



Protecting Australia from future emerging arboviral threats

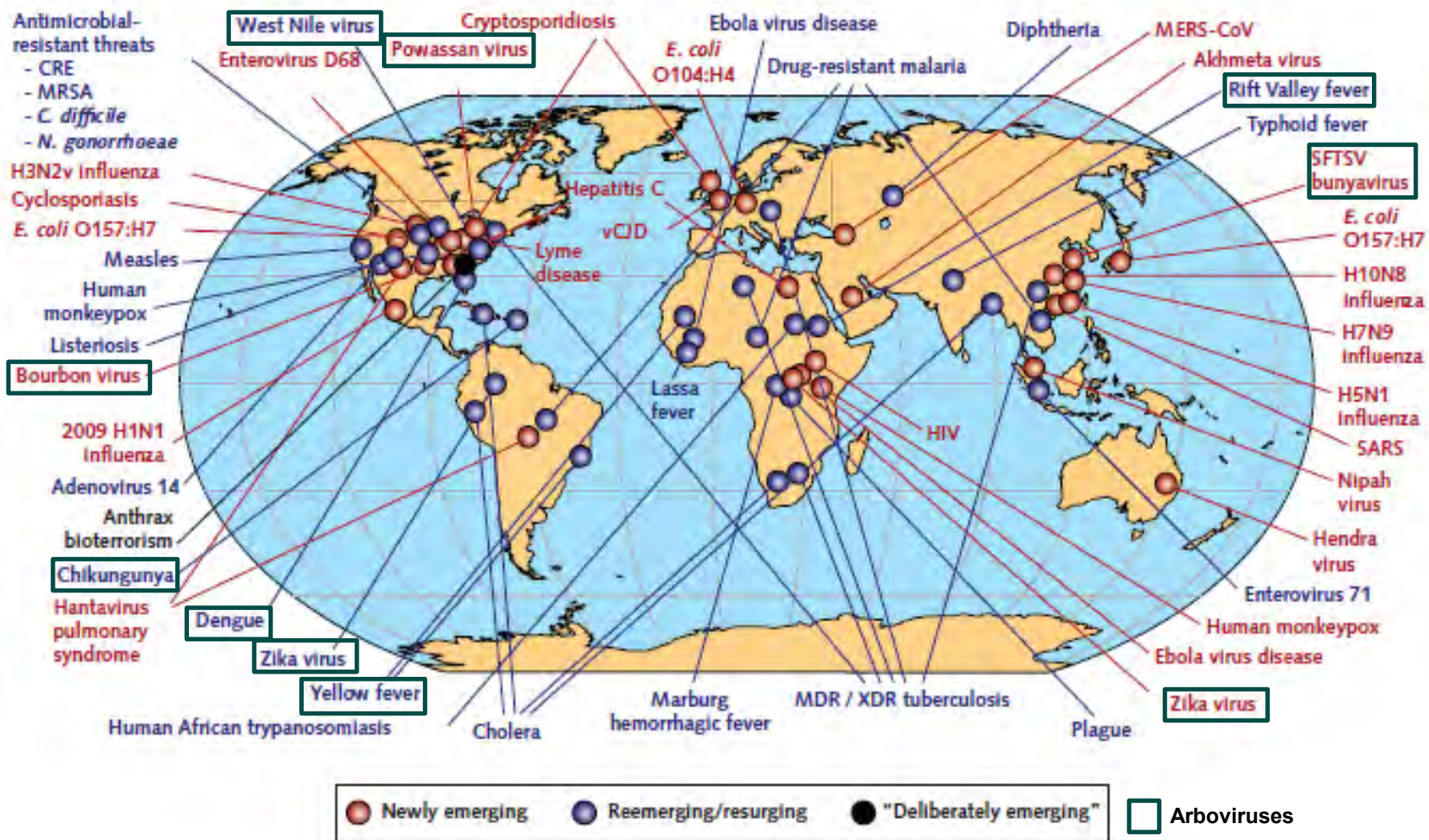
David Warrilow, Forensic and
Scientific Services, Brisbane



“Emerging infectious diseases are diseases of infectious origin whose incidence in humans has increased within the recent past or threatens to increase in the near future. These also include those infections that appear in new geographic areas or increase abruptly. The new infectious diseases and those which are re-emerging after a period of quiescence are also grouped under emerging infectious diseases.”

WHO (SEARO). Combating Emerging Infectious Diseases. New Delhi, 2005.

Emerging pathogens: recent examples



Catharine I. Paules, MD; Robert W. Eisinger, PhD; Hilary D. Marston, MD, MPH; and Anthony S. Fauci. What Recent History Has Taught Us About Responding to Emerging Infectious Disease Threats (2017) *Annals of Internal Medicine*.

Severity

Symptoms

Examples

Mild

Asymptomatic

Fever & rash

Arthralgia & myalgia

Haemorrhage

Severe

Encephalitis



- Ross River virus
- Barmah Forest virus
- Chikungunya

- Dengue fever virus
- Crimean-Congo haemorrhagic fever

- Murray Valley encephalitis virus
- Japanese encephalitis virus

10 Genera of arboviruses

RNA (+)

alphaviruses
flaviviruses



RNA (-)

vesiculoviruses
ephemerovirus



Rhabdoviridae

Monopartite genome

Segmented genome

Bunyaviridae

phleboviruses
orthobunyaviruses
nairoviruses
thogotoviruses



dsRNA

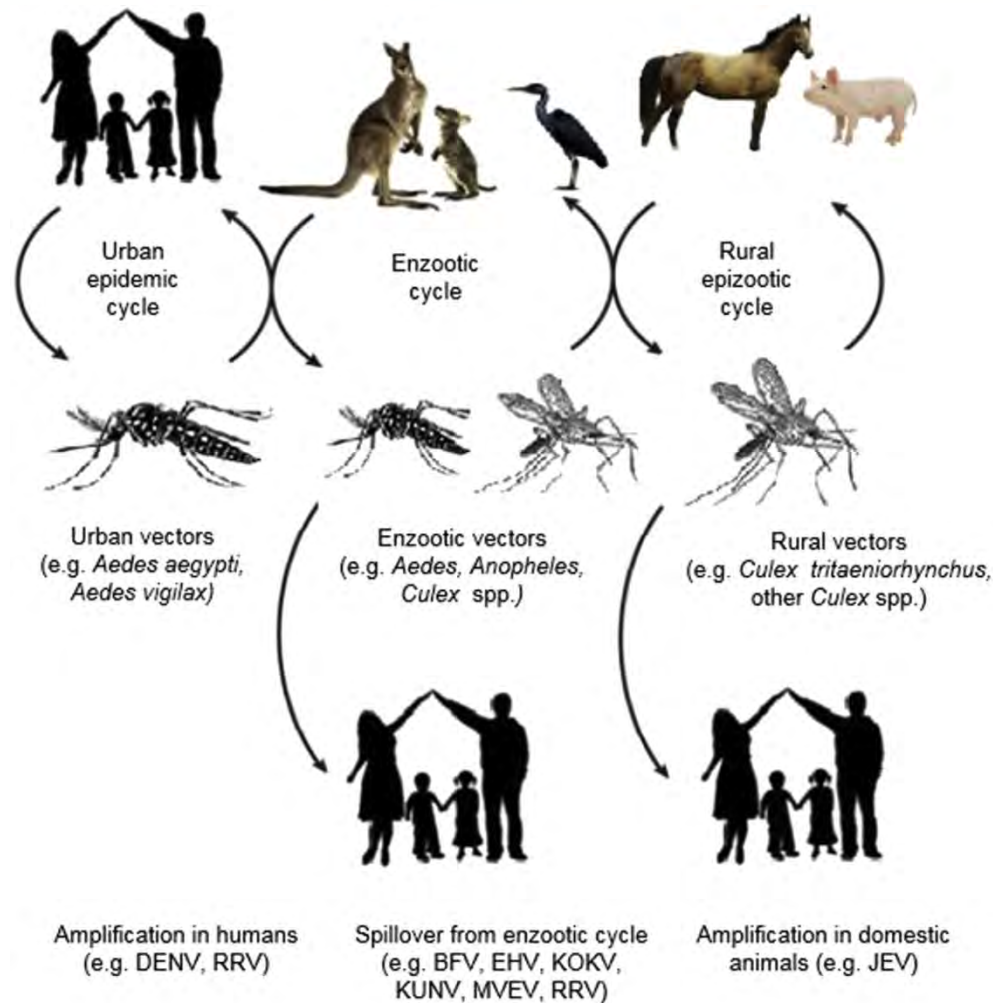
orbiviruses
seadornaviruses



Reoviridae

Medical and veterinary
Veterinary

Mosquito-host transmission cycles



Arboviruses – BFV: Barmah Forest; DENV: Dengue; EHV: Edge Hill; JEV: Japanese encephalitis; KOKV: Kokobera; KUNV: Kunjin; MVEV: Murray Valley encephalitis; RRV: Ross River

Arboviruses of significance in the Pacific region

Virus	Human disease syndrome	Area of activity	Amplifying hosts	Primary mosquito vectors
Ross River	Polyarthrititis	Australia, Papua New Guinea, Pacific islands	Macropods, Humans	<i>Ae. vigilax</i> , <i>Ae. camptorhynchus</i> , <i>Cx. annulirostris</i>
Barmah Forest	Polyarthrititis	Australia	Marsupials, birds	<i>Ae. vigilax</i> , <i>Ae. camptorhynchus</i> , <i>Cx. annulirostris</i>
Japanese encephalitis	Encephalitis	Northern Australia, Papua New Guinea	Wading birds (Herons, Egrets), pigs	<i>Cx. annulirostris</i> , <i>Cx. gelidus</i>
Chikungunya	Polyarthrititis	Papua New Guinea, Solomon Islands, Pacific islands	Humans (& non-human primates)	<i>Ae. aegypti</i> , <i>Ae. albopictus</i>
Murray Valley encephalitis	Encephalitis	Australia, Papua New Guinea	Wading birds (Herons, Egrets)	<i>Culex annulirostris</i>
West Nile (Kunjin subtype)	Encephalitis	Australia, Papua New Guinea	Wading birds (Herons, Egrets)	<i>Culex annulirostris</i>
Dengue	Shock & haemorrhage syndromes	Australia, Papua New Guinea, Solomon Islands, Pacific islands	Humans (& non-human primates)	<i>Ae. aegypti</i>
Zika	Birth defects & G.B. syndrome	Pacific islands	Humans (& non-human primates)	<i>Ae. aegypti</i> , <i>Ae. albopictus</i> <i>Aedes hensilli</i>

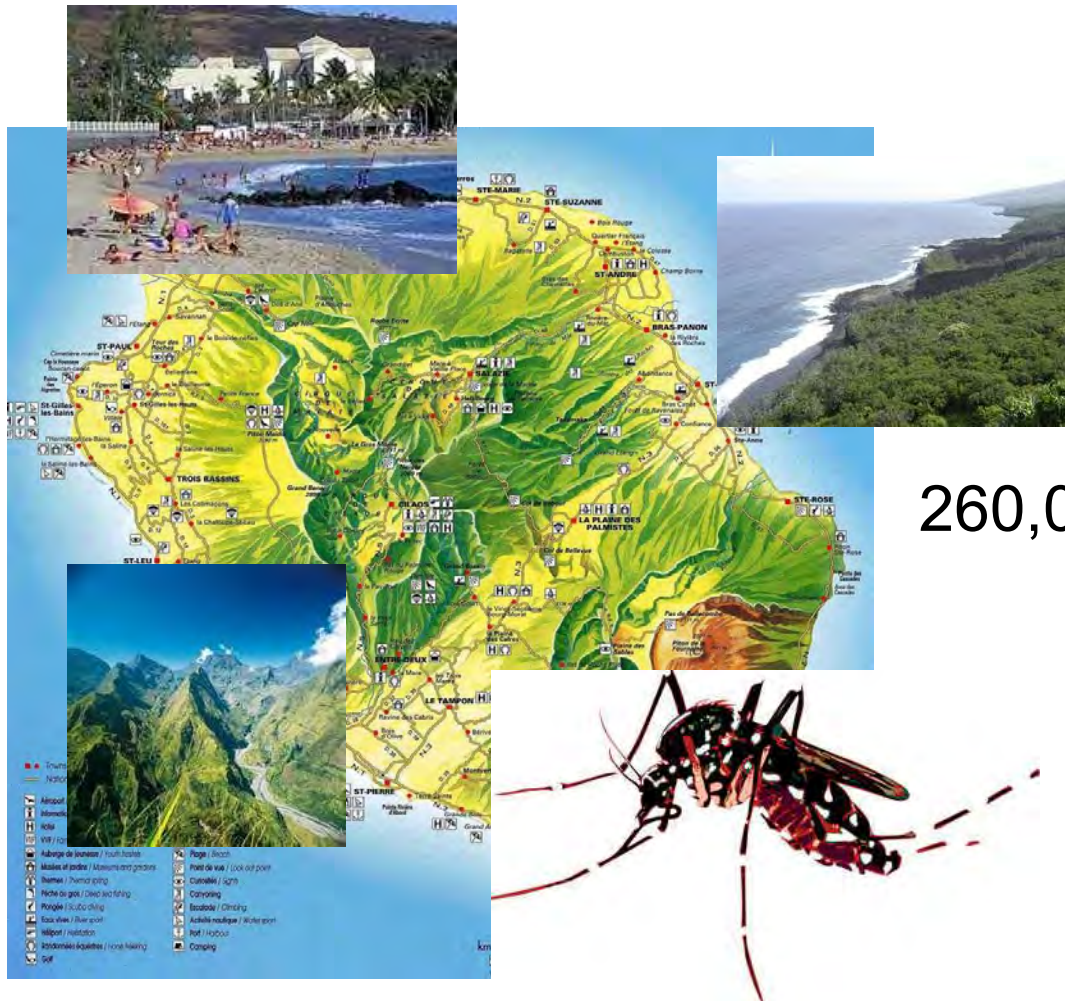


Important arboviruses of Australia

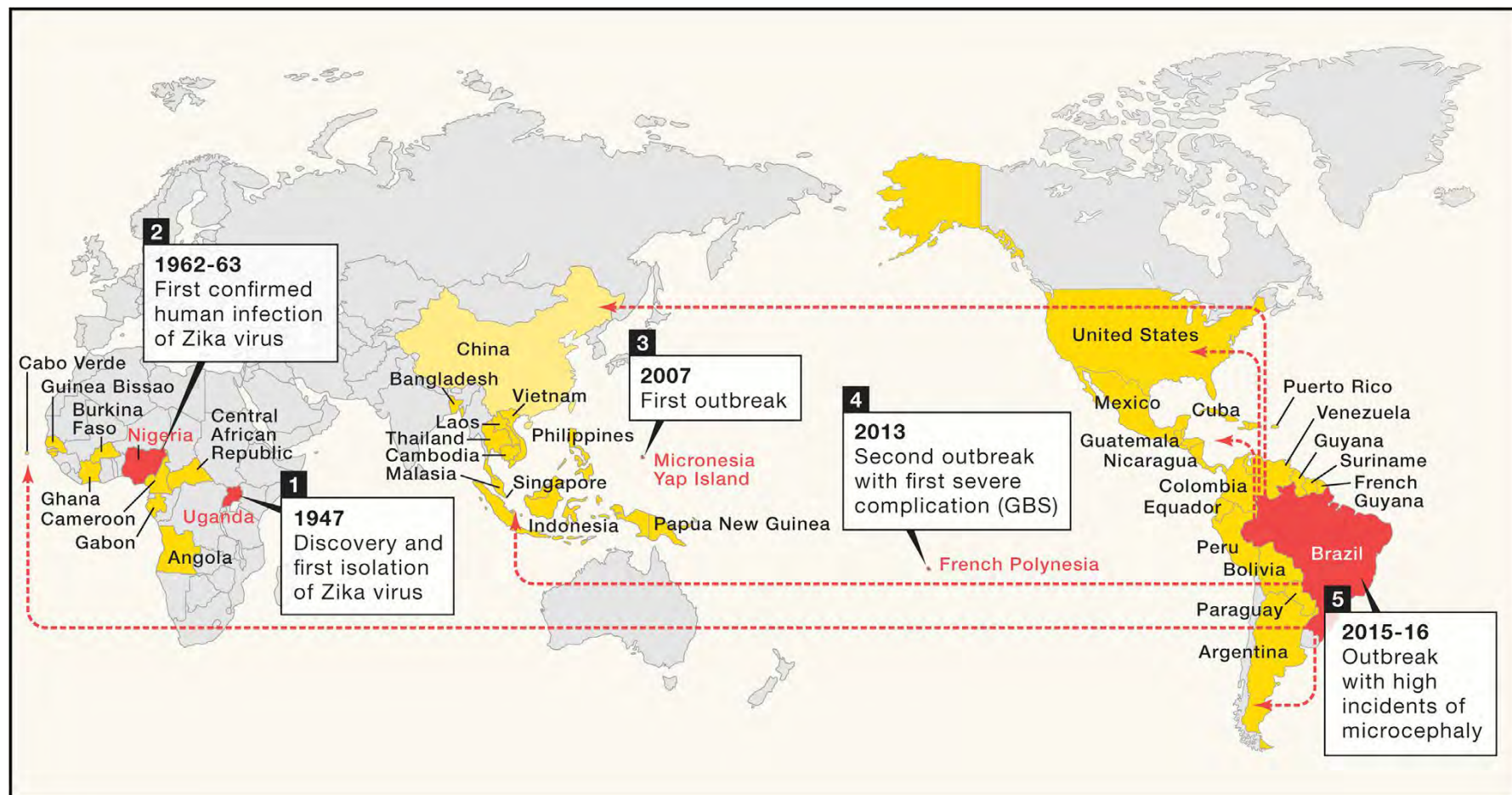
Family	Virus	Clinical features	Endemicity	No. of cases nationally in 2017*
Togaviridae	Ross River	Fever, rash, arthralgia/arthritis	Endemic	6,922
	Barmah Forest	“	Endemic	449
	Chikungunya	“	Not endemic	99
Flaviviridae	Dengue virus	Fever, rash, myalgia, and haemorrhage (rare)	Imported with local transmission	1,108
	Japanese encephalitis	Encephalitis	Not endemic	1
	Kunjin virus	Encephalitis (less severe)	Endemic	6
	Murray Valley encephalitis	Encephalitis	Endemic	0

*National Notifiable Disease Surveillance System (http://www9.health.gov.au/cda/source/rpt_2.cfm)

Explosive outbreaks

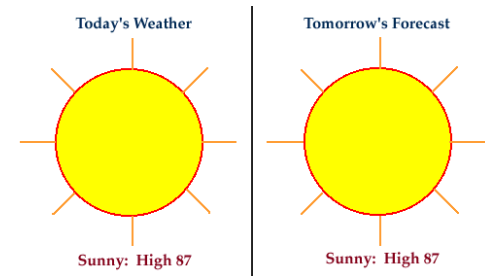


260,000 cases!



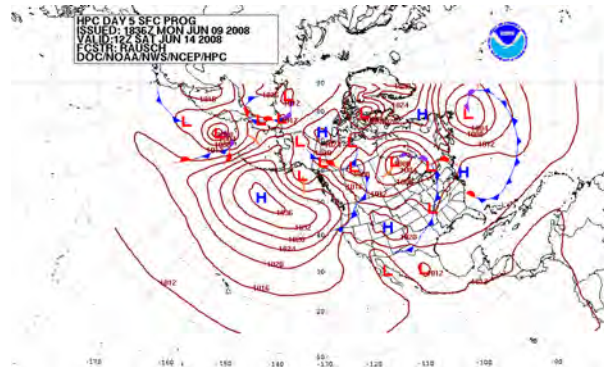
- Predicting the next emerging arbovirus is very challenging.
- Two broad approaches (e.g. weather prediction):
 1. Tomorrow will be like today (persistence).

- Previous experience informs our preparations for the future.



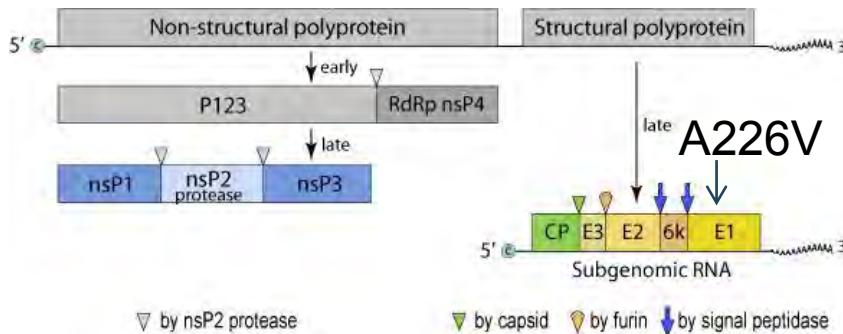
2. Complex models/algorithms (numerical).

- More complex with high chance of error as many variables.
- Only recent accurate prediction for 5-6 days using super-computers.



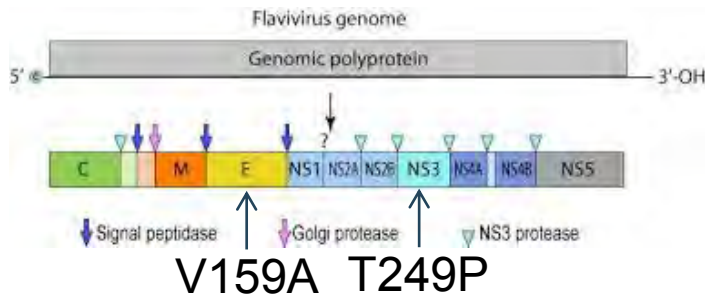


Adaptation of CHIKV to *Ae. albopictus* vector



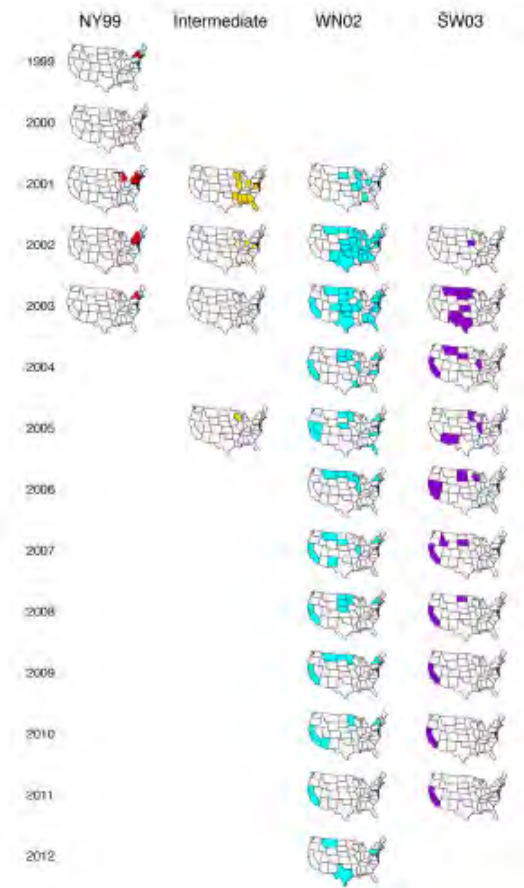
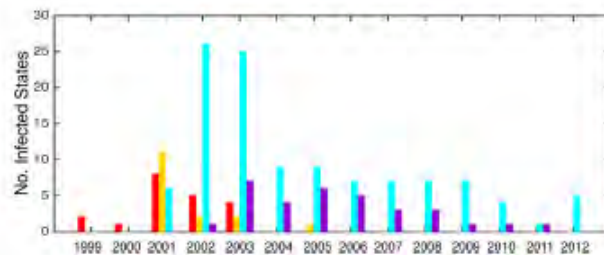
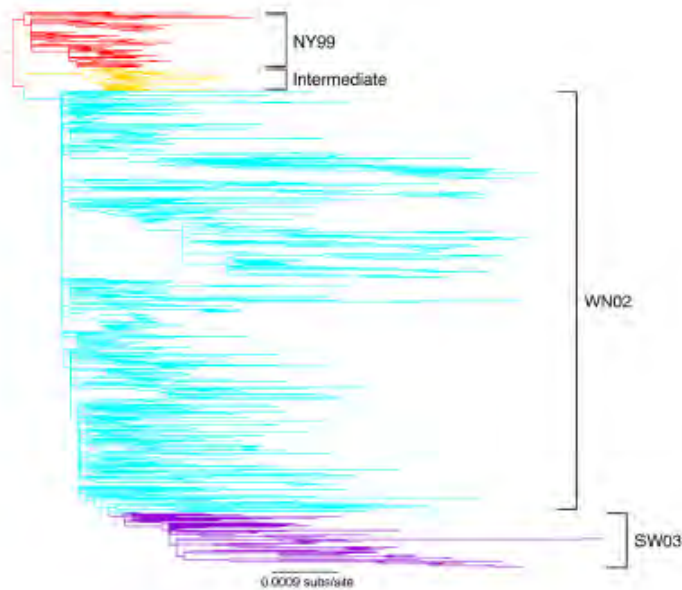
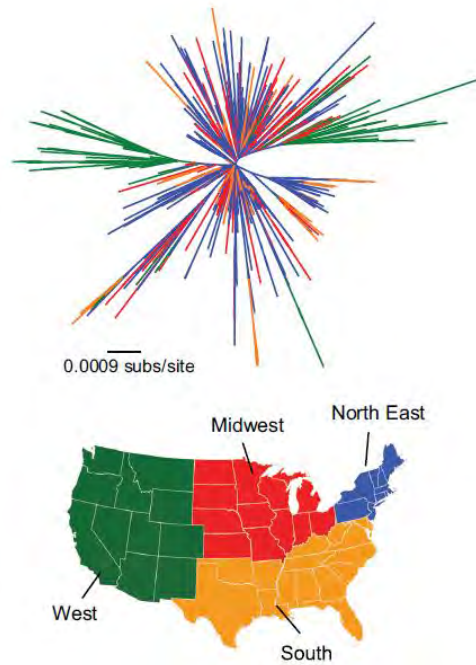
- A226V substitution in E1 – virus better adapted to *Ae albopictus* vector.
- Associated with Reunion Is. outbreak.
- Global spread.

Adaptation of WNV in US outbreak



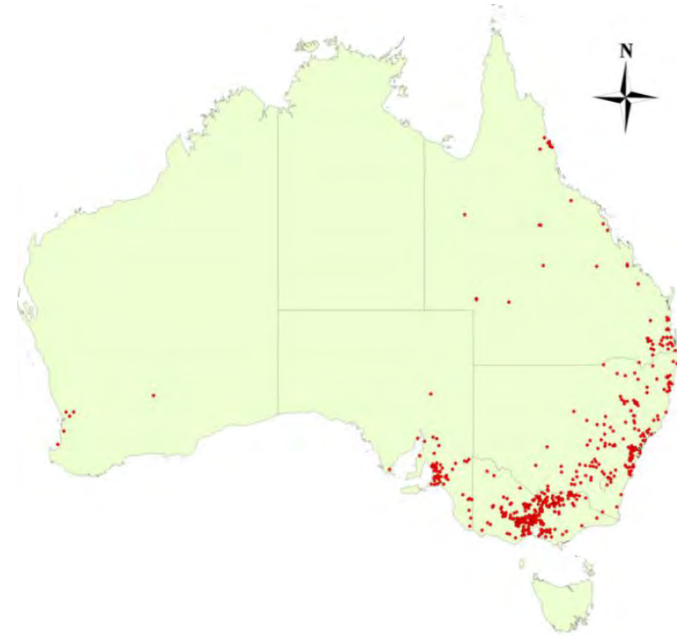
- T249P substitution in NS3 had greater virulence in American crows. Positively selected.
- V159A substitution in E important for adaptation to *Cx pipiens* vector in US.

West Nile virus emergence in the US



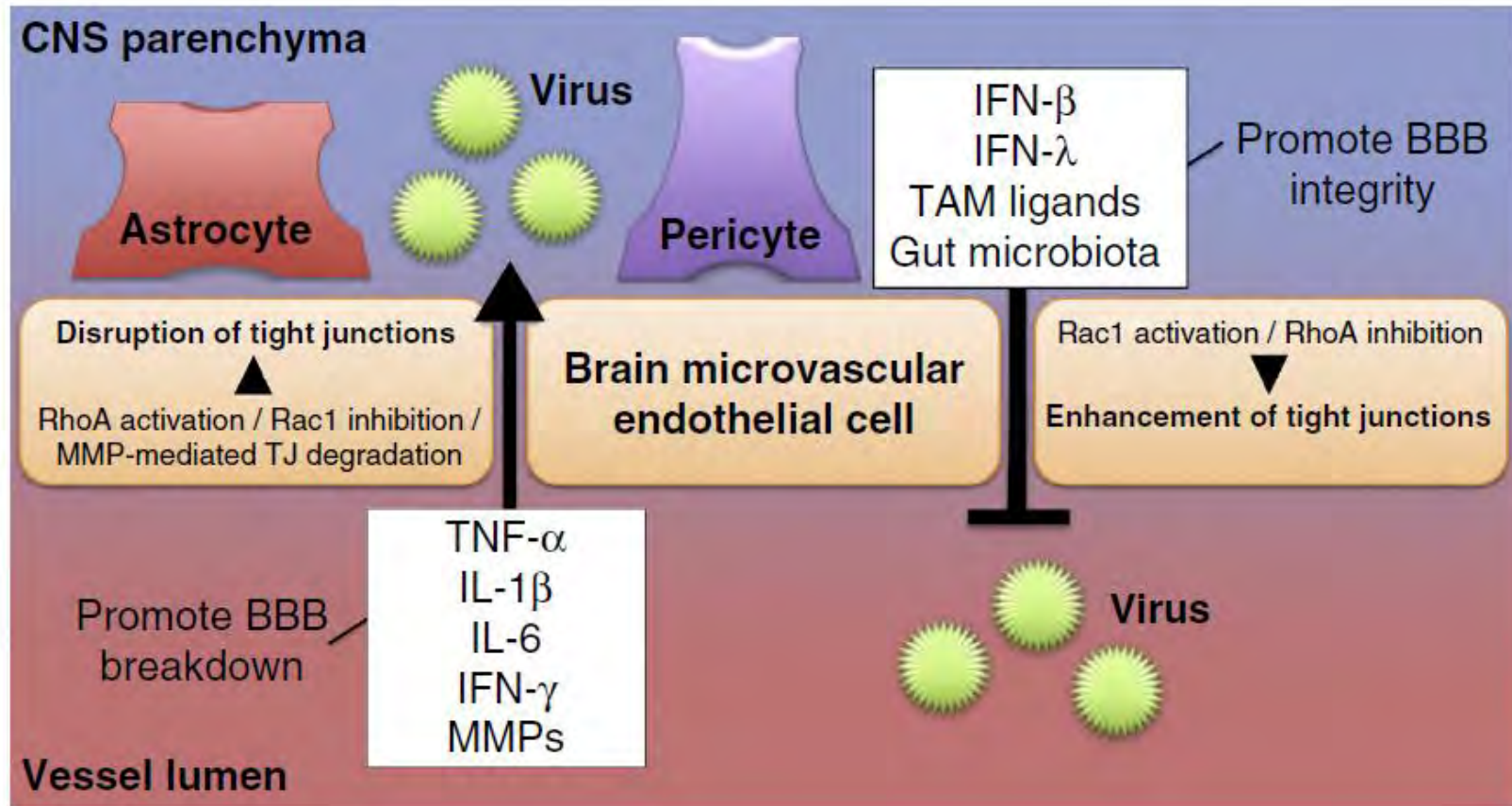
Symptoms

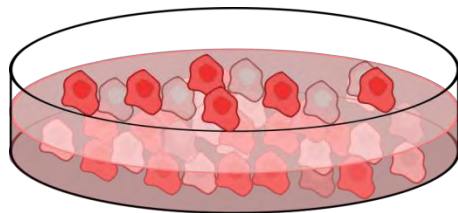
- Facial paralysis
- Muscle tremors/fasciculation
- Hyperaesthesia
- Circling
- Blindness
- Recumbency or inability to stand
- Hind limb weakness
- Multiple limb paralysis
- Altered mental state
- Hypermetria
- Depression
- Death (~10%)



NSW Department of Primary Industries factsheet

Roche et al. Descriptive overview of the 2011 epidemic of arboviral disease in horses in Australia.
Aust Vet J. 2013 Jan-Feb;91(1-2):5-13.





Human neuroblastoma cells
(SK-N-SH)

Time →

Extract
RNA

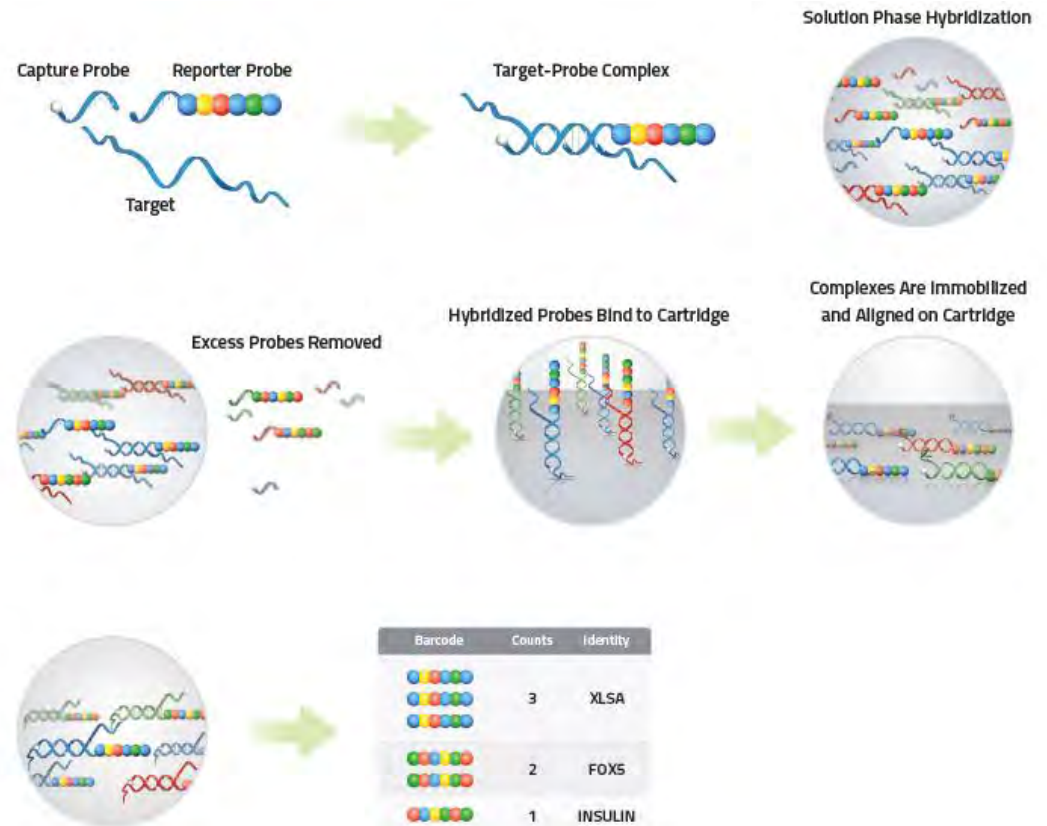


250 human inflammation genes

CXCL9	IL21	MMP9	TGFB2
CXCR1	IL22	MRC1	TGFB3
CXCR2	IL22RA2	MX1	TGFB3R1
CXCR4	IL23A	MX2	TLR1
CYSLTR1	IL23R	MYC	TLR2
CYSLTR2	IL3	MYD88	TLR3
DAXX	IL4	MYL2	TLR4
DDIT3	IL5	NFATC3	TLR5
DEFA1	IL6	NFE2L2	TLR6
ELK1	IL6R	NFKB1	TLR7
FASLG	IL7	NLRP3	TLR8
FLT1	IL8	NOD1	TLR9
FOS	IL9	NOD2	TNF
FXYD2	IRF1	NOS2	TNFAIP3
GNAQ	IRF3	NOX1	TNFSF14
GNAS	IRF5	NR3C1	TOLLIP
GNB1	IRF7	OAS2	TRADD
GNMT1	ITGB2	OASL	TRAF2
GRB2	JUN	OXER1	TREM2
HDAC4	KEAP1	PDGFA	TSLP
HIF1A	KNG1	PIK3C2G	TWIST2
HLA-DRA	LIMK1	PLA2G4A	TYROBP
HLA-DRB1	LTA	PLCB1	
HMGB1	LTB	PPP1R12B	
HMGB2	LTB4R	PRKCA	
HMGN1	LTB4R2	PRKCB PTGDR2	
HRAS	LY96	PTGER1	
HSN2D	MAFF	PTGER2	
HSPB1	MAFG	PTGER3	
HSPB2	MAFK	PTGER4	
IFI44	MAP2K1	PTGFR	
IFIT1	MAP2K4	PTGIR	
IFIT2	MAP2K6	PTGS1	
IFIT3	MAP3K1	PTGS2	
IFNA1	MAP3K5	PTK2	
IFNB1	MAP3K7	RAC1	
IFNG	MAP3K9	RAF1	
IL10	MAPK1	RAPGEF2	
IL10RB	MAPK14	RELA	
IL11	MAPK3	RELB	
IL12A	MAPK8	RHOA	
IL12B	MAPKAPK2	RIPK1	
IL13	MAPKAPK5	RIPK2	
IL15	MASP1	ROCK2	
IL17A	MASP2	RPS6KA5	
IL18	MAX	SHC1	
IL18RAP	MBL2	SMAD7	
IL1A	MEF2A	STAT1	
IL1B	MEF2BNB-MEF2B	STAT2	
IL1R1	MEF2C	STAT3	
IL1RAP	MEF2D	TBXA2R	
IL1RN	MKNK1	TCF4	
IL2	MMP3	TGFB1	

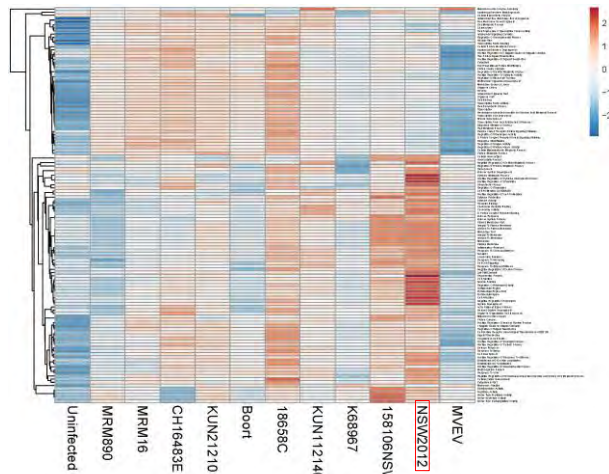
6 reference genes

CLTC
GAPDH
GUSB
HPRT1
PGK1
TUBB

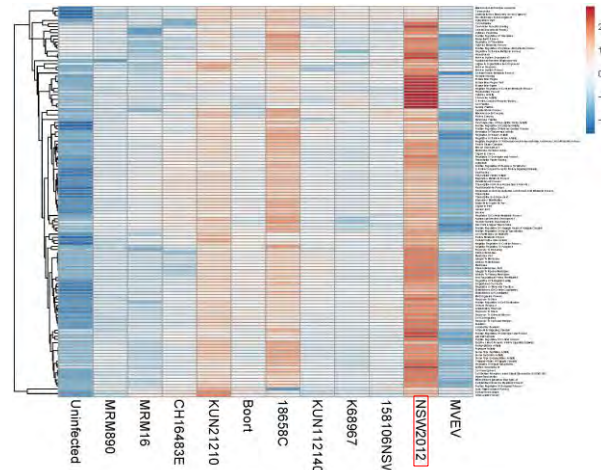


Activation of immune pathways

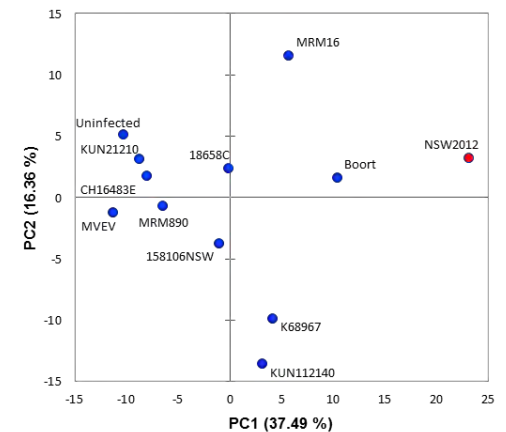
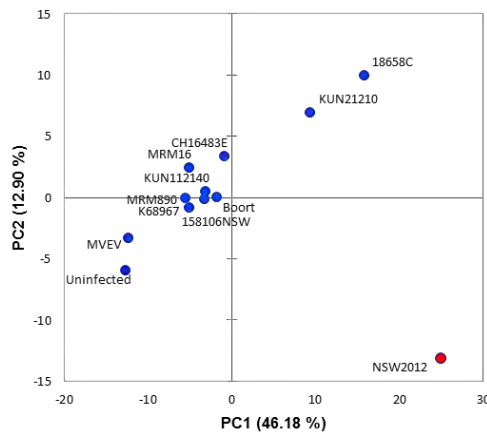
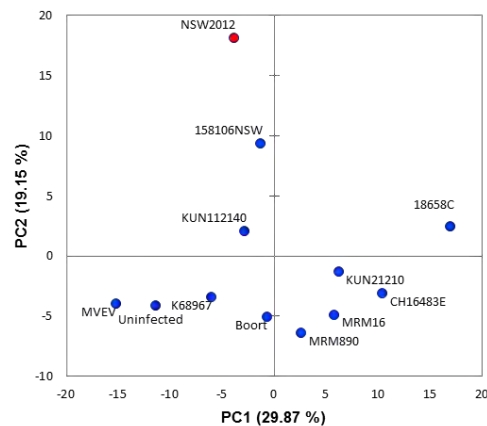
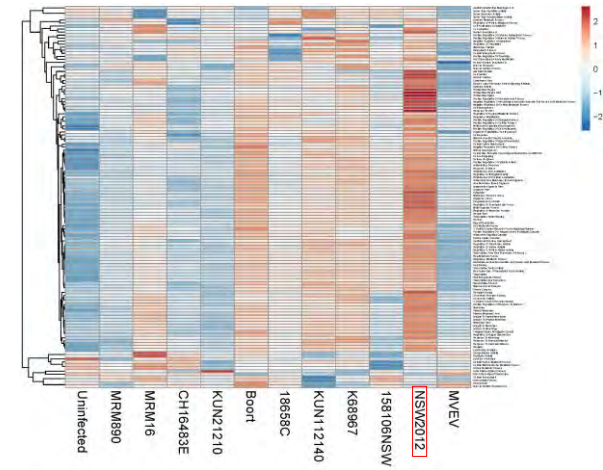
Experiment 1



Experiment 2



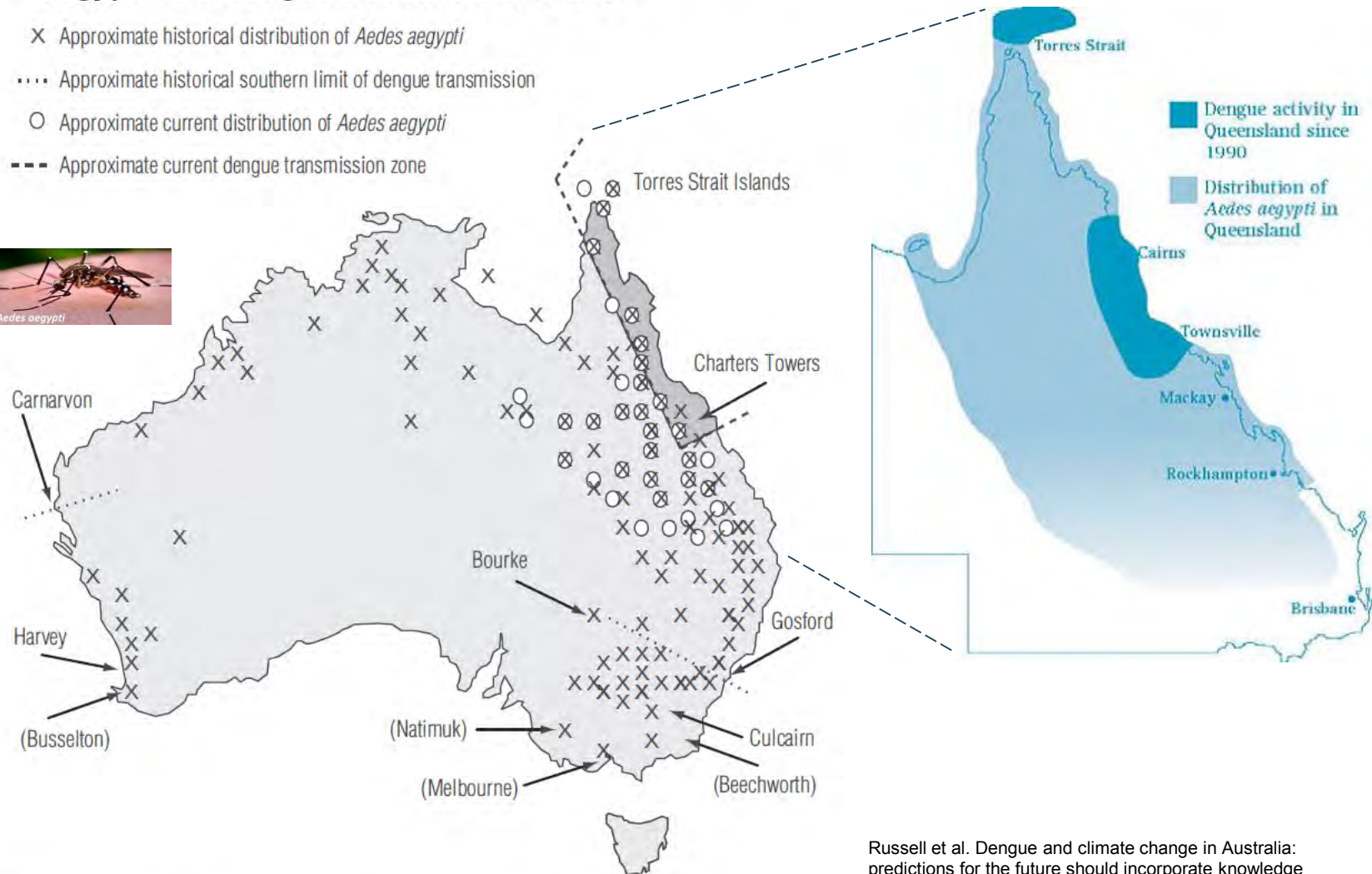
Experiment 3



Current and historical *Ae aegypti* distribution

Past and present (post-1990) distribution of *Aedes aegypti* and dengue transmission zones*

- × Approximate historical distribution of *Aedes aegypti*
- Approximate historical southern limit of dengue transmission
- Approximate current distribution of *Aedes aegypti*
- Approximate current dengue transmission zone



Russell et al. Dengue and climate change in Australia: predictions for the future should incorporate knowledge from the past. Med J Aust. 2009 Mar 2;190(5):265-8.

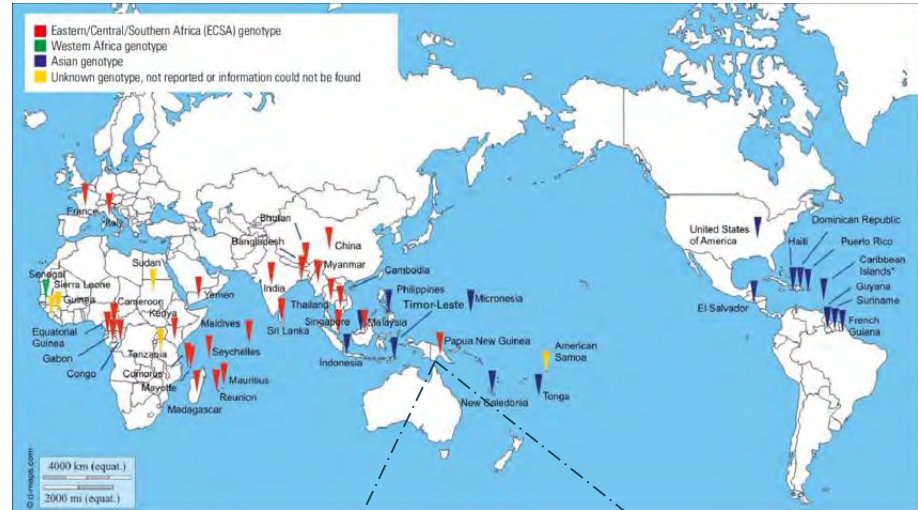
* Parentheses show locations where reports are unconfirmed.

Hypothetical 1: Clear and present danger?

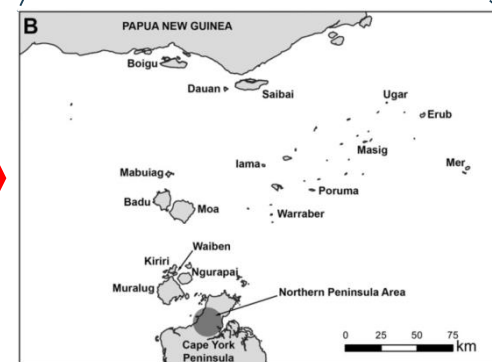
Yellow fever virus



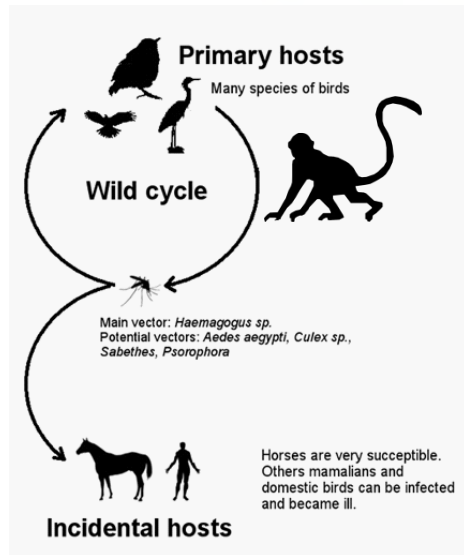
Chikungunya virus



Ae. albopictus in the Torres Strait



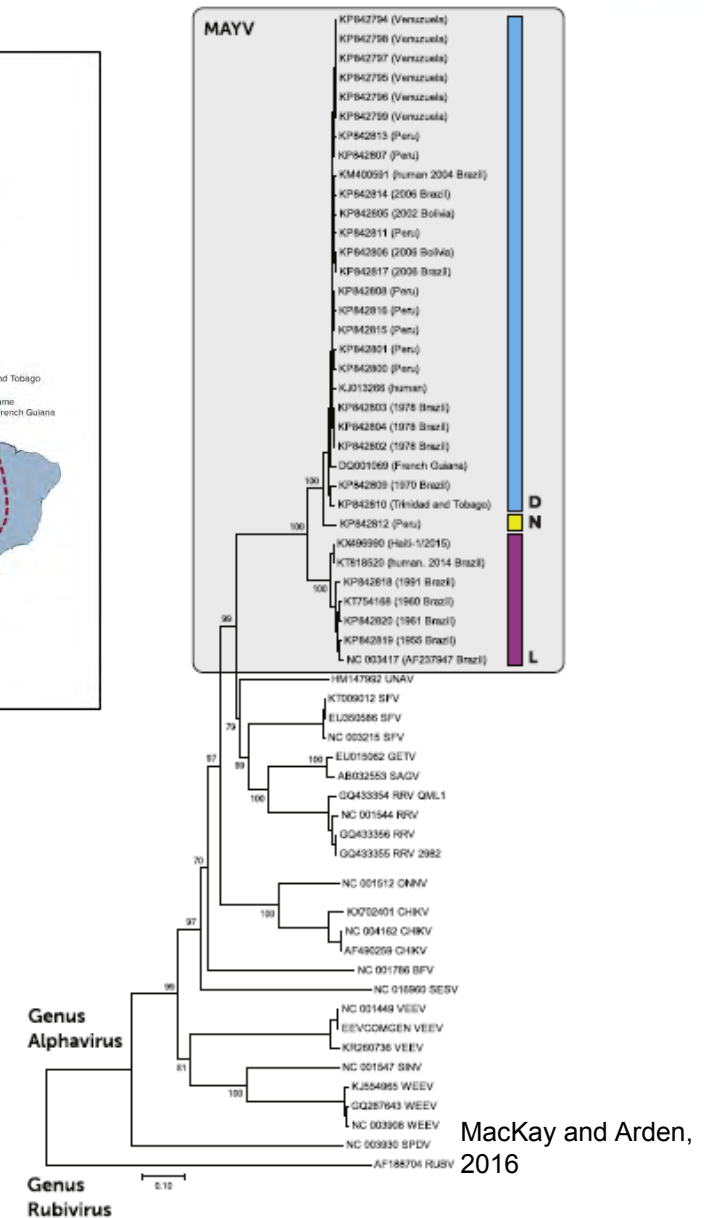
Hypothetical 2: Mayaro virus



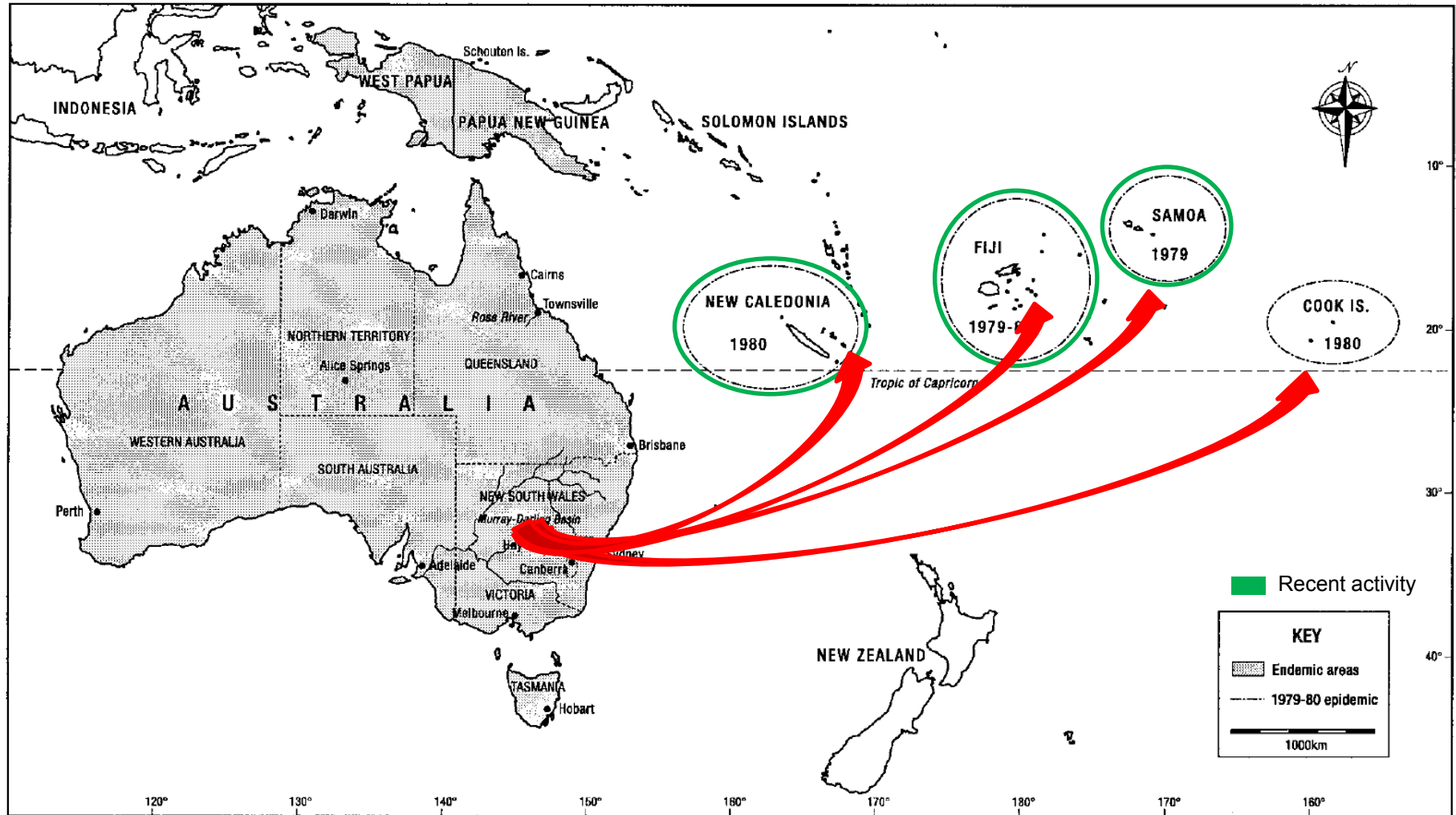
Napoleão-Pego *et al.*, 2014



- Alphavirus.
- Dengue-like symptoms.
- Sylvatic cycle.
- Potential for urban spread.
- Naïve populations.

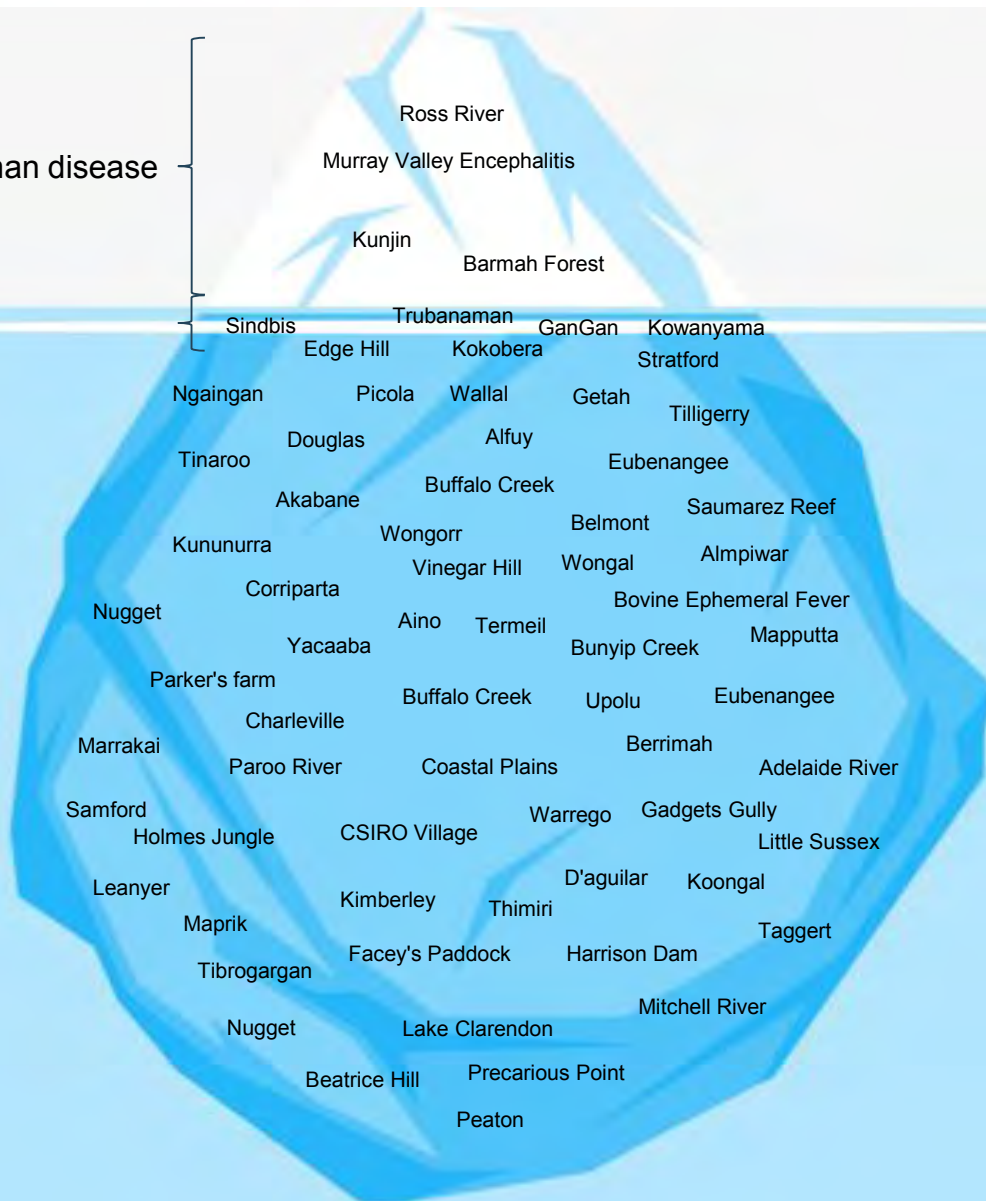


Ross River virus Pacific outbreak 1979-80



Hypothetical 3: Home-grown arboviruses

Arboviruses causing human disease



Australian endemic arboviruses*

Togaviridae	Flaviviridae	Bunyaviridae	Rhabdoviridae	Reoviridae	Unclassified
Ross River	Murray Valley encephalitis	Kowanyama	Bovine ephemeral fever	Corriparta	Beatrice Hill
Barmah Forest	West Nile (Kunjin)	Yacaaba	Almpiwar	Eubenangee	Parker's Farm
Getah	Kokobera/Stratford	Gan Gan		Bluetongue	Little Sussex
Sindbis	Alfuy	Trubanaman		Wongorr	Upolu
	Edge Hill	Akabane			Tzipori
	Saumarez Reef	Taggert			
		Belmont			
		Thimiri			
		Mapputta			
		Maprik			
		Leanyer			

*Not an exhaustive list

[No sequence (study start)]

This study









Australian endemic arboviruses - updated

Togaviridae	Flaviviridae	Bunyaviridae	Rhabdoviridae	Reoviridae	Orthomyxoviridae
Ross River	Murray Valley encephalitis	Kowanyama	Bovine ephemeral fever (Tzipori)	Corriparta	Upolu*
Barmah Forest	West Nile (Kunjin)	Yacaaba	Almpiwar	Eubenangee	
Getah	Kokobera/Stratford	Gan Gan (Salt Ash)	Beatrice Hill	Bluetongue	
Sindbis	Alfuy	Trubanaman (Murrumbidgee)		Wongorr	
	Edge Hill	Akabane			
	Saumarez Reef	Taggert			
		Belmont			
		Thimiri			
		Mapputta*			
		Maprik*			
		Leanyer			
		Parker's Farm			
		Little Sussex			

[*Sequenced by others in the interim]

This study

Emergence factors

Togaviridae	Flaviviridae	Bunyaviridae	Rhabdoviridae	Reoviridae	Orthomyxoviridae
Ross River	Murray Valley encephalitis	Kowanyama 	Bovine ephemeral fever (Tzipori)	Corriparta	Upolu*
Barmah Forest	West Nile (Kunjin)	Yacaaba 	Almpiwar	Eubenangee	
Getah	Kokobera/Stratford	Gan Gan (Salt Ash)  	Beatrice Hill	Bluetongue	
Sindbis	Alfuy	Trubanaman (Murrumbidgee)  		Wongorr	
	Edge Hill	Akabane			
	Saumarez Reef	Taggert vOTU domain			
		Belmont 			
		Thimiri 			
		Mapputta*			
		Maprik*			
		Leanyer			
		Parker's Farm			
		Little Sussex			



Ab (humans)

Mossie human feeder

Mossie rare human feeder

Tick human feeder

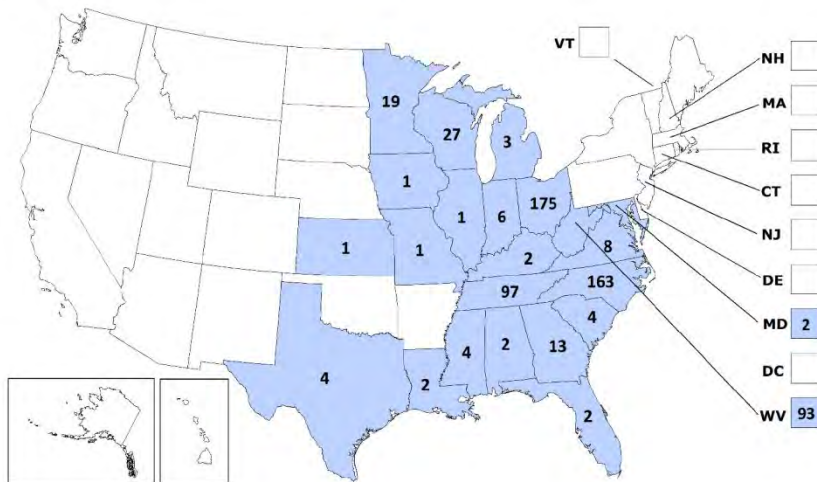


Neurovirulent (mice)

La Crosse virus



La Crosse virus neuroinvasive disease cases reported by state of residence, 2007–2016



California encephalitis virus

California Serogroup Virus Neuroinvasive Disease Cases Reported by State, 1964-2009



Table. Optimal Response to Emerging Infectious Disease Outbreaks: Lessons Learned

Global surveillance to detect outbreaks early

Transparency and communication in response to outbreaks

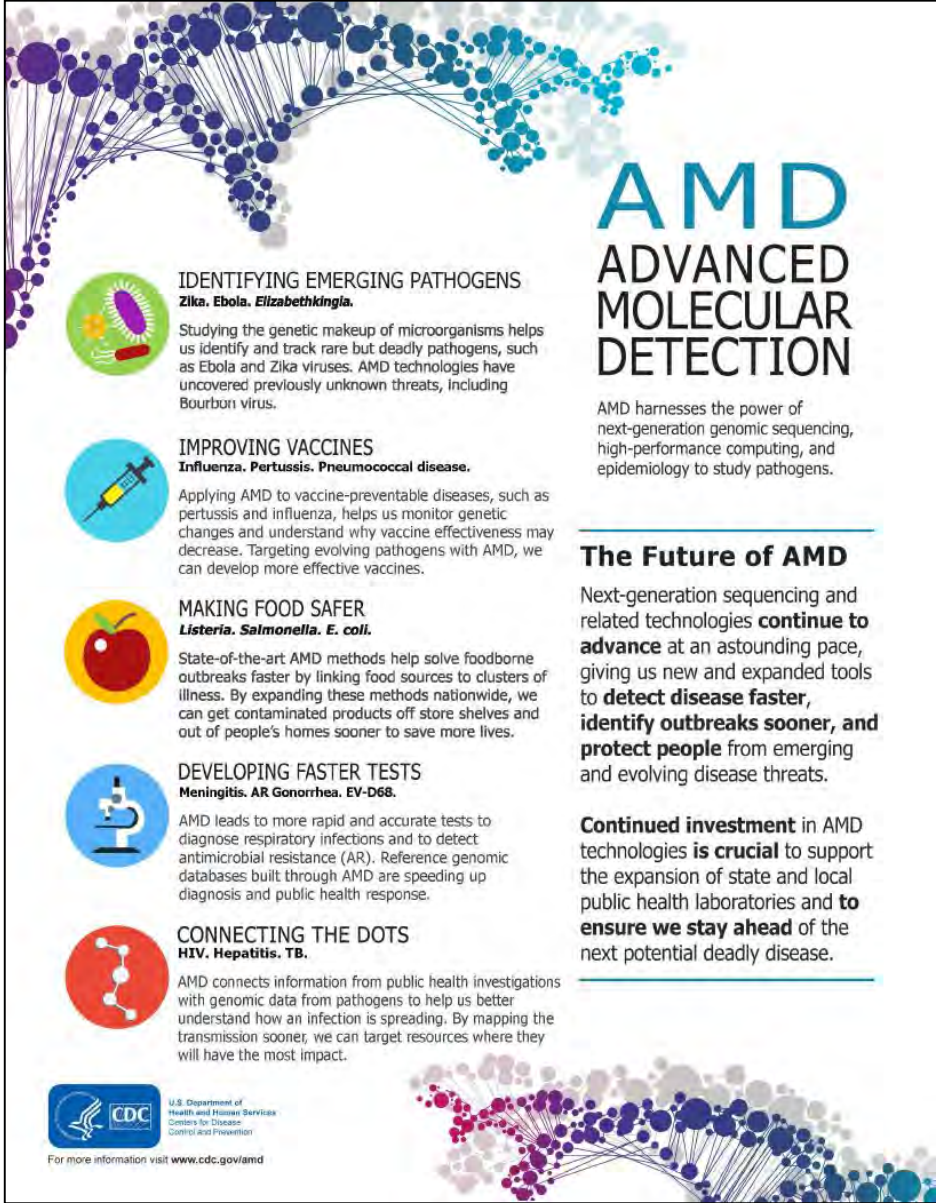
Incorporation of infrastructure and capacity building domestically and internationally in outbreak responses

Conduct of basic and clinical research associated with outbreaks in a coordinated and collaborative manner

Involvement of the afflicted communities in policy decisions

Pursuit and perfection of adaptable platform technologies for vaccines, diagnostics, and therapeutics

Importance of flexible funding mechanisms



AMD ADVANCED MOLECULAR DETECTION

AMD harnesses the power of next-generation genomic sequencing, high-performance computing, and epidemiology to study pathogens.

IDENTIFYING EMERGING PATHOGENS

Zika. Ebola. Elizabethkingia.

Studying the genetic makeup of microorganisms helps us identify and track rare but deadly pathogens, such as Ebola and Zika viruses. AMD technologies have uncovered previously unknown threats, including Bourbon virus.

IMPROVING VACCINES

Influenza. Pertussis. Pneumococcal disease.

Applying AMD to vaccine-preventable diseases, such as pertussis and influenza, helps us monitor genetic changes and understand why vaccine effectiveness may decrease. Targeting evolving pathogens with AMD, we can develop more effective vaccines.

MAKING FOOD SAFER

Listeria. Salmonella. E. coli.

State-of-the-art AMD methods help solve foodborne outbreaks faster by linking food sources to clusters of illness. By expanding these methods nationwide, we can get contaminated products off store shelves and out of people's homes sooner to save more lives.

DEVELOPING FASTER TESTS

Meningitis. AR Gonorrhea. EV-D68.

AMD leads to more rapid and accurate tests to diagnose respiratory infections and to detect antimicrobial resistance (AR). Reference genomic databases built through AMD are speeding up diagnosis and public health response.

CONNECTING THE DOTS


HIV. Hepatitis. TB.

AMD connects information from public health investigations with genomic data from pathogens to help us better understand how an infection is spreading. By mapping the transmission sooner, we can target resources where they will have the most impact.

The Future of AMD

Next-generation sequencing and related technologies **continue to advance** at an astounding pace, giving us new and expanded tools to **detect disease faster, identify outbreaks sooner, and protect people** from emerging and evolving disease threats.

Continued investment in AMD technologies **is crucial** to support the expansion of state and local public health laboratories and to **ensure we stay ahead** of the next potential deadly disease.



U.S. Department of Health and Human Services
Centers for Disease Control and Prevention

For more information visit www.cdc.gov/amd

Sentinel animals



Problems

- Expensive
- Serological cross-reactive
- Can be amplifying hosts

Mosquito traps



Problems

- Expensive
- Logistical problems
- Sample degradation





Hall-Mendelin et al. (2010) PNAS.

Ruralweekly 5

Mosquito virus risk in horses

HORSE owners are urged to be aware of the potential for mosquito-borne viruses to cause illness in horses following the detection of Kunjin virus in mosquitoes in the Central Highlands area.

The identification was made as part of a routine surveillance program run by Queensland Health.

Kunjin virus is endemic in parts of Australia and can cause disease in humans and horses spread by an infected mosquito. Clinical signs in horses include reluctance to move, depression and neurological signs such as stumbling, incoordination and facial paralysis.

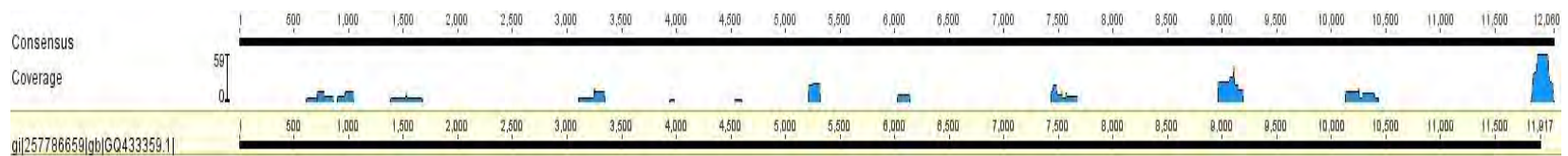
Most clinical cases recover gradually over one to three weeks. However, approximately 10 per cent of affected horses will die or be euthanased for welfare reasons.

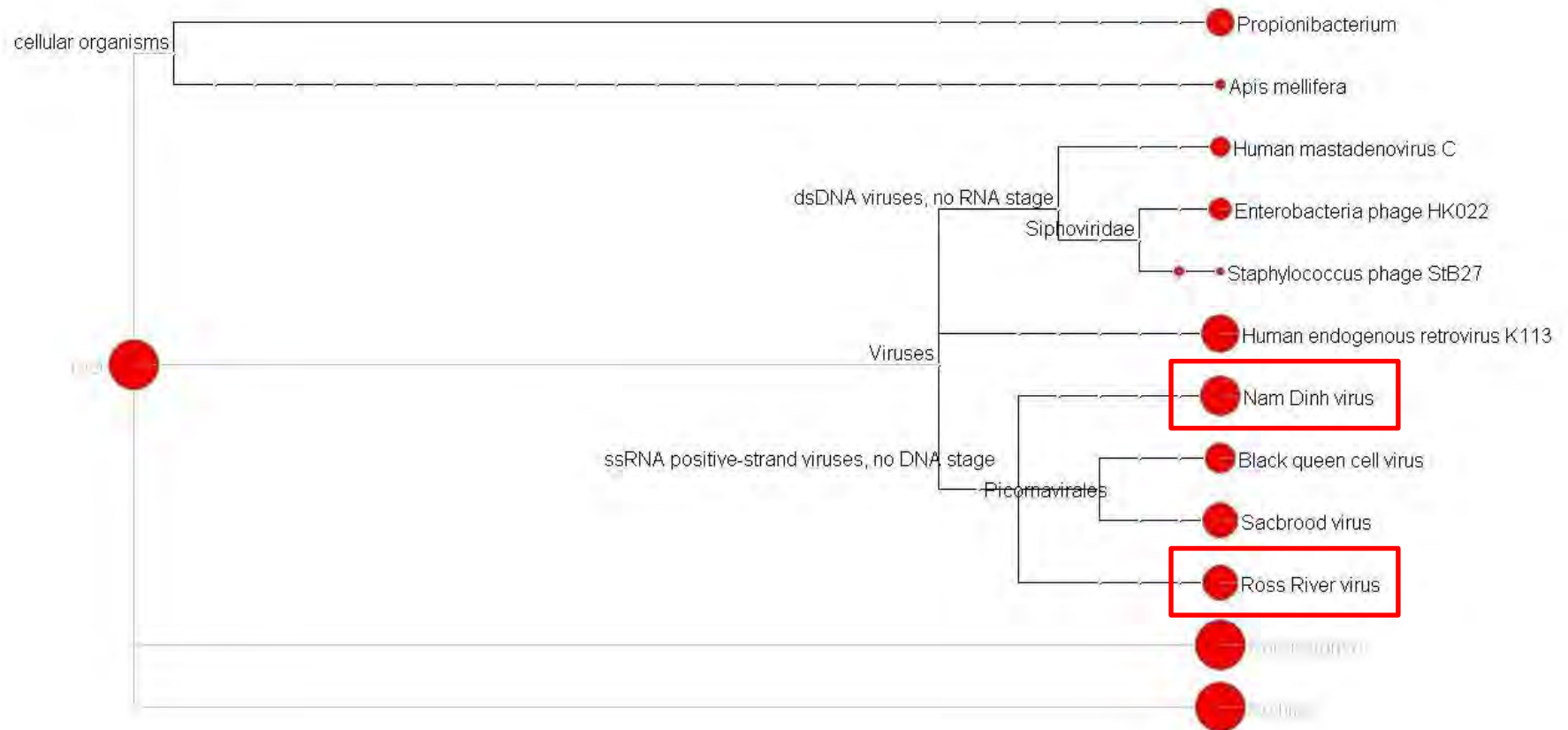
Horse owners can reduce the exposure of their animals to insect bites by rugging, using fly masks and removing sources of stagnant water.

It is important to consider Hendra virus infection in horses showing neurological signs and implement stringent biosecurity and hygiene measures when dealing with sick horses. Horse owners should contact their private vet if their horse is unwell.

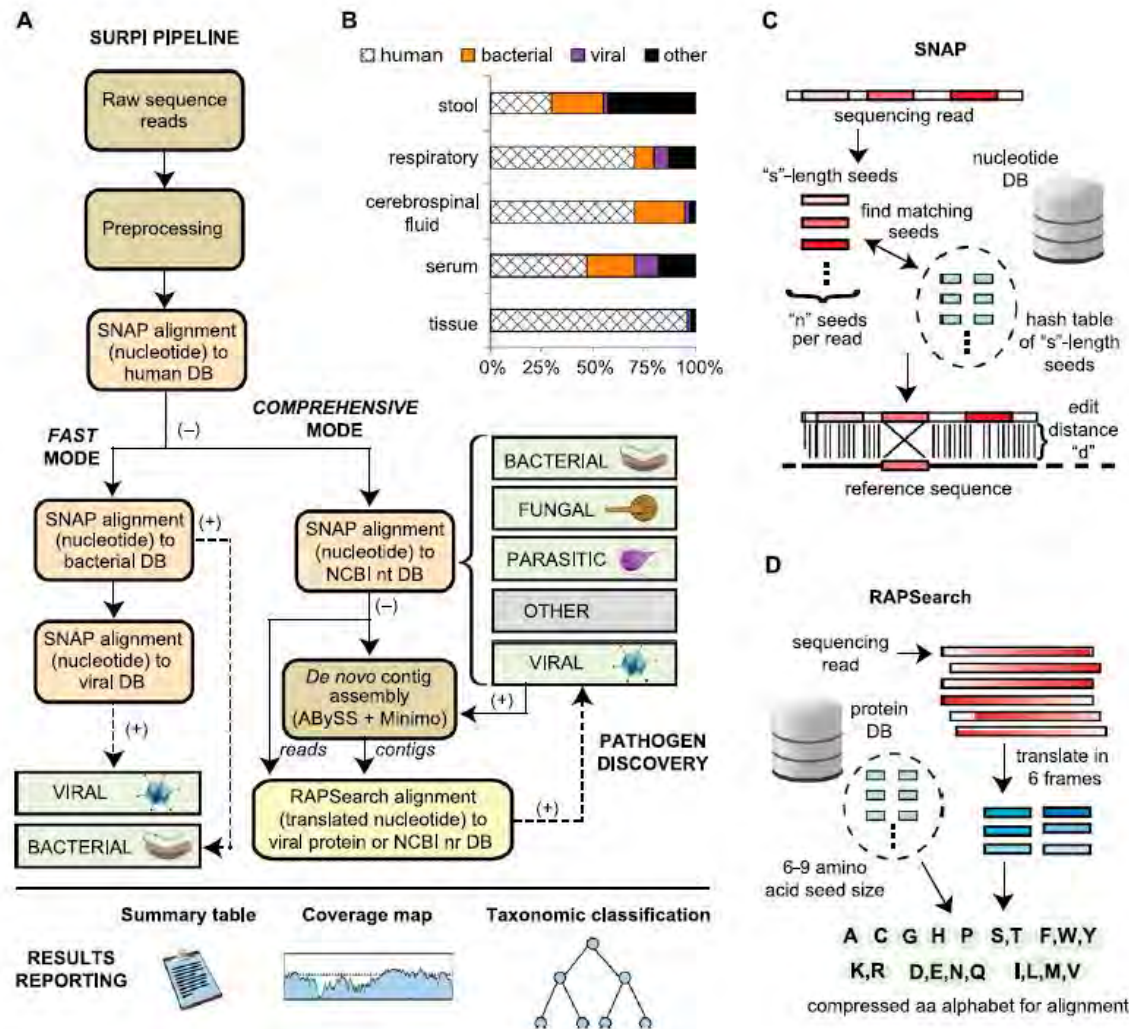


- 187 reads matched to RRV.
- FTA card (Ct~31) by TaqMan.
- 3 years old.
- Frozen and thawed several times.





The Future: Hypothesis-free testing

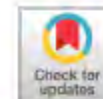




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MINIREVIEW



Towards a Universal Molecular Microbiological Test

Richard J. N. Allcock,^{a,b} Amy V. Jennison,^c David Warrilow^d

School of Biomedical Sciences, University of Western Australia, Nedlands, Western Australia, Australia^a; Translational Cancer Pathology Laboratory, Pathwest Laboratory Medicine WA, QEII Medical Centre, Nedlands, Western Australia, Australia^b; Public Health Microbiology Laboratory, Queensland Health Forensic and Scientific Services, Archerfield, Queensland, Australia^c; Public Health Virology Laboratory, Queensland Health Forensic and Scientific Services, Archerfield, Queensland, Australia^d

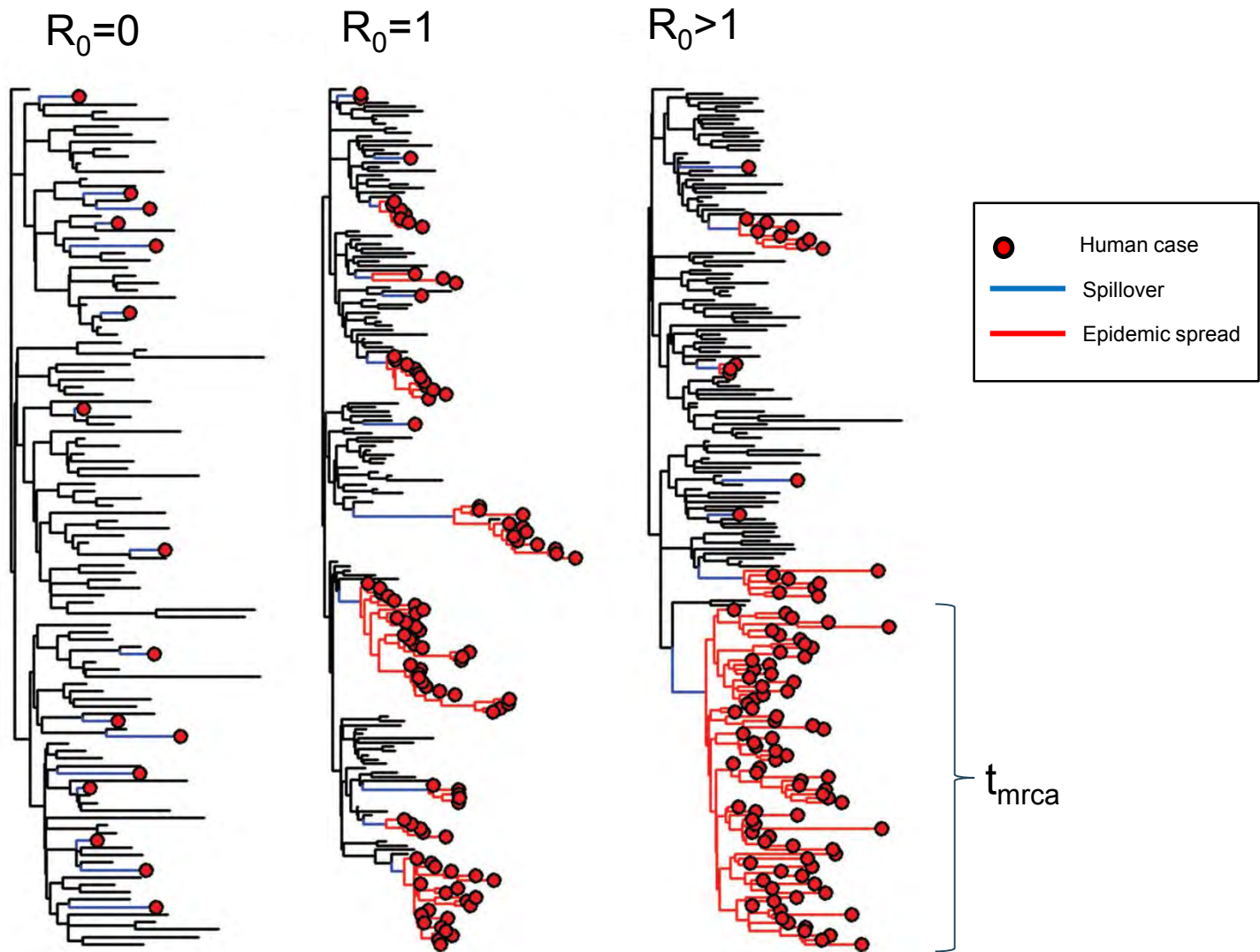
ABSTRACT The standard paradigm for microbiological testing is dependent on the presentation of a patient to a clinician. Tests are then requested based on differential diagnoses using the patient's symptoms as a guide. The era of high-throughput genomic methods has the potential to replace this model for the first time with what could be referred to as "hypothesis-free testing." This approach utilizes one of a variety of methodologies to obtain a sequence from potentially any nucleic acid in a clinical sample, without prior knowledge of its content. We discuss the advantages of such an approach and the challenges in making this a reality.

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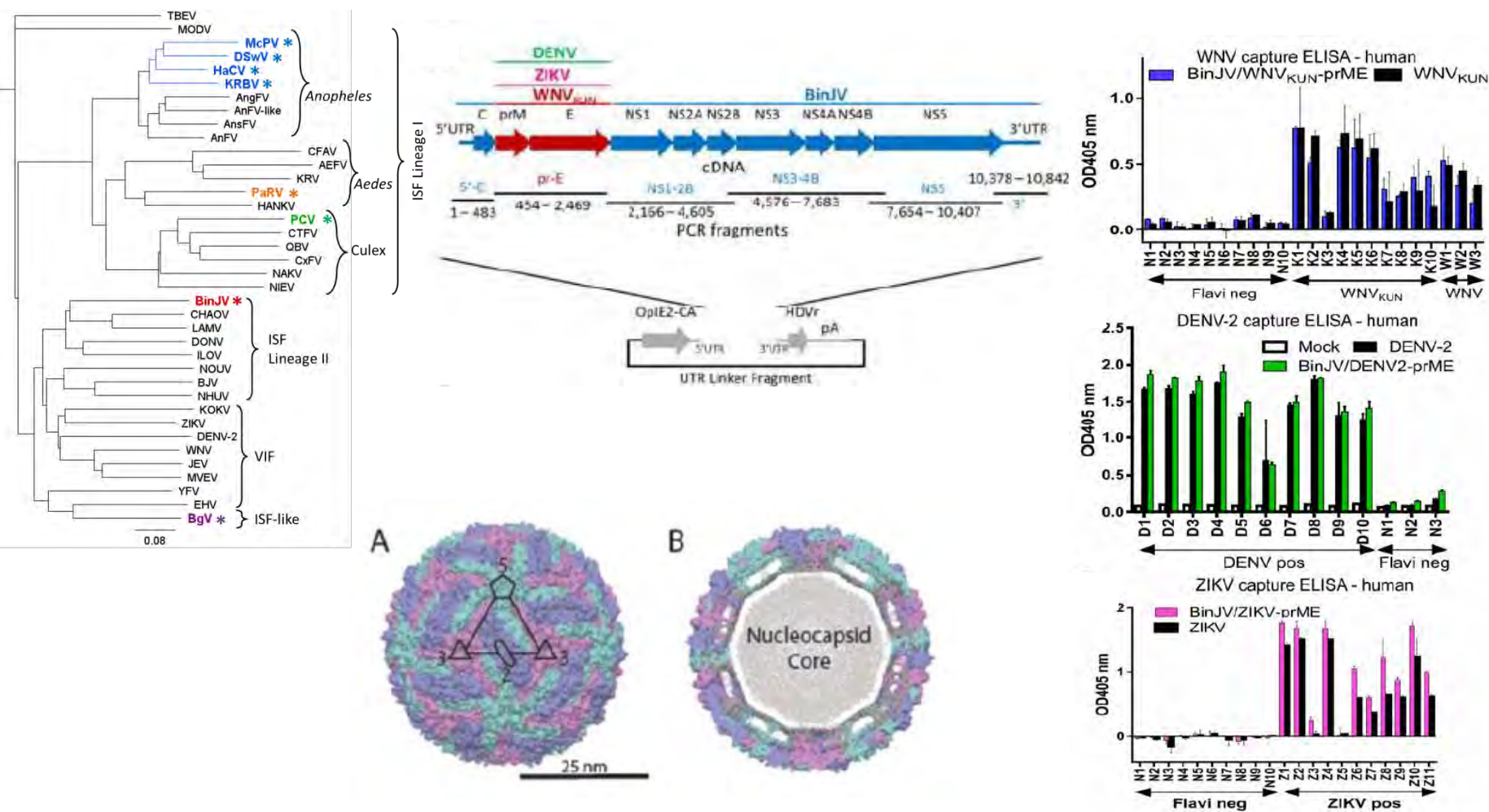
Citation Allcock RJN, Jennison AV, Warrilow DJ. 2017. Towards a universal molecular microbiological test. *J Clin Microbiol* 55:3175–3182. <https://doi.org/10.1128/JCM.01155-17>.

Editor Colleen Suzanne Kraft, Emory University
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Spillover to rapid spread

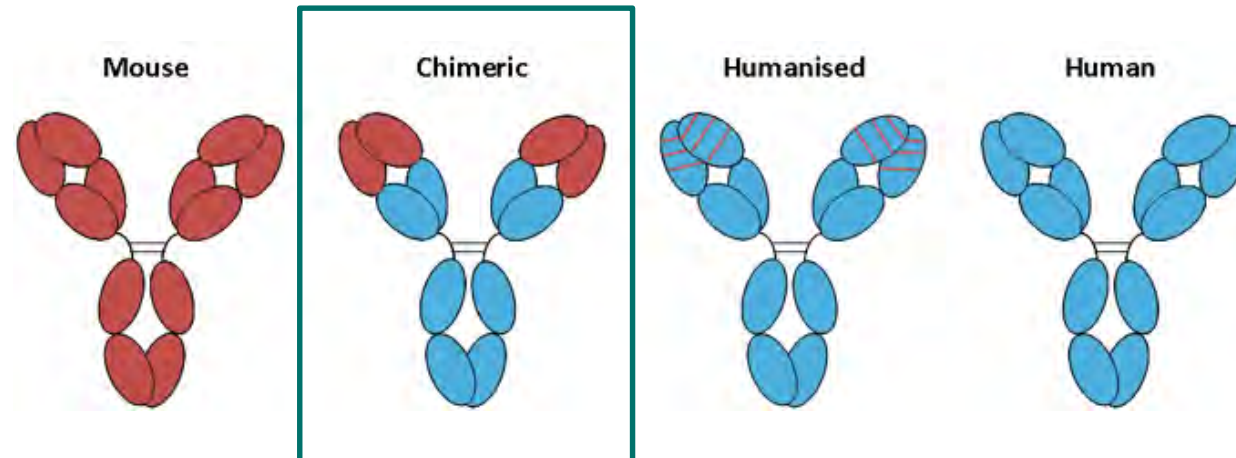
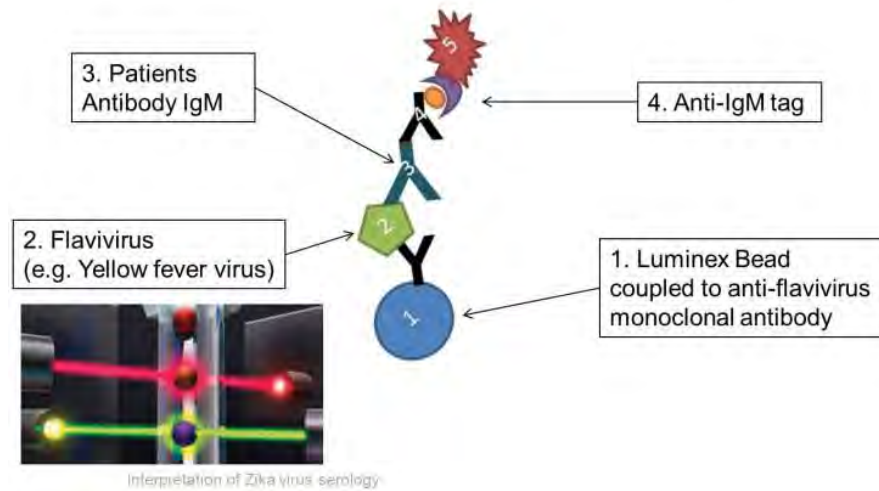


Chimeric antigens



Hobson-Peters et al. A new insect-specific flavivirus provides a recombinant vehicle for efficient production of safe, structurally authentic flavivirus diagnostic antigens. Submitted *Nature Communications*.

Flavivirus IgM MIA



General aspects of licensed vaccines and most advanced vaccine candidates against yellow fever, dengue, chikungunya and Zika viruses*

Virus Family	Main difficulty	Vaccines Licensed Identification/ seed			Under development		Current status	Strategy
			Manufacturer ¹	Strategy	Identification/seed	Manufacturer		
Yellow fever	–	YFV-17DD	Bio-Manguinhos (Fiocruz)	Attenuation by passage in animal, tissue and cell culture	XRX-001	Xcellerex ²	Phase I	YFV-17D inactivated
Flaviviridae		YFV-17D-204	Sanofi Pasteur Pasteur Institute Chiron/Novartis	Attenuation by passage in animal, tissue and cell culture	–	–	–	–
		YFV-17D-213	Federal State Unitary Enterprise of Chumakov Institute	Attenuation by passage in animal, tissue and cell culture				
Dengue Flaviviridae	Multiple serotypes	CYD-TVD	Sanofi Pasteur	Live attenuated chimeric	TV003	NIH/NIAID/Butantan Institute	Phase III (In progress) ³	Live attenuated chimeric
					TDV	Inviragen/Takeda	Phase III (In progress) ⁴	Live attenuated chimeric
Chikungunya	High virulence	–	–	–	TSI-GSD-218	USAMRIID/SalkInstitute	Phase II	Attenuation by passages in cell culture
Togaviridae					VRCeCHKVLP059-00-VP	for Biological Studies NIAID	(Completed) Phase II (In progress) ⁵	VLP
Zika Flaviviridae	GBS ¹⁰ Neurotropism ZVCS ¹¹	–	–	–	GLS–5700	Inovio	Phase I (In progress) ⁶	DNA
					VRC 5288 (ZKADNA085-00-VP)	NIAID	Phase I (In progress) ⁷	DNA
					VRC 5283 (VRC-ZKADNA090-00-VP)	NIAID	Phase I ⁸ /II ⁹ (In progress)	DNA

* Word Health Organization prequalified vaccine (Monath, 2005; Barret et al., 2017).

² Originally developed by Xcellerex. GE Healthcare acquired the intellectual property for the investigational product through its acquisition of Xcellerex in 2012. In 2016, PnuVax acquire GE's intellectual property. See at <http://www.biopharminternational.com/ge-healthcare-sells-rights-inactivated-yellow-fever-vaccine-pnuvax>.

³ Completed in December 2022 (NCT02406729).

⁴ Completed in December 2021 (NCT02747927).

⁵ Completed in December 2017 (NCT02562482).

⁶ In healthy adults: completed in December 2017 (NCT02809443)/in Dengue virus seropositive adults: completed in June 2018 (NCT02887482).

⁷ Completed in December 2018 (NCT02840487).

⁸ Completed in December de 2018 (NC T02996461).

⁹ Completed in January 2020 (NCT03110770).

¹⁰ Guillain-Barré

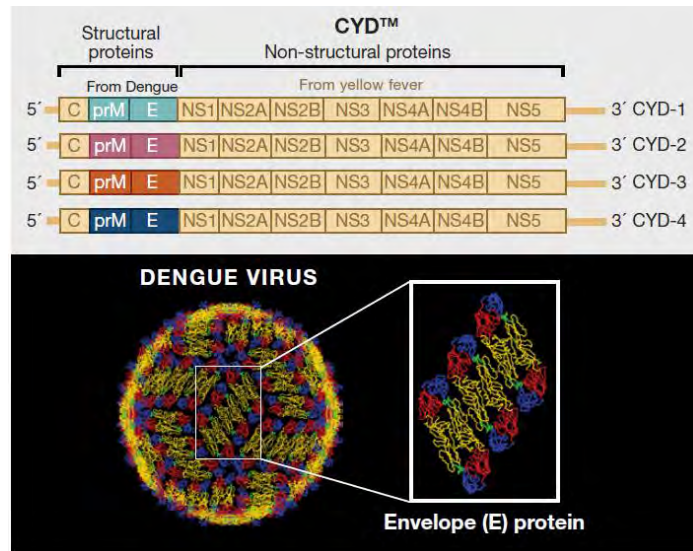
Syndrome.

¹¹ Zika Virus Congenital Syndrome.

Silva IV, Jr, Lopes TB, Oliveira Filho EF, Oliveira RAS, Durães Carvalho R, Gil LHVG. Current status, challenges and perspectives in the development of vaccines against yellow fever, dengue, Zika and chikungunya viruses. Acta Trop. 2018 Jun;182:257-263. doi:

10.1016/j.actatropica.2018.03.009. Epub 2018 Mar 15.

First licensed vaccine - Dengvaxia



Phase III trial results

Asia (Malaysia, Philippines, Thailand and Vietnam)

~75% D3&4 > 50% D1 > 35% D2 (n.s.)

Latin America

~75% D3&4 > 50% D1 > 42% D2

APPROVED for Persons 9–45 years of age living in highly dengue endemic regions of Mexico, the Philippines, Brazil, and El Salvador. WHO SAGE recommends vaccine use in places with high dengue endemicity (seroprevalence 70% in the age group targeted).

INFECTIOUS DISEASE

Safety concerns derail dengue vaccination program

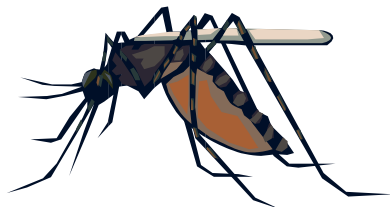
Other vaccine candidates likely to face increased scrutiny

By **Dennis Normile**

The effort to deploy a pioneering vac-

Health Sciences in Bethesda, Maryland, who was among those who warned of possible dangers early on. Its problems will certainly

Science. 2017 Dec 22;358(6370):1514-1515.
doi: 10.1126/science.358.6370.1514.

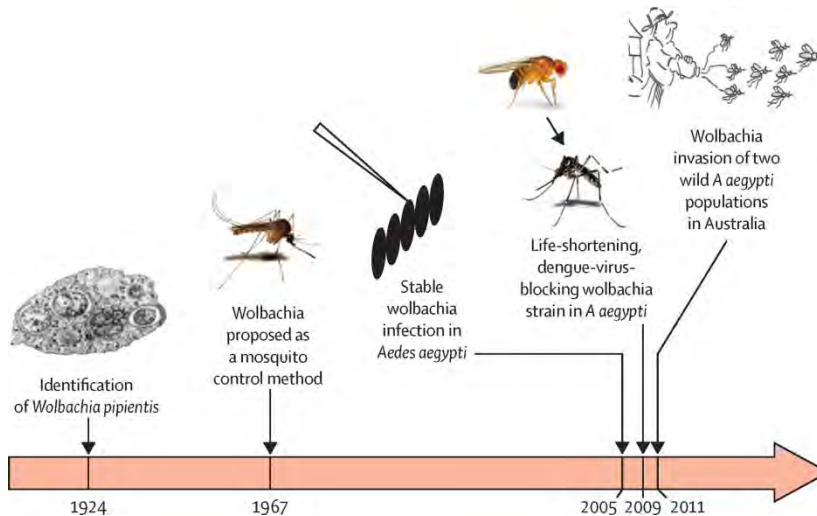
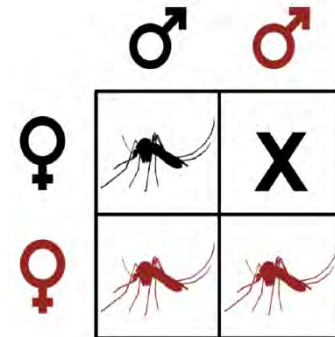


Aedes aegypti



Drosophila melanogaster

Trans-infection



“Eliminate Dengue” program



Cairns Field Trial Update June 2013

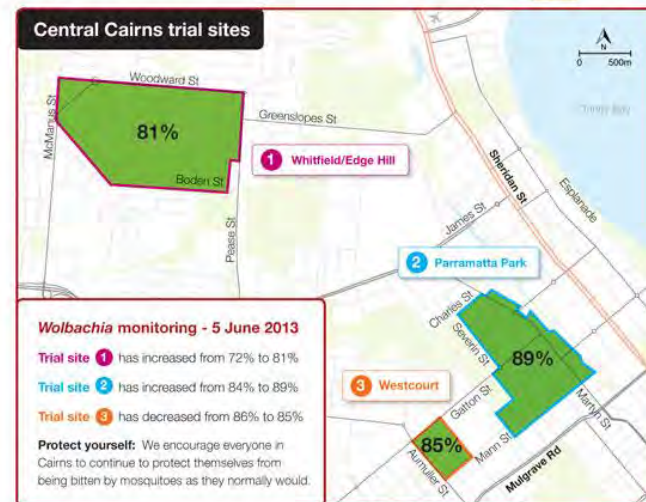


The Eliminate Dengue research program began its third field trial in January 2013 to see if *Wolbachia* – a natural bacterium that reduces a mosquito's ability to transmit dengue – could establish in the inner suburbs of Cairns.

From January to April, we released *Aedes aegypti* mosquitoes that carried *Wolbachia* in areas of Edge Hill, Whitfield, Westcourt and Parramatta Park. We have seen promising results in all three field trial sites, as more than 80% of mosquitoes we have collected carry *Wolbachia*.

The onset of the dry season means our traps are now catching low mosquito numbers across all field trial sites, making accurate results harder to obtain. For this reason, we may not publish our next field trial update until the end of the year, when mosquito numbers increase with the return of the wet season. In the meantime, we will continue our *Wolbachia* monitoring program.

For more information or to join our mailing list, email us at cairns@eliminatedengue.com or call 1800 811 054.



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Opinion

Mission Accomplished? We Need a Guide to the 'Post Release' World of *Wolbachia* for *Aedes*-borne Disease Control

Scott A. Ritchie,^{1,2,*} Andrew F. van den Hurk,³ Michael J. Smout,² Kyran M. Staunton,^{1,2} and Ary A. Hoffmann⁴

Historically, sustained control of *Aedes aegypti*, the vector of dengue, chikungunya, yellow fever, and Zika viruses, has been largely ineffective. Subsequently, two novel 'rear and release' control strategies utilizing mosquitoes infected with *Wolbachia* are currently being developed and deployed widely. In the incompatible insect technique, male *Aedes* mosquitoes, infected with *Wolbachia*, suppress populations through unproductive mating. In the transinfection strategy, both male and female *Wolbachia*-infected *Ae. aegypti* mosquitoes rapidly infect the wild population with *Wolbachia*, blocking virus transmission. It is critical to monitor the long-term stability of *Wolbachia* in host populations, and also the ability of this bacterium to continually inhibit virus transmission. Ongoing release and monitoring programs must be future-proofed should political support weaken when these vectors are successfully controlled.

Highlights

The use of *Wolbachia* to control populations of *Ae. aegypti* mosquitoes, and to reduce transmission of dengue, Zika, and other *Aedes*-borne disease viruses, is growing rapidly. Transinfection that introduces virus-blocking *wMel* *Wolbachia* into *Ae. aegypti* is leading the way, with operational releases in ten countries.

The use of *Wolbachia* to effectively induce male sterility is being used to reduce populations of *Ae. aegypti*, *Aedes polynesiensis*, and *Aedes albopictus* in several international programs.

The authors of this manuscript are: Scott A. Ritchie, Andrew F. van den Hurk, Michael J. Smout, Kyran M. Staunton, and Ary A. Hoffmann.

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