review of RCTs shows no benefit; RCT evidence &gt; 20 years old; case series provide limited evidence of possible benefit in severe dengue</td>
</tr>
<tr>
<td>Immunoglobulins</td>
<td>Single RCT shows no benefit for thrombocytopenia; no evidence available for other manifestations</td>
</tr>
</tbody>
</table>
Therapy: fluid resuscitation for DHF

- World Health Organization (WHO) protocol
  - Graded, dynamic algorithm
  - Mild cases treated with oral rehydration, observation
  - Moderate – Severe cases with IVF, intensive monitoring of vital signs and Hct

- Wills, *et al*, found Ringer's lactate (LR) equivalent to colloids in children with moderate shock
  - Dextran and hetastarch equivalent in children with severe shock, but dextran caused more hypersensitivity reactions
  - LR untested in severe shock
  - *N Engl J Med.* 2005 Sep 1;353(9):877-89
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

**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.

**Monitoring**
Monitor:
- temperature pattern
- volume of fluid intake and losses
- urine output (volume and frequency)
- warning signs
- HCT, white blood cell and platelet counts.
Figure 2.2 Algorithm for fluid management in compensated shock

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

- **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 ↑ 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 ↓
  - Consider significant occult/overt bleed
    - Initiate transfusion with fresh whole blood
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 ↑ or high**

Administer 2nd bolus fluid (colloid)
10-20 ml/kg over ½ to 1 hour

**Improvement**

**YES**

Consider significant occult/overt bleed
Initiate transfusion with fresh whole blood

**NO**
Consider occult / overt bleed

- Improvement
  - YES
  - NO
  - Repeat 2nd HCT

- HCT↑ or high
  - Administer 3rd bolus fluid (colloid)
    - 10–20 ml/kg over 1 hour
  - Improvement
    - YES
    - NO
    - Repeat 3rd HCT
**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>
Dengue Vaccine Development
Dengue Vaccine Challenges

• Each DENV serotype capable of causing DF/DHF
  – Presumed requirement for tetravalent immunity

• Incomplete understanding of pathophysiology
  – Immunopathogenic mechanisms underlying DHF pose theoretical risk

• No validated experimental model of disease
  – NHP develop viremia and immune response, but not disease

• No validated immune correlate of protection
  – Unclear exactly which readout
  – Specific issues with respect to measurement, technique
Dengue Vaccine Pipeline 2013

- **Pre-clinical**
  - SP-Chimerivax
  - WRAIR/GSK - LAV
  - NIH/JHU - Δ30 mut
  - NMRC – Tetra DNA + Vaxf.
  - WRAIR/GSK – PIV + AS
  - Inviragen – PDK53
  - Merck / HBI – r80E + ISCO
  - GenPhar – Cad-Vax
  - NMRC/Genvec – Adv5_DNA
  - Carolina – alphavirus vector
  - VaxInnate – flagellin E
  - Altravax – flagellin E
  - Arbovax - mutant

- **Phase 1**
  - Development halted
  - Tetravalent product to endemic regions
  - Development halted
  - US and Puerto Rico studies - PoConcept
  - Ongoing trial in PR, other endemic
  - Tetravalent study ongoing

- **Phase 2**
  - Exploring future
  - NHP studies completed
  - DENV-3 component derailed initial effort, reformulating
  - NHP study completed, no further development
  - NHP study completed
  - NHP study underway
  - NHP study underway

- **Phase 2b**
  - Development halted

- **Phase 3**
  - 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

Various: Domain III antigen
Sanofi Pasteur’s Dengue Vaccine Candidate Successfully Completes Final Landmark Phase III Clinical Efficacy Study in Latin America

• Second, large-scale phase III study successfully meets primary endpoint with overall vaccine efficacy of 60.8 percent and shows efficacy against each of the four dengue serotypes
• Additional observation of the results shows a significant reduction of the risk of hospitalization by 80.3 percent confirming the potential public health impact of the vaccine
• Initial safety data are consistent with the favorable safety profile documented in all previous studies (phase I, II, III)
Protective efficacy of the recombinant, live-attenuated, CYD tetravalent dengue vaccine in Thai schoolchildren: a randomised, controlled phase 2b trial

- Observer-blind, randomized, controlled, monocenter
- Ratchaburi, Thailand (hyper-endemic for dengue)
- Healthy Thai children, 4-11 years, N = 4,002
- Randomized 2:1 (vaccine N=2669 : control N=1333)
- Injections were administered at months 0, 6, and 12
- Active case finding until month 25
- Dengue cases = Fever + viremia or antigenemia
  - RT-PCR or NS1 ELISA
US Army Goals?

• Immunogenicity
  – Vaccine effect noted compared to control
  – Seroconversion rates & quantitative antibody response
    • Nearly 100% SC for each DENV type
    • 3.4-12.7 fold rise in GMT following dose 3
    • GMT >100 across all DENV types after dose 3
    • GMT >300 for DENV-2 (lowest efficacy) after dose 3

• Efficacy
  – DENV type specific and variable (D4>D3>D1>D2)
  – Failed to meet primary efficacy endpoint (30.2%)
"Preventing or reducing dengue virus transmission depends entirely on control of the mosquito vectors or interruption of human–vector contact."

- *Ae. aegypti* proliferates in household containers
  - domestic water storage and for decorative plants,
  - rain-filled habitats – used tires, discarded containers, blocked gutters, *etc*

- Typically do not fly > 100 m from site of emergence

- Feeding behavior
  - Anthropophagic
  - mainly during daylight
  - both indoors and outdoors

---

### Integrated vector management (IVM)

- **5 key elements**
  - advocacy, social mobilization and legislation
  - collaboration within the health sector and with other sectors
  - integrated approach to disease control
  - evidence-based decision-making
  - capacity-building

"Control of *Ae. Aegypti* mainly achieved by eliminating container habitats that are favorable oviposition sites"
Case Presentation

• 18 year old native from Thailand (moved to US at 10 years of age) presents on day 2 of illness with fever, headache, eye pain, bone pain, and nausea 3 days after returning from a vacation in Puerto Rico. Dengue is suspected and he is treated symptomatically as an outpatient with NSAIDS and po fluid intake is encouraged. He returns 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 best next step and rationale for the same?
  A. Continue close follow up as outpatient, encourage po fluid intake, this is the natural history of a resolving dengue infection
  B. Admit to the hospital, approach as a critically ill patient, this is a severe dengue virus infection
  C. Admit to the hospital, evaluate for another infectious disease process, these new symptoms represent a new medical problem
  D. Prescribe doxycycline, he probably has leptospirosis
Conclusions

• 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?