Arboviruses in the Southern European Region

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Potential Conflict of Interest disclosure

• Research grants from:
  – Roche Molecular Diagnostics
  – BioMerieux
  – Novartis Diagnostics

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  – TerumoBCT
  – Roche Molecular Diagnostics
  – BioMerieux
  – Novartis Diagnostics - Grifols
ARBOviruses

- Heterogenous group of viruses
  - ARthropods (ticks – mosquitoes - sandflies)
  - BOrne
    - Flaviviruses
      - WNV
      - DENV
      - TBEV
      - USUV
    - Alphaviruses
      - CHIKV
    - Bunyaviridae
      - CCHFV (Nairoviruses)
      - TOSV (Phleboviruses)
Flamingo Nat’l Park, FL. 2010
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West Nile virus (WNV), a mosquito-borne flavivirus in the Japanese encephalitis antigenic group, has caused sporadic outbreaks in humans, horses and birds throughout many of the warmer regions of Europe for at least 20 years. Occasional cases of West Nile encephalitis have also been associated with infected blood transfusions and organ donation. Currently, WNV appears to be expanding its geographical range in Europe and causing increasing numbers of epidemics/outbreaks associated with human morbidity and mortality. This brief review report is on the current epidemic situation regarding WNV in Europe, highlighting the clinical, diagnostic and preventive measures available for controlling this apparently emerging human pathogen.
West Nile Virus (WNV)

- *Flaviviridae (Flavivirus)*
- ss (+) RNA
- Virion (spherical shape), 40-50 nm
- *envelope* (proteins E and prM)
- Capsid (protein C 20-30 nm)
West Nile Cycle

- Vertical transmission
- Dead-end hosts
- Rural cycle
- Mosquito movement
- Bird movement/migration
- Urban cycle
- Culex tarsalis (other Culex spp.)
- Culex pipiens (other Culex & Aedes spp.)
- Blood transfusion, organ transplantation
Human Transmission

- Mosquito
  - Laboratory acquired
- Blood transfusions and SOT
  - Transplacental transmission
  - Breast feeding
Theoretical Depiction of WNV Human Viremia & Immune Response

- Virus Assays
- Serology Assays
- ELISA

WN viremia

IgM

IgG Neutralizing Ab

Days Post Onset

CNS illness

cpfu/ml

250

20

2
Fever, weakness, headache, myalgia, nausea, vomiting

Encephalitis (meningoencephalitis)

Incubation period: 3-14 days

1 CNS disease case = ~150 total infections

~20% “West Nile Fever”

~80% Asymptomatic

<1% CNS disease

Very crude estimates
Flaviviruses in Europe: Complex Circulation Patterns and Their Consequences for the Diagnosis and Control of West Nile Disease

Int. J. Environ. Res. Public Health 2013, 10, 6049-6083;
Heterogeneity of West Nile virus genotype 1a in Italy, 2011
Giada Rossini, Fabrizio Carletti, Roberto Rigoli, Sandro Piga, Patrizia Bagnarelli, Paolo Gaibani, Anna Pierro, Alessandro Nanni Costa, Paolo Grossi, Giuseppe Ippolito, Maria Paola Landini, Antonino Di Caro, Maria Rosaria Capobianchi and Vittorio Sambri

Emergence of WNV Lineage 2

Detection of West Nile virus lineage 2 in mosquitoes during a human outbreak in Greece

A. Papa¹, K. Xanthopoulou¹, S. Gewehr² and S. Mourelatos²
1) Department of Microbiology, Medical School, Aristotle University of Thessaloniki, Thessaloniki and 2) Eco-Development, S.A., Thessaloniki, Greece

Clinical Microbiology and Infection, Volume 17 Number 8, August 2011

Human case of autochthonous West Nile virus lineage 2 infection in Italy, September 2011

West Nile virus in Europe: emergence, epidemiology, diagnosis, treatment, prevention.
V. Sambri et al. CMI 2013, in press
The Complex Epidemiological Scenario of West Nile Virus in Italy

L. Barzon et al.  

Int. J. Environ. Res. Public Health 2013, 10, 4669-4689;
Seroprevalence of West Nile Virus–Specific Antibodies in a Cohort of Blood Donors in Northeastern Italy

A. Pierro et al.

- IgG and IgM levels against West Nile virus (WNV) were measured in 20,033 serum samples that were obtained between October 2008 to September 2009 from 9913 blood donors in the district of Ferrara, northeastern Italy.
- As confirmatory test, a microneutralization assay was used to detect the presence of neutralizing antibodies against WNV.
- Sixty-eight subjects (0.69%) were positive for anti-WNV by immunofluorescence assay.
- Large differences in the prevalence of antibodies to WNV were noted between towns in the area evaluated.

Persistence of Anti-West Nile Virus-Specific Antibodies Among Asymptomatic Blood Donors in Northeastern Italy

- The development and persistence of anti-West Nile Virus (WNV) immunoglobulin G (IgG)- and IgM-specific antibodies were investigated in 68 asymptomatic blood donors (BDs) previously tested as positive between October, 2008, and September, 2009, and living in northeastern Italy.
- Our study showed that WNV-specific IgG titers became negative (41%) or decreased (33%), while they increased in a smaller percentage (10%); 16% of BDs showed no titer variation.
- **Reversion to seronegative status within a short time frame suggests that WNV surveillance should be maintained year after year.**
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Dengue in Europe

Several imported cases (travellers) 2010-2013: autochthonous transmission in Southern France, Croatia, Madeira

First cases of autochthonous dengue fever and chikungunya fever in France: from bad dream to reality!
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Detection of Usutu Virus Within a West Nile Virus Surveillance Program in Northern Italy

Marco Tamba, Paolo Bonilauri, Romeo Bellini, Mattia Calzolari, Alessandro Albieri, Vittorio Sambri, Michele Dottori, and Paola Angelini
Comparative Genomic and Phylogenetic Analysis of the First Usutu Virus Isolate from a Human Patient Presenting with Neurological Symptoms

Paolo Gaibani¹*, Francesca Cavrini¹, Ernest A. Gould², Giada Rossini¹, Anna Pierro¹, Maria Paola Landini¹, Vittorio Sambri³

First evidence of simultaneous occurrence of West Nile virus and Usutu virus neuroinvasive disease in humans in Croatia during the 2013 outbreak

T. Vilibe-Cavlek · B. Kaic · L. Barbic · I. Pem-Novosel · V. Slavic-Vrizic · V. Lesnikar · S. Kurecic-Filipovic · A. Babic-Erceg · E. Listes · V. Stevanovic · I. Gjenero-Margan · G. Savini
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Chikungunya

- Spherical virions, about 70 nm diameter with envelope
- Nucleocapsid icosaedric (40nm, nucleoprotein C).
- Envelope: spikes (glycoproteins gpE1 e gpE2)
- ssRNA (+), fully sequenced
Chikungunya: epidemiology

www.chikungunya.net
Bordi et al., Clin Infect Dis 2008.
Number of cases by lab results Emilia-Romagna

Index case lab confirmed 1
Lab confirmed 247

In total, 248 (247+1 case index) persons affected, from four provinces (Ravenna, Forlì/Cesena, Rimini, Bologna)
Cluster of CHIKV Infection Emilia Romagna August – November 2007
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      – TOSV (Phleboviruses)
Sandfly-borne phleboviruses of Eurasia and Africa: Epidemiology, genetic diversity, geographic range, control measures

Cigdem Alkan a, Laurence Bichaud a, Xavier de Lamballerie a,b, Bulent Alten c, Ernest A. Gould a,b, Rémi N. Charrel a,b,*

Antiviral Research 100 (2013) 54–74
Toscana virus infections: A case series from France

J. Dupouey a,b,c, L. Bichaud a,b,c, L. Ninove a,b, C. Zandotti a,b, L. Thirion-Perrier a,b, X. de Lamballerie a,b, R.N. Charrel a,b,*

Table 1  Epidemiological, clinical and biological data of the TOSV infection cases.

<table>
<thead>
<tr>
<th>Patient</th>
<th>Sex</th>
<th>Age</th>
<th>Onset</th>
<th>Time</th>
<th>Clinical data</th>
<th>CSF analysis</th>
<th>Acute phase</th>
<th>Convallescent phase</th>
<th>OMS criteria</th>
<th>Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>IF</td>
<td>MN</td>
<td>PCR</td>
<td></td>
</tr>
<tr>
<td>P1</td>
<td>H</td>
<td>47</td>
<td>Aug 2009</td>
<td>5 days</td>
<td>AM, severe headache and vomiting</td>
<td>230/93%</td>
<td>G+M+</td>
<td>POS</td>
<td>—</td>
<td>NEG</td>
</tr>
<tr>
<td>P2</td>
<td>H</td>
<td>39</td>
<td>Aug 2009</td>
<td>3 days</td>
<td>AM</td>
<td>178/70%</td>
<td>G+M+</td>
<td>POS</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>P3</td>
<td>H</td>
<td>17</td>
<td>Jun 2010</td>
<td>5 days</td>
<td>AM with fever</td>
<td>840/70%</td>
<td>G+M+</td>
<td>NEG</td>
<td>POS</td>
<td>—</td>
</tr>
<tr>
<td>P4</td>
<td>H</td>
<td>38</td>
<td>Aug 2008</td>
<td>—</td>
<td>AM and hepatomegaly</td>
<td>120/90%</td>
<td>G+M+</td>
<td>POS</td>
<td>G+M+</td>
<td>1/40</td>
</tr>
<tr>
<td>P5</td>
<td>F</td>
<td>20</td>
<td>Jul 2008</td>
<td>—</td>
<td>AM</td>
<td>65/50%</td>
<td>G+M+</td>
<td>NEG</td>
<td>POS</td>
<td>—</td>
</tr>
<tr>
<td>P7</td>
<td>F</td>
<td>50</td>
<td>Jul 2008</td>
<td>—</td>
<td>Fever with general asthenia and left lower limb fascitis</td>
<td>—</td>
<td>G–M+</td>
<td>NEG</td>
<td>G+M+</td>
<td>—</td>
</tr>
<tr>
<td>P9</td>
<td>F</td>
<td>4</td>
<td>Sep 2009</td>
<td>7 days</td>
<td>Encephalitis</td>
<td>54/59%</td>
<td>G–M–</td>
<td>eneg</td>
<td>G+M+</td>
<td>NEG</td>
</tr>
<tr>
<td>P11</td>
<td>F</td>
<td>33</td>
<td>Nov 2009</td>
<td>13 days</td>
<td>Encephalitis with fever and drowsiness</td>
<td>150/90%</td>
<td>G+M+</td>
<td>NEG</td>
<td>G+M+</td>
<td>1/20</td>
</tr>
<tr>
<td>P12</td>
<td>H</td>
<td>61</td>
<td>Jul 2010</td>
<td>—</td>
<td>Fever with myositis</td>
<td>—</td>
<td>G+M+</td>
<td>1/80</td>
<td>NEG*</td>
<td>G+M–</td>
</tr>
<tr>
<td>P13</td>
<td>H</td>
<td>43</td>
<td>Jul 2004</td>
<td>—</td>
<td>AM</td>
<td>28/95%</td>
<td>G–M+</td>
<td>—</td>
<td>1/40</td>
<td>—</td>
</tr>
<tr>
<td>P14</td>
<td>H</td>
<td>76</td>
<td>Jul 2007</td>
<td>—</td>
<td>Left upper limb hemiparesis with headache and confusion</td>
<td>—</td>
<td>G–M+</td>
<td>1/40</td>
<td>—</td>
<td>G+M+</td>
</tr>
<tr>
<td>P15</td>
<td>H</td>
<td>64</td>
<td>Jun 2009</td>
<td>1 day</td>
<td>Encephalitis, seizures with fever and stiff neck</td>
<td>46/0%</td>
<td>G+M+</td>
<td>1/160</td>
<td>—</td>
<td>G+M+</td>
</tr>
<tr>
<td>P16</td>
<td>F</td>
<td>42</td>
<td>Mar 2010</td>
<td>1 day</td>
<td>Fever with cervical lymphadenopathy</td>
<td>—</td>
<td>G+M+</td>
<td>—</td>
<td>NEG</td>
<td>—</td>
</tr>
<tr>
<td>P17</td>
<td>H</td>
<td>30</td>
<td>Apr 2007</td>
<td>—</td>
<td>Fever with headaches and sudden right facial paralysis</td>
<td>&lt;5</td>
<td>G–M+</td>
<td>NEG</td>
<td>G+M–</td>
<td>—</td>
</tr>
</tbody>
</table>

AM: Aseptic meningitis.
CSF analysis: Leukocytes (/mm³) – Proportion of lymphocytes (%), in cerebral spinal fluid (CSF).
PCR: Polymerase chain reaction targeting Toscana virus genome in CSF (* in blood).
—: Not available.
Dad, there's a mosquito in my room.

Hunh? It's okay, Zoe, go back to bed.

Okay. I won't worry about West Nile virus, or malaria...

...or encephalitis, or dengue fever, or...

I miss the boogeyman.