



Next-generation Sequencing in Emerging Infections

16th Annual Scientific Meeting
Stanley Ho Centre for Emerging Infectious Diseases

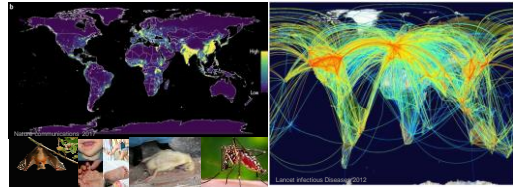
11th June 2019

Le Van Tan, PhD

Oxford University Clinical Research Unit, Viet Nam



Relative risk distribution of (zoonotic) emerging infectious diseases



The diagnostic challenges posed by emerging infectious diseases

- Viruses evolve quickly. As such primers need to be regularly updated. But how?
- For novel viruses, which primers to use?
- For some diseases (respiratory and brain infections), so many targets to cover.
- Viral load might be low.
- **There is an urgent need to explore novel diagnostic approaches.**



Next-generation sequencing for (emerging) infectious disease diagnosis: the principle



Case study #1

September 16th 2016

A previously healthy 16-year-old boy in Southern Vietnam suddenly developed fever, back pain, diarrhea and limb weakness

September 18th 2016

Admitted to our hospital for tropical diseases in Ho Chi Minh City with high fever, neck stiffness and flaccid paralysis of 4 limbs.

Acute cerebrospinal fluid (CSF) lab results:

- Pleocytosis
- Normal glucose
- Normal lactase
- Elevated protein



Open Forum Infectious Diseases, 2017



Initial laboratory investigations

Routine diagnosis

- Gram stain
- Bacterial culture
- Cryptococcal antigen test (LFA)
- Acid-fast bacilli examination
- Herpes simplex virus PCR
- NS1 Dengue
- Japanese encephalitis virus IgM
- Enterovirus RT-PCR
- Dengue RT-PCR
- Zika RT-PCR

All negative



CSF
Plasma
Urine
Rectal swab

Viral metagenomics results

| | CSF | Rectal swab | Plasma |
|----------------------|-----------|-------------|-----------|
| No of reads | 550,644 | 597,032 | 754 |
| Pathogen | Not found | Not found | Not found |
| No of pathogen reads | Not done | Not done | Not done |

Article Navigation

Was the detection of JEV in urine just a contamination?

Japanese Encephalitis Virus RNA Not Detected in Urine

Hui Zhao , Yu-Guang Wang, Yong-Qiang Deng, Ke-Yu Song, Xiao-Feng Li, Hong-Jiang Wang, Chao-Min Zhu, E-De Qin, Cheng-Feng Qin

Clinical Infectious Diseases, Volume 57, Issue 1, 1 July 2013, Pages 157–158,
<https://doi.org/10.1093/cid/cit169>

Published: 26 March 2013

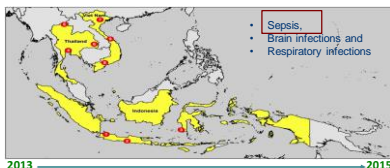
Additional JEV testing

| Test | Admission samples | | | Second samples (15 days later) | | |
|-----------------|-------------------|--------|-------|--------------------------------|-------|-------|
| | CSF | plasma | Urine | CSF | Serum | Urine |
| JEV IgM ELISA | Neg | Neg | ND | Pos | Pos | ND |
| Deep Sequencing | Neg | Neg | Pos | ND | ND | ND |
| JEV RT-PCR | Neg | Neg | Pos | ND | ND | Neg |

- JEV was responsible for the cause of paralysis.
- 9th November 2016: transferred to rehab hospital, marked with weakness of all limbs.

Japanese encephalitis virus (JEV)

- Vaccine: Available
- > 50,000 cases annually
- No antiviral drug available
- Mortality/sequelae: >40%
- 'Gold standard' for diagnosis:
 - CSF IgM: late sample/convalescent
 - CSF PCR (not useful)
 - Can urine be useful for early JEV diagnosis?

Case study #2:
Pathogen discovery (disease X, WHO)

Burden of sepsis

- 30 million cases and 6 million deaths per year

Aetiology

- Unknown in ~50% of the patients

Causes and outcomes of sepsis in southeast Asia:
a multinational multicentre cross-sectional study

Southeast Asia Infection Disease Clinical Research Network*

