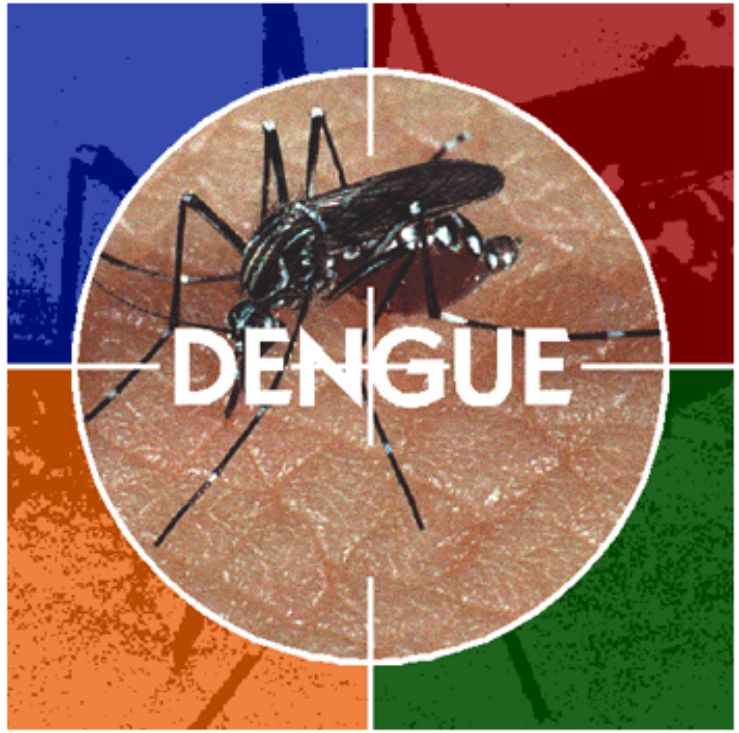

Dengue

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Objectives

- Virus
- Vector
- Epidemiology
- Pathogenesis
- Diagnosis
- Management
- Prevention

1. Virus

Dengue virus

- Family: Flaviviridae
- Genus: Flavivirus
- Species: Dengue virus

- Transmitted by mosquitoes
- Composed of single-stranded RNA
- Has 4 serotypes (DEN-1, 2, 3, 4)

Dengue virus

- Arbovirus
  - Arthropod borne -Mosquito

- It is a pathogen that causes
  - Dengue fever (DF)
  - Dengue hemorrhagic fever (DHF)
    - Can lead to Dengue shock syndrome (DSS)

Dengue Viruses

- Each serotype provides specific lifetime immunity, and short-term cross-immunity
- All serotypes can cause severe and fatal disease
- Genetic variation within serotypes
- Some genetic variants within each serotype appear to be more virulent or have greater epidemic potential

2. Vector

Aedes aegypti/ albopictus

- Dengue transmitted by infected female mosquito
- Primarily a daytime feeder
- Lives in and around human habitation
- Lays eggs and produces larvae preferentially in artificial containers

Aedes aegypti Mosquito

Aedes aegypti Breeding Sites

Life cycle

blood meal by female mosquito

Extrinsic incubation 8-10 days

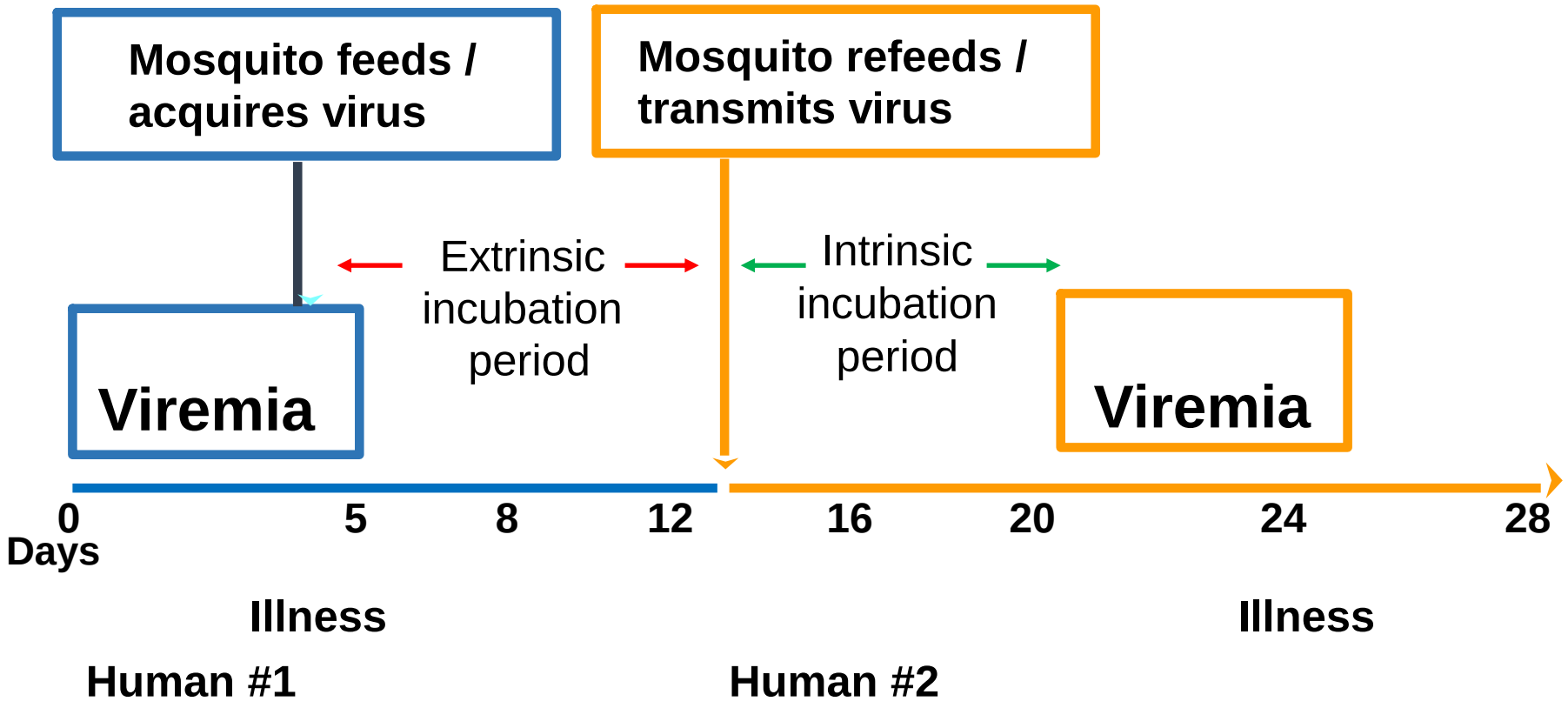
Replicate in mosquito (in salivary glands)

Introduction of virus to human during a blood meal with saliva

Intrinsic incubation 4-6 days

Replicate in human

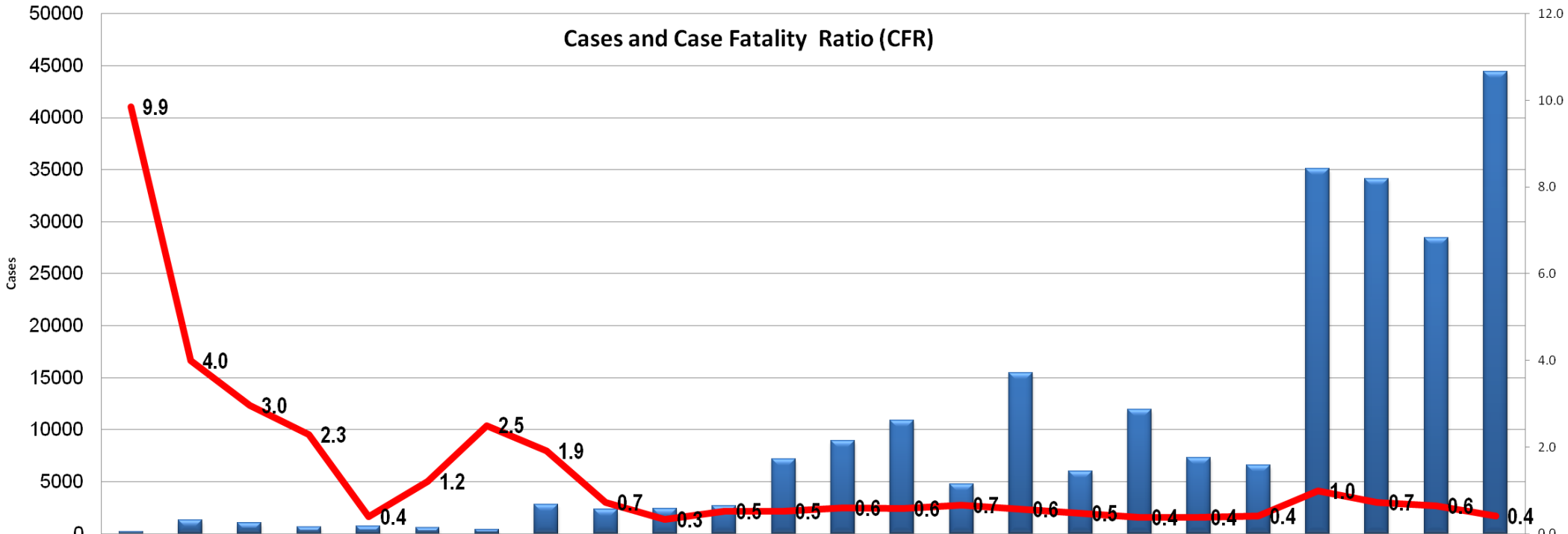
Transmission of Dengue Virus



4. Epidemiology

Dengue in SL

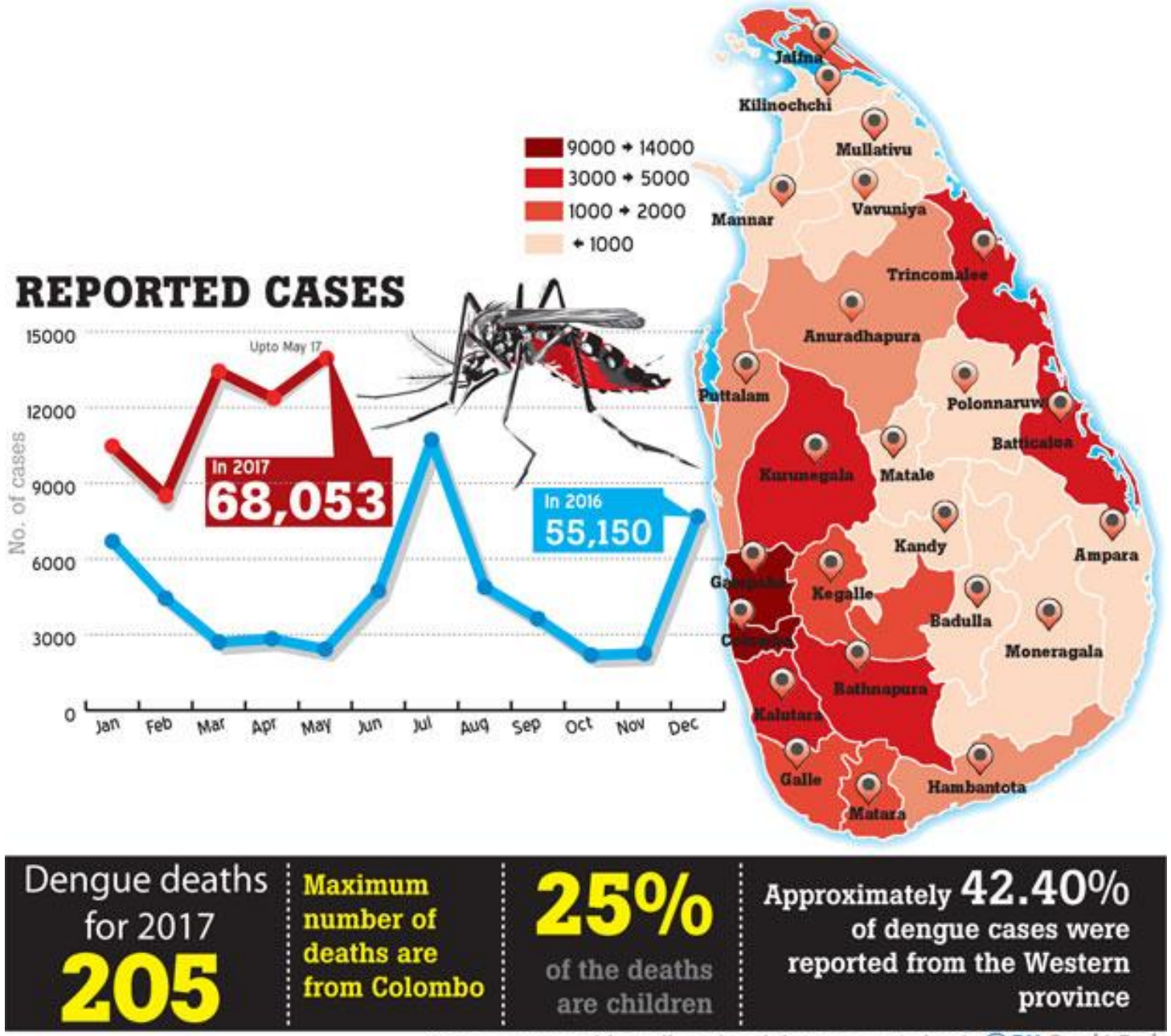
- First serologically confirmed case was reported in 1962
- First documented dengue outbreak occurred in 1965- 1966
- There had being continued dengue transmission with all four dengue serotypes circulating during 1970s and 1980s with periodic epidemics
- Sri Lanka experienced first epidemic of Dengue Haemorrhagic fever / Dengue Shock Syndrome caused by DEN- 3 sero type in 1989 – 1990
- Since then regular epidemics have occurred mainly with DEN 3 and DEN 2



	1989	1990	1991	1992	1993	1994	1995	1996	1997	1998	1999	2000	2001	2002	2003	2004	2005	2006	2007	2008	2009	2010	2011	2012
Cases	203	1350	1048	656	756	582	440	2824	2332	2419	2668	7213	8931	10933	4805	15463	5994	11980	7332	6607	35095	34105	28473	44456
CFR	9.9	4.0	3.0	2.3	0.4	1.2	2.5	1.9	0.7	0.3	0.5	0.5	0.6	0.6	0.7	0.6	0.5	0.4	0.4	0.4	1.0	0.7	0.6	0.4

Dengue in SL- recent epidemic 2017

- During the last 12 month of the year 2017, 185,969 suspected dengue cases have been reported to the Epidemiology Unit from all over the island.
- This was three times higher than 2016.
- The dengue outbreak followed heavy rains and subsequent flooding and landslides
- Approximately 41.53% of dengue cases were reported from the Western province.



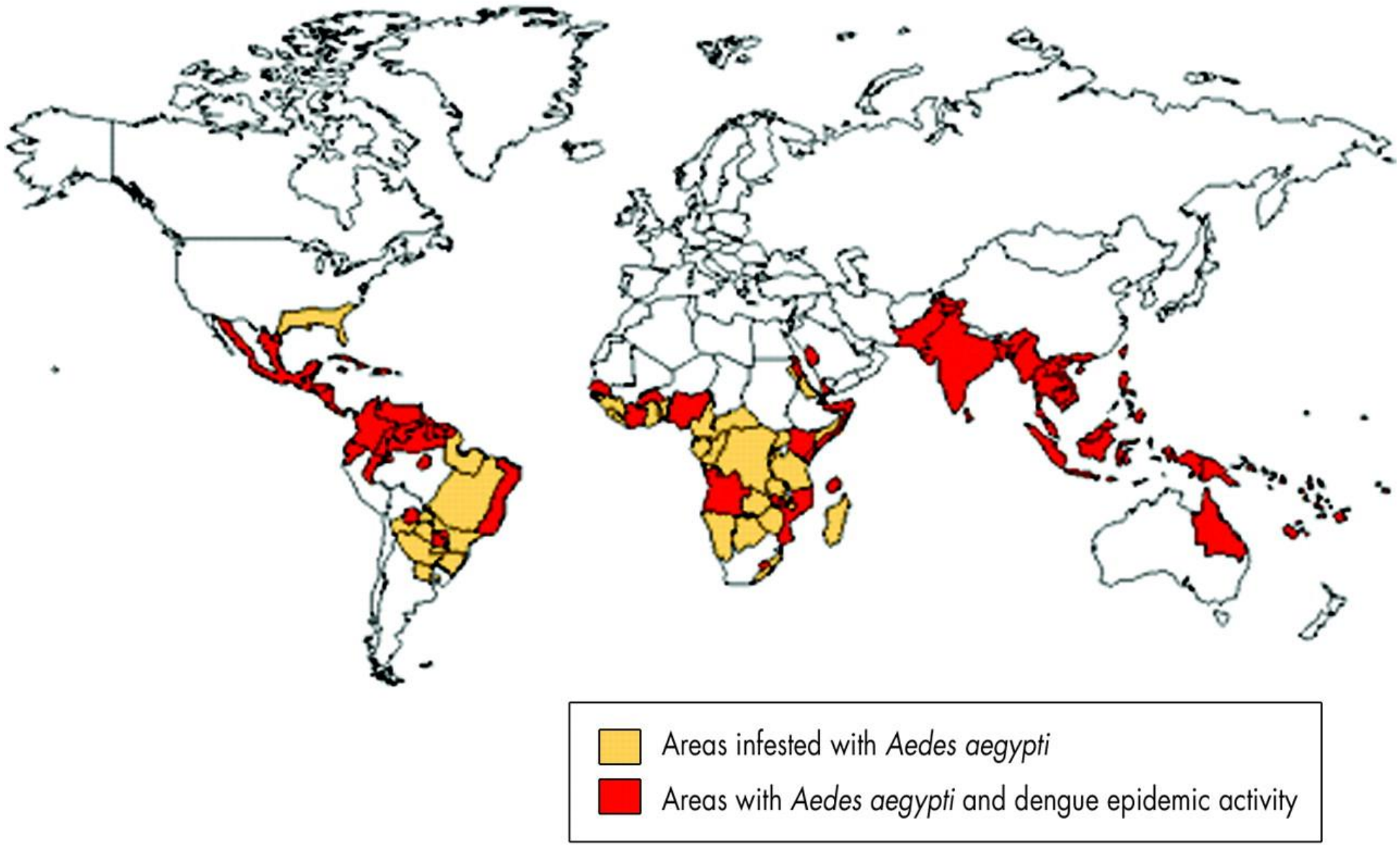
Dengue deaths for 2017
205

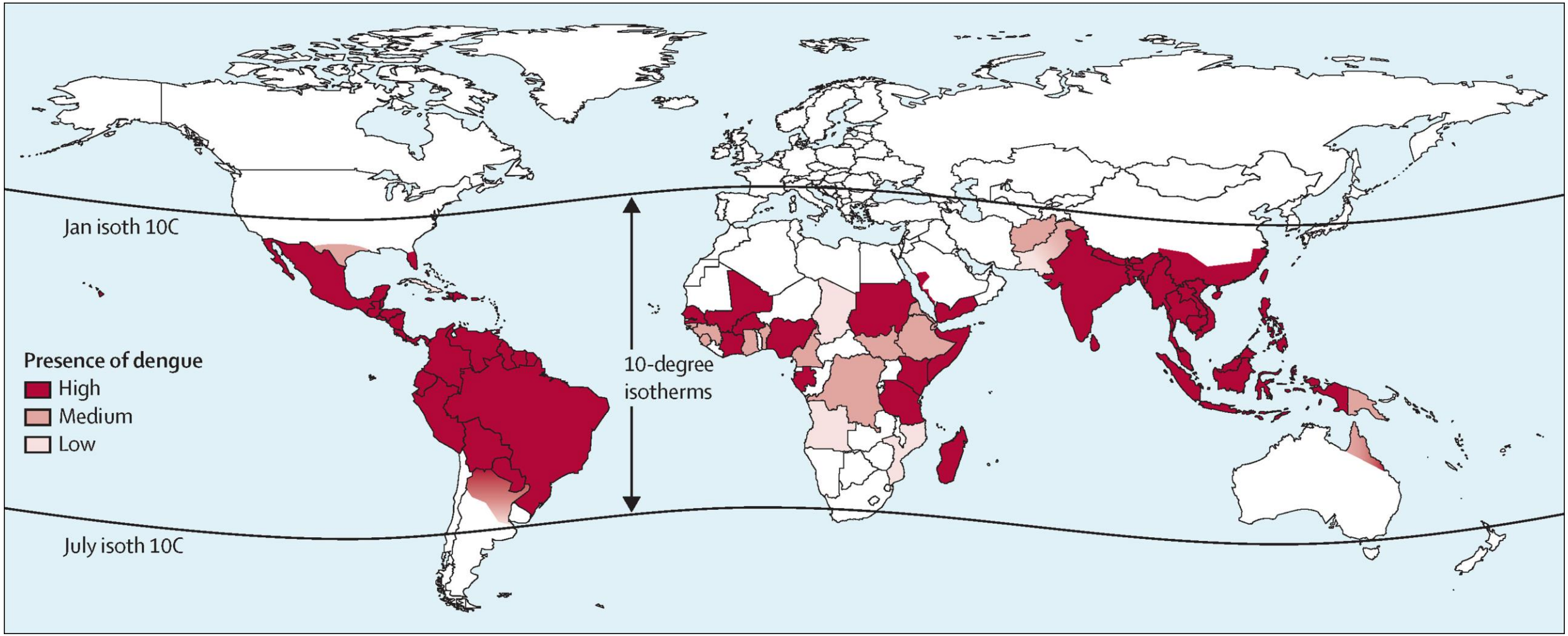
Maximum number of deaths are from Colombo

25%
of the deaths are children

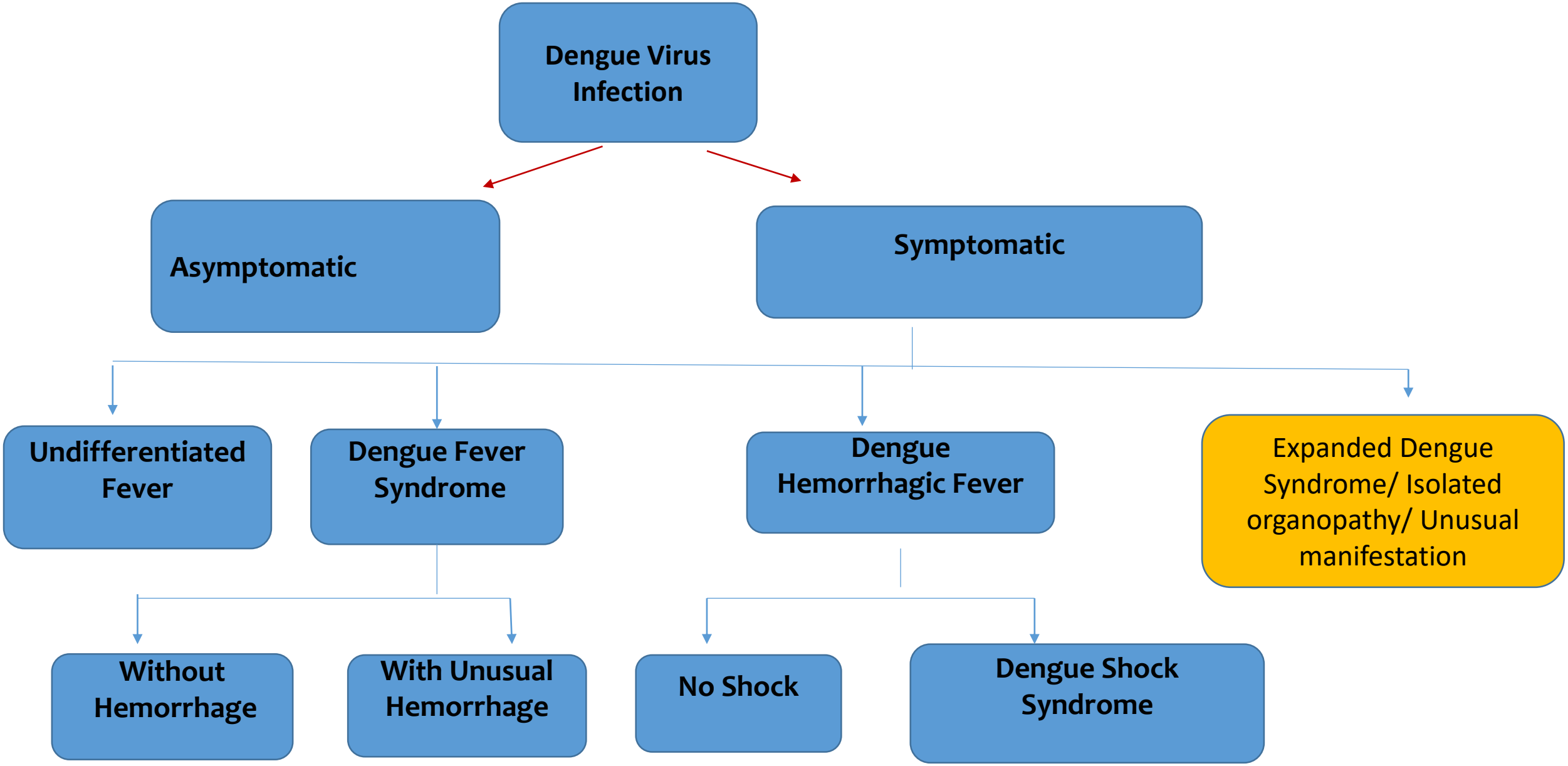
Approximately **42.40%** of dengue cases were reported from the Western province

Source: www.epid.gov.lk national dengue control unit ©DM GraphicDesk


Areas infested with *Aedes aegypti*
Areas with *Aedes aegypti* and dengue epidemic activity


Jan isoth 10C
Presence of dengue
High
Medium
Low
10-degree
isotherms
July isoth 10C

5. Clinical presentation


Dengue Virus Infection
Asymptomatic
Symptomatic
Undifferentiated Fever
Dengue Fever Syndrome
Dengue Hemorrhagic Fever
Expanded Dengue Syndrome/ Isolated organopathy/ Unusual manifestation
Without Hemorrhage
With Unusual Hemorrhage
No Shock
Dengue Shock Syndrome

Undifferentiated fever

- Usually follows a primary infection but may also occur during a secondary infection
- Clinically it is indistinguishable from other viral infections

Dengue fever

- May occur either during primary or secondary infection
- The onset is sudden with high fever, severe headache, arthralgia, myalgia, anorexia, abdominal discomfort, and sometimes a macular papular rash and flushing
- The fever may be biphasic and tends to last for 2–7 days.
- Although, haemorrhagic manifestations are uncommon in dengue fever, petechiae/pupura, gastrointestinal bleeding, epistaxis, and gingival bleeding have been observed in some individuals

WHO case definition for DF:

Acute Febrile illness with 2 or > of the following:

- Headache
- Retro-orbital pain
- Myalgia
- Arthralgia
- Rash
- Hemorrhagic manifestations
- Leukopenia
- Hepatomegaly common

DHF: WHO Criteria for diagnosis

- Often occurs with defervescence of fever
- All of the following must be present:
  - Fever
  - Hemorrhagic tendencies:
    - +ve tourniquet test
    - Petichiae, ecchymosis or purpura
    - Bleeding from other sites
  - Thrombocytopenia (<=100,000/cu mm)
  - Evidence of plasma leak
    - Rise in hematocrit > 20% above average
    - Drop in Hct after volume-replacement treatment
    - Pleural effusion/ascites/hypoproteinemia

DSS: WHO Criteria for diagnosis

All of the above + evidence of circulatory failure:

- Rapid, weak pulse
- Narrow pulse pressure < =20 mm hg
- Cold clammy skin
- Restlessness

Often present with abdominal pain; mistaken for acute abdominal emergency

EXPANDED DENGUE SYNDROME

- Organopathy in the setting of acute dengue virus infection

- Liver failure
- CNS dysfunction
- encephalopathy and seizures
- acute pure motor weakness
- myocardial dysfunction

- Mononeuropathies
- Polyneuropathies
- Guillain-Barré syndrome
- transverse myelitis
- myocarditis

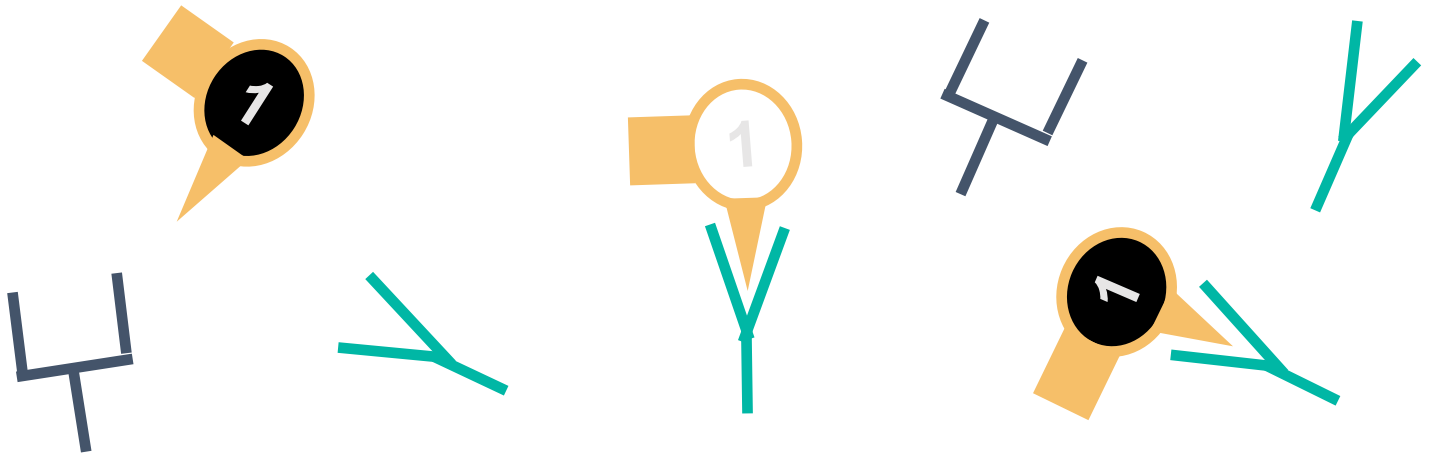
Grading of DV infection

DF/DHF	Grade	Symptoms	Lab
DF		**Fever with 2 or > of: headache/retro-orbital pain, myalgia, arthralgia**	**Leukopenia, occasionally thrombocytopenia, no evidence of plasma leak**
DHF	**I**	**Above + +ve tourniquet test**	**Platelets < 100,000, Hct rise > 20%**
DHF	**II**	**Above + spontaneous bleeding**	**,,**
DHF	**III/DSS**	**Above + s/o circulatory failure**	**,,**
DHF	**IV/DSS**	**Profound shock with undetectable BP and pulse**	**,,**
			Lab evidence of DV infection

Hypothesis on Pathogenesis of DHF

- Antibody-dependent enhancement
  - is the process in which certain strains of dengue virus, complexed with non-neutralizing antibodies, can enter a greater proportion of cells of the mononuclear lineage, thus increasing virus production

Homologous Antibodies neutralizes the virus



- Dengue 1 virus
- Neutralizing antibody to Dengue 1 virus
- Non-neutralizing antibody
- Complex formed by neutralizing antibody and virus

Pathogenesis of DHF

- In a subsequent infection, the pre-existing heterologous antibodies form complexes with the new infecting virus serotype, but do not neutralize the new virus

- Virus-antibody complexes taken up by monocytes
- Virion multiplication in human monocytes is promoted
- Activation of CD4+ and CD8+ lymphocytes → release of cytokines
- Complement system activated with depression of C3 & C5

Heterologous Antibodies Form Infectious Complexes

Dengue 2 virus

Non-neutralizing antibody to Dengue 1 virus

Complex formed by non-neutralizing antibody and virus

DHF: Pathophysiology

- Activation of complement → Increased vascular permeability →loss of plasma from vascular compartment → hemoconcentration & shock
- Disorder of haemostasis involving thrombocytopenia, vascular changes and coagulopathy
- Severe DHF with features of shock : DSS

INJECTED PHARYNX

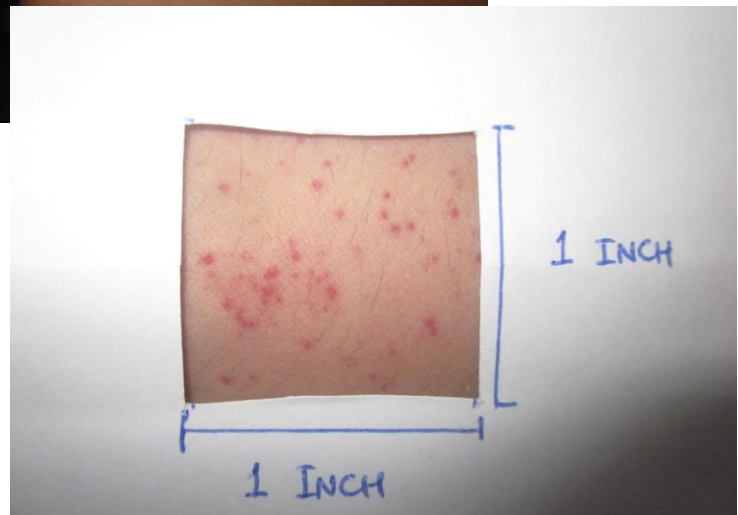
PETECHIAL RASH

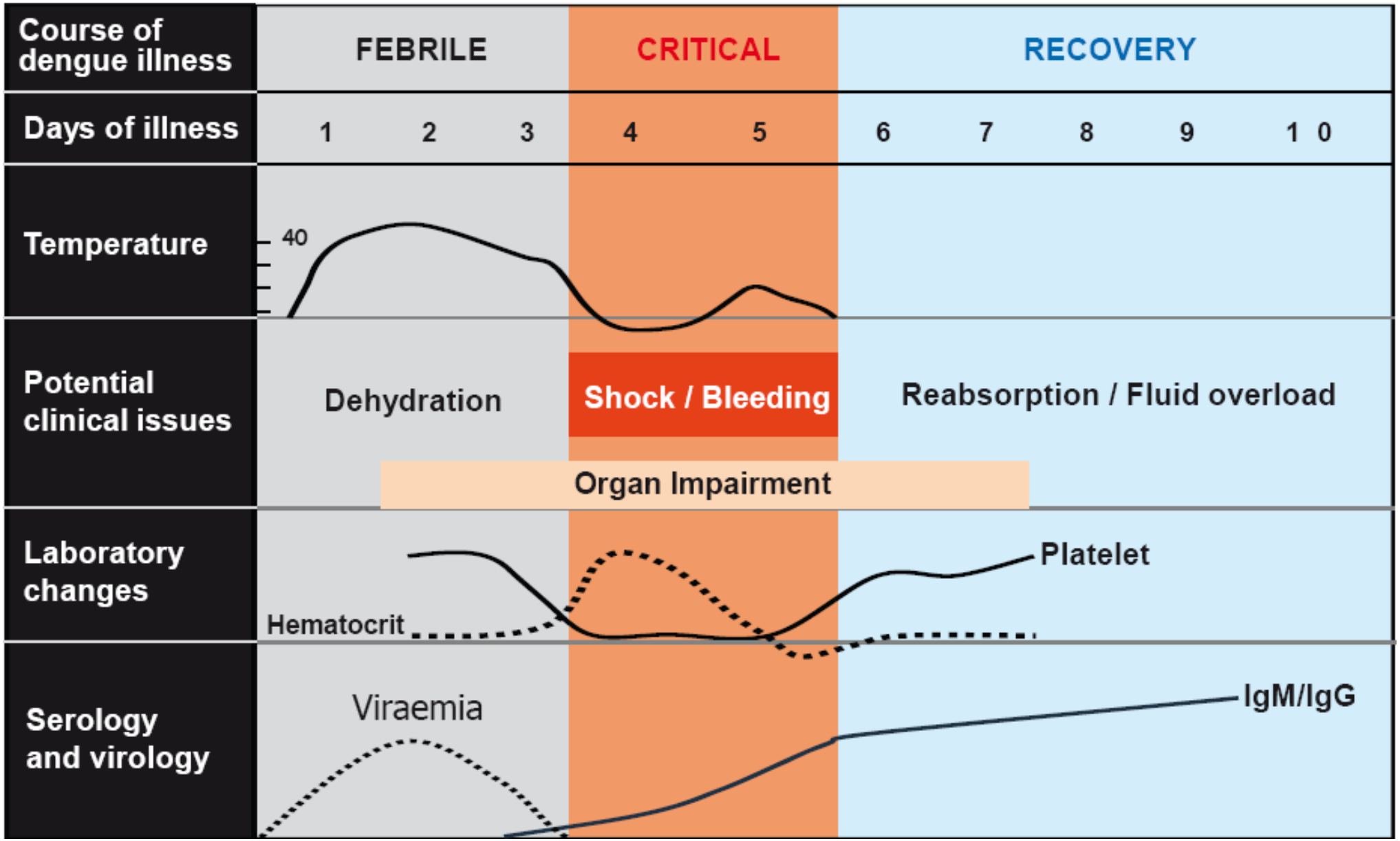
SUBMUCOSAL HEMORRHAGE

BRUISING

CAPILLARY REFILL TIME

TOURNIQUET TEST

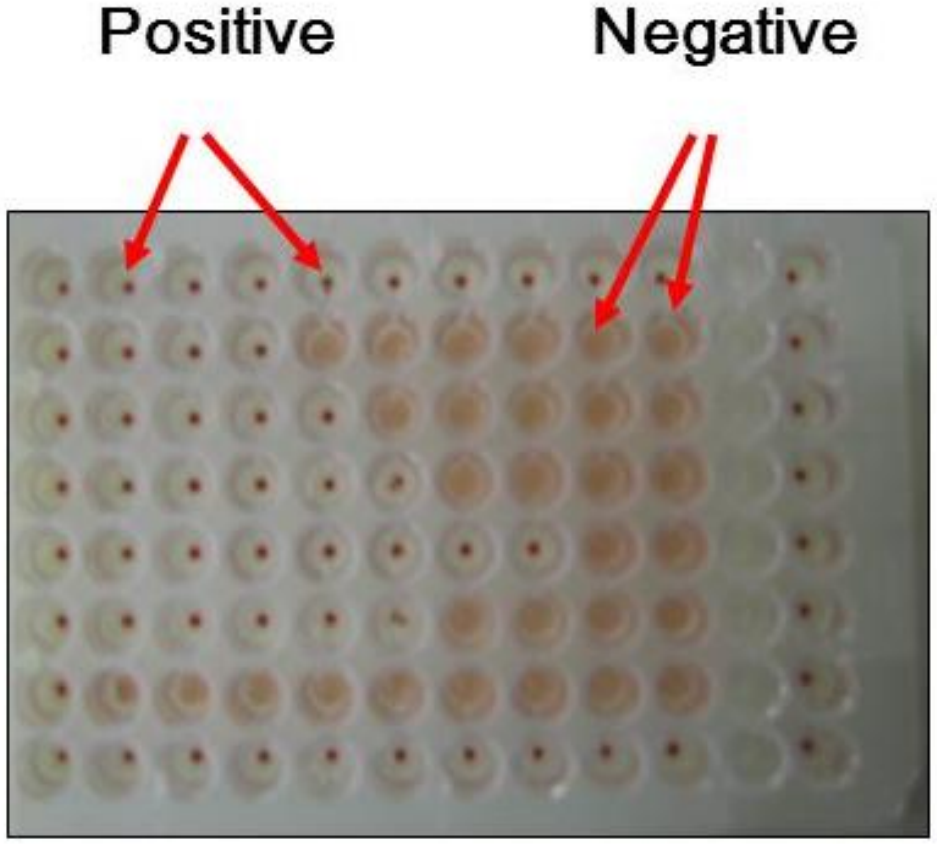



Course of dengue illness
FEBRILE
CRITICAL
RECOVERY
Days of illness
1
2
3
4
5
6
7
8
9
10
Temperature
40
Potential clinical issues
Dehydration
Shock / Bleeding
Reabsorption / Fluid overload
Organ Impairment
Laboratory changes
Platelet
Hematocrit
Serology and virology
Viraemia
IgM/IgG

6. Diagnosis

Methods

1. Serology
   - ELISA
   - Immuno-chromatography
   - HAI
   - CF
   - PRNT (plaque reduction neutralization technique)



Hemagglutination inhibition for detection of Dengue antibodies

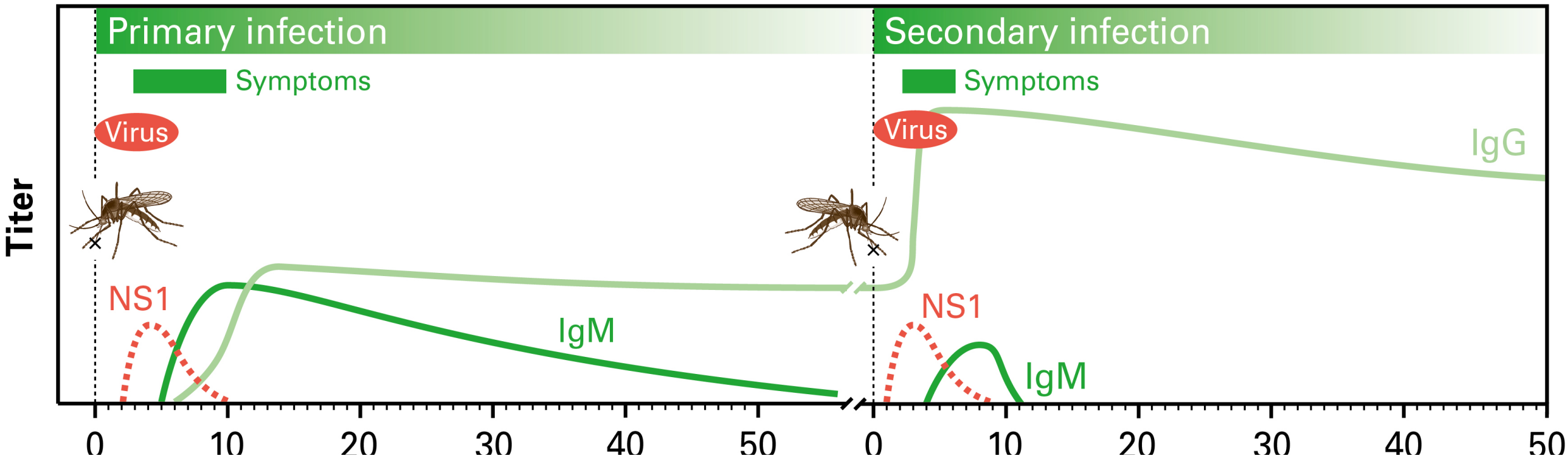
1. Serology

- IgM
  - Appear in 3-5 days
  - Peak at 2 weeks
  - Undetectable after 3 months
  - Evidence of recent infection
  - Venous blood/ serum sample should be collected after 5^{th} day
- IgG
  - Appear after 7 days in primary infection
  - Higher titre and earlier appearance in secondary infection
  - Needs paired serum to detect acute infection (> 4fold rise in titre)
    - First sample in acute stage < 7 days
    - Convalescent sample 10-14 days later
- Total Antibodies

2. Antigen detection

- ELISA/ Dot blot/ immunochromatograpy
- Detect NS1 antigen
- Samples – venous blood/ serum, liver/ lung (autopsy)
  - collected within first 5 days of fever

  (Positive up to 9 days in both primary and secondary infection)
- Early diagnosis
- NS1 antigen ELISA – sensitivity 86% and specificity 100%
- Less sensitive in secondary infections
  - Interaction with IgG in previous infection
- Newer assays to type the virus


Primary infection
Secondary infection
Symptoms
Symptoms
Virus
Virus
IgG
NS1
IgM
NS1
IgM
Titer
0
10
20
30
40
50
0
10
20
30
40
50

3. Genome detection

- Real time RT- PCR
- Used to quantitate viral RNA and using primer pairs and probes that are specific to each dengue serotype
- Rapid turnaround time
- Sample collected within first 5 days
- Transported in ice

4. Virus isolation

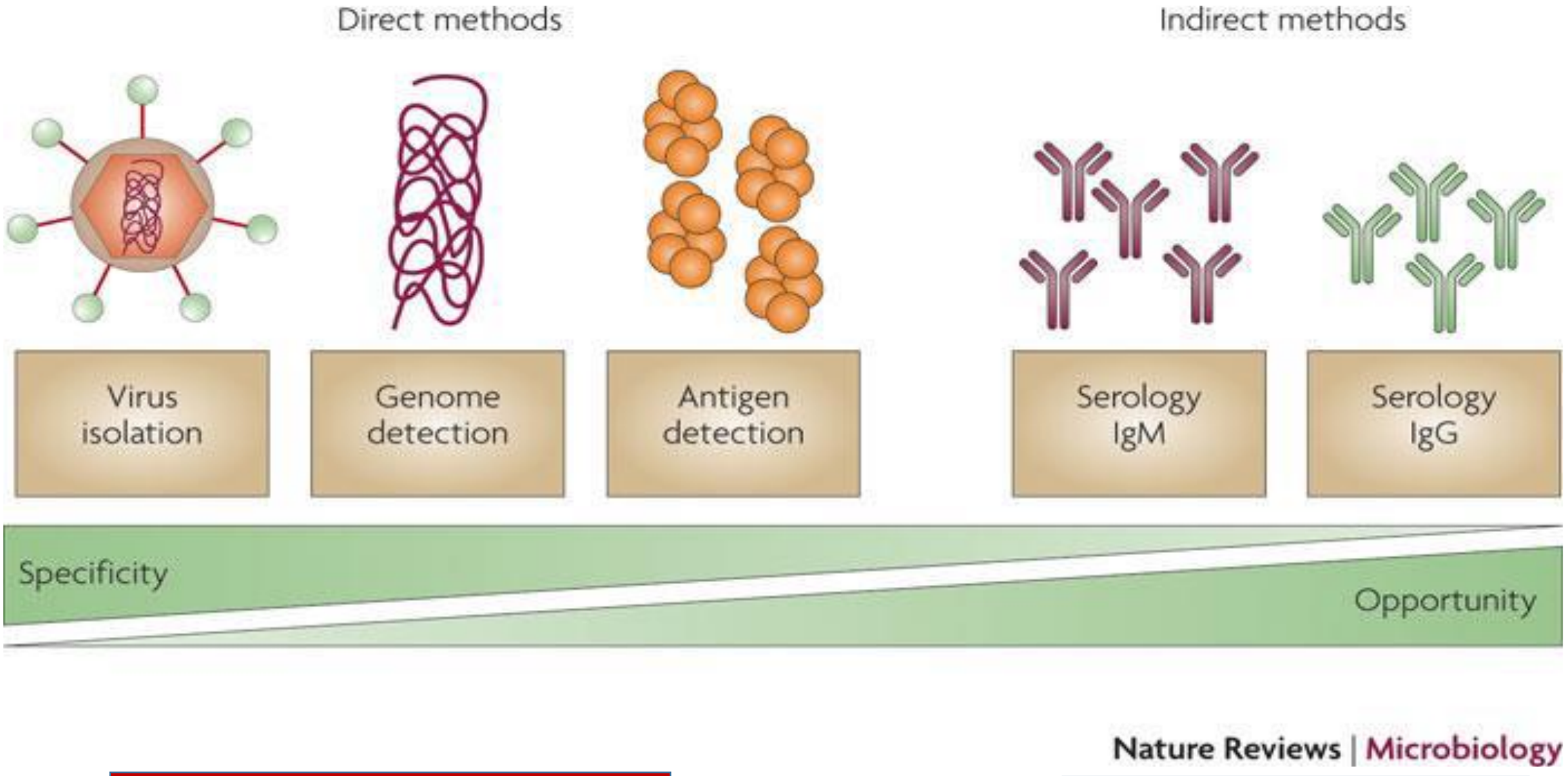
- Used for epidemiological studies and research
  - Inoculation into mosquitoes (adult/larvae)
  - Inoculation into mosquito cell lines (C6/36)
  - Mammalian cell lines (vero)

- Slow results (Days) vs. PCR (Hours)

- Samples- serum, plasma, CSF, autopsy

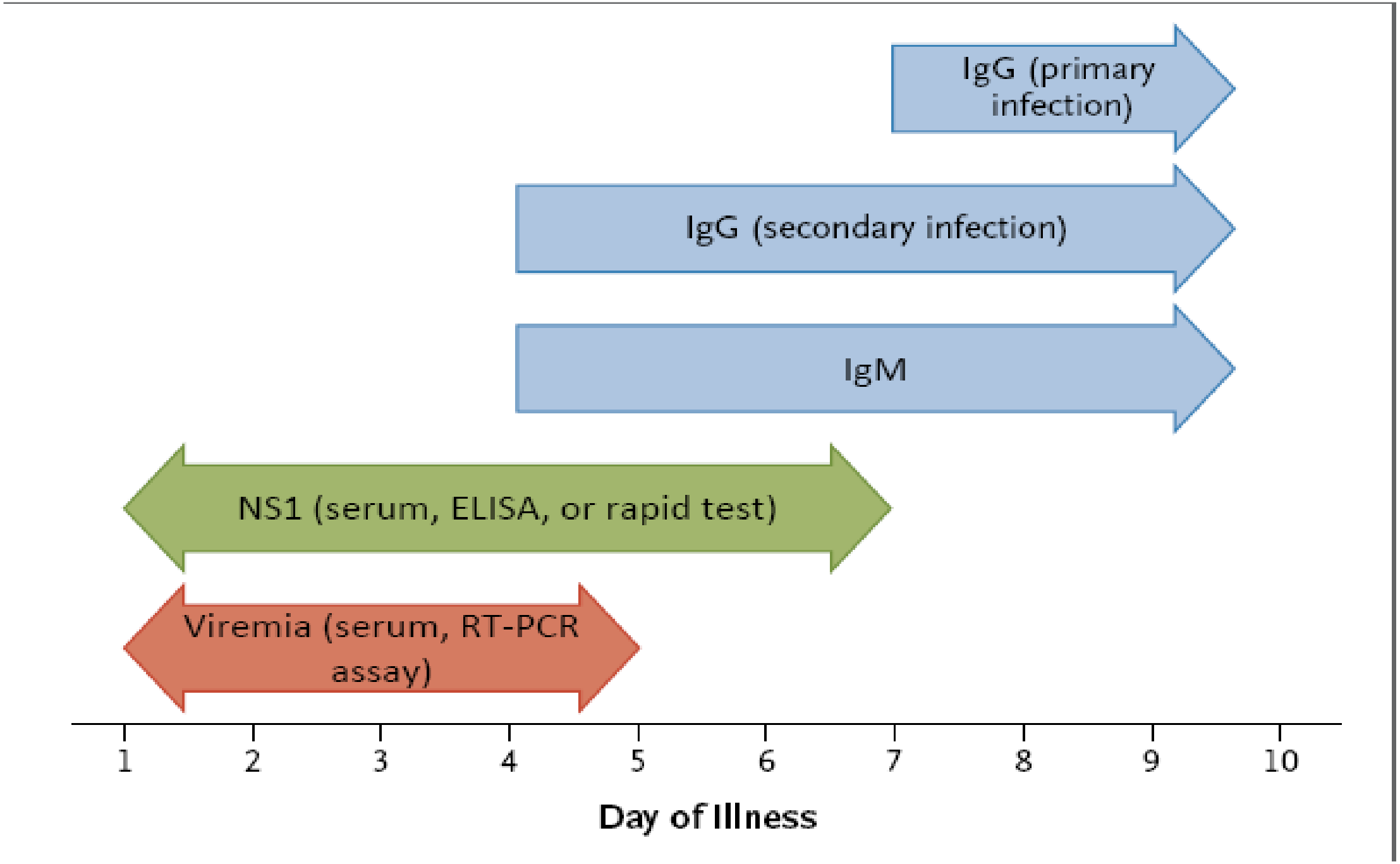
  - Sample collected within first 5 days

- Transported in ice (at +4-8 ^{0}C)



Less than five days

More than five days


IgG (primary infection)
IgG (secondary infection)
IgM
NS1 (serum, ELISA, or rapid test)
Viremia (serum, RT-PCR assay)
1
2
3
4
5
6
7
8
9
10
Day of Illness

Other investigations

- FBC
  - WBC
  - Platelet
- Hematocrit/ PCV
- USS

Thank you