Dengue Virus Infection

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Global epidemiology

- 50 – 100 million infections each year
- 500,000 hospitalizations
- Important cause of fever in tropical countries
Vector: *Aedes aegyptae*
Results of immunoglobulin G (IgG) ELISA for antiflavivirus antibodies among patients exhibiting fever, Gonaïves, Haiti, October 2004 (n = 105)

[Diagram showing age distribution and IgG positivity/negativity]
DF and DHF cases reported by the Guanan health district, Zacapa, Guatemala 1999-2000

Number of cases

Epidemiological week

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SIGSA. Ministerio de Salud Publica, Guatemala
Dengue Virus: RNA flavivirus

E dimer

M protein

Capsid protein

Genomic RNA

T=3-like organization of surface dimers

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Swiss Institute of Bioinformatics

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Antibody dependent enhancement

- DENV
- FcγR
- Monocyte
- Increased viral load
- Disease

Heterotypic Ab from previous infection

Nature Reviews Microbiology
Mean Dengue virus titers (serotype 2) per day of fever according to severity. Bangkok 1994-96

Dengue
DHF grado I-II

Viremia titer (logMID50/mL)

Day of Fever

Defervescence

0

-5

-4

-3

-2

-1

0

Immune Response to Dengue

Primary Infection
Onset of symptoms

Secondary Infection
Onset of symptoms

Antibody & Antigen level

Virus
NS1
IgM

IgG
Kinetics of antibodies against Dengue after primary or secondary infection

IgG Titer

- IgM- and PCR-
- PCR +
- IgM+

Days of Illness

Laue T. J Clin Microbiol 1999
Host risk factors

- Age: infants > children > adults
Age distribution of Dengue hospitalizations of 126 children in Central Havana, Cuba 1981

Number of cases

Age in years

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Host risk factors

- Age: infants > children > adults
- Gender: females
- Ethnicity: African descent protective?
- Chronic underlying conditions
Dengue Clinical Manifestations

- 80% of infections asymptomatic
- Less common but important
  - Hepatitis (11% AST >10X)
  - Encephalitis
  - Pancreatitis 40%
  - Pleural Effusion 80%

Additional symptoms:
- Fever
- Rash
- Muscle and joint pains
Dengue: Skin rashes
Severity of Pleural Effusion in Dengue infected children according to the type and serotype of infection

Pleural Effusion index* = Profundidad del DP/Diametro Hemitorax Der x100

DEN Primary
DEN Secondary
DEN 1 y 3 Primary
DEN 1 y 3 Secondary


*PE deepness/Right Thoracic diameter x100

DEN-1
DEN-2
DEN-3
DEN-4

*PE deepness/Right Thoracic diameter x100
Dengue: a clinical approach

Without Signs of Alarm

With Signs of Alarm

1. Severe leakage of plasma
2. Severe hemorrhage
3. Organs severely affected

Presumptive Diagnosis
- Fever
- Anorexia and nausea
- Rash
- Body pains
- ≥ Signs of alarm
- Leucopenia
- Tourniquet test +

Signs of alarm
- Abdominal pain (or painful palpation)
- Persistent vomiting
- Clinical accumulation of fluids
- Bleeding in mucosae
- Lethargy; restlessness

History of dengue in the neighborhood or travel to a dengue endemic area

1. Severe leakage of plasma leading to:
   - Shock (DSS - Dengue Shock Syndrome)
   - Accumulation of fluids with breathing difficulty
2. Severe bleeding according to a clinical criterion
3. Organs severely affected
   - Liver: GOT or GGT > 1000
   - CNS: affection of conscience
   - Heart or other organs

* Requiring strict observation and medical intervention

Source: Formulated by Eric M. Torres.

Figure 4 – Classification of dengue.

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## Dengue definitions WHO and CDC

<table>
<thead>
<tr>
<th>Type</th>
<th>Clinical Case Definition</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Dengue Fever</strong></td>
<td><strong>Probable DF:</strong> Fever for 2 to 7 days and one of the following: cefalea, arthralgia, retrorbital pain, rash, leucopenia or consistent serology</td>
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<td></td>
<td><strong>Confirmed DF:</strong> Virus (+), PCR (+), confirmatory serology, dengue Ag</td>
</tr>
<tr>
<td><strong>Dengue Hemorragic Fever (DHF)</strong></td>
<td>All the following criteria:</td>
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<tr>
<td></td>
<td>1. Fever for 2-7 days (sometimes biphasic)</td>
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<td>2. Hemorrhagic manifestations</td>
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<td></td>
<td>3. Thrombocytopenia (&lt;100,000)</td>
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<td></td>
<td>4. Evidence of plasma effusions</td>
</tr>
<tr>
<td><strong>Dengue Shock</strong></td>
<td>DHF + circulatory collapse:</td>
</tr>
<tr>
<td></td>
<td>1. Fast and weak pulse or hypotension</td>
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<tr>
<td></td>
<td>2. Altered conscious status, renal failure</td>
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</table>
**Tourniquet Test**

Rumpel-Leede Capillary-Fragility Test or simply a capillary fragility test

- Blood pressure cuff inflated at MAP for 5 min
- **Positive = 10 petechiae per square inch**
- Tourniquet test positive in only 39.1% of all DHF cases in **India**
  

- **Vietnam:** Sen 41.6% Spe 94.4%, PPV 98.3% NPV 17.3%.
  
  **Trop Med Int Health.** 2002 Feb;7(2):125-32
Virus isolation from plasma, blood or tissues

4X increase in antibody titers IgM or IgG

Dengue antigen in serum

PCR from blood, tissue or pathology
Diagnostic Serology

- **Serology**
  - **Hemmaglutination**
    - Sensitive, easy, specific
    - Ideal for sero-epidemiology
  - **MAC-ELISA**
    - Day 5: 80% cases + (Puerto Rico)
    - Day 6-10: 93% cases +
    - Day 10-20: 99% +
    - Persistence of IgM for 2-3 months reported
  - **IgG ELISA**
    - Non specific, cross reactions
Approach to the Child with Dengue Fever

• Rest
• Acetaminophen or paracetamol
• No aspirin or AINEs
• No antibiotics
• Oral Rehydration solution (OMS)
  • 50 mL/kg for 4-6 hrs
  • Maintenance 80-100 mL/kg/day
• Alert signs
Transaminases TGP > 60 mg/mL

Leucopenia < 5000 cel/mm3

Low neutrophil counts < 3000 cel/mm3
Dengue Shock Algorithm

Ringer lactate / 5% GNS (10-20 ml/kg/hr) as fast as possible

Shock persists

Colloidal plasma solution / plasma expander (10-20 ml/kg/hr)

No improvement – Suggestive of significant internal bleeding

Fresh whole blood

Improvement

Reduce IV fluids

*Central venous pressure should be monitored in case of severe shock

6 ml/kg/hr fluids

Hematocrit / vitals / urine output (UO)

Improvement

(Hematocrit, Pulse, BP, UO stabilizes)

5 ml/kg/hr

No improvement

(Pulse pressure < 20 m Hg, reduced urine output)

10 ml/kg/hr

3 ml/kg/hr

*Continue for 24-48 hrs.
*Stop IVF when Vitals/hematocrit Stabilizes

15 ml/kg/hr

(Unstable vital parameters)

High Hematocrit

Low hematocrit

Colloids

Blood transfusion

Improve
Mortality rate in children given colloid vs. Cristaloid solutions during DHF

<table>
<thead>
<tr>
<th>Study or subgroup</th>
<th>Events/total</th>
<th>Adequate sequence generation</th>
<th>Peto fixed odds ratio (95% CI)</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>Colloids</td>
<td>Crystallloids</td>
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<tr>
<td>Sepsis</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Upadhyaya 2005</td>
<td>9/29</td>
<td>9/31</td>
<td>1.1 (0.4 to 3.3)</td>
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<tr>
<td>Malaria</td>
<td></td>
<td></td>
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<tr>
<td>Maitland 2005</td>
<td>2/56</td>
<td>11/61</td>
<td>0.23 (0.07 to 0.74)</td>
</tr>
<tr>
<td>Maitland 2005</td>
<td>4/23</td>
<td>3/20</td>
<td>1.2 (0.24 to 5.9)</td>
</tr>
<tr>
<td>Maitland 2003</td>
<td>0/20</td>
<td>4/32</td>
<td>0.18 (0.02 to 1.4)</td>
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<tr>
<td>Dengue</td>
<td></td>
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<tr>
<td>Wills 2005</td>
<td>1/384</td>
<td>0/128</td>
<td>3.8 (0.04 to 350)</td>
</tr>
<tr>
<td>Cifra 2003</td>
<td>1/11</td>
<td>3/16</td>
<td>0.48 (0.06 to 4.0)</td>
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</tbody>
</table>
Dengue hospital discharge criteria for children

- No fever for 24 hrs. without antipyresis
- Return of appetite
- Visible clinical improvement
- Good urinary output
- Minimum 3 days after shock
- No respiratory distress from effusions
- Platelet count > 50,000
Dengue preventive strategies

- Protective clothing during all day during vector season
- Impregnated bed nets
- Mosquito repellents
- Drainage of stagnant water reservoirs
- Insecticide spraying in the communities
Vaccines for Dengue: the ultimate prevention?

- Should be safe and immunogenic
- Protection before age of exposure (<3 years)
- High protection against 4 serotypes
- Long term immunity
- Current schedule: 0-6-12 months
Chimeric technology from flavivirus vaccines

Yellow fever 17D or Dengue genome

5'  C  prM  E  Non-structural genes  3'

Exchange for envelope genes from wild type dengue 1,2,3,4

prM  E

5'  C  Non-structural genes  3'

Chimeric cDNA -> transcribes RNA

5'  3'

mRNA transfer

Envelope = heterologous virus
Replicon = YF 17D or DENV

Growth of virus in cell culture

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Vaccines for Dengue: where are we now

- Phase III: sanofi-Pasteur vaccine
  - Balanced immune response achieved
  - Safe – minimal viremia after dose 1
  - Thailand phase III follow up 5 years
- Phase II: Inviragen and NIH
  - Age-dose range studies
Summary

• Dengue is a common and potentially severe infection with no treatment available (only supportive)
• More common after heavy rainfall and floods where vector is present
• Early detection and management is key
• Prevention is difficult but achievable