Triple Threat in the Tropics

Beata Casanas DO, FACP FIDSA
Associate Professor of Internal Medicine
Division of Infectious Diseases
University of South Florida, Morsani College of Medicine
Arboviruses

- Any virus species transmitted by mosquitoes, ticks, or other arthropods
- E.g.
  - *Culex* spp. - West Nile Virus, St. Louis & Eastern Equine Encephalitis
  - *Aedes* spp. – Dengue Virus, Chikungunya Virus, Zika Virus, Yellow Fever Virus
Many to Potentially Talk About

- But will we focus on three recent global infections with focus on Caribbean
  - Chikungunya
  - Dengue
  - Zika
Chikungunya

- Mosquito-borne viral infection
- Epidemics resembling Chikungunya recorded as early as 1824 in India and elsewhere
- 1st isolated virus in 1952-53 in Tanzania
- Prior outbreaks in countries in Africa and Asia
- In 2013, 1st locally-acquired cases in the Americas reported in the Caribbean
- In 2014, cases in Florida noted acquired locally
Chikungunya virus disease cases reported by state – United States, 2014

www.cdc.gov/chickungunya/geo/united-states-2014.html
Chikungunya virus disease cases reported by state – United States, 2015

www.cdc.gov/chikungunya/geo/united-states-2015.html
Chikungunya virus disease cases reported by state – United States, 2016 (as of September 20, 2016)

www.cdc.gov/chikungunya/geo/united-states-2016.htm
Countries and territories where chikungunya cases have been reported* (as of April 22, 2016)

www.cdc.gov/chikungunya/geo/index.html
Reported Chikungunya Cases in Florida, 2014 - 2018

Acquired in FL
Acquired in USA (not in FL)
Acquired outside USA
Unknown (FL or another state/county)
Mode of Transmission

- Mosquito vectors
  - Predominately *Aedes aegypti* and *Aedes albopictus* which also transmit dengue
  - Aggressive daytime biters and widely distributed
- Other modes of transmission very rare
  - *In utero* resulting in miscarriage
  - Intra-partum to child
  - Theoretical concern in blood transfusion or transplantation
Clinical Manifestations

- Incubation period ~ 3-7 days
- Majority are clinically symptomatic
- Abrupt fever and polyarthralgias predominate
  - Joint pain symmetric most commonly in hands and feet and can be severe and debilitating
- Other signs and symptoms include headache, myalgia, conjunctivitis, nausea/vomiting, and maculopapular rash
Clinical Outcomes

- Acute symptoms typically resolve in 7-10 days
- Mortality quite rare except in elderly
- Some can have relapse of rheumatologic symptoms in months following acute illness
  - Some with persistent joint pains for months to years
Diagnostic Testing

- Initial labs may reveal thrombocytopenia, lymphopenia, elevated creatinine and LFTs
- Specific diagnostic tests
  - Viral culture (< 3 days of symptoms in BSL 3 conditions)
  - RT-PCR (< 8 days of symptoms)
  - Serology for IgM
- Specific diagnostic testing at CDC and many state health department labs and few commercial labs
- Should rule out dengue
Treatment and Prevention

- Supportive care – fluids and rest
- No specific antiviral therapy
- Acetaminophen or NSAIDS for fever and acute pain
- No vaccine available
- Primary prevention is to reduce mosquito exposure
Dengue

- > 50 million infections per year
- ~ 1/3 of human population in dengue endemic areas
- Resurgence due poor urban planning
  - Poor sanitation and potable water supply
  - Rain water collection with increased breeding sites
  - Concentration of population in proximity of mosquitoes
  - Eroded mosquito control programs
American Countries with laboratory confirmed dengue hemorrhagic fever, prior to 1981 and from 1981 to 2003

Prior to 1981

1981 - 2003

Source: WHO/PAHO/CDC, Aug. 2004
Distribution of dengue, worldwide, 2016

The boundaries and names shown and the designations used on this map do not imply the expression of any opinion whatsoever on the part of the World Health Organization concerning the legal status of any country, territory, city or area or of its authorities, or concerning the delimitation of its frontiers or boundaries. Dotted lines on maps represent approximate border lines for which there may not yet be full agreement. © WHO 2016. All rights reserved
Dengue

- 4 known serotypes
- Fever, retro-orbital HA, rash, N/V, aches and pains
- Positive tourniquet test
- Repeated infection with another serotype poses risk for Dengue Hemorrhagic Fever (DHF)
  - Severe capillary leak and bleeding at “critical time” - ~3-7 days when defervesce occurs
  - Labs reveal hemo-concentration and thrombocytopenia
  - Transaminitis and CNS effects
Dengue in the U.S.

- Last dengue outbreak in Florida in mid-1930s
- Last US outbreak in Louisiana in 1945
- Dengue is endemic in Puerto Rico
- Hawaii now? – 300 locally transmitted cases since 2015, Big Island
- Limited local transmission in the Keys 2009
  - 28 cases confirmed
  - ~10% of FL Keys residents surveyed in 2009 had dengue antibodies
Reported Dengue Fever Cases in Florida, 2008-2018

- **Acquired in FL**
- **Acquired in USA (not in FL)**
- **Acquired outside USA**
- **Unknown (FL or another state/county)**

<table>
<thead>
<tr>
<th>Year</th>
<th>FL</th>
<th>USA (not FL)</th>
<th>Outside USA</th>
<th>Unknown</th>
</tr>
</thead>
<tbody>
<tr>
<td>2008</td>
<td>32</td>
<td>0</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>2009</td>
<td>21</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>2010</td>
<td>64</td>
<td>131</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>2011</td>
<td>9</td>
<td>12</td>
<td>50</td>
<td>0</td>
</tr>
<tr>
<td>2012</td>
<td>3</td>
<td>18</td>
<td>103</td>
<td>0</td>
</tr>
<tr>
<td>2013</td>
<td>1</td>
<td>24</td>
<td>80</td>
<td>1</td>
</tr>
<tr>
<td>2014</td>
<td>7</td>
<td>78</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>2015</td>
<td>1</td>
<td>75</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>2016</td>
<td>3</td>
<td>57</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>2017</td>
<td>1</td>
<td>25</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>2018</td>
<td>20</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>
Primary Dengue Infection
Dengue Diagnostic Process

Days Post-onset

0 1 2 3 4 5 6 7 8 9 10

Acute Serum
RT-PCR

- Undetermined cause of illness
+ Identification of DENV-1, -2, -3 or -4

Convalescent Serum

IgM ELISA

- No evidence of recent infection
+ Recent infection

IgM -

IgM +

IgG -/+ 4 x (IgG +)
Laboratory Diagnosis of Dengue

- **PCR** – Best early in illness (< 1 week)
- **MAC ELISA**
  - IgM which useful after 5 days of illness
  - Cross-reactivity with other flaviviruses
- **IgG ELISA** – Useful if acute and convalescent titers
- **NS1ELISA** – Useful in acute infections without cross-reactivity
- **PRNT (Plaque Reduction Neutralization Test)**
  - Can determine serotype of infection
Zika Virus

- RNA flavivirus transmitted by mosquitoes
- Initially described in Uganda in 1947 infecting rhesus monkeys (named after Zika forest)
- Human illness first described in 1953 in Nigeria
- Only 13 cases reported in following 57 years
- In 2007, outbreak in Yap Islands of Micronesia (5,000)
- In 2013-14, outbreak in French Polynesia (32,000)
- Only sporadic infections in Southeast Asia
Zika Virus in the Americas

- March 2015, 1st outbreak in Brazil
- By December 2015, estimates by Brazil Ministry of Health that greater than 1.3 million infected
  - September 2015 investigators noted increase in babies born with microcephaly (real vs over-reporting)
- October 2015, Colombia reported initial locally transmitted case
- Rapid spread throughout Central and South America
How Zika virus spread from Africa

1947: Discovered in Uganda
1977-78: Pakistan, Malaysia, Indonesia
2007: Yap, Micronesia
2013: French Polynesia
2015: Brazil

Source: Lancaster University
## Florida Zika Case Counts
(As of 8/17/18)

<table>
<thead>
<tr>
<th>Infection Type</th>
<th>Infection Count 2016</th>
<th>Infection Count 2017</th>
<th>Infection Count 2018</th>
</tr>
</thead>
<tbody>
<tr>
<td>Travel-Related Infections of Zika</td>
<td>1122</td>
<td>225</td>
<td>62</td>
</tr>
<tr>
<td>Locally Acquired Infections of Zika</td>
<td>300</td>
<td>2</td>
<td>0</td>
</tr>
<tr>
<td>Undetermined</td>
<td>49</td>
<td>35</td>
<td>0</td>
</tr>
<tr>
<td>Pregnant Women with Lab Evidence of Zika</td>
<td>299</td>
<td>136</td>
<td>46</td>
</tr>
<tr>
<td>Babies Born with Congenital Zika Syndrome</td>
<td>4</td>
<td>3</td>
<td>0</td>
</tr>
</tbody>
</table>

*Note: These categories are not mutually exclusive and cannot be added together.
Source: [www.zikafreefl.org](http://www.zikafreefl.org) accessed on 8/17/18*
World Map of Areas with Risk of Zika

International areas and US territories
- Area with risk of Zika infection (below 6,500 feet)*
- Area with low likelihood of Zika infection (above 6,500 feet)*
- Areas with no known risk of Zika infection

United States areas
- State previously Reporting Zika
- No Known Zika

*Mosquitoes that can spread Zika usually live in places below 6,500 feet. The chances of getting Zika from mosquitoes living above that height are very low.

### CDC: Zika Virus in the US, States (As of 8/1/2018)

<table>
<thead>
<tr>
<th>Infection Type</th>
<th>2015</th>
<th>2016</th>
<th>2017</th>
<th>2018</th>
</tr>
</thead>
<tbody>
<tr>
<td>Travel-Related Infections of Zika</td>
<td>62</td>
<td>4897</td>
<td>437</td>
<td>34</td>
</tr>
<tr>
<td>Locally Acquired Infections of Zika</td>
<td>0</td>
<td>224</td>
<td>7</td>
<td>0</td>
</tr>
<tr>
<td>Sexual Transmission</td>
<td>0</td>
<td>45</td>
<td>7</td>
<td>0</td>
</tr>
<tr>
<td>Lab Transmission</td>
<td>0</td>
<td>1</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Other/Unknown</td>
<td>0</td>
<td>1</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Pregnant Women with Lab Evidence of Zika (2015 – 7/2018)</td>
<td></td>
<td></td>
<td></td>
<td>2474</td>
</tr>
</tbody>
</table>

## CDC: Zika Virus in the US, Territories (As of 8/1/2018)

<table>
<thead>
<tr>
<th>Infection Type</th>
<th>2015</th>
<th>2016</th>
<th>2017</th>
<th>2018</th>
</tr>
</thead>
<tbody>
<tr>
<td>Travel-Related Infections of Zika</td>
<td>1</td>
<td>145</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Locally Acquired Infections of Zika</td>
<td>9</td>
<td>36,367</td>
<td>665</td>
<td>74</td>
</tr>
<tr>
<td>Other</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Pregnant Women with Lab Evidence of Zika (2015 –7/2018)</td>
<td></td>
<td></td>
<td>4900</td>
<td></td>
</tr>
<tr>
<td>Babies Born with Congenital Zika Syndrome (2015-7/2018)</td>
<td></td>
<td></td>
<td>167</td>
<td></td>
</tr>
<tr>
<td>(Plus 8 Pregnancy losses with Zika-associated birth defects)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Zika Transmission

- Primarily through bite of infected mosquito
  - *Aedes aegypti* > *Aedes albopictus*
  - Daytime biter, often biting multiple humans in blood meal, and lives close to human habitation
Sexual transmission—has occurred just before and after symptoms. Viral RNA found in sperm up to 69 days after infection with median duration of 12 days.
Transmission

- Good evidence of vertical transmission – found in amniotic fluid, placenta and in the brain of infants
- Postnatal transmission - Venezuelan case suggests transmission via breastfeeding (3/2016) – NO change in breastfeeding recommendations
- Theoretically could occur with blood transfusion and organ transplantation
Estimated range of *Aedes aegypti* and *Aedes albopictus* in the United States, 2016
Global distribution of Aedes mosquitoes

Aedes aegypti and Aedes albopictus can spread the Zika virus if infected with it

Aedes aegypti mosquito

Probability of occurrence
- Highest
- Lowest

Aedes albopictus mosquito

Probability of occurrence
- Highest
- Lowest

Predicted global distribution of each species based on statistical distribution models

Source: ELife 2015
Clinical Manifestation

- Most are asymptomatic (~20% with symptoms)
- Incubation period is unknown but suspect < 1 week
- In those with symptoms:
  - Macular or papular rash
  - Fever
  - Arthritis or arthralgias
  - Non-purulent conjunctivitis
- Less common symptoms – headache, myalgias, retro-orbital pain, edema, and vomiting
- Symptoms resolve in 2-7 days
Who Gets Symptomatic Zika?

- Women more than men (43% vs 14%)
- <40 years old more than >40 y/o (62% vs 18%)
- Higher symptomatic rates but not higher overall infection rates
- Reporting bias?
Diagnosis

- If within 7 days of symptoms – RT PCR of serum
  - Only brief window of viremia 3-7 days
  - Negative results do not exclude diagnosis
  - Should consider testing for dengue and chikungunya
- Now urine RT PCR increases window to 2 weeks
- Those > 4 days after symptoms – Zika virus IgM and neutralizing antibodies
  - Unfortunately at times difficult to discern due to some cross-reactivity with dengue and other flaviviruses
  - Plaque-reduction neutralization testing
Current Testing in Pregnancy

- Nucleic acid testing (RT-PCTR) of symptomatic pregnant women within 12 weeks of illness onset
- Pregnant women have about a 3-fold longer estimated median detection of RNA in serum
- Test at least once a trimester, for pregnant women who live in or frequently travel to areas with Zika transmission
- Should be tested within 2 weeks if travel to area of Zika – whether or not symptoms
Pre-Conception Guidelines

- If trying to get pregnant and recently infected
  - Wait 8 weeks after infection to try to get pregnant
  - Men should wait 6 months before trying to conceive if symptomatic infection (~ 8 weeks if asymptomatic)
Treatment and Prevention

- No specific treatment – focus on symptoms
- No vaccine currently exists
- Prevention of exposure to mosquito vector
- Reduce sexual transmission
- If infected, defer blood donation for at least 1 month
- Control mosquito population
Zika and Microcephaly

- First noted in Brazilian outbreak
- RNA found in amniotic fluid and brain tissue of fetuses with microcephaly
- Intracranial calcifications and poly-malformations
- High rates of microcephaly in those whose mother had infection during pregnancy – especially 1st trimester
- Increase in fetal loss also noted in mothers infected
- No associated noted in YAP Islands outbreak however
- Zika virus appears neurotropic (also associated with Guillain-Barre syndrome in some adults infected)
Guillain Barre and Zika Virus

Several countries in the Americas have reported unusual increases in cases of Guillain-Barré syndrome (GBS) in parallel with the ongoing Zika virus outbreak.

During the Zika virus outbreak in French Polynesia (2013 to 2014):
- 74 patients had presented neurologic or autoimmune syndromes after the manifestation of symptoms consistent with Zika virus infection.
- 42 were classified as GBS.
Blood transfusion and Zika Virus

- Zika virus is transmissible via blood products
- Original recommendations:
  - Deferral of blood donors for one month following Zika Virus infection/ exposure
  - Individuals who have donated blood and subsequently develop symptoms consistent with Zika virus infection within 14 days should notify the donation side so the product can be quarantined
Blood transfusion and Zika Virus

- FDA recommendations Feb 2016:
  - Broadened 28 day deferral policy to include anyone who has had symptoms suggestive of a Zika infection in the prior 4 weeks
  - Anyone who has had sexual contact with a person who has traveled to, or lived in, an area of active Zika transmission in the prior 3 months
Blood transfusion and Zika Virus

Current recommendations:

- The FDA:
  - No testing of individual blood donations ($5.3 mln for each positive test)
  - “unless there is an increased risk of local mosquito-borne transmission of Zika virus in a specific geographic area that would trigger individual testing in that location”
Transplant and Zika Virus

- There is no routine screening policy for organ donors or recipients
  - There are reports of possible Zika virus transmission by blood transfusion and it is probable that infection may also be transmitted by organ transplantation

- This virus has a longer duration of viremia compared by other flaviviruses
  - Might be reasonable to screen the donors coming from areas affected by outbreak, with probable exposure in the previous 10-14 days
Transplant and Zika Virus

- The impact of immunosuppression on the natural history of Zika virus infection is unknown.

- Donor characterization includes a full recent travel history.
  - When the potential donor has travelled to Latin America or other affected areas, the SNOD should enquire whether the donor had been bitten by mosquitos and about any associated illness.
  - Should be documented on the Donor Characterization Form.

- Recipient clinicians should balance the risk and possible consequences of a donor-transmitted infection and the risks of harm by declining the organ.
Genetically engineered/modified mosquitoes and Zika Virus

- In mid-2012, British biotech company Oxitec released the super bugs with the aim of reducing the overall mosquito population that spreads dengue fever, the Zika virus, and chikungunya in northeast Brazil.

- The first cases of Zika in the Americas were reported last May after spending decades working its way from Africa through Asia.

- The aim of Oxitec's GM program was to release only male Aedes mosquitoes into the wild and they would in turn produce offspring with their virus carrying female counterparts.
Genetically engineered/modified mosquitoes and Zika Virus

- Tetracycline, which is often used to treat teenage acne, can be found in nature too, showing up in soil, surface water, and food, with some research stating that the GMM survival rate could potentially increase to 15%
How GM mosquitoes work

Life cycle of wild male and female mosquitoes produces blood-sucking females.

- Male and female mate
- Female lays eggs
- Eggs hatch into larva
- Larva develops into pupa
- Pupa grows into adult

Introduction of GM males breaks this cycle as faulty gene causes offspring to prematurely die.

More GM males are created in the lab by adding tetracycline to larvae to allow development.
Future Developments

- Highest resolution image of Zika virus can guide vaccine development
- Compared the glycoprotein interactions and surface properties of Zika with the structures of other flaviviruses (Dengue, Japanese Encephalitis, West Nile and Yellow Fever) – most significant differences were seen at glycosylation loop linked to receptor binding
- Identified likely drug-binding pockets on the surface of the virus
Vaccines

- Novel Human Type 5 Adenovirus-vectored vaccine expressing the premembrane and envelope proteins of Zika Virus
- 2 vaccines studied in mice: expressing ZIKV-Sig-prM-Env and ZIKV-Env proteins
- Both vaccines elicited rapid-onset, robust, and durable antibody response
Innovative Approaches

- Efforts to rapidly identify and manage epidemics are limited by: short diagnostic window, cross-reactivity among flaviviruses, lack of point-of-care diagnostic tools
- Rate-limiting step – access to reference materials and clinical samples
- International Reference Laboratory – knowledge in public domain
DENV  |  CHIKV  |  ZIKV
---|---|---
- A.aegypti/ablopictus
- After 3-14 days, high fever, *severe HA*, retro-orbital pain, severe arthralgias, myalgias, low BP
- Facial flushing (~sunburn), transient morbilliform rash
- High viremia, low WBC
- + tourniquet test
- HEMORRHAGIC FEVER 3-7d as fever drops, 1-3%
  - 20% mortality → 1% with supportive care
- A.aegypti/ablopictus
- After 1-12 days, temp 102F+, *severe arthralgias*, myalgias
  - ~60% morbilliform rash 3+ days into fever
  - HIGH viremia early
  - Lasts ~ 7-10 days
  - “epidemic polyarthritis”
  - Arthralgias may last for MONTHS
- A.aegypti/?ablopictus
- NO SYMPTOMS 80%
- After 3-12 days, 20%: Temp 102F+, *conjunctivitis*, arthralgias, myalgias
  - Diffuse or morbilliform rash
  - LOW viremia early
  - Lasts < 7 days
  - CNS & other fetal anomalies, Guillain-Barré Syndrome
- A.aegypti/ablopictus
- After 1-12 days, temp 102F+, *severe arthralgias*, myalgias
  - ~60% morbilliform rash 3+ days into fever
  - HIGH viremia early
  - Lasts ~ 7-10 days
  - “epidemic polyarthritis”
  - Arthralgias may last for MONTHS
- A.aegypti/?ablopictus
- NO SYMPTOMS 80%
- After 3-12 days, 20%: Temp 102F+, *conjunctivitis*, arthralgias, myalgias
  - Diffuse or morbilliform rash
  - LOW viremia early
  - Lasts < 7 days
  - CNS & other fetal anomalies, Guillain-Barré Syndrome
Questions?
References

- Centers for Disease Control
  www.cdc.gov/chikungunya/
  www.cdc.gov/Dengue
  CDC.Zika in infants and children.

References

References

- Guo Q, at al, A Novel Human Type 5 Adenovirus – Vectored Vaccine for Zika Virus. JID 2018;218:365-77
- Chaves B, at al, Coinfection with Zika Virus (ZIKV) and Dengue Virus Results in Preferential ZIKV Transmission by Vector Bite to Vertebrate Host. JID 2018;218:563-571