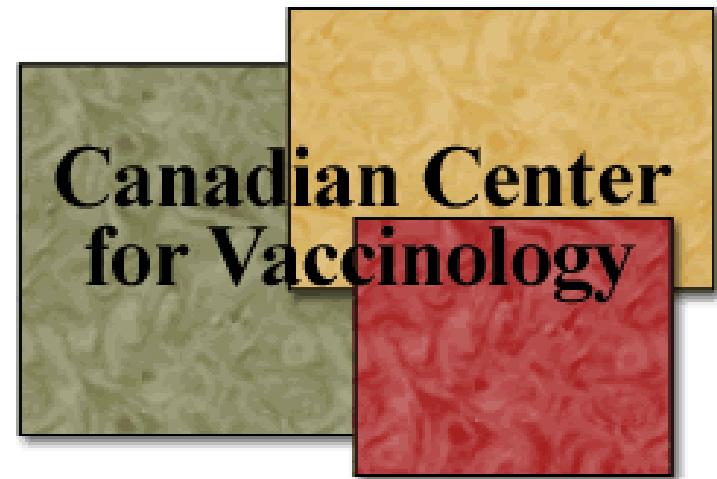


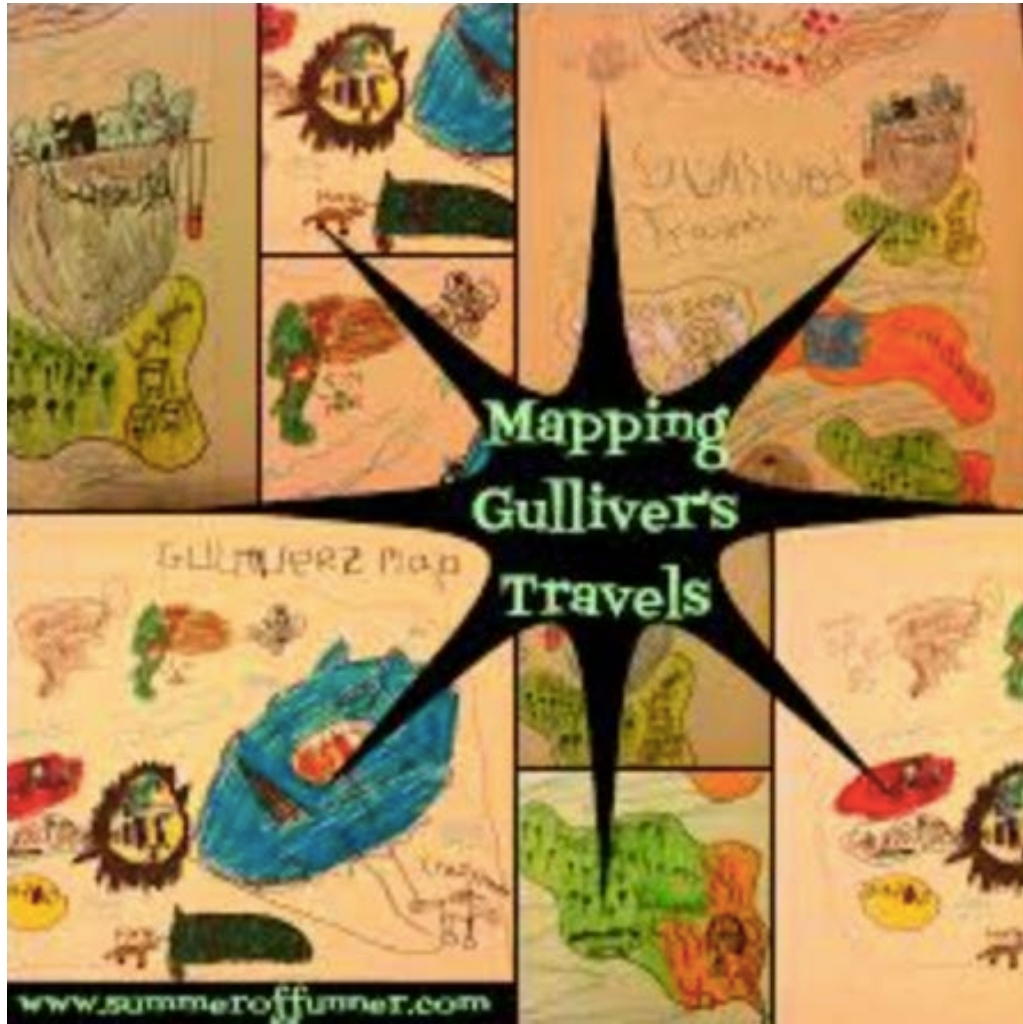
Pathogenesis and vaccinology of dengue

Robert Anderson, Dalhousie University
& Canadian Center for Vaccinology
Halifax, Nova Scotia, Canada

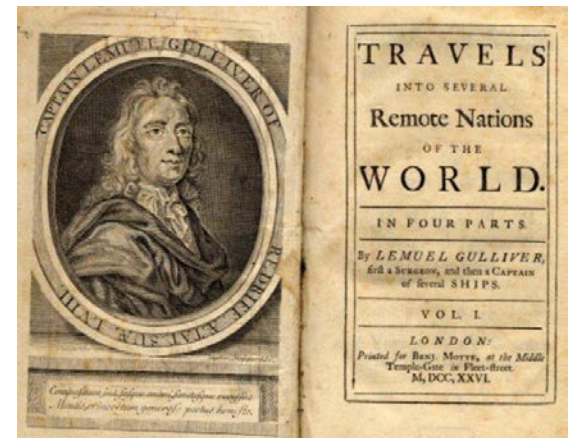
TropMed, Mahidol University
August 28 2018



Gulliver's Travels



Gulliver's Travels
Jonathan Swift (1726)



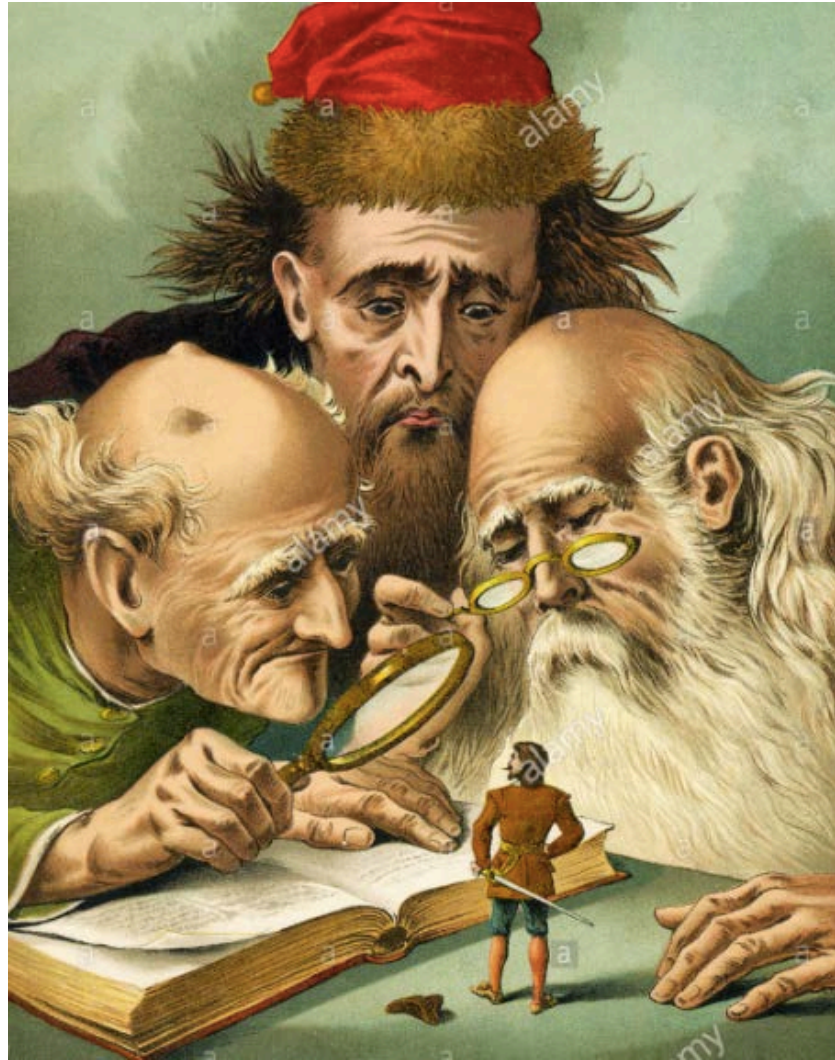
Some Gulliverian perspectives on global health



Some Gulliverian perspectives on global health



Some Gulliverian perspectives on global health



Dengue Research

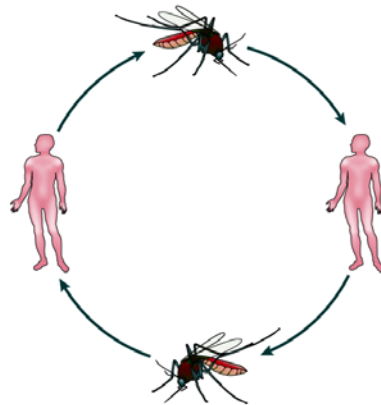
Armed Forces Research Institute of Medical Sciences
(AFRIMS)
Bangkok, Thailand

AFRIMS, Bangkok, Thailand



Dengue

- 390 million cases worldwide each year
- Fever to severe hemorrhaging and shock
- No effective vaccine
- “Tropical” disease on the move



Dengue virus-associated disease

- Dengue fever (DF)
- Dengue hemorrhagic fever (DHF), dengue shock syndrome (DSS)
- Vascular endothelial permeability, thrombocytopenia
- Antibody-dependent enhancement (ADE) as risk factor



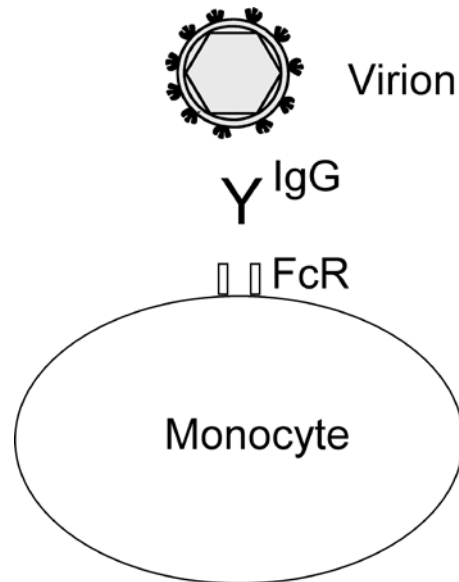
Dengue research questions

how does dengue virus cause disease?

how can it be stopped?

Dengue virus uses antibody to enhance infection

- Antibody dependent enhancement (ADE)

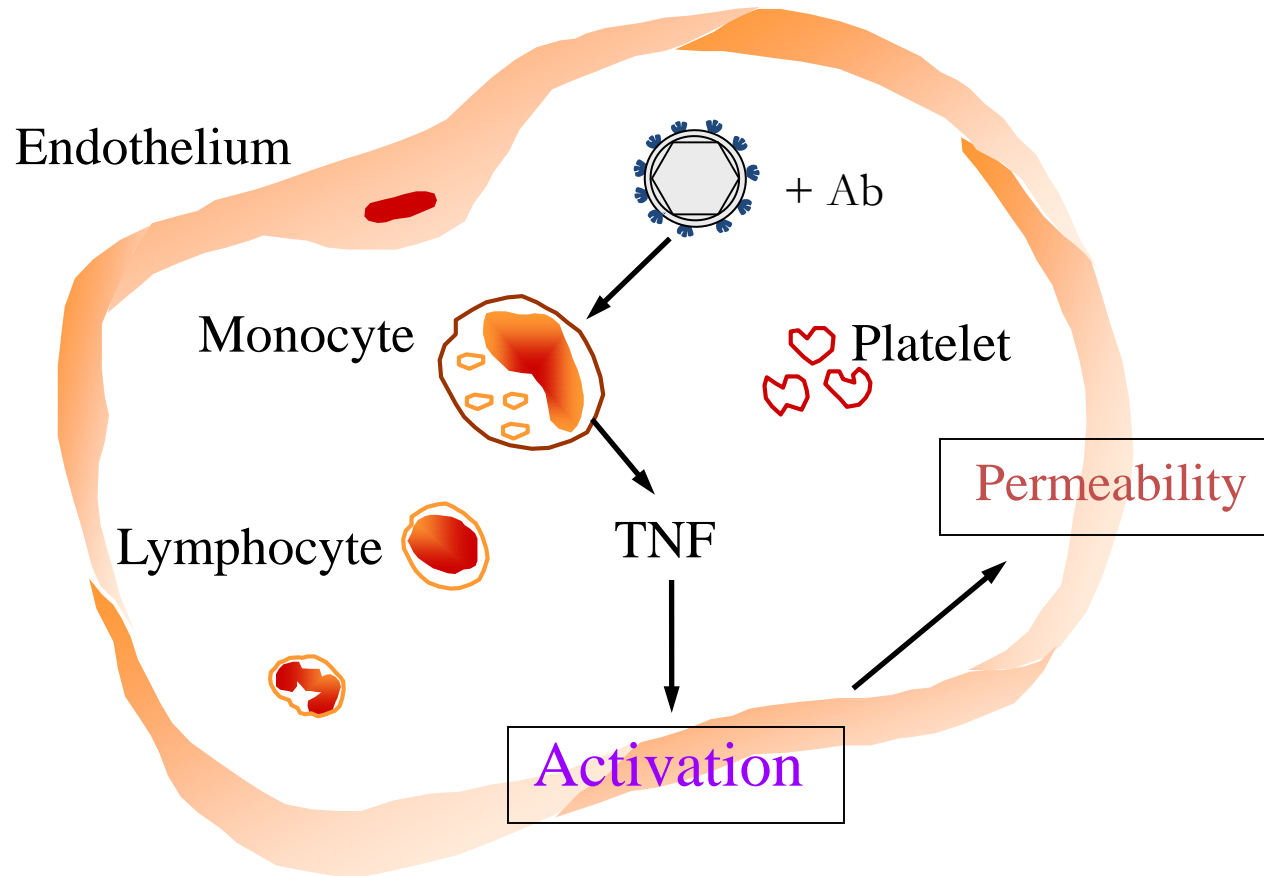


Prior immunity enhances risk for severe disease

4 dengue serotypes with poor neutralizing but strong enhancing cross-reactivity

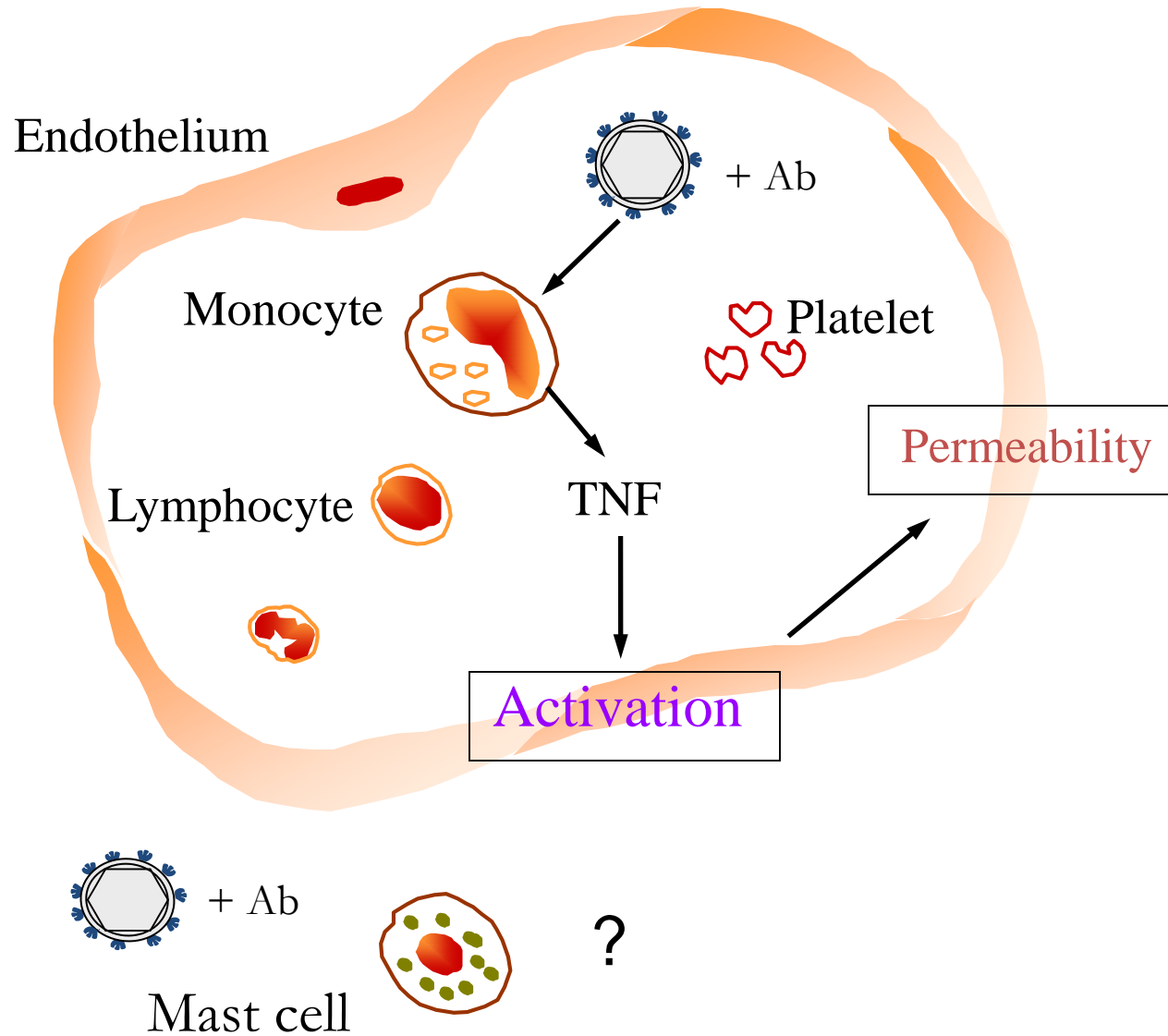
“Virus Factory”

Intravascular responses in dengue pathogenesis



Extravascular cell targets for dengue-induced endothelial perturbation ?

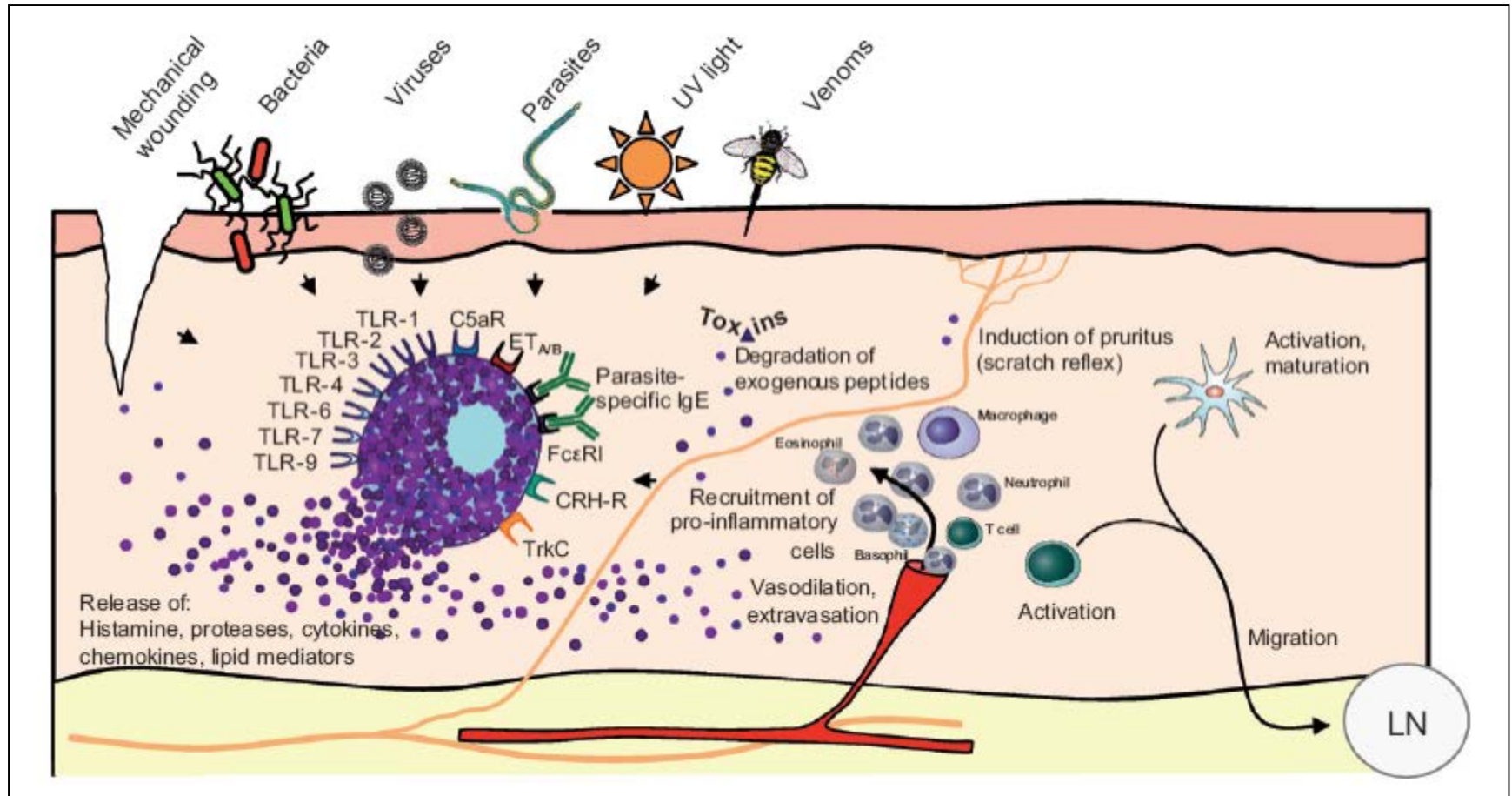
Intravascular responses in dengue pathogenesis



Dengue Life in Canada



Immune functions of human mast cells



Metz *et al.* (2008) Immunobiology

Evidence for mast cells in dengue disease

- Mast cell-like cells in dengue autopsy specimens
(Bhamarapravati et al 1967)
- Increased urinary histamine levels in DHF patients
(Tuchinda et al., 1977)
- Increased blood histamine levels in DHF patients
(Phan et al., 2000)
- Increased mast cell VEGF and proteases in DSS patients
(Furuta et al., 2012)

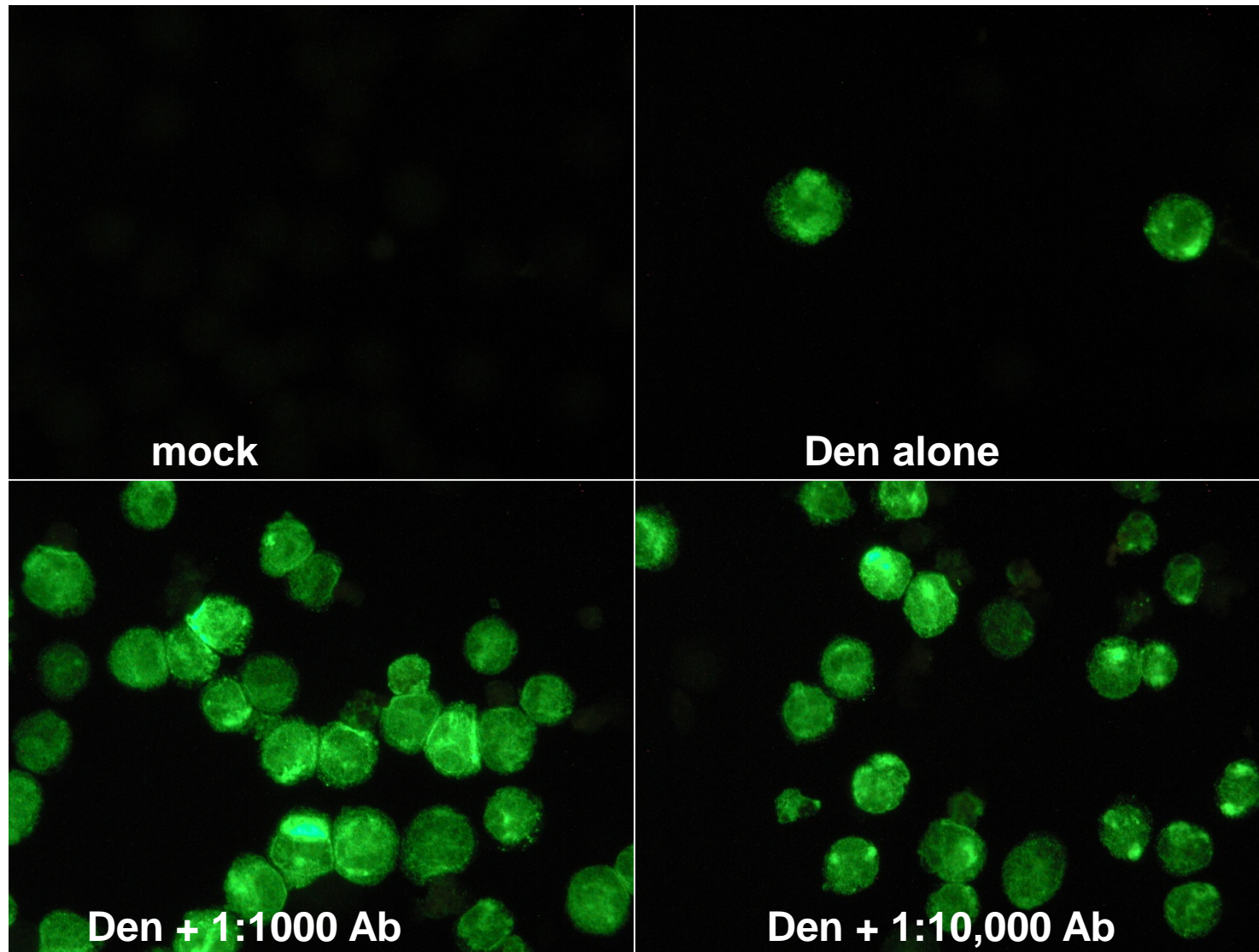
Mast cells are susceptible to dengue infection, cytokine/chemokine release and apoptosis

Mast cell sources:

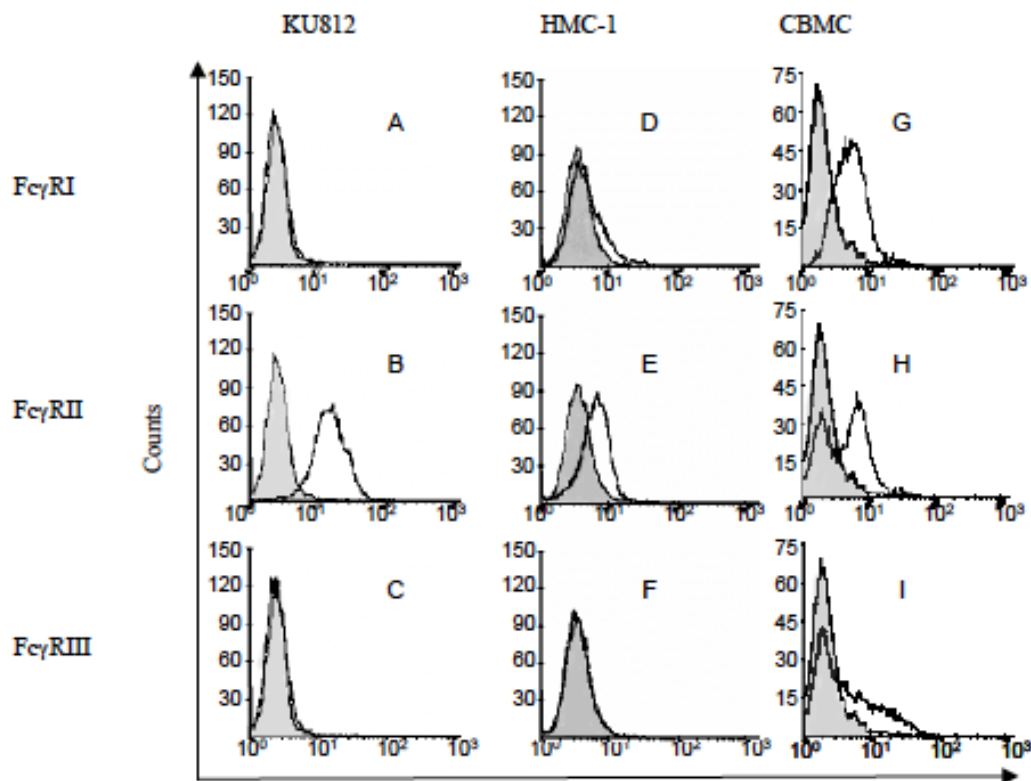
- KU812 (human mast cell/basophil precursor cell line)
- HMC-1 (human mast cell line)
- cord blood derived mast cells (CBMCs)

(King et al, J Virol 2000, 2002; Brown et al, J Leuk Biol 2006, 2009; J Virol 2011; PLoS One 2012)

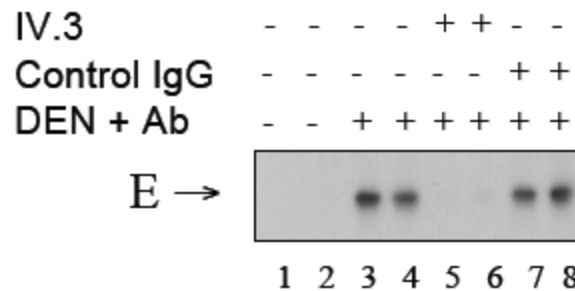
Antibody-enhanced dengue virus infection of KU812 cells



Antibody-enhanced dengue infection of human mast cells is mediated by Fc γ RII



Virus-cell binding assay



What are the consequences of antibody-enhanced dengue virus infection of mast cells?

Cytokine production, eg. TNF

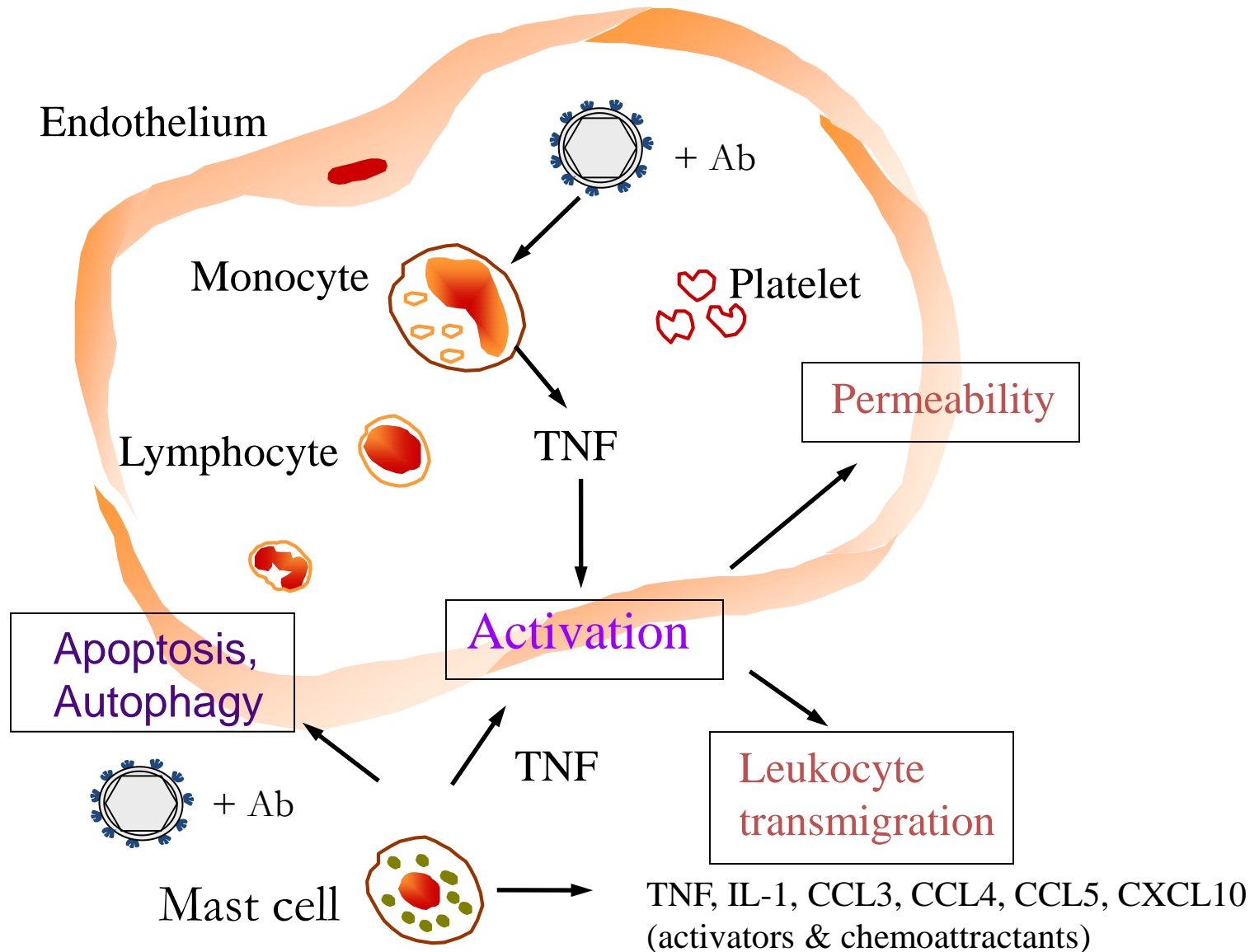
Apoptosis

Autophagy

Chemokine production, eg. CCL4, CCL5, CCL10

Degranulation

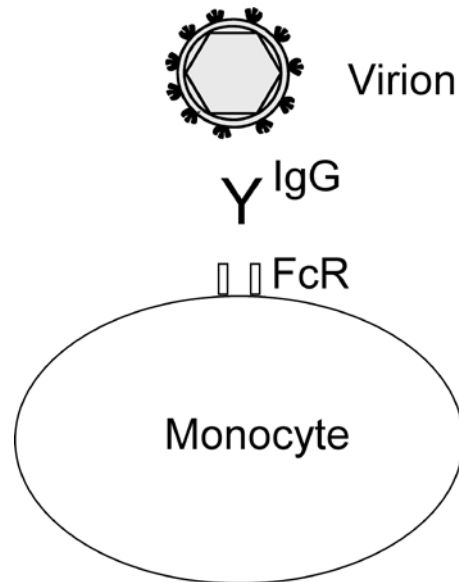
Intra & extravascular responses in dengue pathogenesis



Towards a dengue vaccine

Dengue vaccine – why so elusive?

- Antibody dependent enhancement (ADE)



Prior immunity enhances risk for severe disease

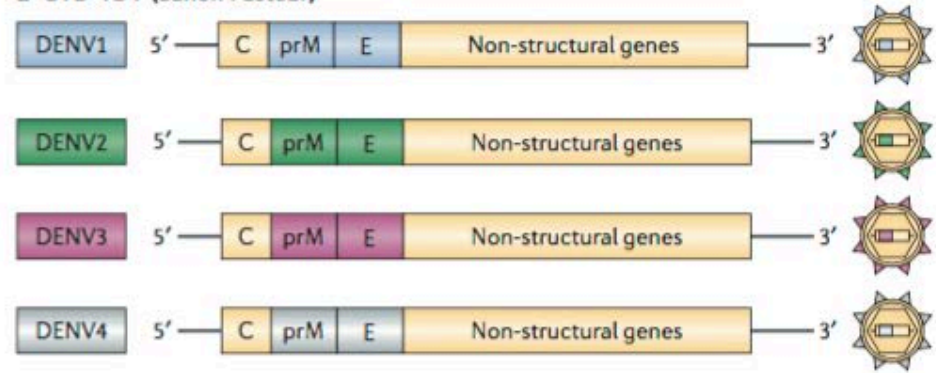
4 dengue serotypes with poor neutralizing but strong enhancing cross-reactivity

“Virus Factory”

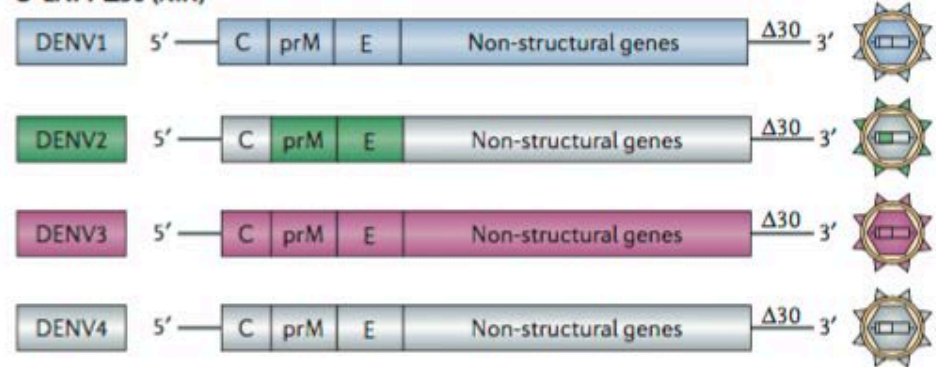
Current dengue vaccine trials

Screaton et al [2015]
Nat Rev Imm 15, 745

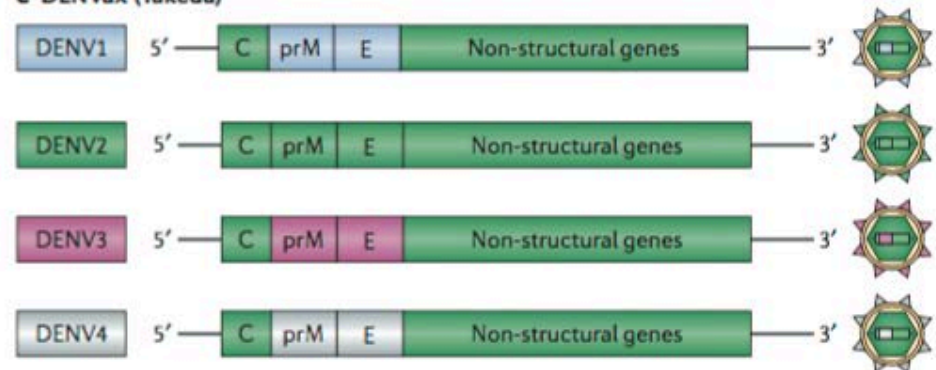
a CYD-TDV (Sanofi Pasteur)



b LATV $\Delta 30$ (NIH)



c DENVax (Takeda)



Current dengue vaccine trials

Dengue vaccine found to worsen disease symptoms



By **Ben Tinker**, CNN

🕒 Updated 11:31 AM ET, Tue December 5, 2017

Story highlights

The Philippines has halted a massive dengue fever vaccination effort

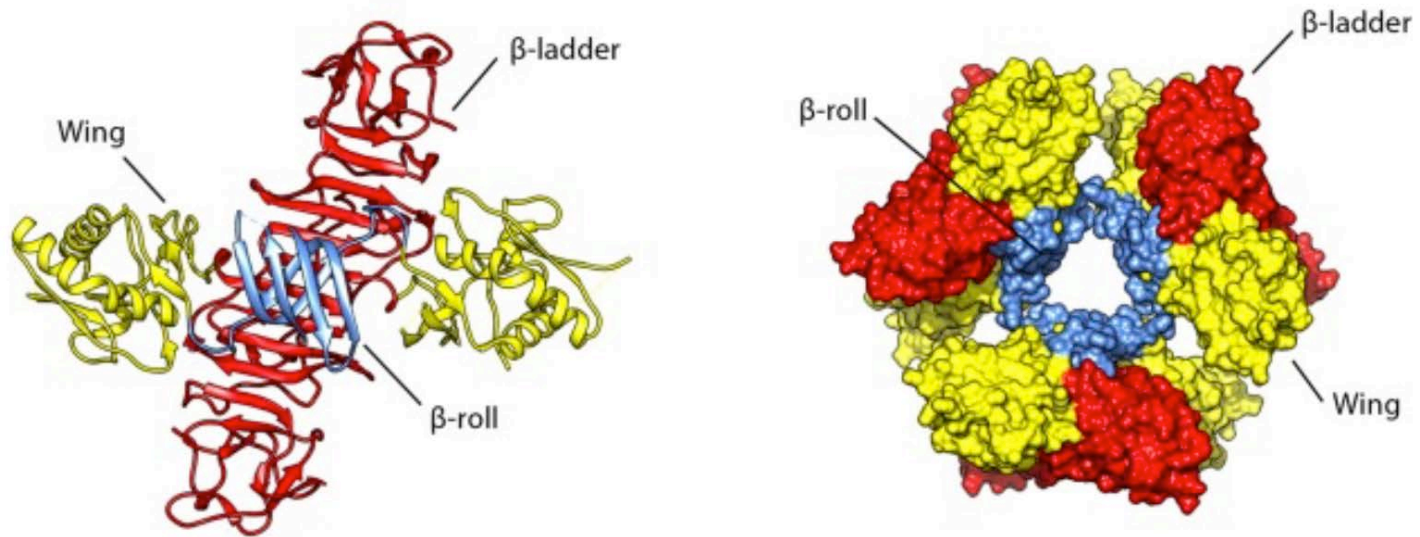
The country has launched an investigation into the approval of Dengvaxia

(CNN) — The Philippines has suspended its large-scale dengue vaccination effort amid the surprising results of a new study conducted by the vaccine's manufacturer.

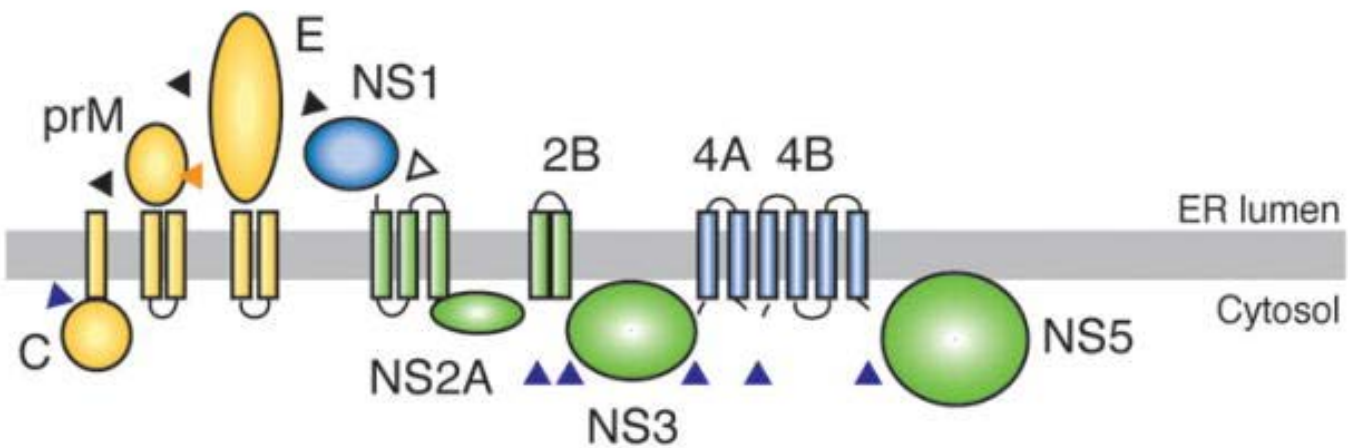
On Wednesday, French pharmaceutical giant Sanofi Pasteur published clinical trial data showing that its dengue vaccine, Dengvaxia, could have unintended consequences in patients who had never been infected with the mosquito-borne virus.

"The analysis confirmed that Dengvaxia provides persistent (protective) benefit against dengue fever in those who had prior infection," [Sanofi said in a statement](#). "For those not previously infected by dengue virus, however, the analysis found that in the longer term, more cases of severe disease could occur following vaccination upon a subsequent dengue infection."

Enter the dengue NS1 protein....



Scaturro et al [2015]
PLoS Path 11, e1005277



National Cheng Kung University - Medical College, Tainan, Taiwan



National Cheng Kung University - Dept Microbiology & Immunology



National Cheng Kung University (NCKU) Collaboration

MOU signed 2008

Research collaboration on dengue

22 peer-reviewed publications since 2008

Towards an NS1 vaccine for dengue

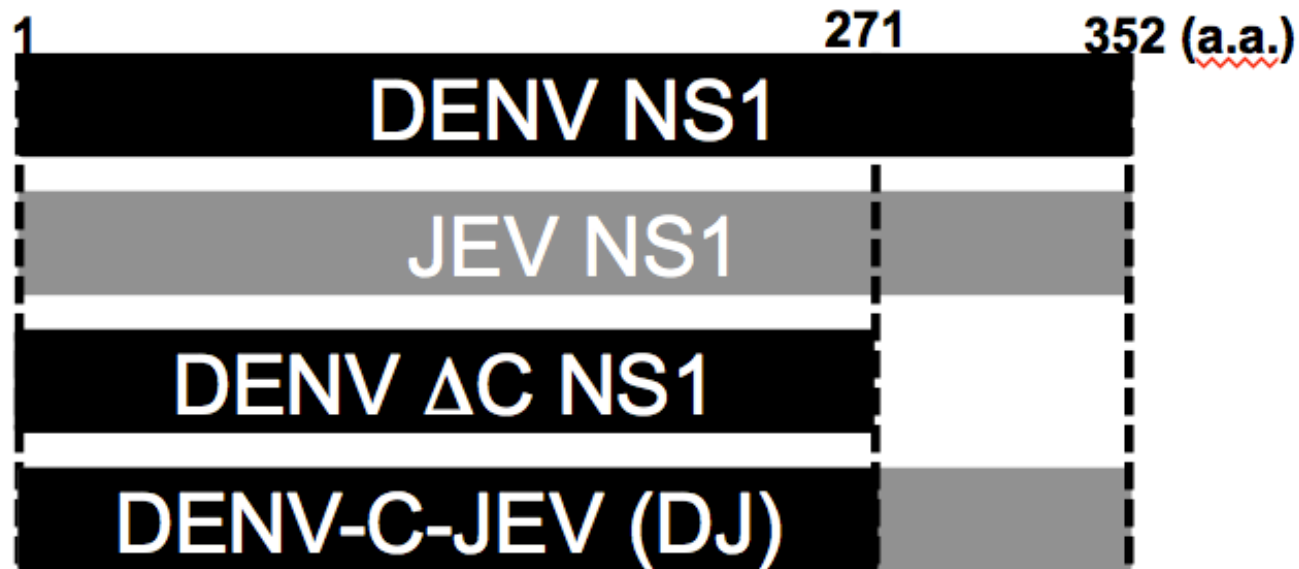
Pros

- NS1 Ab is protective
- highly conserved among 4 dengue serotypes
- target for Abs (CDC, ADCC) and CTLs
- no risk of inducing enhancing antibodies (ADE)

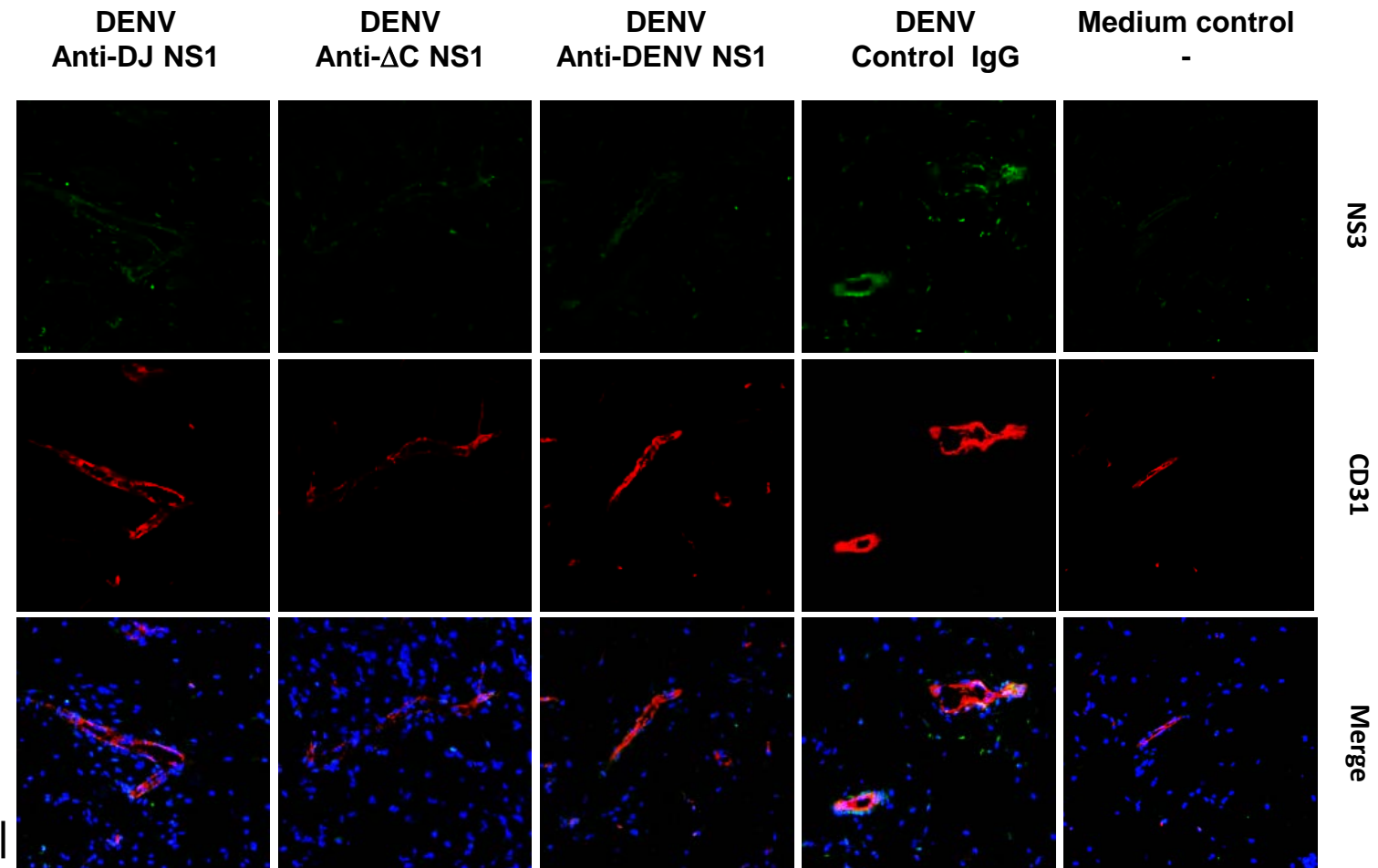
Cons

- C-terminal sequence cross-reactive with host cell proteins (eg. platelets, endothelial cells, coag factors)

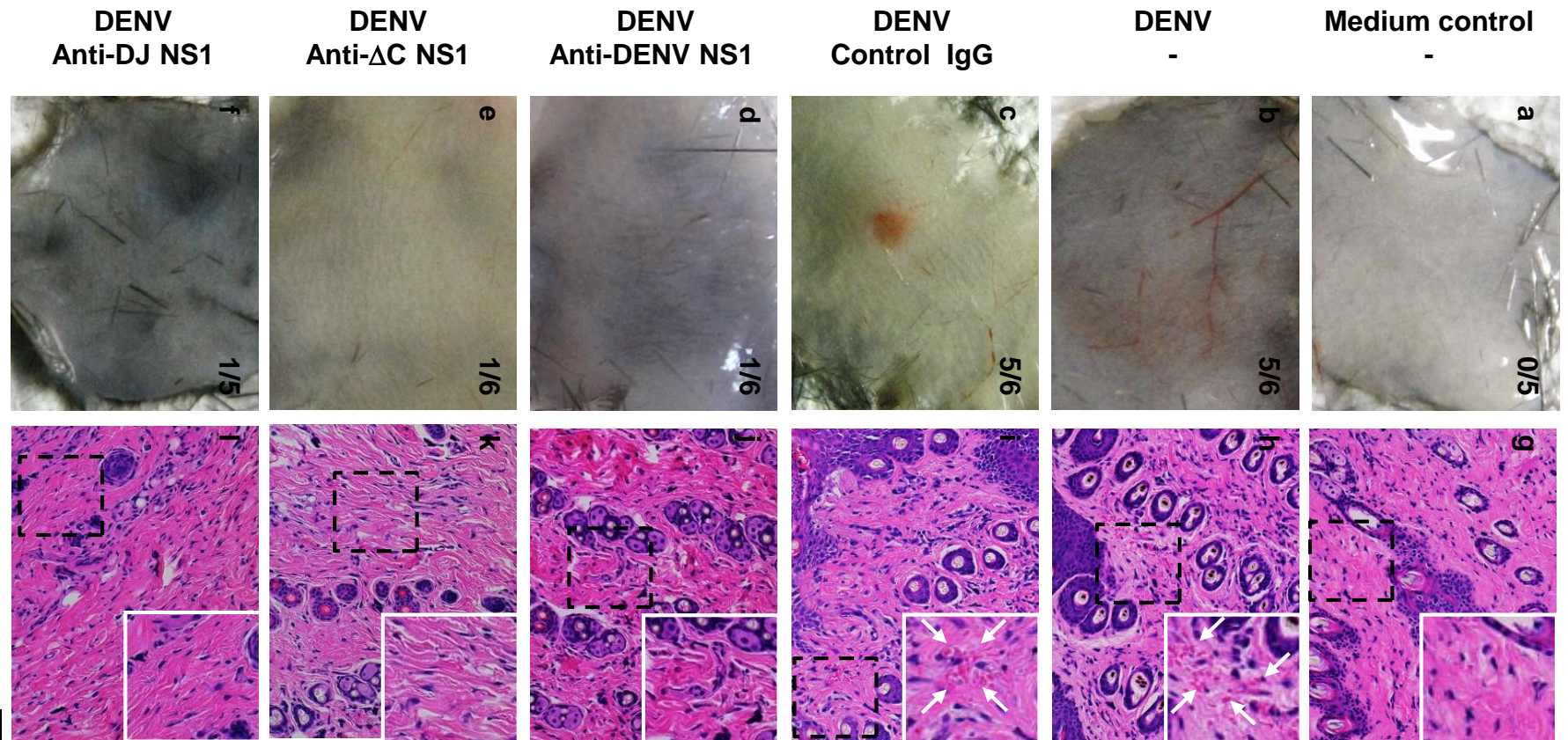
NS1 and C-terminal modified NS1 proteins



Passive immunization of mice with anti-NS1 reduces DENV replication after challenge



Passive immunization of mice with anti-NS1 reduces dengue-induced tissue hemorrhage



Summary

Dengue virus can perturb vascular endothelium by multiple mechanisms, including:

- direct virus infection
- vasoactive factors from intravascular cells (eg. monocytes, lymphocytes)
- vasoactive factors from extravascular cells (eg. mast cells, macrophages)
- cross-reactive immune responses

NS1 is a promising dengue vaccine candidate which elicits protective immunity and avoids risk of ADE

Acknowledgments

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