Facilitating Dengue Vector Surveillance for Disease Control: A pilot study

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What would you say is the deadliest animal on Earth?
The World’s Deadliest Animals
Number of people killed by animals, 2015

- 830,000 Mosquito
- 580,000 Human

- 60,000 Snake
- 24,200 Sandfly
- 17,400 Dog
- 8,000 Kissing bug
- 4,400 Freshwater snail
- 3,500 Scorpion
- 3,500 Tsetse fly
- 1,600 Tapeworm
- 500 Hippopotamus
- 100 Elephant
- 60 Bee
- 40 Jellyfish
- 6 Shark

Global Burden of Dengue

Endemic in >100 countries; 390 million infections, 96 million clinical cases annually

Sources:
Background: Dengue Virus

• Mosquito-borne viral infection

• Confirmed serotypes: DENV1-4 (DENV-5 is emerging)

• Range of disease - asymptomatic to flu-like symptoms and severe, fatal disease

Female Ae. aegypti mosquito in the process of acquiring a blood meal from her human host. Source: Centers for Disease Control and Prevention

Dengue Vector Control Strategies

### Existing Methods

#### Immature control

- **Major categories**
  - Container cleaning (bleach/wash/dump)
  - Container manipulation (treated covers/polystyrene beads)
  - Container treatment
  - Social campaigns (education, source reduction)
  - Environmental Management
  - Legislation

#### Adult control

- **Major categories**
  - Space spraying
  - Indoor residual spraying
  - Personal protection

- **Space spraying**
  - Truck ULV, Low-flying aircraft, hand-held portables, perifocal treatment

- **Personal protection**
  - DEET
  - Picaridin
  - Bed nets
  - Consumer products

- **Indoor residual spraying**

### Methods under Development

- NEW entomopathogenic fungi
- Molecular insecticides, medea/HEGs, new insecticides

### Source

Diagnostic Limitations

- **PCR**
  - Expensive
  - Time consuming
  - Specialized laboratory

- **ELISA**
  - Cross-reactivity
  - Seroprevalence only

- **PRNT**
  - Expensive
  - Time consuming
  - Specialized laboratory

VecTOR dipstick

- Immunochromatographic wicking dipstick assay
- Works by antigen-antibody sandwich principle
- Developed by VecTOR Test Systems

Study Purpose

- **Goal**
  Improve dengue vector surveillance system at local level

- **Objective**
  Evaluate a rapid diagnostic tool to detect DENV in mosquitoes

- **Aims**
  - Quantify the specificity and sensitivity of the VecTOR dipstick assay to detect DENV in field-collected mosquitoes and control samples
  - Assess the acceptability of the VecTOR distick assay among laboratory personnel via questionnaire
Study Sites

Nakhon Pathom Province, Sam Phran District

Bangkok Metropolis, Sai Mai district

Field Activities

- Household selection
- Indoor collection (A)
  - Purposive sampling
  - Prokopack aspirator
- Outdoor collection (B)
  - Simple Random Sampling
  - BG-Sentinel traps
- Mosquito storage

Mosquito collection. Source: Personal Archive
Laboratory Test Work Flow

- 50 dipsticks
  - 30 (pooled mosquitoes)
    - 30 samples
  - 4 (preliminary research)
  - 16 (training)
    - 8 positive control
    - 8 negative control
## DENV Detection Summary

<table>
<thead>
<tr>
<th>Site of collection</th>
<th>Culex and Anopheles</th>
<th>Aedes Female</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sai Mai district</td>
<td>57</td>
<td>7</td>
</tr>
<tr>
<td>Sam Phran district</td>
<td>947</td>
<td>260</td>
</tr>
<tr>
<td>KU campus</td>
<td>21</td>
<td>14</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td>1025</td>
<td><strong>281</strong></td>
</tr>
</tbody>
</table>

Out of 281 pooled mosquitoes into 30 tubes

<table>
<thead>
<tr>
<th>Sample Type</th>
<th>PCR</th>
<th>VecTOR dipstick</th>
</tr>
</thead>
<tbody>
<tr>
<td>DENV+ (n=1)</td>
<td>n=1</td>
<td>N=0</td>
</tr>
<tr>
<td>DENV- (n=29)</td>
<td>n=29</td>
<td>N=29</td>
</tr>
<tr>
<td>DENV+ control samples (n=8)</td>
<td>n=8</td>
<td>N=4</td>
</tr>
<tr>
<td>DENV- control samples (n=8)</td>
<td>n=8</td>
<td>N=8</td>
</tr>
</tbody>
</table>

56.3% Cohen’s Kappa agreement between PCR and VecTOR dipstick
Questionnaire

- Distributed to laboratory personnel

- 15 closed-ended questions

- Three sections (5 questions each)
  - Ease-of-use
  - Practicality
  - Integration

- Six-point Likert rating scale
  - “0” (not applicable)
  - “1” (strongly disagree)
  - “5” (strongly agree)

VecTOR dipstick assay training. Source: Personal Archive
Questionnaire Summary

Section Scores
(VecTOR Dipstick)

Ease of Use: 4.47
Practicality: 3.29
Integration: 3.20
Summary

• Goal of dengue surveillance systems is to control spread of virus within human populations

• Dengue diagnostic tests are an essential component of disease surveillance and management systems - need for rapid, inexpensive, point-of-care tools with high sensitivity and specificity

• VecTOR dipstick assay has potential as a ‘user-friendly’ tool where capacity for PCR testing is limited:
  – High specificity (100%)
  – Fair sensitivity (44%)
  – Considered easy to use

• Practicality and integration were questionable
Recommendations and Next Steps

• Train other lab personnel and community members

• Test in other regions (field-collected mosquitoes) - other flaviviruses and arboviruses to assess cross-reactivity

• Larger sample size (for sensitivity and specificity)

• Further research on practicality of the assay
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  – The Department of Fishery Science, Kasetsart University
  – The Head Office of Sam Pran District, Nakhon Pathom Province
  – All community workers of Sam Pran District and Saimai District
Thank You for Your Attention!