Susceptibility Status of Insecticides Used for Mosquito Control in Asia

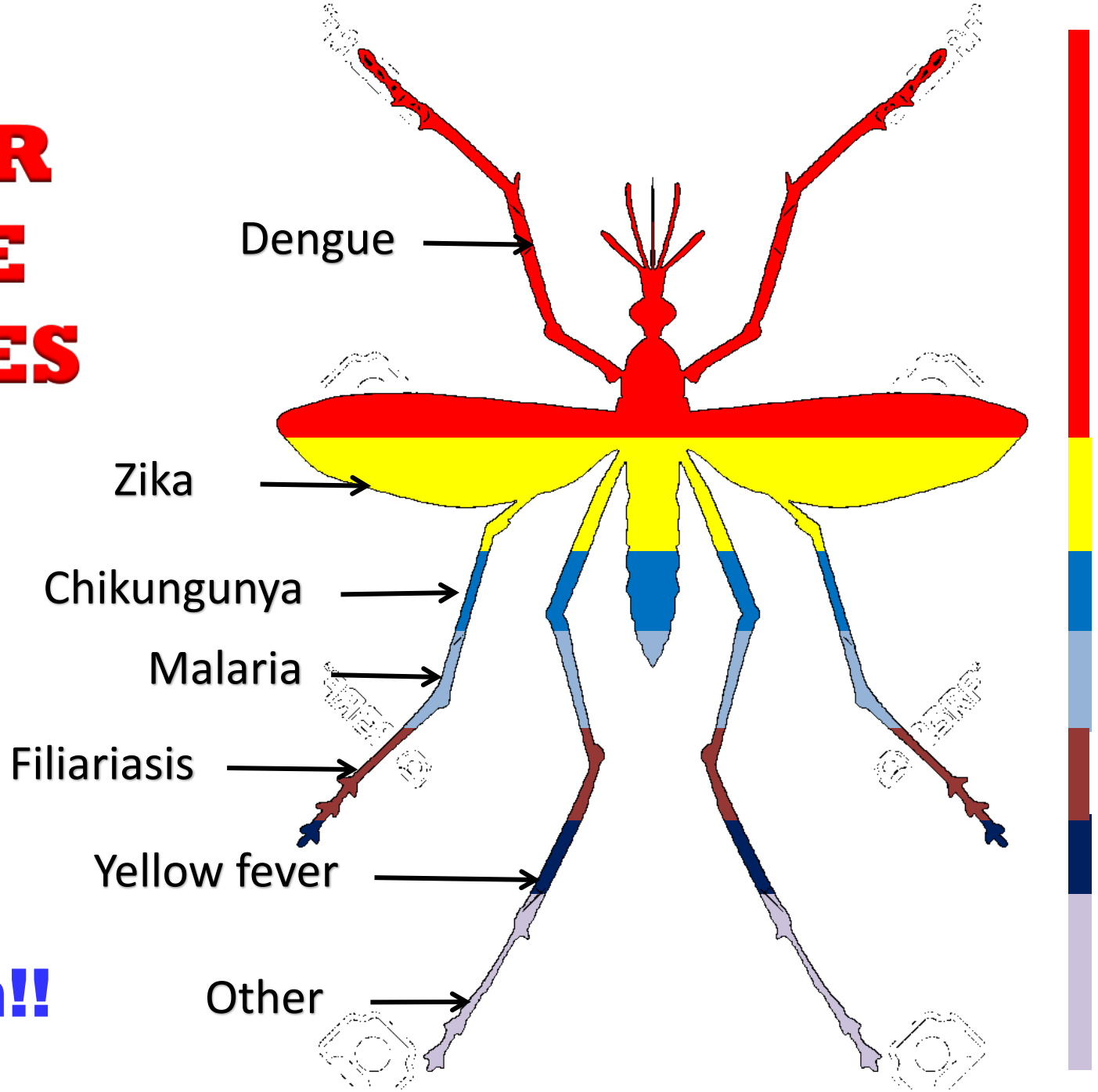
NAZNI WA

Medical Entomology Unit &

WHOCC Vectors, GLP Laboratory IMR

VECTOR BORNE DISEASES

Burden human population!!



Important MOSQUITOES

Mansonia

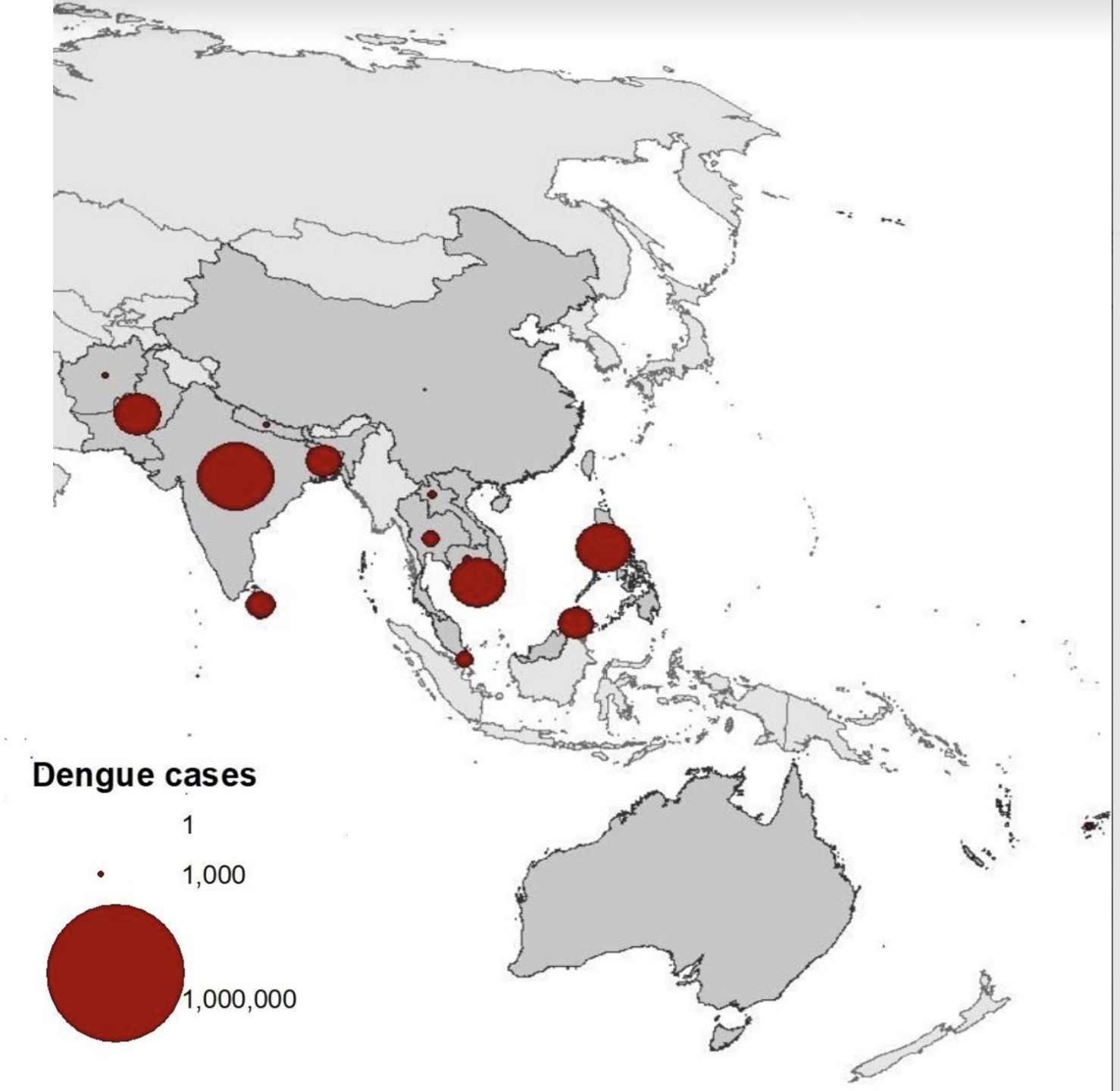
Anopheles

Culex

Aedes

Armigeres

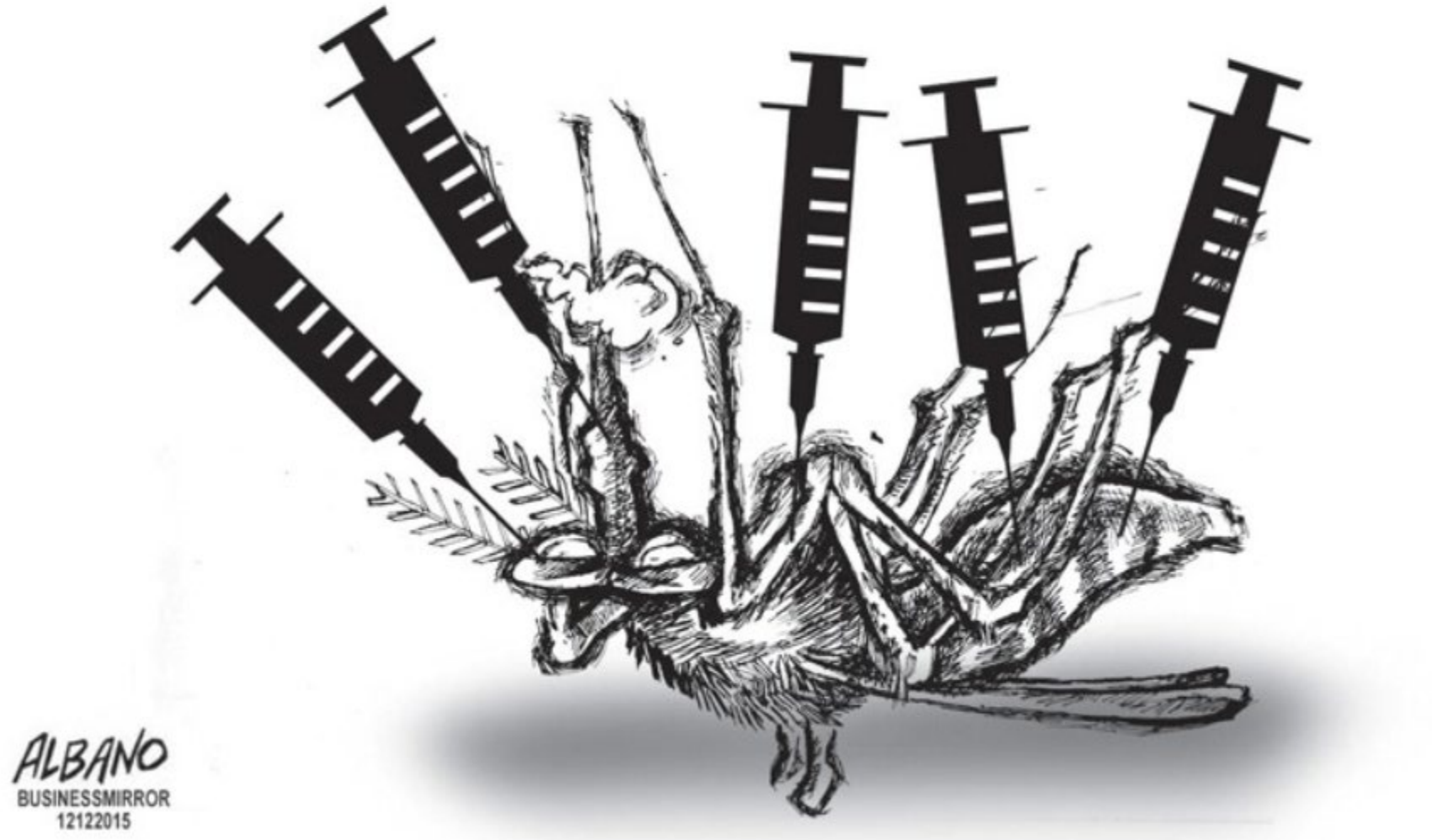
Dengue Burden In Asia



European Centre for Disease Prevention and Control 7Jan 2022

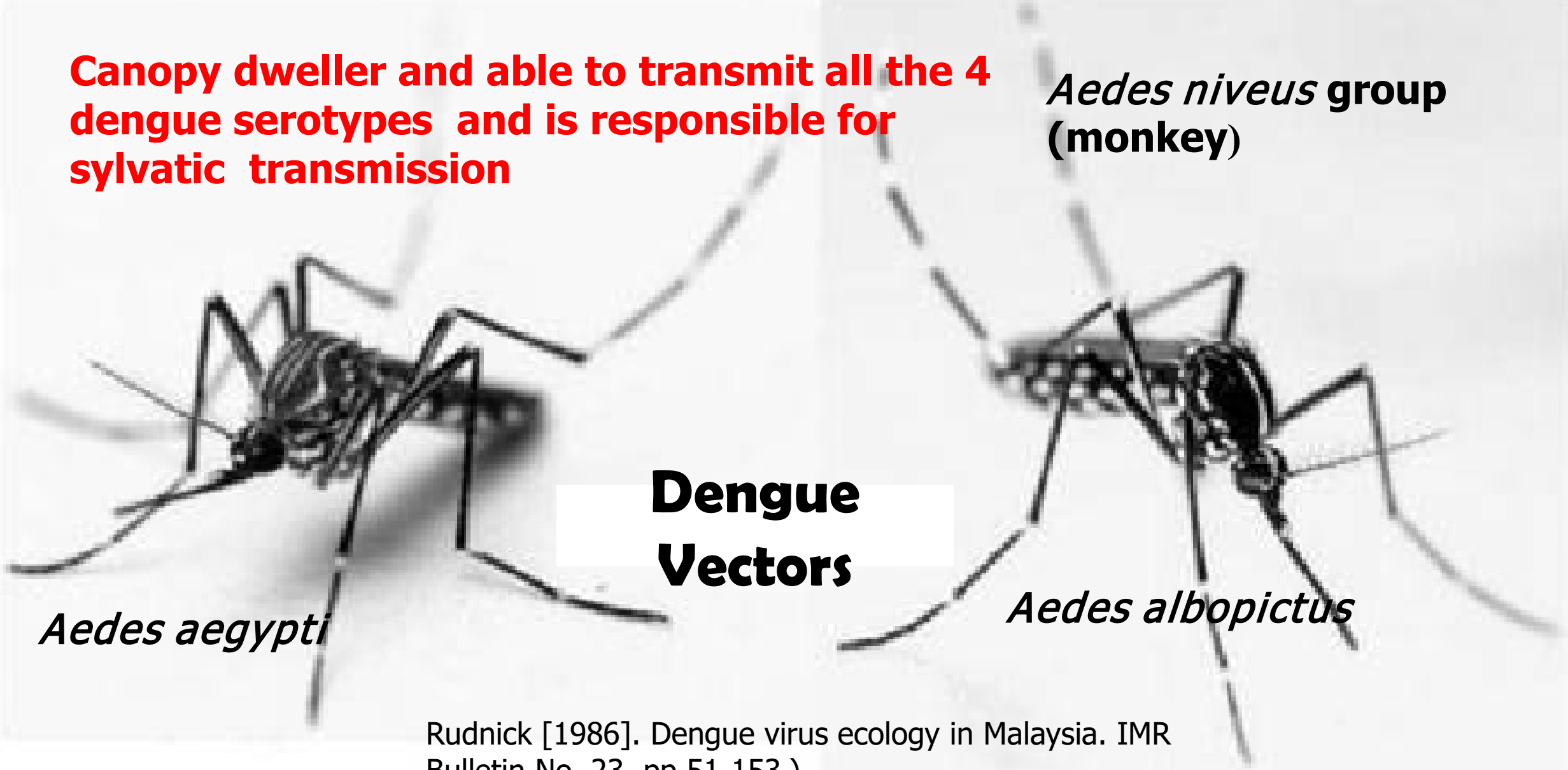
Killing dengue

Opinion by BusinessMirror Editorial - December 12, 2015



Canopy dweller and able to transmit all the 4 dengue serotypes and is responsible for sylvatic transmission

***Aedes niveus* group (monkey)**



Dengue Vectors

Aedes aegypti

Aedes albopictus

Rudnick [1986]. Dengue virus ecology in Malaysia. IMR Bulletin No. 23, pp 51-153.)


NOW EVERYONE CAN FLY
AFF

August 4, 2017 • In the Lab • By **Asian Scientist Newsroom**

Dengue's Spread In Asia Linked To Air Travel

Examining data from Asia over 59 years beginning in 1956, scientists in China show that air traffic is associated with the spread of the dengue virus across Asia.

PUBLIC RELEASE: 3-AUG-2017

Air travel respo dengue throug

PLOS

SHARE

While the incidences of many othe infectious diseases have declined past decade, the number of cases outbreaks of dengue virus have co to increase. The spread of dengue areas is likely due in large part to t air travel, researchers now report *Neglected Tropical Diseases*.

Dengue virus affects as estimated million people around the globe e and can cause symptoms ranging mild fever and headache to severe

In the new work, Huaiyu Tian and Bing Xu, both of Beijing Normal University, China, together with colleagues from the University of Oxford and elsewhere analyzed the spread of dengue viruses in Asia from 1956 to 2015. They used 2,202 genetic sequences of dengue viruses, collected in 20 countries or regions of Asia over the 59 years, to determine how different strains were related. They also investigated trends in air travel, maritime mobility, migration, and socio-economics to determine what factors impact the spread of dengue.

The spread of three different dengue virus serotypes, **DENV-1, -2, and -3, is associated with air traffic more so than any other factors,** the data rev

Tian H, Sun Z, Faria NR, Yang J, Cazelles B, Huang S, et al. (2017) Increasing airline travel may facilitate co-circulation of multiple dengue virus serotypes in Asia. **PLoS Negl Trop** Dis 11(8): e0005694. https://doi.org/10.1371/journal.pntd.0005694

Health & Science

How a tiny mosquito became one of the world's 'most efficient killers'

Female Aedes aegypti mosquitoes rest on the forearm of a health technician in a lab conducting research on preventing the spread of the Zika virus in Guatemala City. (Josue Decavele/Reuters)

World Health Organisation Day

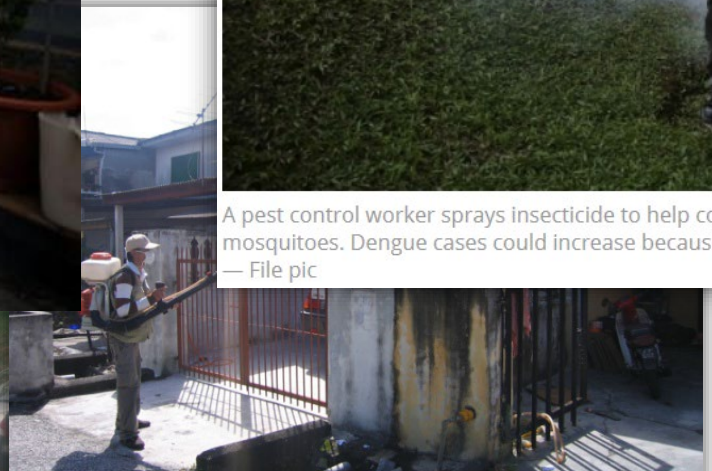
DENGUE Key Issues

- No specific antiviral medication
- No effective vaccine likely to be available for public use
- Current strategy for prevention is community based, integrated mosquito control
- As low as 2-3 female *Aedes aegypti* emerging everyday in a locality of 100 people is sufficient for dengue transmission

Although still widely used are chemical, biological and environmental management techniques, the overall problem remains very

CHALLENGING

A pest control worker sprays insecticide to help control the spread of dengue fever carried by mosquitoes. Dengue cases could increase because of the El Nino phenomena causing the heat wave. — File pic



Dengue control when a single case is reported
thermal fogging for 200 m radius (12.56 ha)

Dengue outbreak:

- Thermal fogging (200 m radius)
- Truck mounted ULV (400m radius)
- Same insecticide used for thermal fogging and ULV
- Repeat fogging 7-10 days later

Cost Estimation of Current Dengue Control Measures

Type of Control	Average Cost per Case (RM)	No of Dengue Cases per District	Total Cost of Dengue per District (RM)	Total Cost of Dengue in Selangor [4 Districts] (RM)
Current Control	**5432**	**1034**	**5.6 million**	**22.4 million**

Packierisamy et al. 2015. The Cost of Dengue Vector Control Activities in Malaysia by Different Service Providers. *46th APACPH Conference Supplement. DOI: 10.1177/1010539515589339*

IDEAL DENGUE Vector Control Tool

1. Scientifically Proven
2. Affordable
3. Sustainable

9 Mei 2017

Chemical Pesticides have been used for **Vector Control** for at least **75 years**.

Insectides for mosquito control

Years	WHO approved insecticides			
1940-45	DDT			
1946-50	Lindane			
1951-55	Malathion			
1956-60				
1961-65	Fenitrothion	Propoxur		
1966-70	Chlorpyrifos-methyl			
1971-75	Pirimiphos-methyl	Bendiocarb	Permethrin	
1976-80	Cypermethrin			
1981-85	Alpha-cypermethrin	Cyfluthrin		
	Lambda-cyhalothrin	Deltamethrin	Bifenthrin	
1986-90	Etofenprox			
1991-95				
1996-00				
2001-05				

Organochlorines
Organophosphates
Carbamates
Pyrethroids

Only a limited number of insecticide classes are available for adult mosquito control. No new malaria mosquito adulticide has been approved by the WHO in the last 15 years.

Annual insecticide use in vector control operations in 2010–2019 and in 2000–2009, by region and insecticide class, expressed in metric tonnes of active ingredient (WHO 2021)

WHO region	2000–2009					2010–2019				
	OC	OP	C	PY	NN	OC	OP	C	PY	NN
African	805	20	19	36	0	337	390	335	34	35
Americas	0	455	4	230	0	0	1104	287	76	0
Eastern Mediterranean	0	47	5	22	0	0	15	47	25	1
European	0	3	0	2	0	0	1	0	0	0
South-East Asia	3623	547	2	45	0	2977	92	3	31	0
Western Pacific	0	302	0	80	0	0	23	6	29	0
All	4429	1375	30	414	0	3314	1625	677	194	36

C, carbamates; NN, neonicotinoids; OC, organochlorines; OP, organophosphates; PY, pyrethroids

Annual global use of insecticides for vector control operations as reported to WHO, specified per disease (or public health pest) and insecticide class, and expressed in metric tonnes of active ingredient. Data were weighted and pooled over the period 2010–2019.

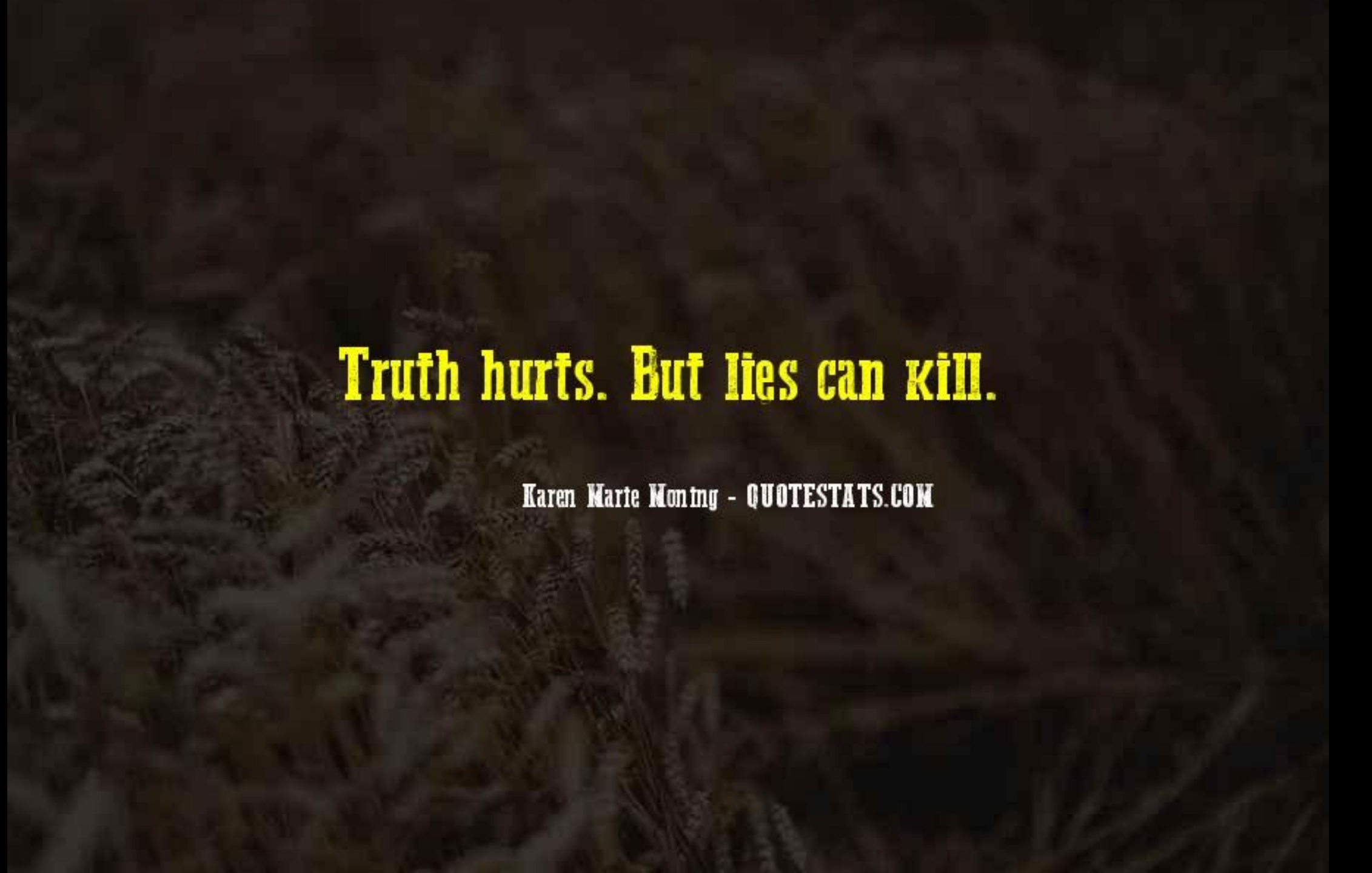
Disease or pest	Vector	Insecticide class								Total
		OC	OP	C	PY	NN	BL	IGR	SP	
Disease										
Malaria	Anopheline mosquitoes	2619	526	358	96	36	23	3	3	3662
Dengue	Aedine mosquitoes	0	1097	173	71	0	61	17	13	1433
Leishmaniasis	Phlebotomine sandflies	695	0	16	14	0	0	0	0	725
Chagas disease	Triatomine bugs	0	0	118	9	0	0	0	0	127
Lymphatic filariasis	Mosquitoes	0	0	12	0	0	0	0	0	12
Flea- and lice-borne diseases	Fleas, lice	0	0	0	3	0	0	0	0	3
Zika virus disease	Aedine mosquitoes	0	1	0	1	0	0	0	0	1
Chikungunya	Aedine mosquitoes	0	1	0	0	0	0	0	0	1
Japanese encephalitis	Culicine mosquitoes	0	0	0	1	0	0	0	0	1
Onchocerciasis	Simuliid blackflies	0	0	0	0	0	0	0	0	0
Schistosomiasis	Snails	0	0	0	0	0	0	0	0	0
Tungiasis	Sand fleas	0	0	0	0	0	0	0	0	0
Tick-borne diseases	Ticks	0	0	0	0	0	0	0	0	0
Pest										
Nuisance mosquitoes	Not applicable	0	23	5	16	0	2	0	1	47
Flies and other pests	Not applicable	0	0	3	0	0	0	0	0	3

BL, bacterial larvicides; C, carbamates; IGR, insect growth regulators; NN, neonicotinoids; OC, organochlorines; OP, organophosphates; PY, pyrethroids; SP, spinosyns


Institute For Medical Research: 1900 -

Insecticides

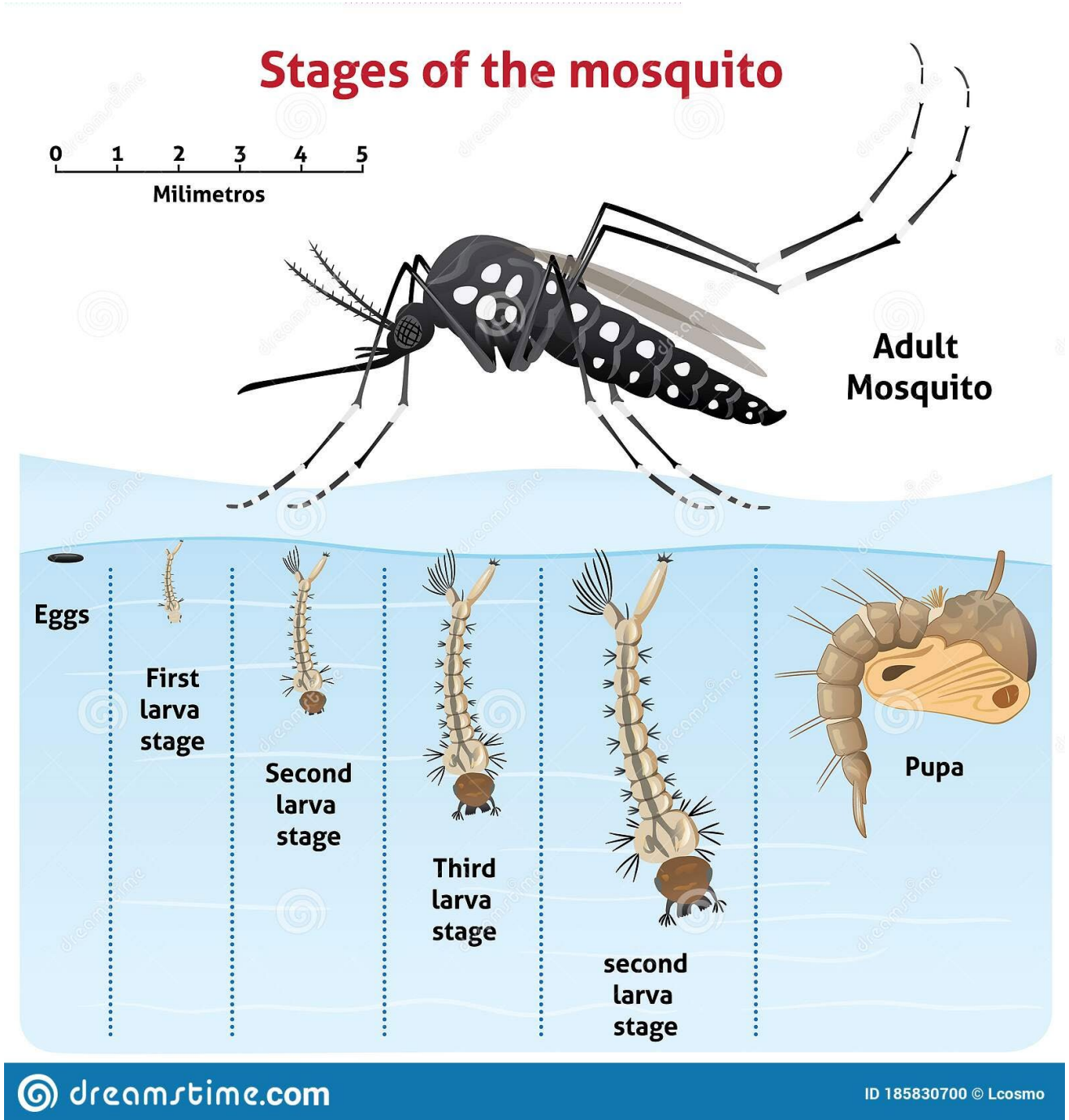
RESISTANCE


Truth hurts. But lies can kill.
Karen Marie Moning - QUOTESTATS.COM

Misuse and Overuse

Under and overdosing, bad spray coverage, extended spraying intervals of pesticides on a large scale for insect-vector and agricultural pest control has led to resistance development.

Resistance is <u>NOT</u> restricted to one or the other life stage



A population may be termed **resistant** when its **LC50** has increased by **10 times** (Knipling, 1950).

The **larval test** is **more sensitive** than the **adult test** in **detecting changes in susceptibility level**

Adult LC50	Larval LC50
2 X increase	**10 X increase**

Brown, 1986

Table 1. Insecticide resistance status and biochemical mechanisms involved in *Aedes aegypti*. (Bharati *et al*., 2021)

Tested Insecticide	Resistance status and region	Mechanism of resistance
DDT	R (Sri Lanka[12], Malaysia[28-30], Pakistan[31-33], India[36-38])	Elevated activity of detoxifying enzymes[28]
		Target site modified via kdr mutation[28,38]
Dieldrin	R (Malaysia[28])	*
	S (Malaysia[28], India[36])	
	IR (Malaysia[30])	
Temephos	R (Malaysia[28], Indonesia[46-49], India[37,54], Sri Lanka[55])	Elevated activity of detoxifying enzymes[28,48]
	S (India[36,54,56], Thailand[59])	
	IR (Pakistan[33], India[54], Singapore[62])	
Malathion	R (Sri Lanka[12], Malaysia[28,65], Pakistan[32,33], Indonesia[47,49,64])	Metabolic detoxification through enzymes[12,28,48]
	S (Sri Lanka[12], Malaysia[28,29], Pakistan[31], India[37], Indonesia[48])	
	IR (Malaysia[30], India[36])	Resistance against malathion related to CCEs[12]
Fenitrothion	R (India[36])	*
	S (Malaysia[29])	
Primiphos methyl	S (Malaysia[22], Singapore[69])	*
Dichlorvos	R (India[70])	*
Lambdacyhalothrin	R (Malaysia[22], Pakistan[31,32], India[70], China[71])	*
	S (Pakistan[32])	
	IR (Pakistan[31], India[36], China[71]	
Deltamethrin	R (Malaysia[28, 30], Pakistan[31,32], India[36,38], Sri Lanka[55], Singapore[69])	Elevated activity of metabolic enzymes[28,30]
	S (India[37])	Target site resistance through prevalent kdr mutations {28,38}
	IR (Sri Lanka[55],Thailand[59])	
Permethrin	R Malaysia[28-30], Pakistan[33], India[38], Indonesia[48,75], Thailand[59], Singapore[62,69])	Metabolic detoxification through enzymes[28,30,48]
	IR (Pakistan[31], India[36], Thailand[59])	Target site resistance through prevalent kdr mutations[28]
Cyfluthrin	IR (Malaysia[29], India[36])	*
Cypermethrin	IR (China[71])	*
Etofenprox	R (Singapore[62,69])	*
Propoxur	R (Sri Lanka[12], Malaysia[29], India[56])	*
	S (Sri Lanka[12], India[36])	
Bendiocarb	R (Malaysia[28-30], Pakistan[31,56], , India[36])	Metabolic detoxification through enzymes[28,30]
	IR (Pakistan[32])	

Note: R: Resistant, IR: Incipiently resistant, S: Susceptible, Kdr mutation: Knockdown resistance mutation, CCEs:Carboxylesterases, *: Mechanism not found/studied

Summary for *Ae.aegypti*: Resistant & Incipient Resistant

Tested Insecticide	Country							
	China	India	Indonesia	Malaysia	Pakistan	Singapore	Sri Lanka	Thailand
DDT			✓	✓	✓		✓	
Dieldrin				✓				
Temephos		✓	✓	✓	✓	✓	✓	
Malathion		✓	✓	✓	✓		✓	
Fenitrothion		✓						
Dichlorvos		✓						
Lambdacyhalothrin	✓		✓	✓	✓			
Deltamethrin		✓		✓	✓	✓	✓	✓
Permethrin		✓	✓	✓	✓	✓		✓
Cyfluthrin		✓		✓				
Cypermethrin	✓							
Etofenprox						✓		
Propoxur		✓		✓			✓	
Bendiocarb		✓		✓	✓			

Table 2. Insecticide resistance status and biochemical mechanisms involved in *Aedes albopictus* (Bharati *et al*., 2021)

Tested insecticide	Resistance status and region	Mechanism of resistance
DDT	R (Sri Lanka[12], Malaysia[30], Pakistan[31,33], India[36,37,76,77], China[79])	Enhanced activity of insecticide detoxifying enzymes,
	S (Malaysia[28,30])	CYP450s[30], CCEs[76], GSTs[76])
	IR (Malaysia[30])	
Dieldrin	S (Malaysia[28,30], India[36])	*
	R (Malaysia[30])	
	IR (Malaysia[30])	
Temephos	R (India[36,37], Sri Lanka[55], China[79], Malaysia[80])	Elevated activity of insecticide detoxification enzymes, CCEs[7]
	S (India[7], Malaysia[28], India[77])	
	IR (India[7], Pakistan[33], Sri Lanka[55])	
Malathion	R (Malaysia[30], Pakistan[33])	*
	S (India[7], Malaysia[28,30], Pakistan[31], India[36,37,77])	
	IR (Malaysia[30], India[37],	
Fenitrothion	R (India[36])	*
	S (India[36])	
Primiphos methyl	R (Malaysia[22])	*
Dichlorvos	S (China[79])	*
Lambdacyhalothrin	R (Malaysia[22], Pakistan[31])	*
	S (India[7], Pakistan[33])	
	IR (Pakistan[31], India[36])	
Deltamethrin	R Malaysia[28], Pakistan[31], China[79])	Elevated activity of metabolic enzymes[28,30]
	S (India[7,36,37,76,77], Malaysia[28,30], Pakistan[33], Sri Lanka[55])	Target site resistance through prevalent kdr mutations[28]
	IR (Malaysia[30])	
Permethrin	R (Sri Lanka[12], Malaysia[28], Pakistan[31,33], India[36], China[79])	Target site modification via kdr based mechanism[12]
	S (Malaysia[28,30]	Enhanced detoxification via CYP450s[28]
	IR (Malaysia[30])	
Cyfluthrin	IR (India[36])	*
Cypermethrin	R (China[79])	*
Propoxur	R (China[79])	*
	S (Sri Lanka[12], India[36]	
Bendiocarb	R (Malaysia[30], Pakistan[31,33], India[36])	Enhanced activity of detoxifying enzymes through CYP450s[30]
	S (Malaysia[28,30])	
	IR (Malaysia[30], Pakistan[31])	
Note: R: Resistant, S: Susceptible, IR: Incipiently resistant. Kdr mutation: Knockdown resistance mutation. CCEs: Carboxylesterases.		
GSTs: Glutathione S-transferase. CYP450s: Cytochrome P450s. *: Mechanism not found/studied.		

Summary for *Ae.albopictus*: Resistant & Incipient Resistant

Tested Insecticide	Country							
	China	India	Indonesia	Malaysia	Pakistan	Singapore	Sri Lanka	Thailand
DDT	Δ	Δ		Δ	Δ		Δ	
Dieldrin		Δ		Δ				
Temephos	Δ	Δ		Δ	Δ		Δ	
Malathion		Δ		Δ	Δ			
Fenitrothion		Δ						
Primiphos methyl				Δ				
Lambdacyhalothrin		Δ		Δ	Δ			
Deltamethrin	Δ			Δ	Δ			
Permethrin	Δ	Δ		Δ	Δ		Δ	
Cyfluthrin		Δ						
Cypermethrin	Δ							
Propoxur	Δ							
Bendiocarb		Δ		Δ	Δ			

Insecticide Resistance Status in Malaysia

Species	Resistance to Insecticide
Aedes aegypti	DDT Permethrin Deltamethrin Alpha cyfluthrin Temephos
Aedes albopictus	Malathion Fenitrothion
Culex quinquefasciatus	Malathion Fenetrothion Permethrin
Anopheles maculatus	Still susceptible to insecticides

MONITORING RESISTANCE IS NECESSARY TO ENSURE THAT EFFECTIVE INSECTICIDES ARE BEING USED AND THAT CHANGES IN INSECTICIDE POLICY ARE BASED ON SOUND SCIENTIFIC DATA.

- Integrated management of insecticide resistance among dengue vectors using non-insecticidal methods, rotation of insecticides, standardizing contemporary methods for detection and management of resistance, assessing the impact of resistance on dengue prevention programmes, and monitoring the role of unregulated and broad-spectrum usage of insecticides (e.g. agricultural pest, nuisance fly control, household products) and its possible role in insecticide resistance.

Knockdown resistance status of field strains *Aedes aegypti* adults exposed to 0.75% permethrin.

Strain	State/ District	Filial generation	KT_{50} (min)	95% CI	Regression line	KD Resistance Ratio
Ridzuan Condominium	Petaling Jaya, Selangor	F1	293.26	114.77 – 12446.39	Y= 1.16X-5.25	**26.57**
Taman Sungai Jelok	Ulu Langat, Selangor	F1	44.56	40.37 – 50.20	Y= 2.34X-11.72	4.00
Seksyen 15, Bandar Baru Bangi	Selangor	F1	6613.06	N.A.	Y=0.6X-6.93	**598**
Taman Seri Bunga, Kuala Perlis	Kuala Perlis	F1	98.66	78.65 – 145.61	Y=2.54X-8.64	8.85
Flat Paya Nahu Baru, Sungai Petani	Kedah	F1	20.28	18.75 – 21.72	Y=4.96X-15.22	1.8
Bandar Rincing	Selangor	F1	5.7	4.86 – 6.50	Y= 3.21x – 9.49	0.51
Sri Damansara	Selangor	F1	14.72	13.46 – 14.98	Y=9.03X-10.88	1.32
Susceptible strain	IMR lab strain	F1046	11.15	10.17 – 12.10	Y= 4.21X-13.10	-

(Rosilawati et al., 2016)

Insecticide evaluation in an operational setting

If there is continuous increase in dengue cases in the hotspot locality, even after 2 cycle of adulticide treatment, MOH will request IMR to evaluate the efficacy of the insecticide used in that area.

1. WHO Insecticide Test Kit Evaluation

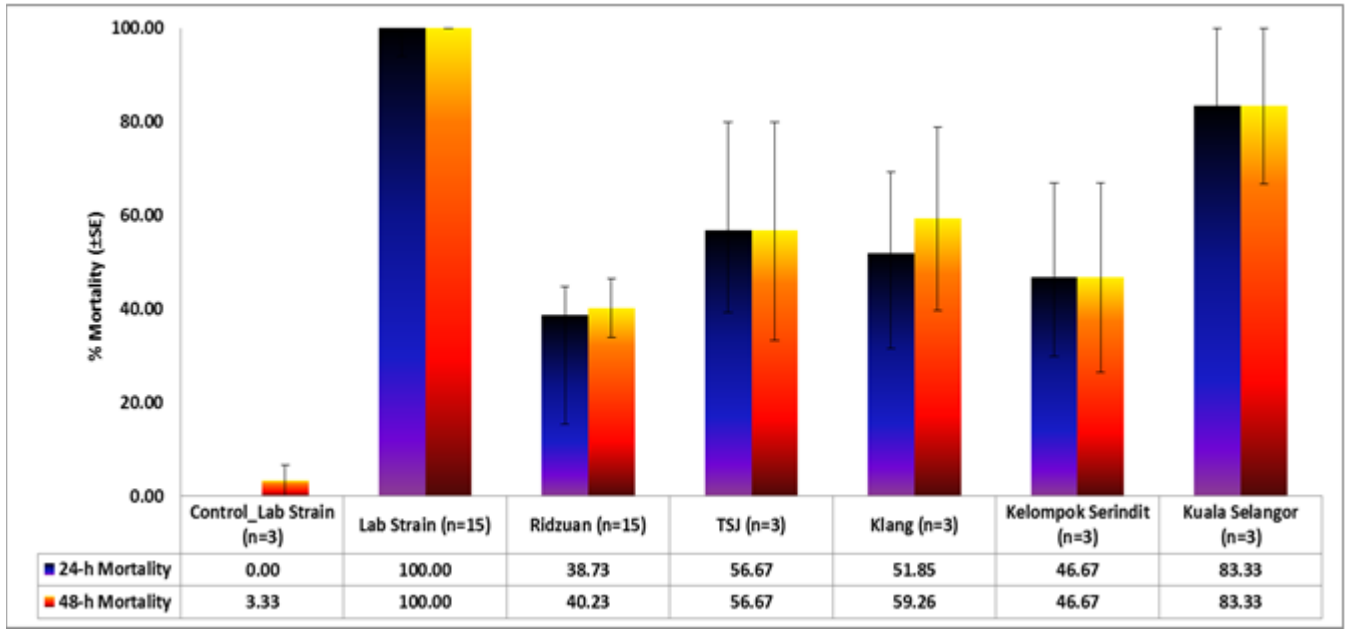
Mortality	Mean Mortality (%)±SE	
	24-h	48-h
Lab Strain (n=12)	75.33±12.28	75.67±12.31
Field Strain (n=3)	26.67±26.67	26.67±26.67
Control (n=3)	0.00±0.00	0.00±0.00

2. Field Evaluation

Cylindrical cages with *Aedes* mosquitoes in the living room

2. Field Evaluation

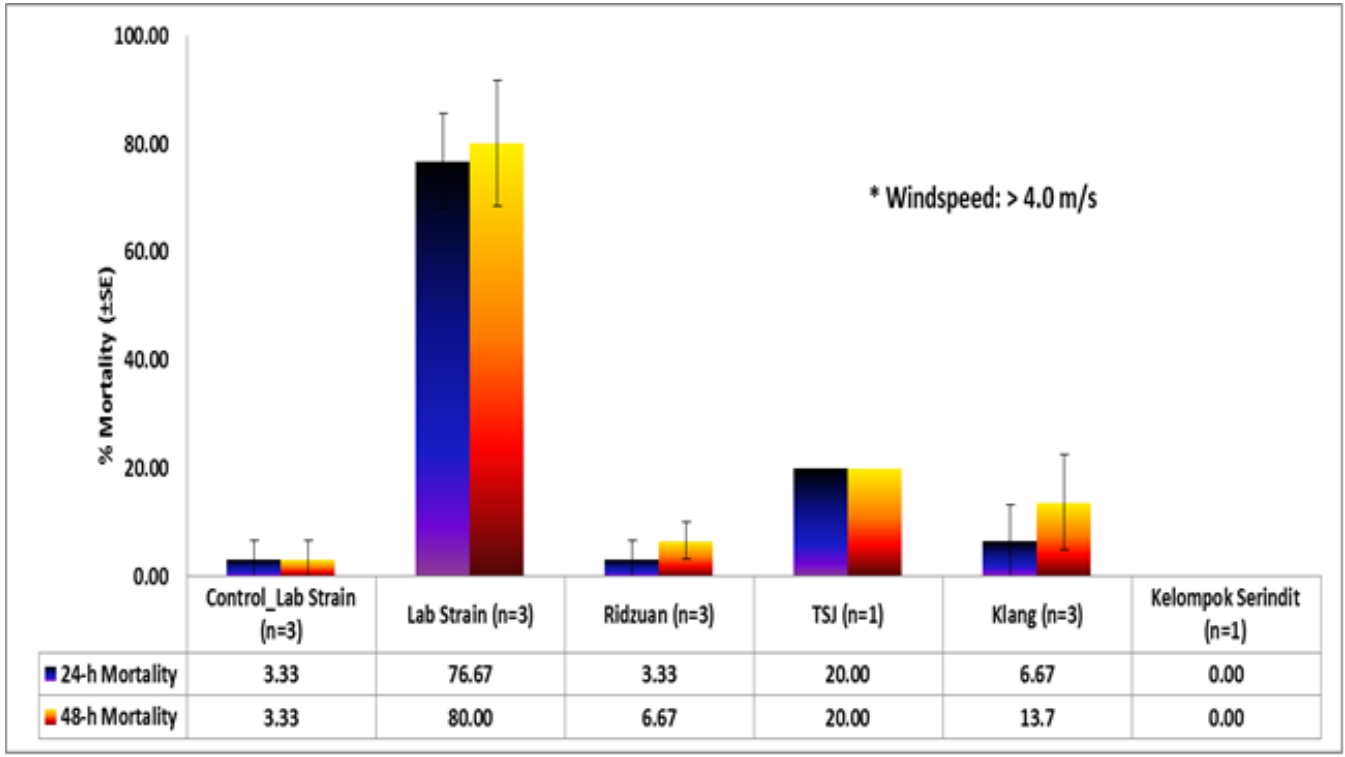
Cylindrical cages with Aedes mosquitoes in the bed room



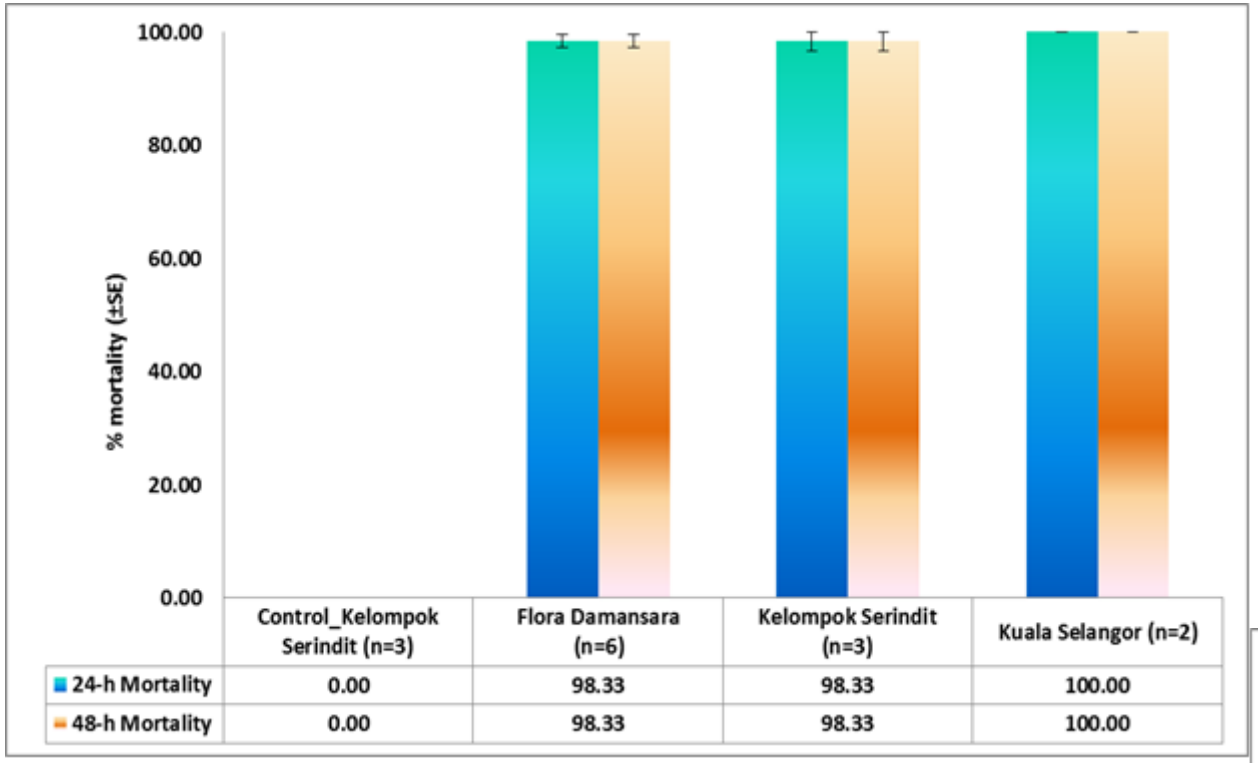
	Control_Lab Strain (n=3)	Lab Strain (n=15)	Ridzuan (n=15)	TSJ (n=3)	Klang (n=3)	Kelompok Serindit (n=3)	Kuala Selangor (n=3)
24-h Mortality	0.00	100.00	38.73	56.67	51.85	46.67	83.33
48-h Mortality	3.33	100.00	40.23	56.67	59.26	46.67	83.33

Mortality of adult female *Ae aegypti* field and lab strains exposed to cyphenothrin via thermal fogging.

Mortality of adult female *Ae aegypti* field and lab strains exposed to cyphenothrin via ULV spraying.

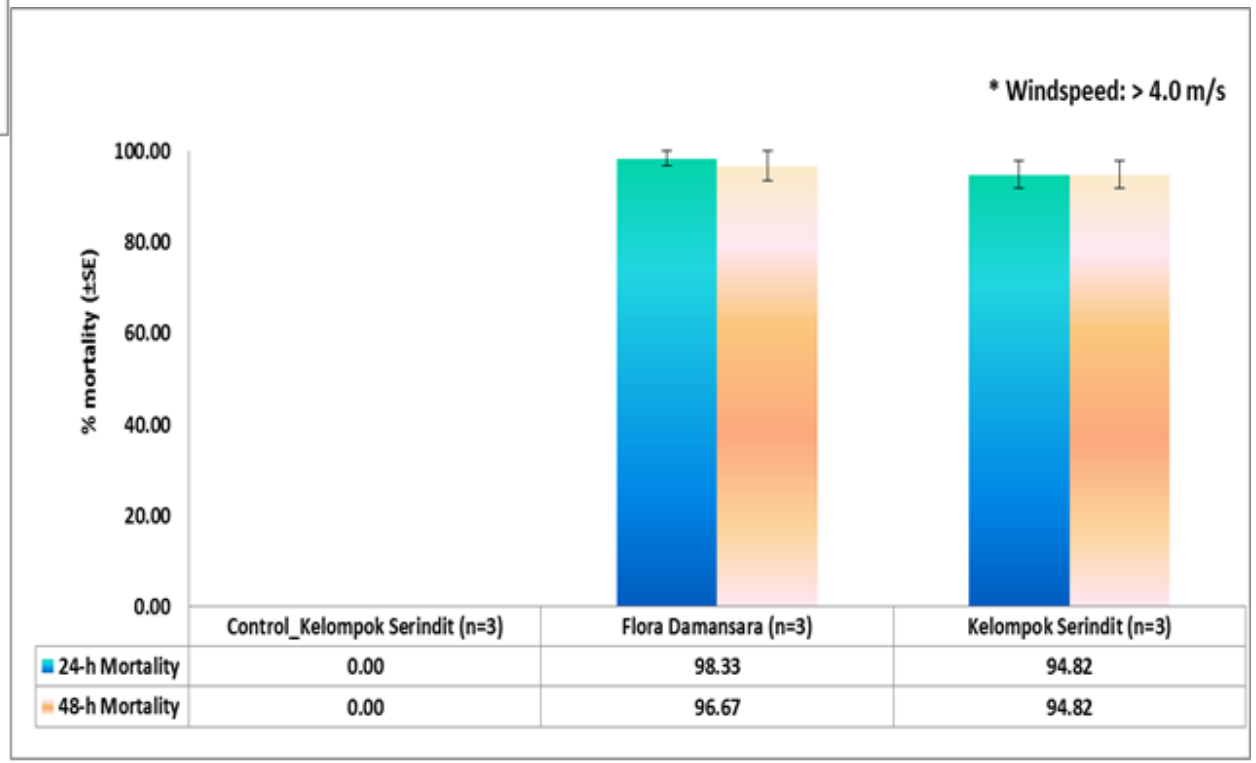


	Control_Lab Strain (n=3)	Lab Strain (n=3)	Ridzuan (n=3)	TSJ (n=1)	Klang (n=3)	Kelompok Serindit (n=1)
24-h Mortality	3.33	76.67	3.33	20.00	6.67	0.00
48-h Mortality	3.33	80.00	6.67	20.00	13.7	0.00



Mortality of adult female *Aedes albopictus* field and lab strains exposed to cyphenothrin via thermal fogging.

Mortality of adult female *Aedes albopictus* field and strains exposed to cyphenothrin via ULV



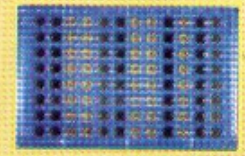
- Resistance surveillance is crucial in providing **baseline data** for control program and pesticide selection
- **Early detection** of resistance in mosquito populations helps in **preventing failure** in disease control and management.

IMR RAPID INESCTICIDE RESISTANCE TEST KIT

Malaysia-PI20024414
USA- 10/321,723
Japan- P2002-371148
EU- 02258794.3
China- 02158931.3
Australia- 2002318878

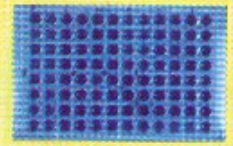
PATENTS FILED

TEST RESULTS



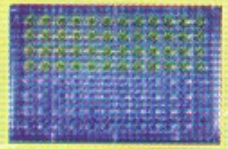
Esterase

COLOUR	SCRORE	RESULT
Colourless / very faint yellow	0	Highly susceptible
Yellowish blue	1	Susceptible
Light Blue	2	Moderately resistant
Dark Blue	3	Highly resistant



Oxidase

COLOUR	RESULT
Light Blue	Susceptible
Dark Blue	Resistant



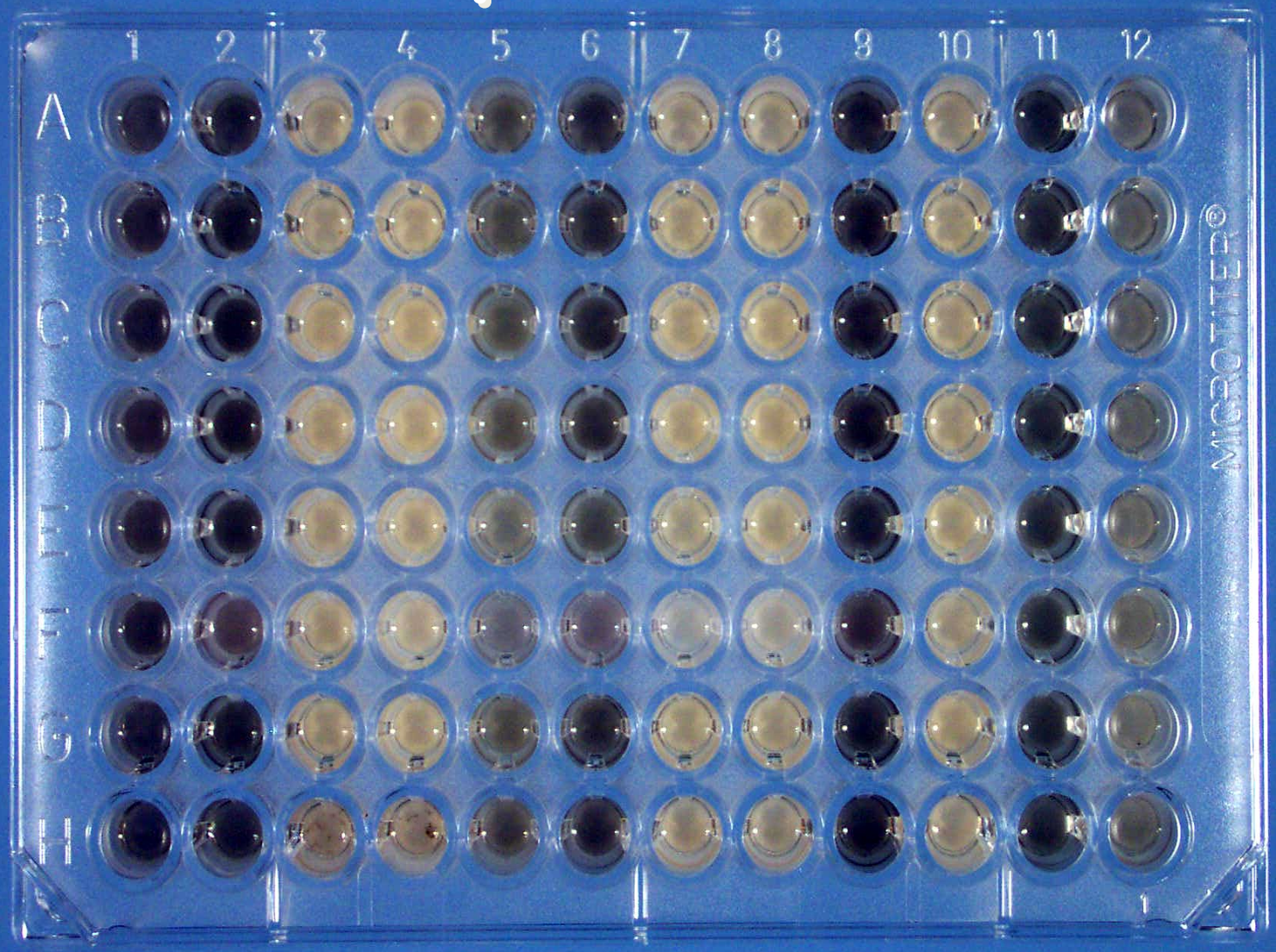
Acetylcholinesterase

COLOUR	RESULT
Colourless	Sensitve
Bright Yellow	Insensitive

TEST PROCEDURES

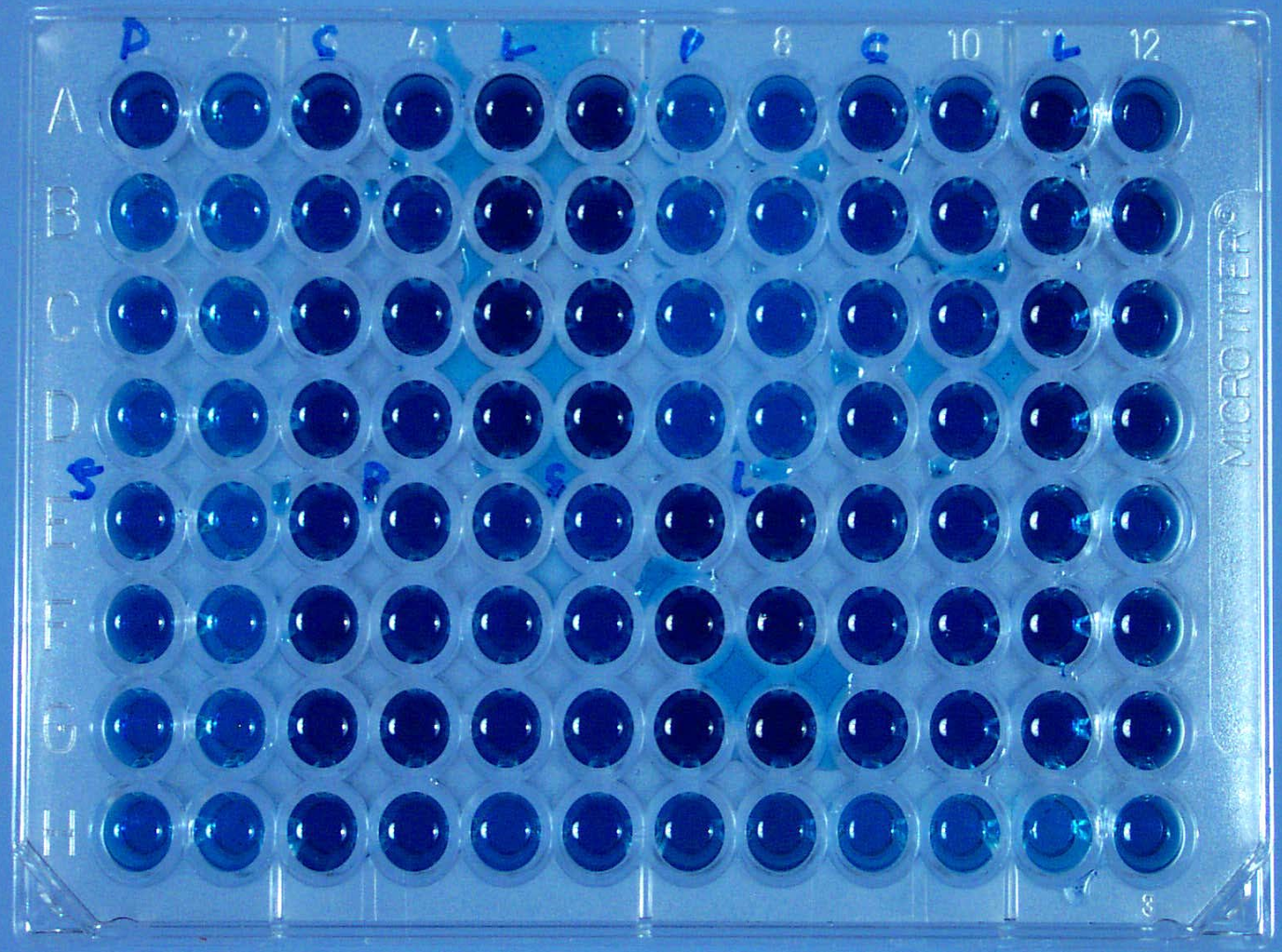
- Homogenise Insect
- Add Substrate
- Add Indicator
- Read (colour code)

Non-specific esterases



Organophosphates/Carbamates

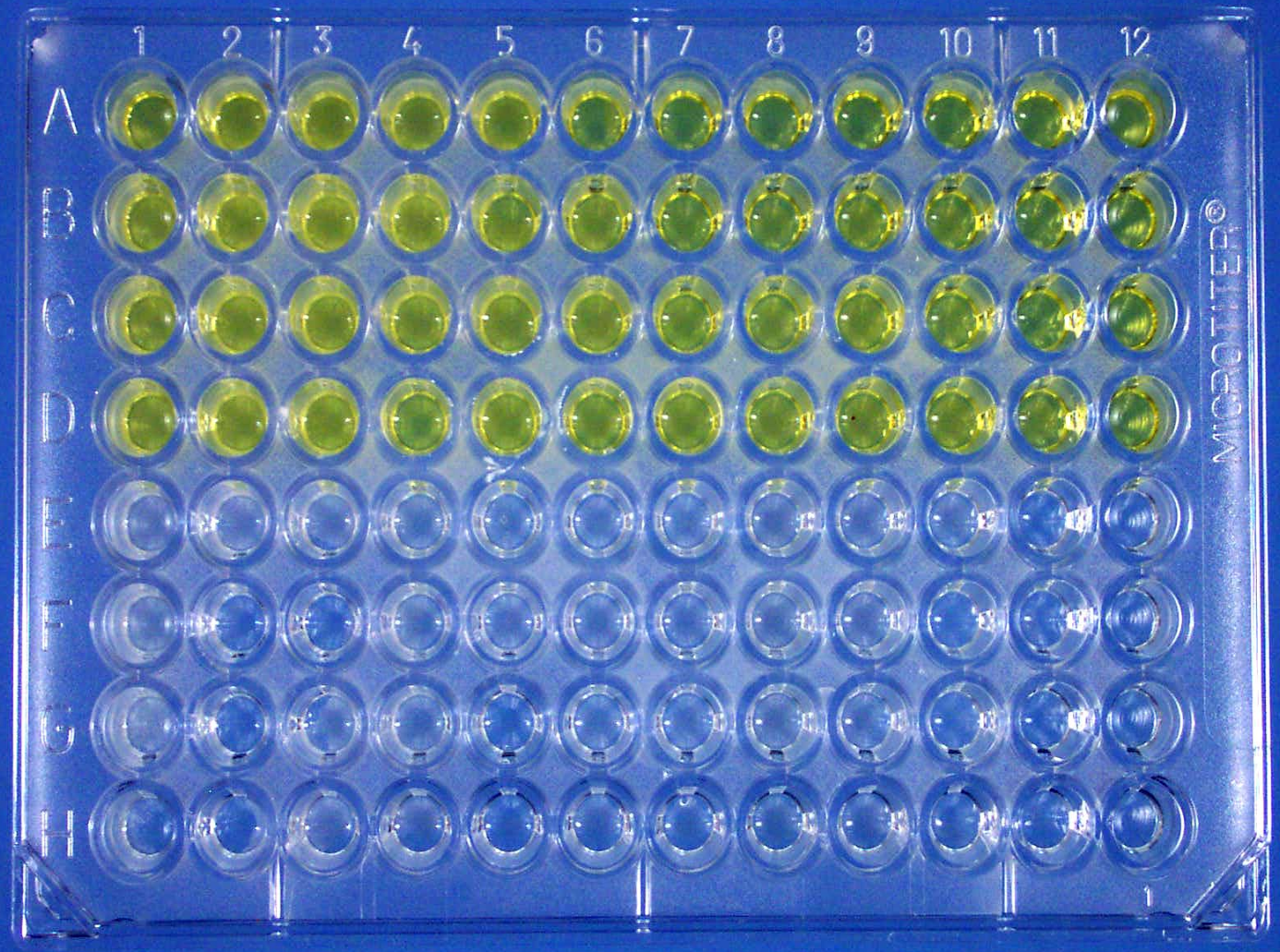
OXIDASES



OP, DDT, Carbamates, Pyrethroids

ACETYLCHOLINESTERASE

INSENSITIVE



Organophosphates,
Carbamates

- **Resistance** does NOT imply that it is **impossible** to control the resistant population or to prevent disease transmission, or that all populations of this species cannot be controlled.

- Thus a **single report** of resistance to an insecticide does not imply that an insecticide is no longer useful either within the local region or globally.

Thank You