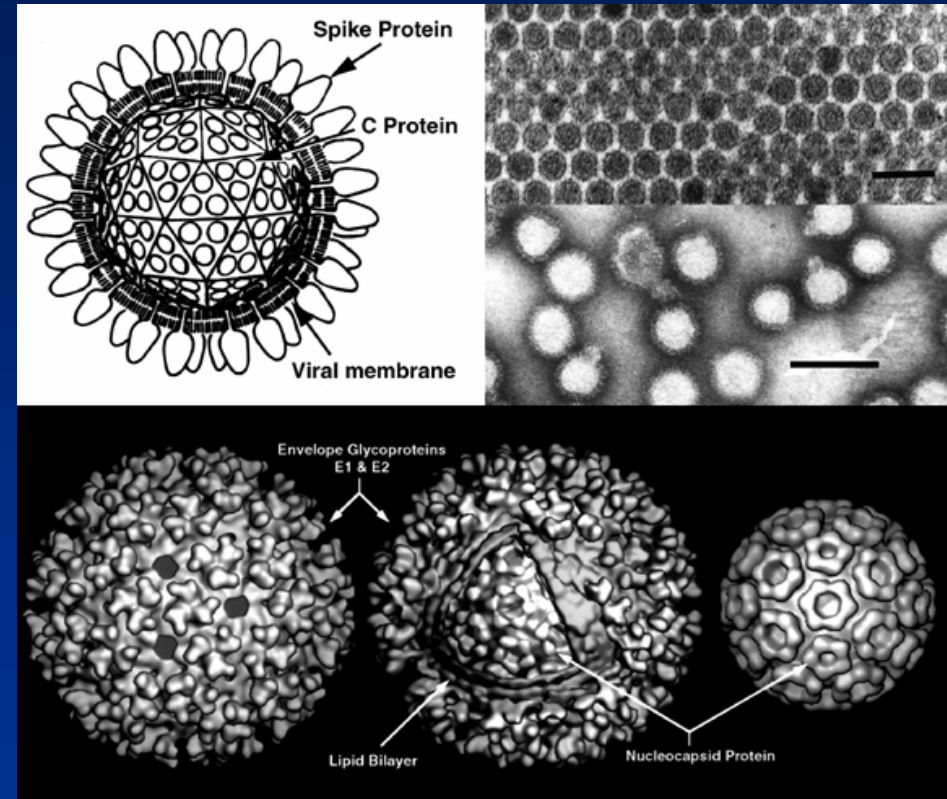


**Recent outbreaks  
of  
chikungunya in Sri Lanka  
and  
the role of “Asian Tigers”**

# Introduction

## CHIK Virus Classification:

- An ARBOVIRUS
- Family - Togaviridae
- Genus – *Alphavirus*



\*\* Enveloped, positive- strand RNA virus.

# Epidemiology



- Chikungunya virus (CHIKV) was first identified in Tanzania in 1953.
- In Asia, first isolated in Bangkok, Thailand in 1958.
- First isolation in India was in 1963.
- Repeated outbreaks in Thailand, India and Indonesia have been reported since then.

# CHIK in Sri Lanka

- Since the last outbreak in 1969, CHIK has been considered as disappeared from Sri Lanka.
- No active or passive surveillance has been carried out in the country until it reappeared in 2006.

# Latest outbreak

- In 2005-2006, epidemics of CHIK occurred in many Indian ocean Islands and countries.
  - Comoros
  - Reunion Islands
  - Mauritius
  - Seychelles
  - India
  - Sri Lanka

# Clinical Features

- High grade fever (up to 40<sup>0</sup> C- for 2-4 days)
- Severe Arthralgia- Not responding to simple anaelgesics
- Arthritis- Polyarthritis /Mainly involving knee joint, ankle joint, elbow joint and small joints
- Myalgia- Involving several muscle groups
- Skin rash – erythematous, maculopapular



# *Clinical Features Cont...*

## Associated symptoms

- Headache
- Nausea and vomiting
- Backache
- Facial swelling
- Photophobia



# *Clinical Features Cont...*

Severe complications

**Rare**, but include

- Meningo-encephalitis
- Hepatic failure

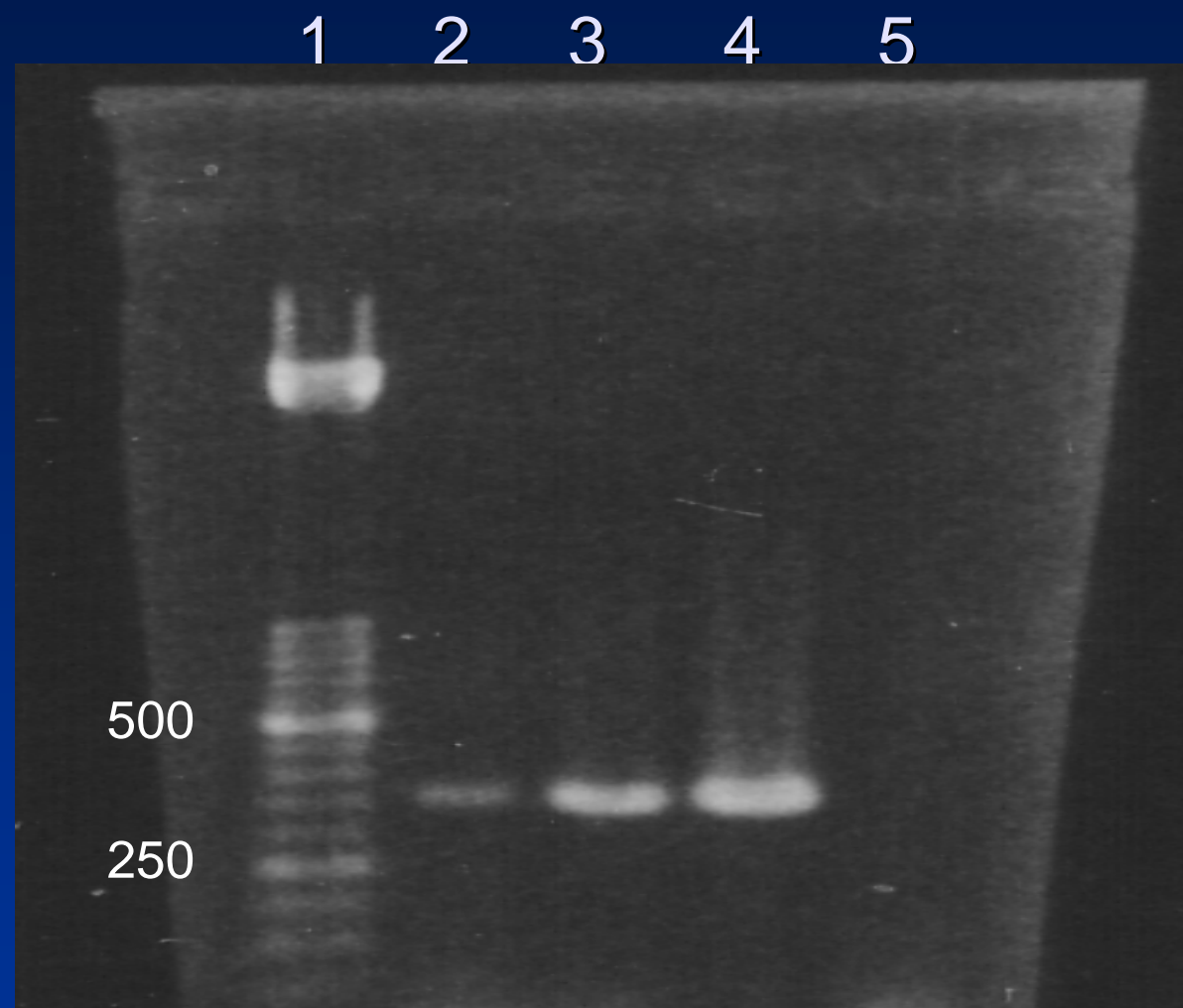


**Confirmation of 2006 outbreak  
by  
RT-PCR**

# Methodology

- Clinical suspicion: Patients with fever < 4 days accompanied by either arthralgia or arthritis or/and myalgia.
- RNA was extracted from serum using QiAmp RNA Extraction kits.
- CHIK RNA was amplified by Reverse Transcriptase polymerase Chain Reaction ( RT – PCR), performed using CHIK specific primers.
- Amplified products (354 bp) were visualized in 1.5 % agarose gel followed by ethidium bromide staining.

# Gel picture showing amplified CHIK RNA



- Lane 1 - 50 bp DNA ladder
- Lanes 2, 3 - CHIK clinical samples
- Lane 4 - (+) Ve control
- Lane 5 - (-) Ve control

# Results

<b>Area</b>	<b>No. tested</b>	<b>No. positive</b>
Hambantota	01	01
Kalmunai	21	16
Ampara	12	03
Colombo	20	12
Gampaha	03	02
Kalpitiya	09	00
	66	34 <b>(51%)</b>

# Results & Conclusions Cont...

- Clinical manifestations were more profound in the early stages of the disease.
- Fever with either arthralgia or arthritis or both were the commonest symptoms observed in PCR positive patients.
- Most commonly affected joints were Knee (67.6%), Ankle (55.9%) and small joints of hands and feet (52.9%).
- Arthritis was more prominent in the ankle than the knee joints.
- Haematological indices of PCR positive and PCR negative patients did not show significant difference.

# “Transmission of Chikungunya”

## Role of Asian Tigers and Genetic Divergence

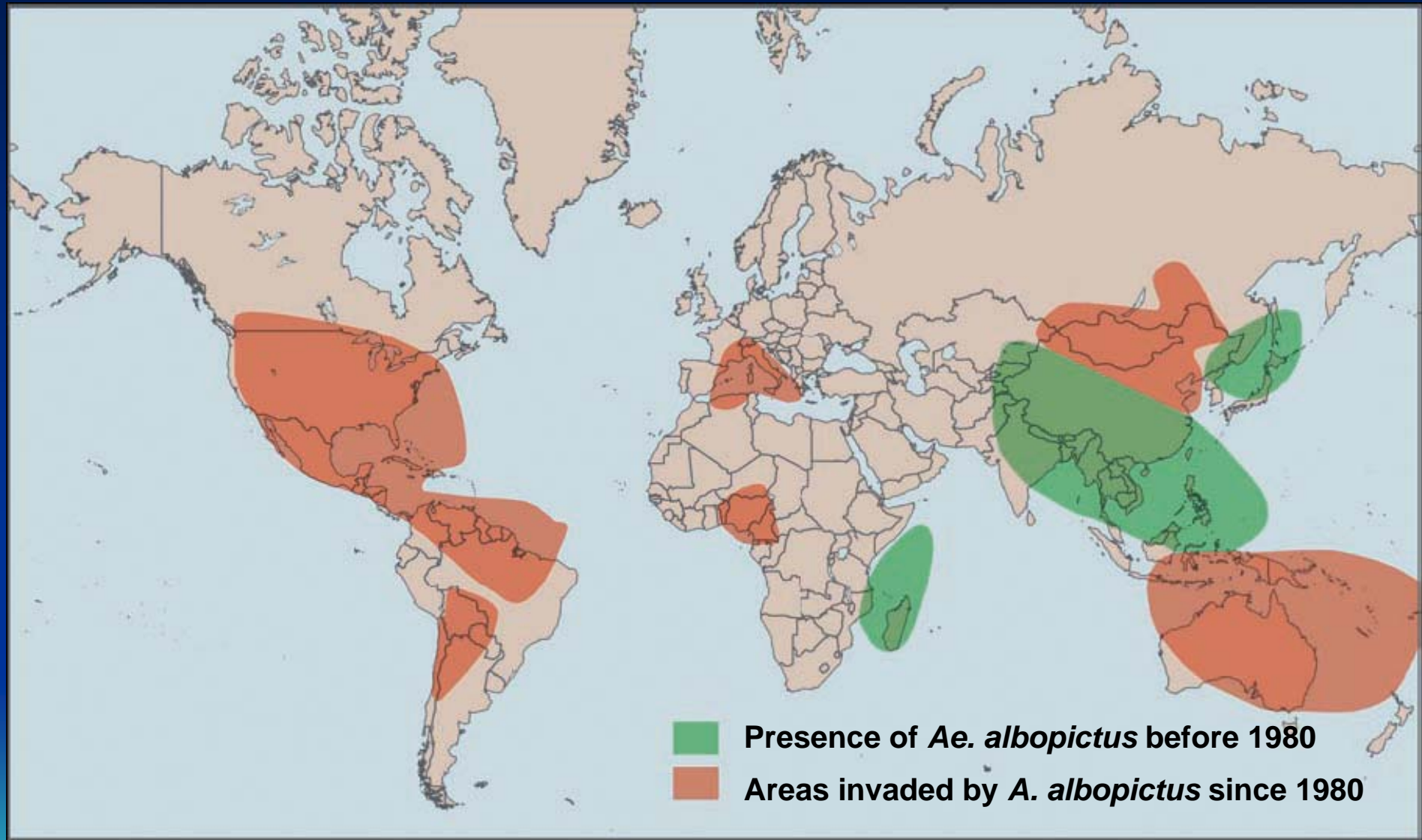


# Introduction

- *Aedes albopictus* is generally considered to have a lower vector capacity for arboviruses than *Ae. aegypti*.
- The expansion of global **air travel** and **seaborne trade** has enabled the insect to move great distances in short periods despite geographic barriers.
- In the past 50 years, the anthropophilic *Ae. albopictus* has spread to all continents and adapted to most climates.



# World Distribution of the *Aedes albopictus* Mosquito



Charrel et al., (2007). *New Eng. J. Med* 356(8): 769-771



# Spread of CHIK in Sri Lanka

- In 2006-2007, CHIK was mainly seen in dengue-endemic urban town areas such as, Western, Central, North Western and Southern Provinces which are inhabited by *Ae. aegypti*.
- A change in the trend was seen in 2007-2008, when CHIK appeared in rural areas: Matale, Mawanella, ( Central Province) Erathna, Kuruwita, Pallebedda, Eheliyagoda and villages in Kagalle disirict (Sabaragamuwa Province) mainly with banana and rubber plantations – mainly inhabited by *Ae. albopictus*.

?? Suggested the possible role of *Ae. albopictus* in CHIK transmission

# Results

Sample collection in 2008 (SL)

Area	Month	No. collected	No. PCR (+)
Pallebedda	March	67	33 (49.3%)
Eheliyagoda	April	153	100 (65.4%)

No. sequenced-in an ongoing collaborative study in Singapore.

SL 2007 – 2

SL 2008 – 55 (Pallebedda = 12, Eheliyagoda = 43)















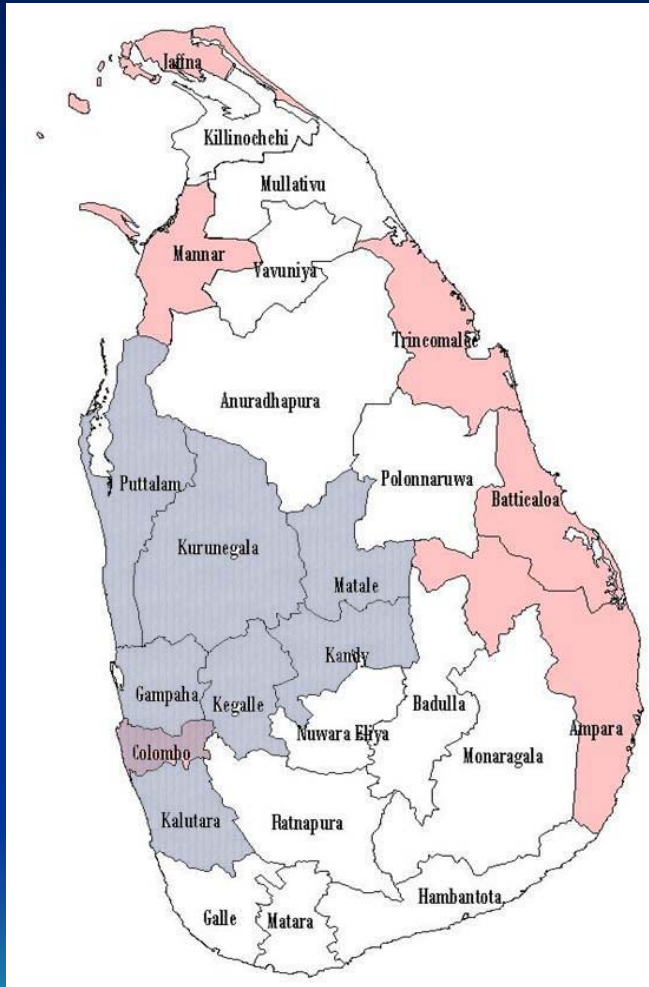




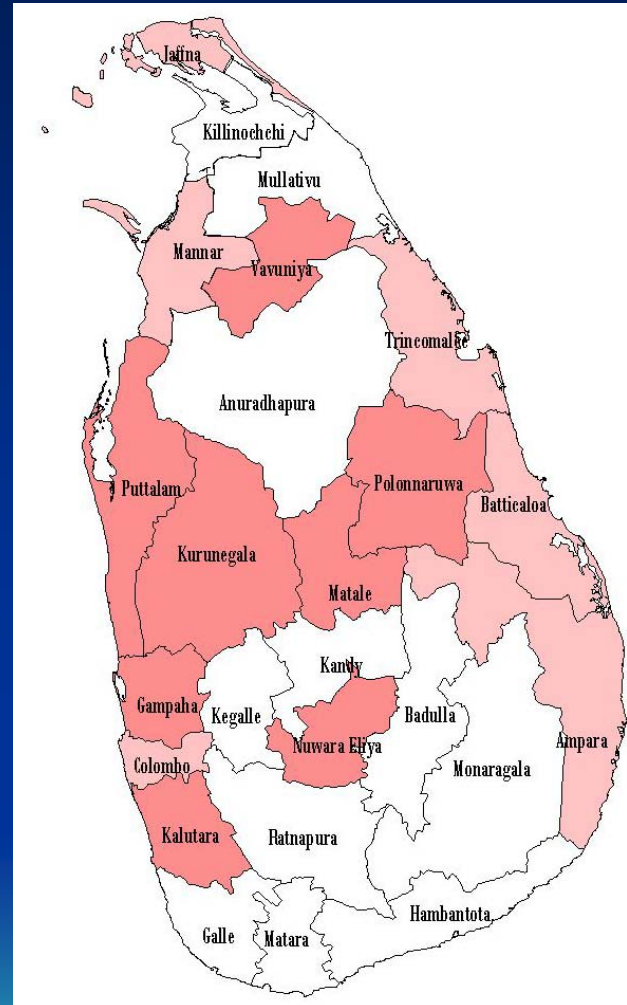




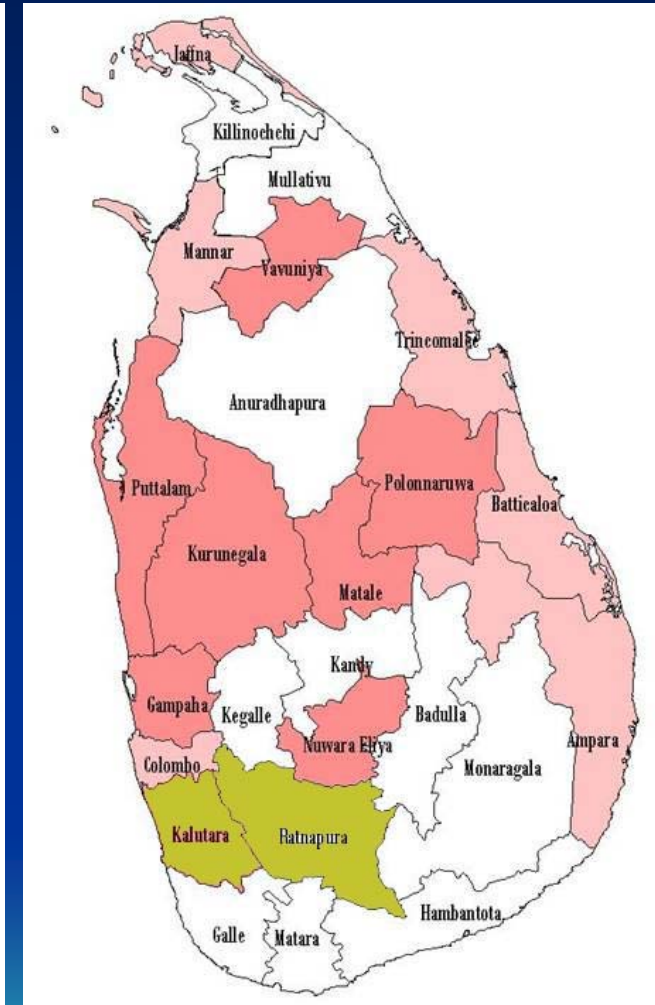
# Map of Sri Lanka showing the spread of chikungunya and dengue fever



2006 – CHIK & DEN




2007 - CHIK



2008 - CHIK



# Role of *A. albopictus* in transmission of chikungunya

- Until 2005, *Ae. aegypti* was considered as the major vector of CHIK.
  - Evidence that *Ae. albopictus* as a highly efficient vector of CHIK was speculated following massive outbreaks in 2005.
    - ✓ In Reunion and Mauritius, *Ae. albopictus* was the primary vector.
    - ✓ It was revealed during CHIK outbreaks in Cameroon (2006) and Gabon (2007) that *Ae. albopictus* has displaced *Ae. aegypti*.
    - ✓ Spread of CHIK in Italy in 2007 was consistent with its presence in the country.
- 

# Chikungunya: No Longer a Third World Disease



“The big surprise of the outbreak at La Réunion was that the infamous Asian tiger mosquito, which is spreading fast across Europe and the United States, proved an excellent vector. This summer, Italy had a small chikungunya outbreak, the first ever in Europe.” Enserink M. (2007). *Science* 318: 1860-1861

# How did it become an efficient vector of CHIK?

- CHIK isolates classically belonged to three phylogroups
  - ✓ West African
  - ✓ Asian
  - ✓ East, Central and South African (ECSA)
- The outbreak in Reunion in 2005 was due to an ECSA virus
- All isolates before 2005 (Even those moved to RU initially) showed Alanine at position 226 (A226) of E1 gene.
- *Ae. aegypti* was the main vector of this genotype.



- Evidence suggests that the **virus adapted to *Ae. albopictus*** through “evolutionary convergence”.
- Adaptation was due to a single point mutation (226V) in CHIK envelope protein (E1) – “**First detected during 2005 outbreak in CHIK history**”
- 226V has been shown to confer survival and transmission advantage to CHIK virus in *Ae. albopictus*.
  - ✓ enhances the viral replication in mosquito midgut.
  - ✓ yields 100-fold higher virus concentrations as compared to *Ae. aegypti* in mosquito salivary glands.

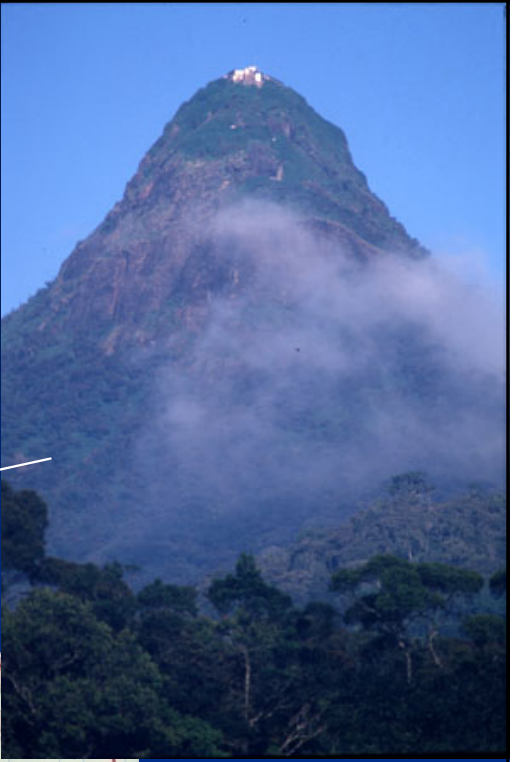
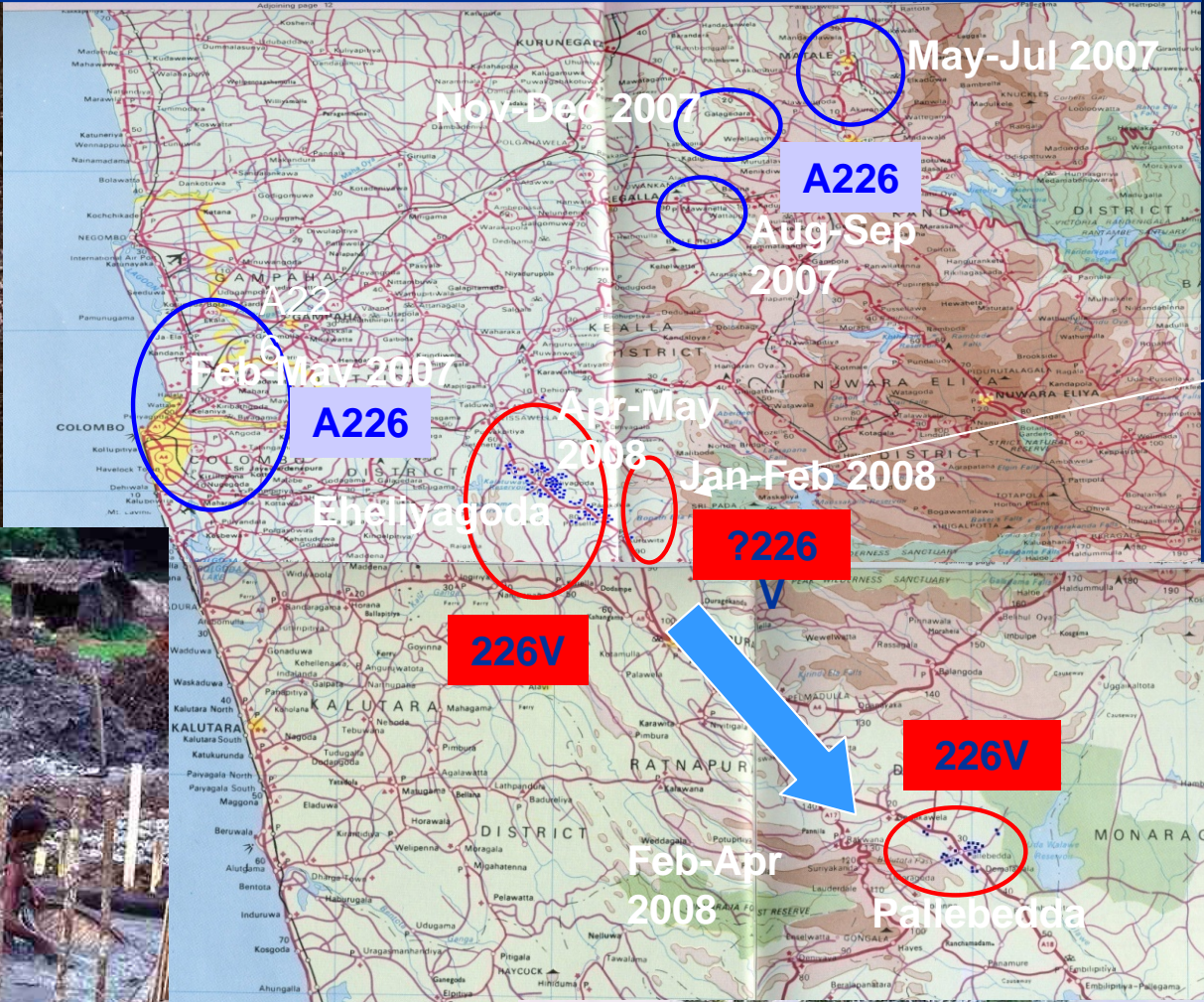


■ 226V has been acquired by the virus as an independent adaptive mutation, at least in 3 occasions, in response to a similar requirement of transmission by *Ae. albopictus* –  
*“Evolutionary Convergence”*

1. *In Reunion Islands (within a year in 2005)*
2. *In India (within 1-2 years in 2006-2007)*
3. *In Cameroon (2006) and Gabon (2007).*



# Recent spread of Chikungunya in Sri Lanka





# Discussion

The majority (98.2%) of 2008 isolates showed 226V irrespective of geographical region.

Both 2007 isolates (collected from Ragama and Matale) showed A226.

In 2008, chikungunya was seen spreading to rural areas with banana and rubber plantations – ideal for *Ae. albopictus*.

This observation is coincident with the shift from A226 to 226V in the viral population.

(A similar observation has been seen in India. The first outbreak of 226V in India was in Kerala in an area with rubber plantation – Kumar et al., 2008)



# Conclusion

Due to widespread of *Ae.albopictus* in the country, with a single mutation (226V) in the virus, CHIK has become a rural infection during 2007-2008.

With the widespread of *Ae.albopictus* the possibility of Chikungunya becoming a global threat in the near future can not be ruled out.



Thank you

