ACIAR-SRA Result-sharing Workshop
Presentation of Results - 1

Occurrence of *Sri Lankan cassava mosaic virus (SLCMV)*

19th July 2017, Phnom Penh, Cambodia

Nami Minato

Outline

- Background – CMD and its distribution in Asia region
- SRA cassava mosaic disease surveillance
- Results and discussion
- Notes for group discussion

Cassava mosaic disease (CMD)

Symptoms & Yield loss

- Cassava mosaic disease infected cassava typically shows mosaic symptoms on leaves and leaf curl.
- Yield losses attributed to CMD range 20 to 95%, and have been reported to be substantially increased following mixed infection.

(Photo by Sophearith Sok)

Transmission

- Viruses causing CMD are transmitted by
  1. Planting materials (cassava cuttings/seeds)
  2. Insect vector, whiteflies

Cassava mosaic geminiviruses (CMGs)

Virus taxonomy

- Viruses causing CMD (cassava mosaic geminiviruses: CMGs) belong to family *Geminiviridae*, genus *Begomovirus*.

- CMD is caused by 11 species of virus, such as
  - African cassava mosaic virus (ACMV)
  - Indian cassava mosaic virus (ICMV)
  - Sri Lankan cassava mosaic virus (SLCMV)

- CMD has been spread in Africa for over 200 years, and in South Asia (India and Sri Lanka).

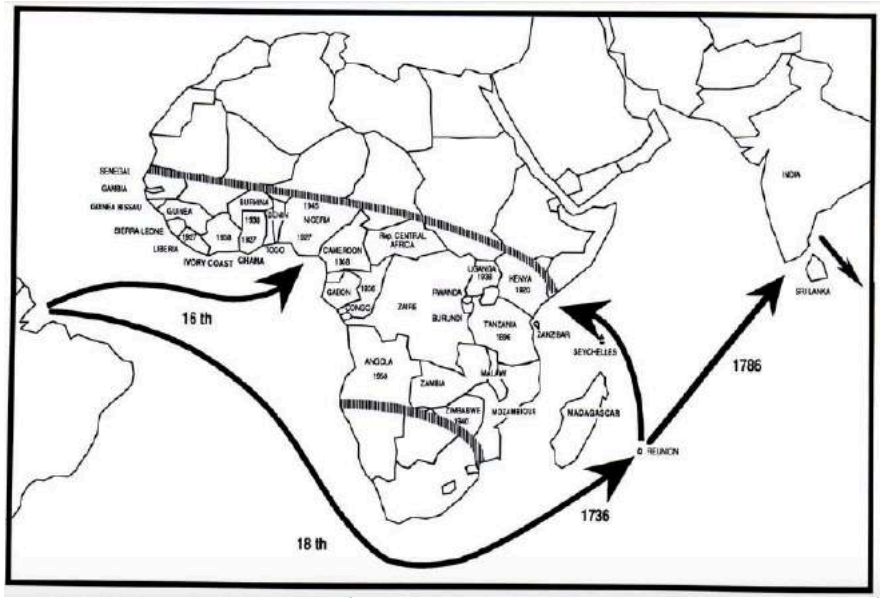
Spatial and temporal distribution

Cassava mosaic disease in Asia

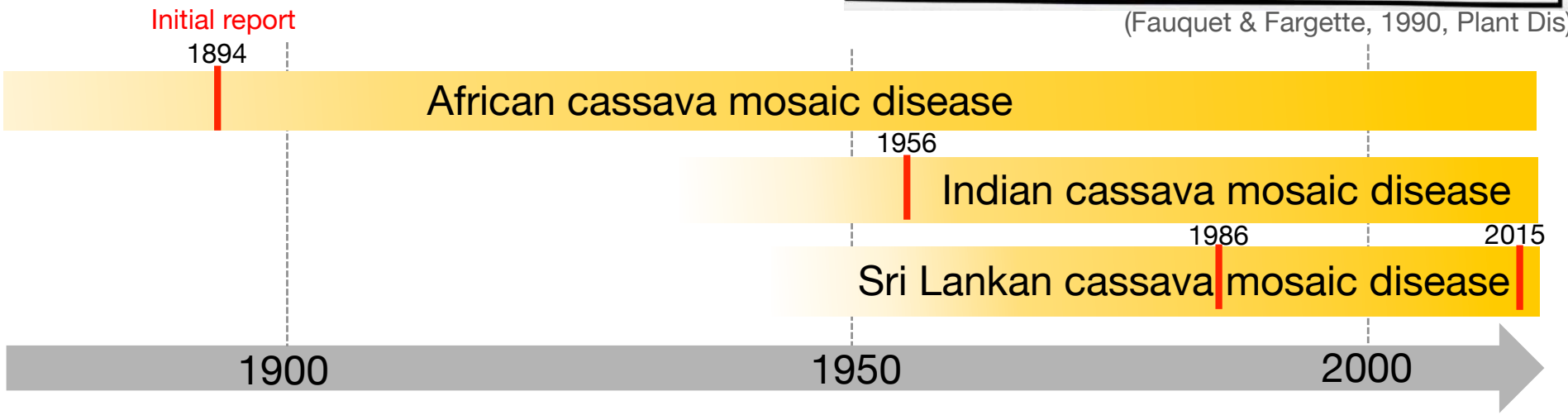
Indian cassava mosaic virus (ICMV)

Sri Lankan cassava mosaic virus (SLCMV)

In December 2015,
SLCMV was reported in Cambodia



(Fauquet & Fargette, 1990, Plant Dis)



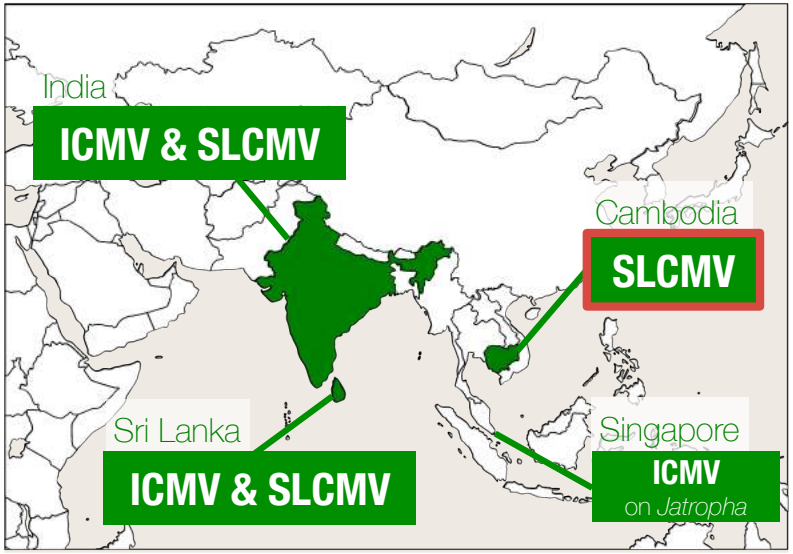
Spatial and temporal distribution

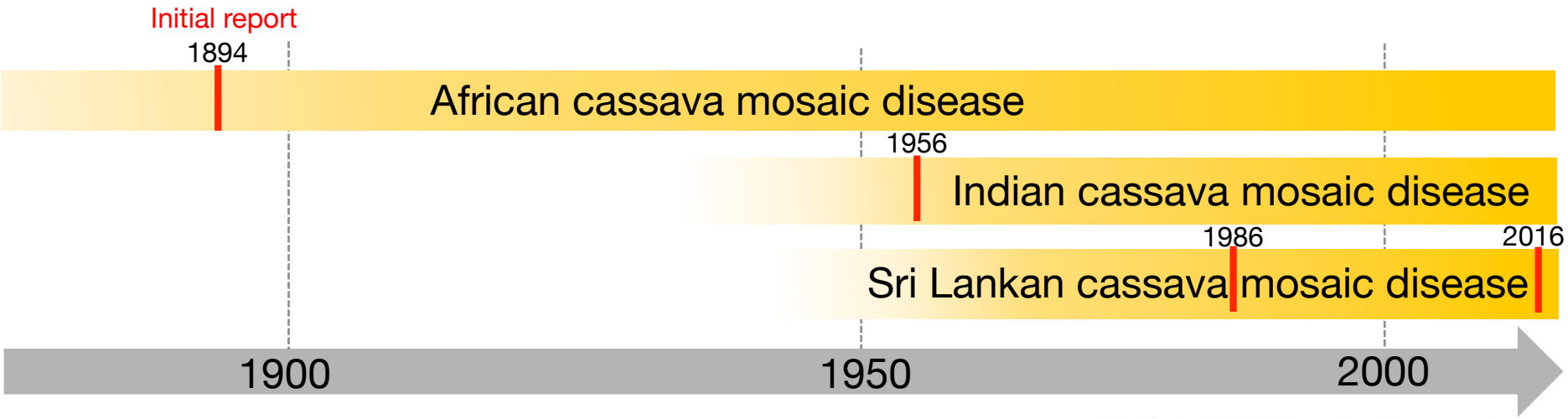
Cassava mosaic disease in Asia

Indian cassava mosaic virus (ICMV)

Sri Lankan cassava mosaic virus (SLCMV)

In December 2015,
SLCMV was reported in Cambodia





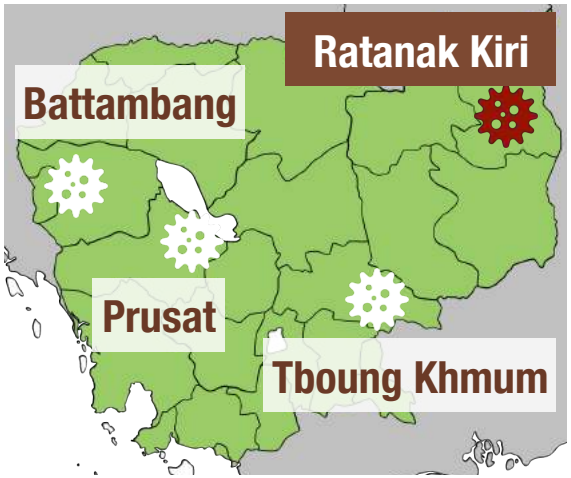
Research Issues

Cassava mosaic disease in Asia

- Non-symptomatic infection
- Response of cassava varieties in Southeast Asia has not been analyzed

→ Need to check the virus infection by molecular biological methods (eg PCR and ELISA)

SLCMV task-force survey in Early 2016



- First report in **Ratanak Kiri**, Eastern Cambodia
- CIAT Report (Rith, Monica & Wilmer) showed positives from **Ratanak Kiri + 3 provinces**
- Randomly picked 9 fields as a quick response

→ Incidence of cassava production area needs to be analyzed to help policy-making

Ongoing CMD surveillance

SLCMV survey in ACIAR-SRA project (2016 – 2017)

"Developing an emergency response and long term management strategy for Cassava Mosaic Virus in Cambodia and Vietnam"

- Target countries:

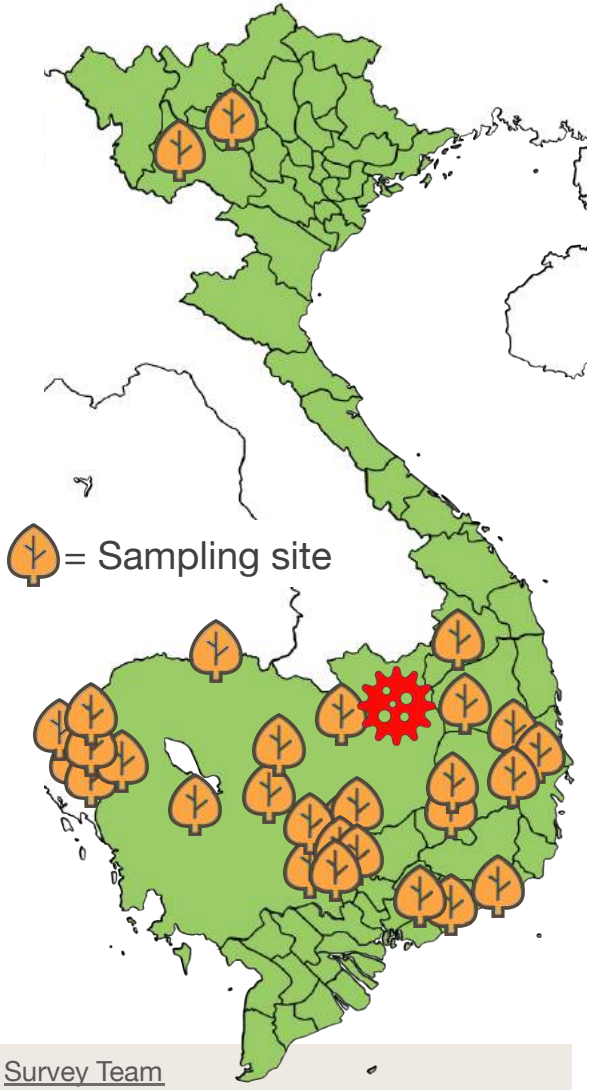
  Cambodia & Vietnam

- Activities:

1) cassava leaf & whitefly sampling and short seed systems interview
2) In-depth seed systems survey & traders interview

to see a current geographical distribution of SLCMV
to check two vectors for spreading, insects & seeds

Big survey to see SLCMV distribution



Survey Team

VN: Plant Protection Research Institute (PPRI) & Plant Protection Department (PPD)

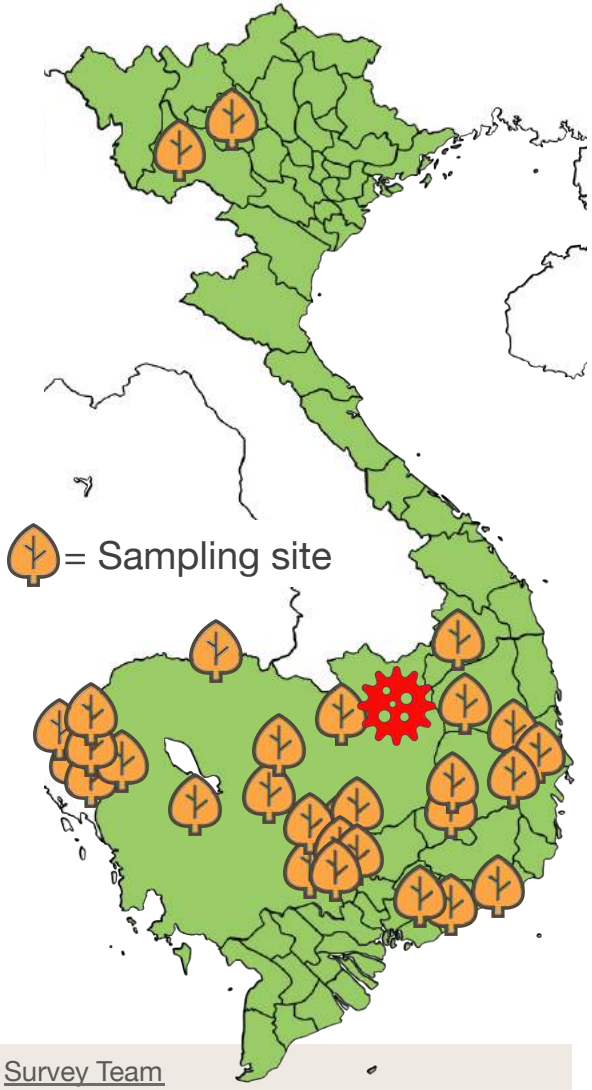
KH: General Directorate of Agriculture (GDA) & Provincial Department of Agriculture (PDA)

- Covering big production areas
  - 15 districts in each country

Country		District
V (VN)	1	Thuan Chau
	2	Van Yen
	3	Chu Prong
	4	Mad Lak
	5	Eak Lak (Eaker?)
	6	Dak Glong
	7	Sa Thay
	8	Krong Pa
	9	Song Hinh
	10	Bac Binh
	11	Ham Tan
	12	Tan Bien
	13	Tan Chau
		Ham Thuan
	14	Nam
	15	Long Thanh

Country		District
K (KH)	1	Anlong Veaeng
	2	Malai
	3	Sala Krau
	4	Pailin
	5	Kamrieng
	6	Phnum Proek
		Rattanak
	7	mondul
	8	Kravanh
	9	Koun Mom
	10	Steung Treng
	11	Snoul
	12	Dambae
	13	Memot
	14	Romeas Haek
	15	Sandan
	16	Baray

Big survey to see SLCMV distribution



Survey Team

VN: Plant Protection Research Institute (PPRI) & Plant Protection Department (PPD)

KH: General Directorate of Agriculture (GDA) & Provincial Department of Agriculture (PDA)

- Covering big production areas
  - 15 districts in each country
- From Nov. to Dec. 2016
- Data: 1) Photographs to check symptoms
  2) Leaf samples to see SLCMV infection
  3) Whiteflies to identify biotypes
  4) Short survey to see seed movements
- PCR: Universal + SLCMV-specific primer sets

Seed systems survey

Individual survey

- Households that have been surveyed cassava fields
  - 15 households/ district;
  - 15-16 districts/ country
- From Nov. to Dec. 2016 (CVN: Feb 2017)

Types of seed trade, purchase & sale:
- Distances, volumes, sources
- Varieties, quality
- Farm characteristics

In-depth survey

- 2 locations/ country
  - Expansion site
  - Established site
- From Feb. to Mar. 2017
- Combined with trader survey

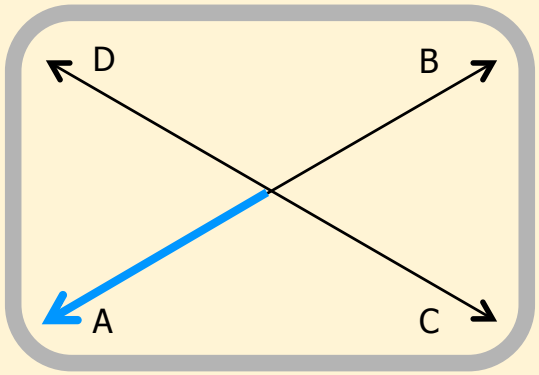
Network analysis of seed movement

+ Univ. of Florida added network analysis of information movement

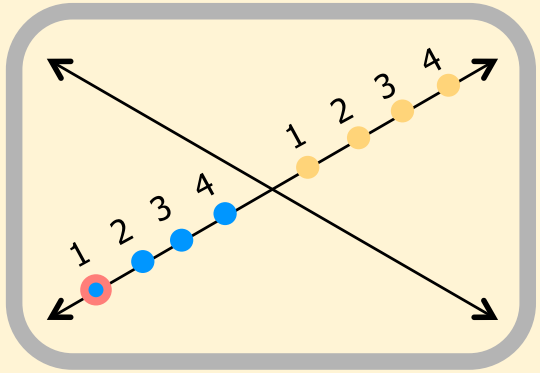
Methods - sampling & diagnostic

Sampling method

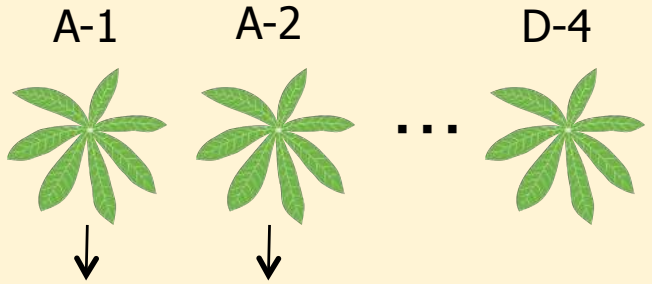
- 15 fields/ district
- 4 transects = 16 plants/ field
- Use dried leaves for diagnosis
  - dried up with silica gel
  - Low cost; stable with tropical weather



Transect A



Plant 1



Dry with silica gel individually

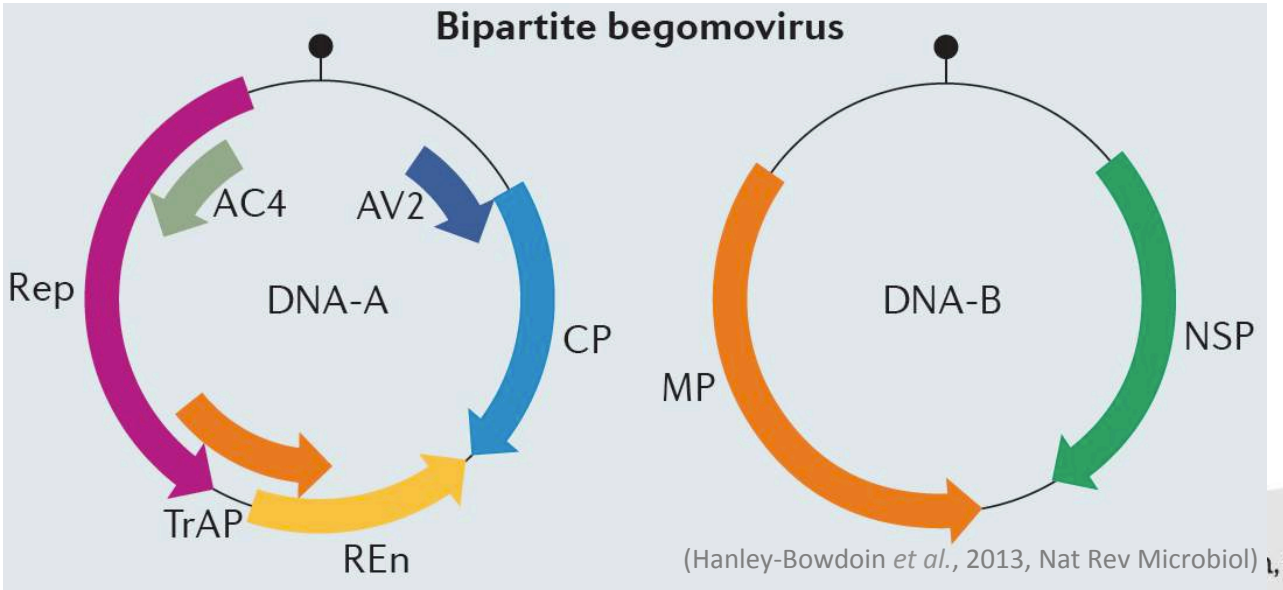
Methods - sampling & diagnostic

Sampling method

- 15 fields/ district
- 4 transects = 16 plants/ field
- Use dried leaves for diagnosis
  - dried up with silica gel
  - Low cost; stable with tropical weather

Diagnostic method

- Using PCR because of its sensitivity
- DNA extraction from dried leaves -> PCR



(Hanley-Bowdoin *et al.*, 2013, Nat Rev Microbiol)

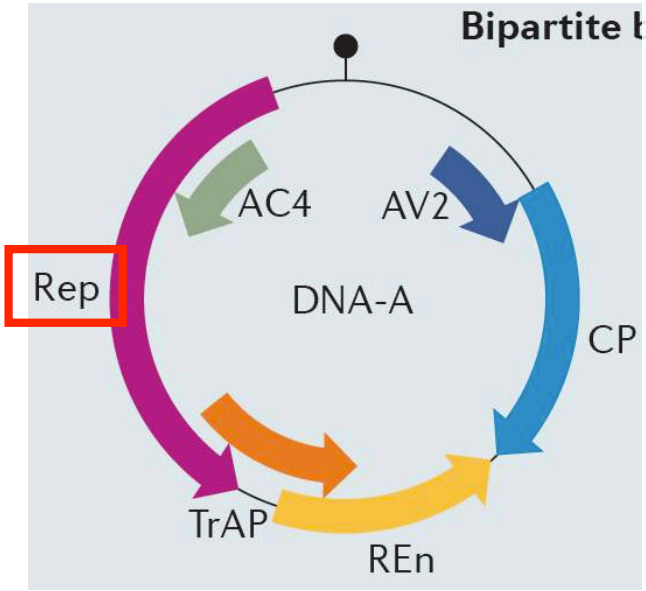
Methods - sampling & diagnostic

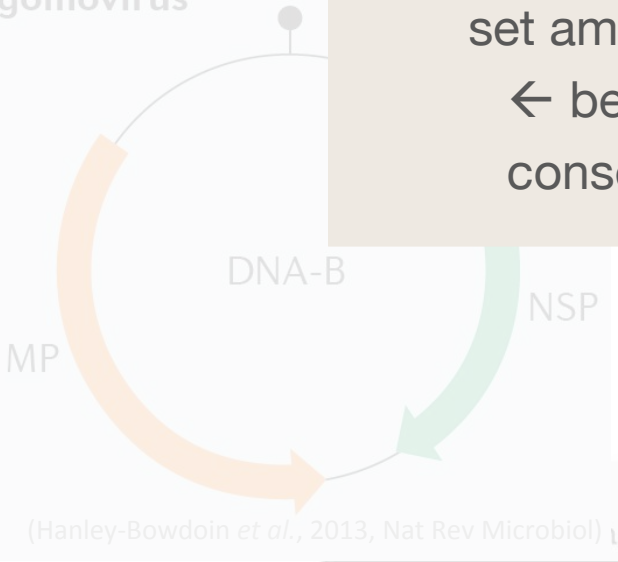
Sampling method

- 15 fields/ district
- 4 transects = 16 plants/ field
- Use dried leaves for diagnosis
  - dried up with silica gel
  - Low cost; stable with tropical weather

Diagnostic method

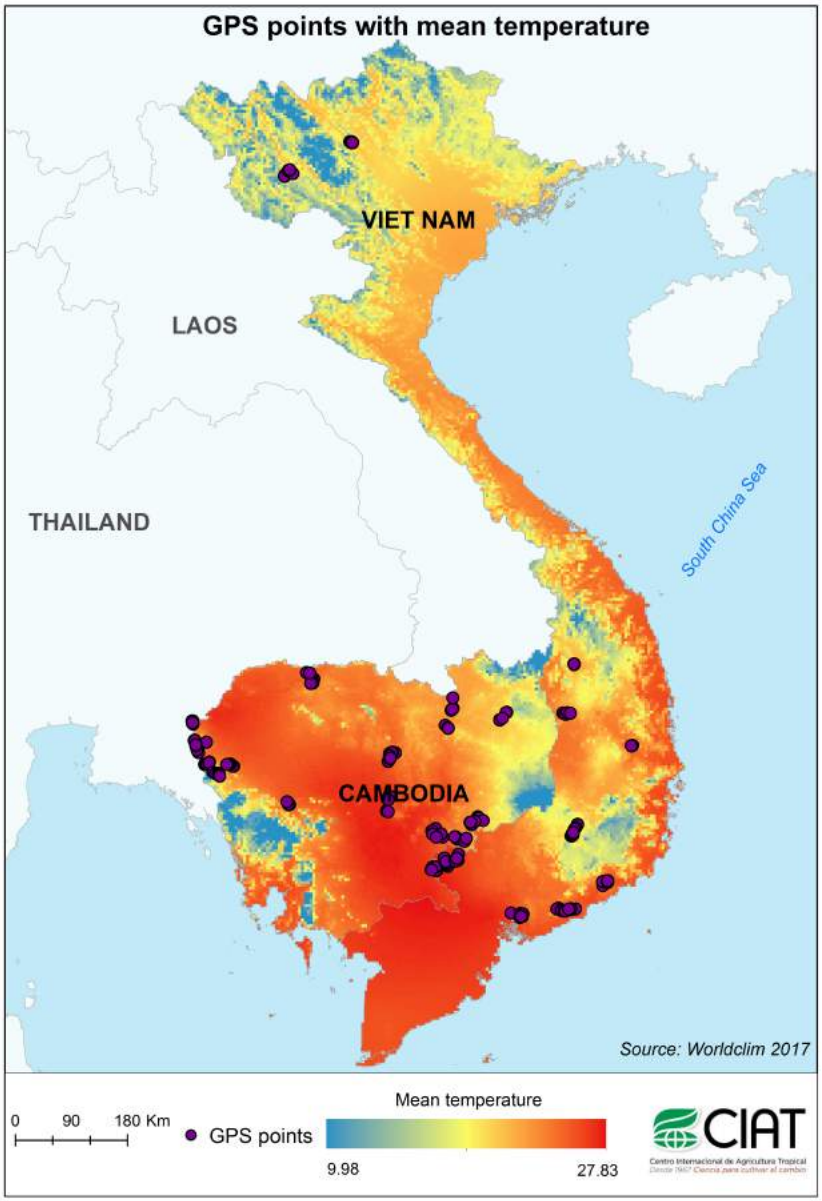
- Using PCR because of its sensitivity
- DNA extraction from dried leaves -> PCR
- Common PCR diagnosis: targeting CP or Rep
- Our diagnostic used the primer set amplifying Rep
  ← because of its highly conserved sequence





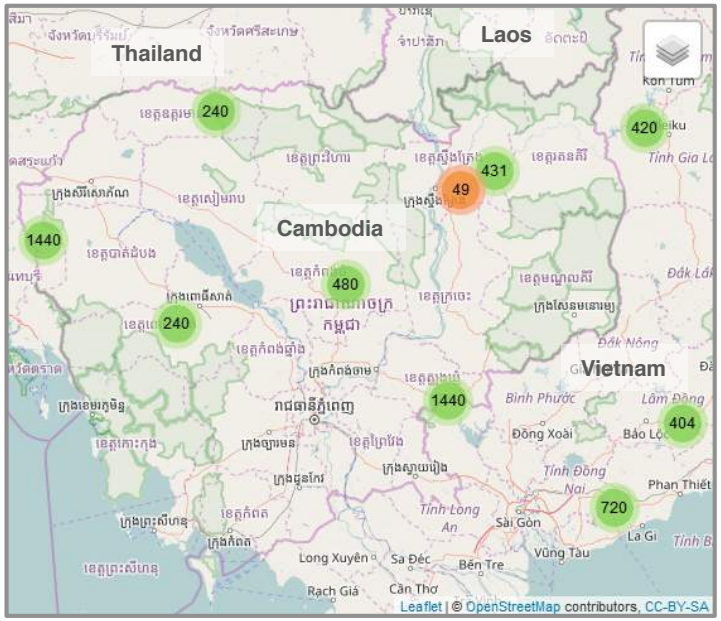
(Hanley-Bowdoin *et al.*, 2013, Nat Rev Microbiol)

Geographical distribution of SLCMV



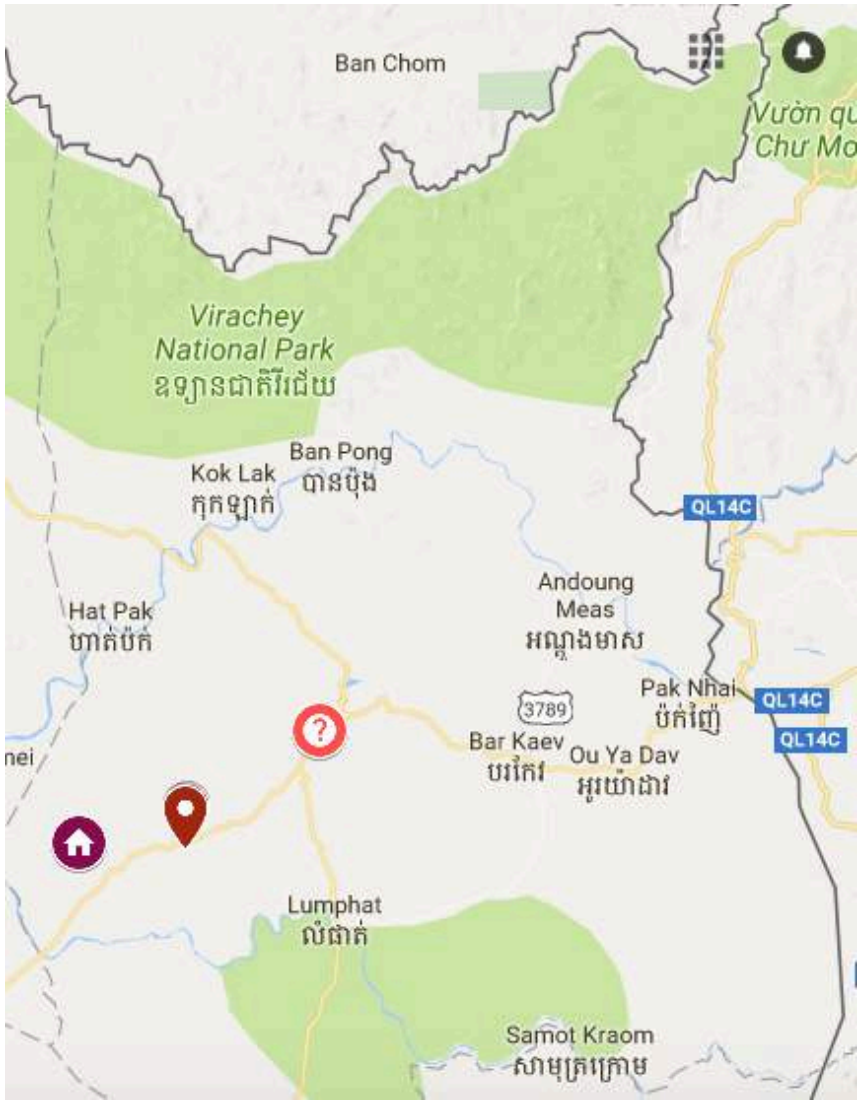
Can be shown as a HTML file

Incidence of SLCMV in Cambodia



Province	District	Field-level incidence (%) (# of Positive/15 fields)
Oddar Meanchey	Anlong Veaeng	0% (0/15)
Banteay Meanchey	Malai	0% (0/15)
Pailin	Sala Krau	0% (0/15)
Pailin	Pailin	0% (0/15)
Battambang	Kamrieng	0% (0/15)
Battambang	Phnum Proek	0% (0/15)
Battambang	Ratonak Mondol	0% (0/15)
Pursat	Kravanh	0% (0/15)
Ratanak Kiri	Koun Mom	13.3% (2/15)
Stung Treng	Stueng Traeng	46.6% (7/15)
Kratie	Snuol	0% (0/15)
Tboung Khmum	Dambae	0% (0/15)
Tboung Khmum	Memot	0% (0/15)
Svay Rieng	Romeas Haek	0% (0/15)
Kampong Thom	Sandan	0% (0/15)
Kampong Thom	Baray	0% (0/15)

SLCMV incidence in Koun Mom, Ratanak Kiri

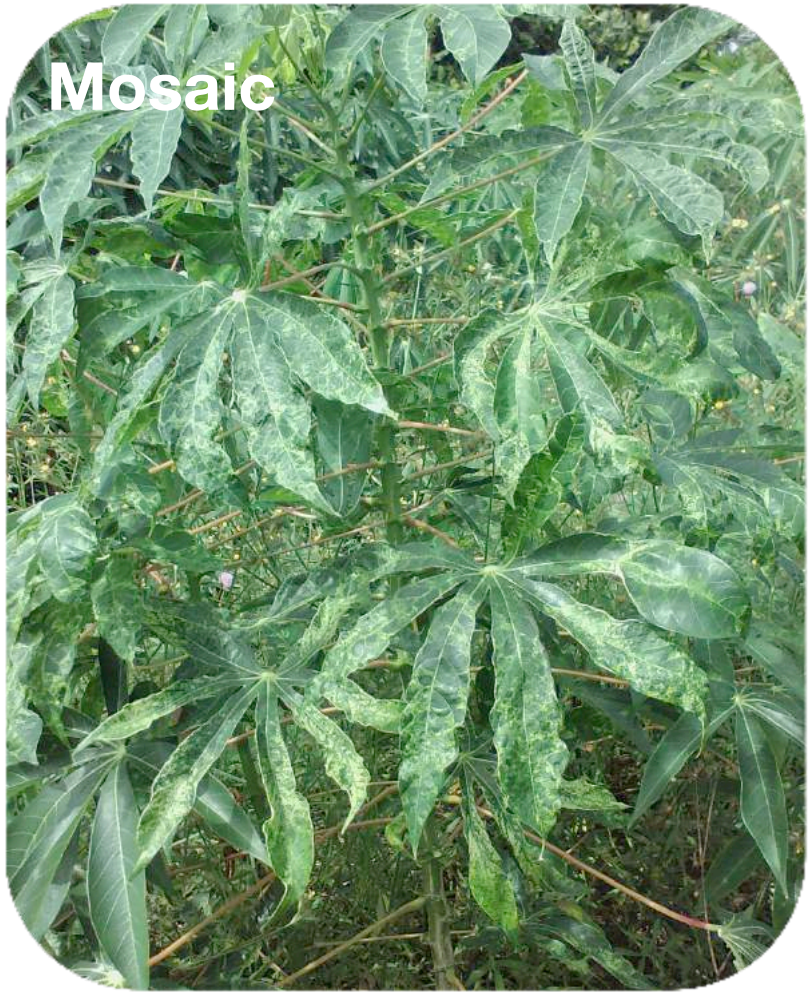


Field Code	# of infected plants	Infection Rate (%)	Infected Transect
Koun Mom District, Ratanak Kiri Province			
K9F1	0/16	0	
K9F2	6/16	37.5%	ACD
K9F3	4/16	25.0%	AD
K9F4	0/16	0	
K9F5	0/16	0	
K9F6	0/16	0	
K9F7	0/16	0	
K9F8	0/16	0	
K9F9	0/16	0	
K9F10	0/16	0	
K9F11	0/16	0	
K9F12	0/16	0	
K9F13	0/16	0	
K9F14	0/16	0	
K9F15	0/16	0	

SLCMV incidence in Stueng Traeng, Stung Treng

Field Code	# of infected plants	Infection Rate (%)	Infected Transect
Stueng Traeng District, Stung Treng Province			
K10F1	4/16	25.0%	ABC
K10F2	7/16	43.8%	ABCD
K10F3	0/16	0	
K10F4	0/16	0	
K10F5	1/16	6.3%	C
K10F6	4/16	25.0%	CD
K10F7	0/16	0	
K10F8	0/16	0	
K10F9	0/16	0	
K10F10	0/16	0	
K10F11	0/16	0	
K10F12	0/16	0	
K10F13	7/16	43.8%	BCD
K10F14	7/16	43.8%	ABCD
K10F15	9/16	56.3%	ABCD

Symptoms of SLCMV in Cambodia



Symptoms of SLCMV in Cambodia

(1) Systemic

Probably the stake was infected

(2) Upper leaves

Insect transmission?

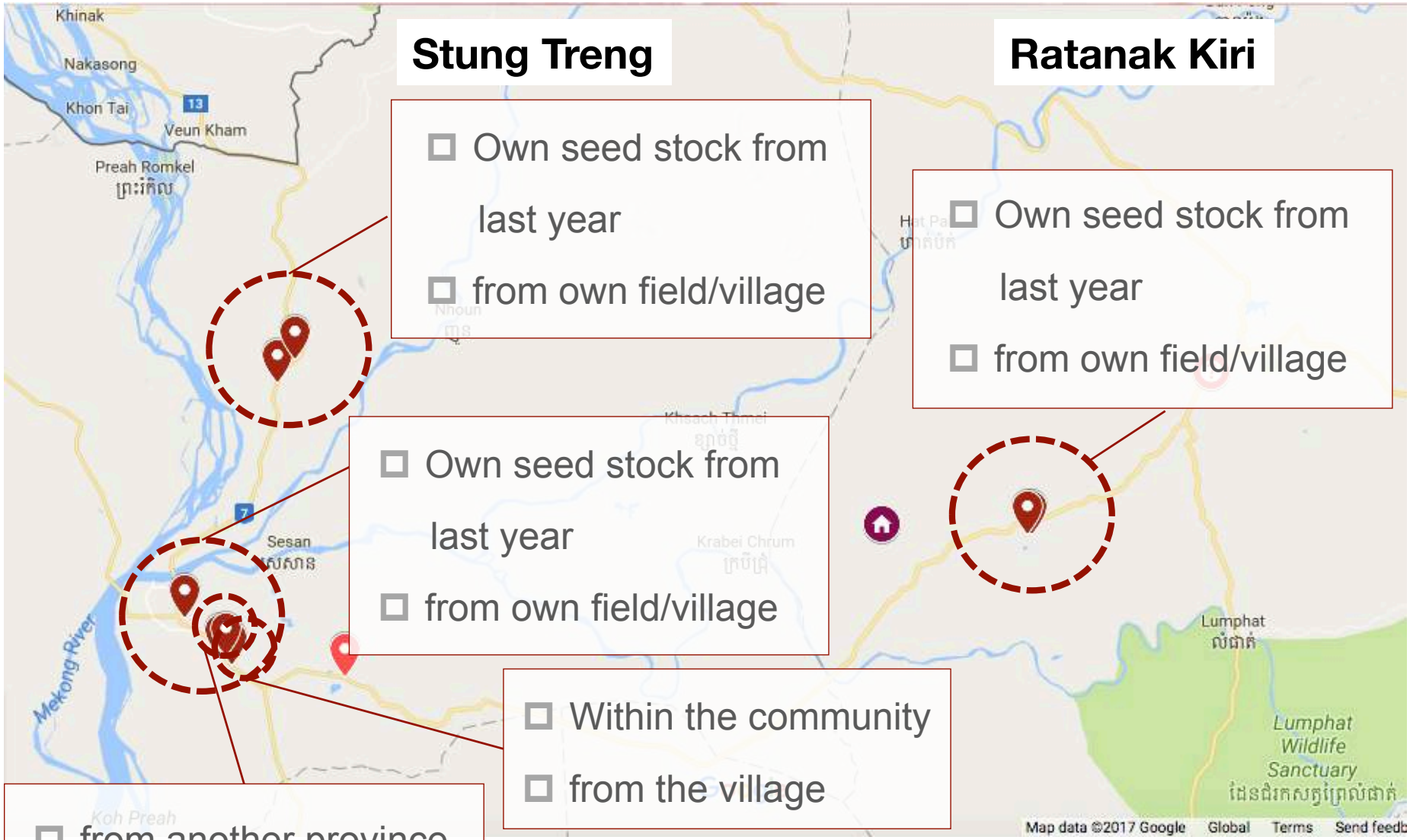
In one field

Asymptomatic infection

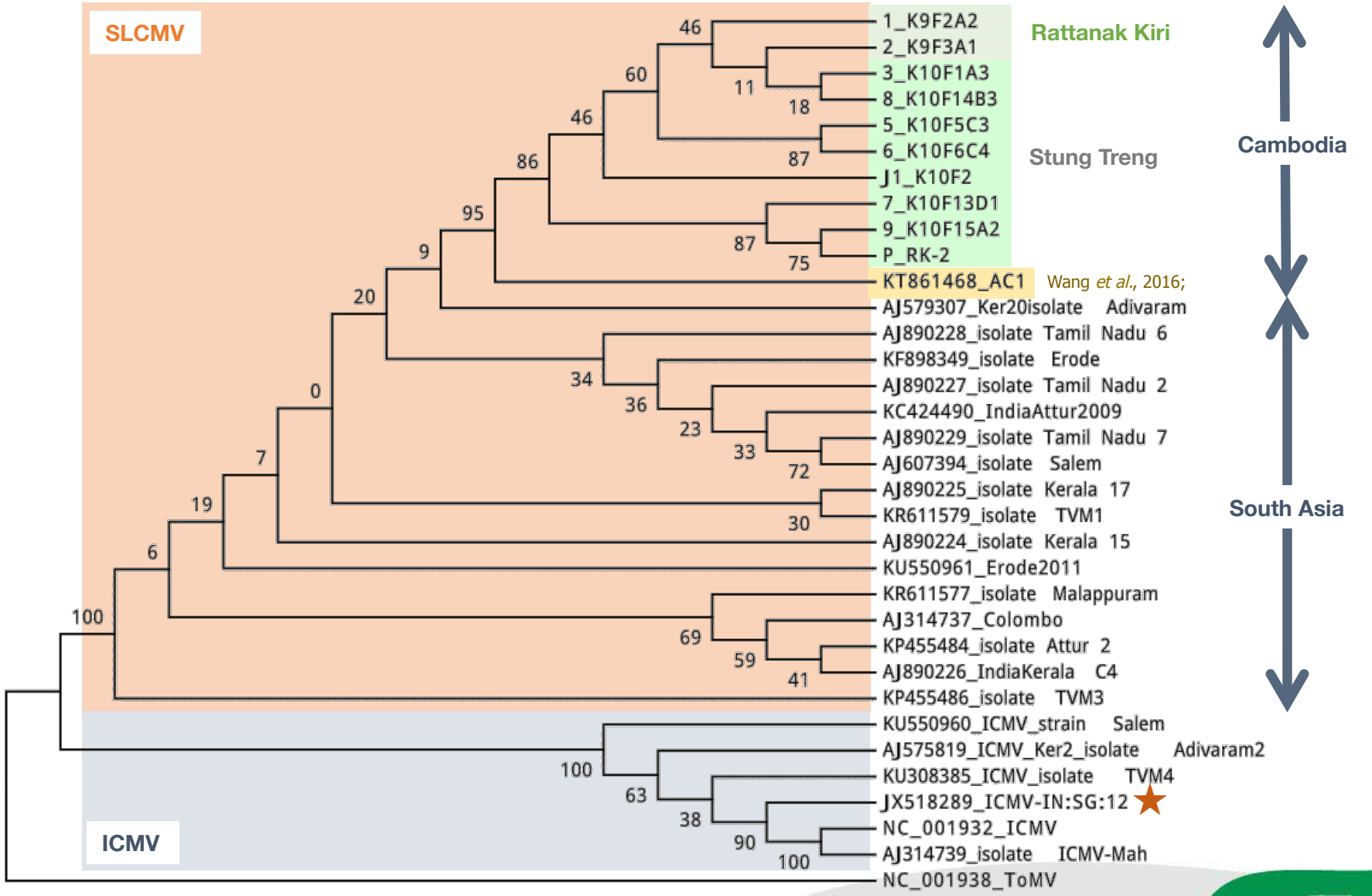
Concurrence of SLCMD and witches' broom

Mosaic + witches' broom symptoms

Seed acquisition in infected fields

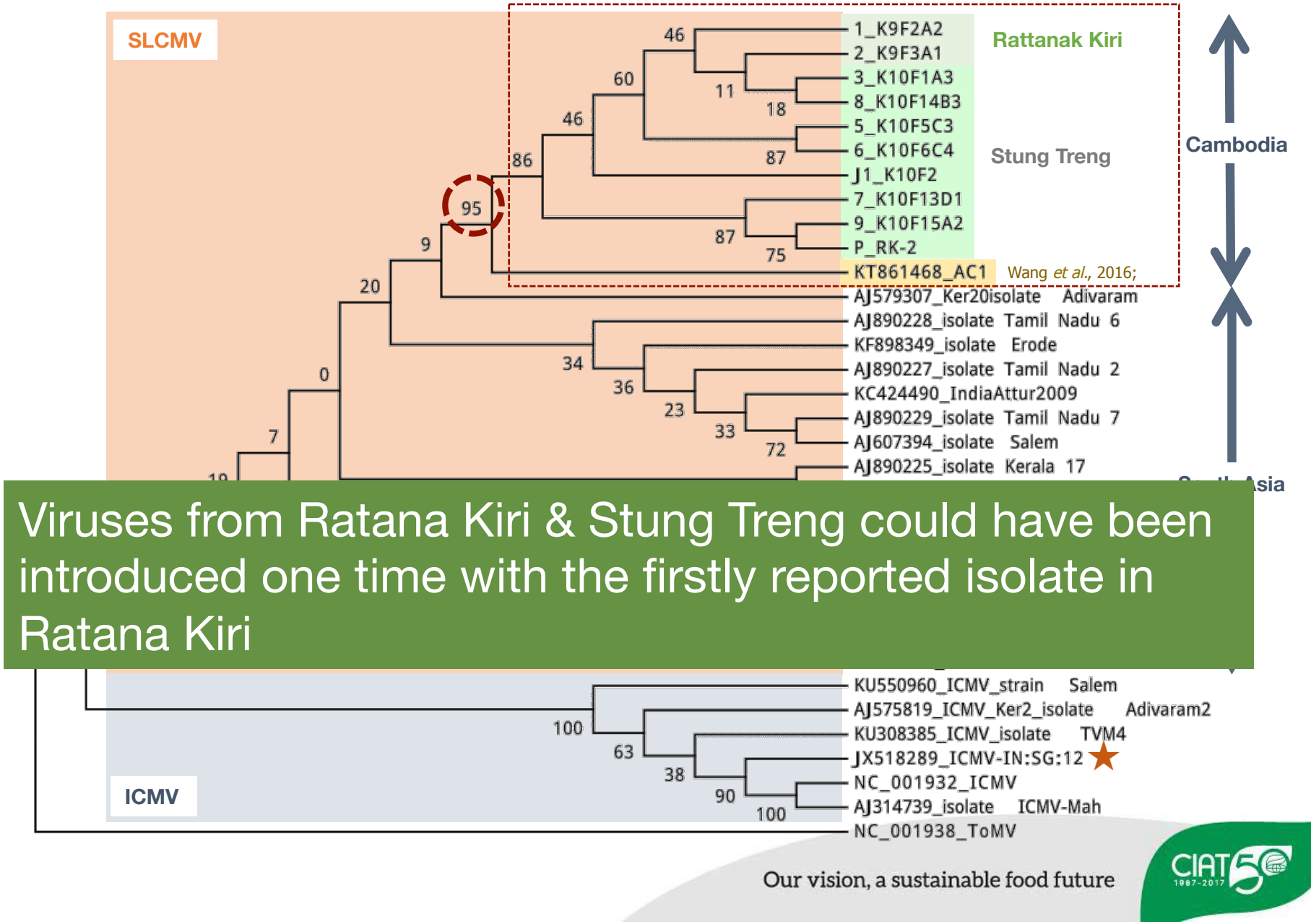


Phylogenetic analysis of SLCMV from KH



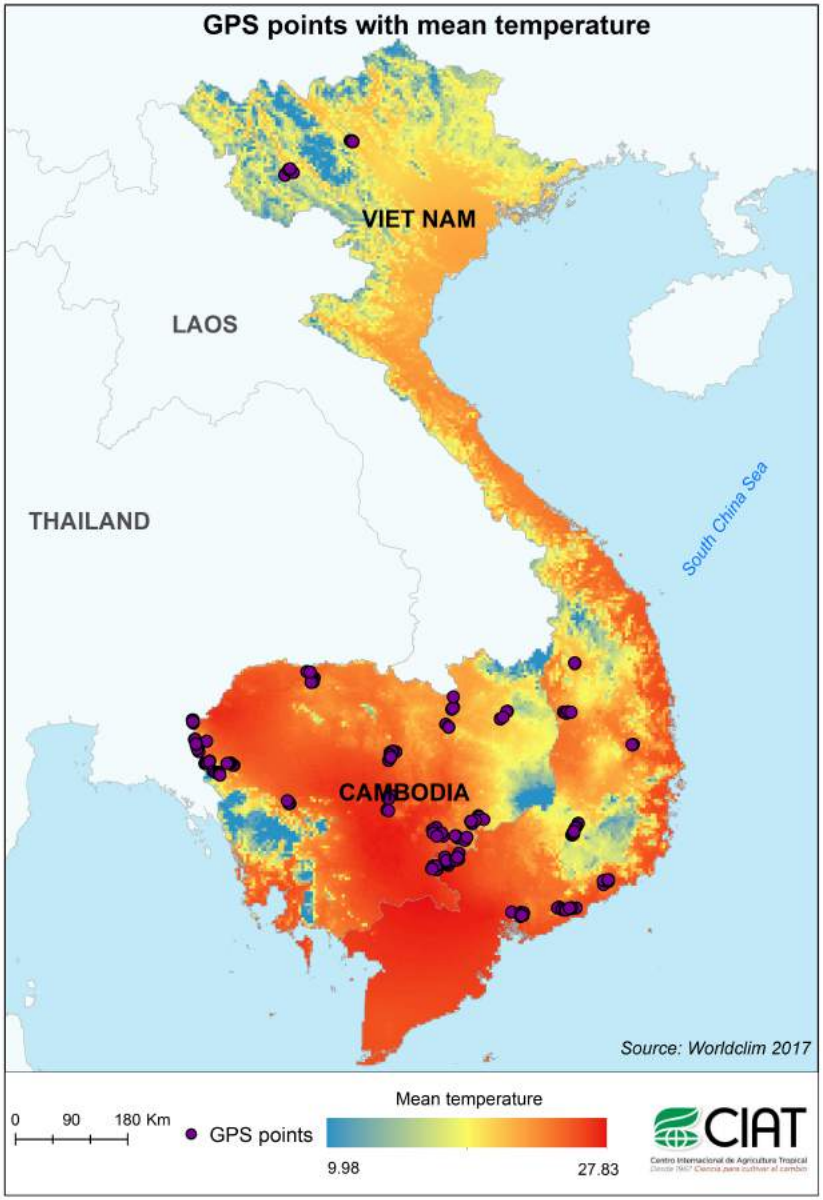
Our vision, a sustainable food future

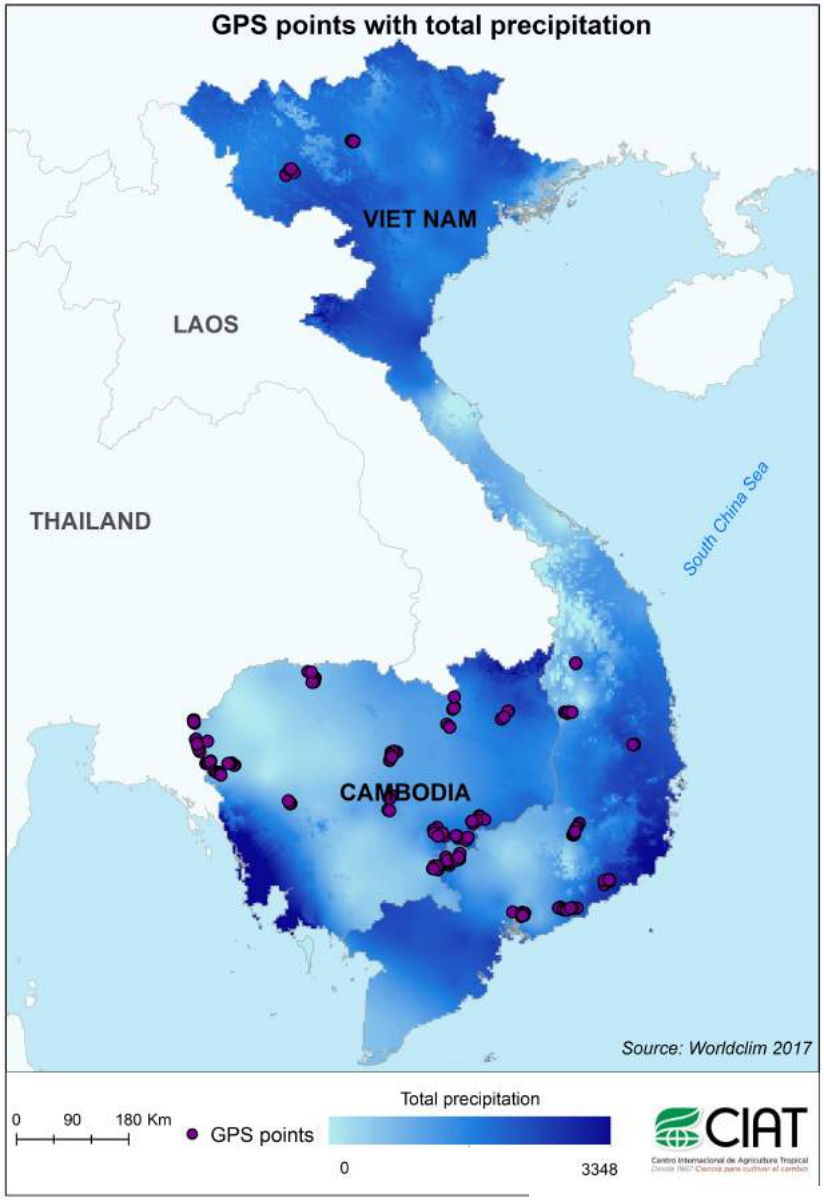
Phylogenetic analysis of SLCMV from KH



Viruses from Ratana Kiri & Stung Treng could have been introduced one time with the firstly reported isolate in Ratana Kiri

Virus distribution of SLCMV & Climate





(Map from Than Nguyen, CIAT)

Notes for group discussion

- High incidence in Stung Treng province
- Alternative hosts:

  Jatropha trees (ICMV); Chaya trees (SLCMV)
- Cassava plants are more susceptible to new infection

  before they age

CIAT

International Center for Tropical Agriculture
Since 1967 Science to cultivate change

CGIAR

A CGIAR Research Center

Headquarters
Km 17 Recta Cali-Palmira C.P. 763537
P.O. Box 6713, Cali, Colombia
Phone: +57 2 445 0000

ciat@cgiar.org
www.ciat.cgiar.org

ciat.ecoefficient

@ciat_cgiar

@CIAT_