



Alliance



Objective 3: Surveillance & Diagnostics

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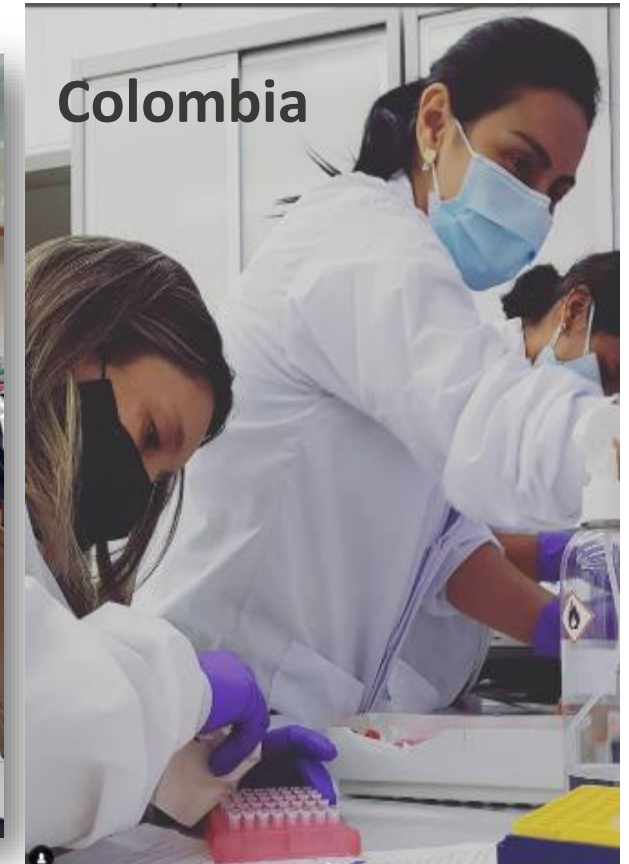
Project Mid-term Review

February 2022



Objective 3

- **Objective 3:** Develop, test and deploy diagnostic protocols, tools, and information platforms fit for purpose in monitoring, surveillance, and certification applications



Close collaboration & communication network

Alliance



Targtes: CMD and CWBD

Leaf mosaic and deformation



Leaf yellowing, short petioles, phyllody



Information platform: data integration and communication

<https://pestdisplace.org>

Name *

Prueba3-2020-CMD-SEA

Purpose

ACIAR Activity 3

Sampling Protocol

CIAT-Virology-Crop Protection Protocol v2.0

Grant Code

Grant Code

GUESTS

PARTNERS

DONORS

REFERENCES

SAMPLES

IMAGES

VIEW MAP

Guest
















User

ADD

Full Name	
Jenyfer Jimenez - ORCID:0000-0001-8149-6615	
Samoul Oeurn - ORCID:0000-0002-0771-2495	
Hoat Hoat - ORCID:0000-0002-2240-3922	
pinkham Vongphachanh - ORCID:0000-0003-4440-5838	
Hang Lee - ORCID:0000-0002-3504-1270	
Phuong Dung Le - ORCID:0000-0002-2147-3697	

Information platform: data integration and communication

<https://pestdisplace.org>

Name *	GUESTS					PARTNERS	DONORS	REFERENCES	SAMPLES	IMAGES	VIEW MAP
Purpose											
Sampling Protocol											
Grant Code											
Guest											
											
											

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<https://pestdisplace.org>

<https://pestdisplace.org>

Name *

Purpose

Sampling Protocol

Grant Code

GUESTS

PARTNERS

DONORS

REFERENCES

SAMPLES

IMAGES

VIEW MAP

F35-S1

F35-S2

Edit Sample

F1-S1

Collection Date

20200602

Visual Symptom

Negative

+ New Diagnostic

Diagnostic Method

Result

Pathogen

Delete

Create a new laboratory diagnostic

Diagnostic Method

Pathogen Name

Result

Elisa

PCR

Confirmed by sequencing

Restriction

qPCR

CANCEL

CONFIRM

Information platform: data integration and communication

<https://pestdisplace.org>

The screenshot displays the pestdisplace.org web application. On the left is a sidebar with fields for 'Name *', 'Purpose', 'Sampling Protocol', 'Grant Code', and 'Guest'. The main content area features a top navigation bar with tabs: GUESTS, PARTNERS, DONORS, REFERENCES, SAMPLES, IMAGES, and VIEW MAP (highlighted with a red circle). Below the navigation bar, there is an 'Edit Sample' section with a plant image and a table of samples. A 'Create a new laboratory' dialog box is open, showing a list of diagnostic methods: Elisa, PCR, Confirmed by sequencing, Restriction, and qPCR. To the right, a map of Southeast Asia is shown with a pie chart overlay. The pie chart has two segments: a green segment representing 39,890 and an orange segment representing 104,110. The map labels include Kazakhstan, Mongolia, Kyrgyzstan, China, Japan, Afghanistan, Pakistan, Nepal, Bangladesh, India, Sri Lanka, Maldives, Laos, Philippines, Malaysia, Indonesia, and Papua New Guinea.

Name *

Purpose

Sampling Protocol

Grant Code

Guest

GUESTS PARTNERS DONORS REFERENCES SAMPLES IMAGES VIEW MAP

Edit Sample

Lab code

F1-S1

F1-S2

F1-S3

F1-S4

F1-S5

Loos

LONG

104,110

Transf

+

New Diagnostic

Diagnostic Method

Create a new laboratory

Diagnostic Method

Elisa

PCR

Confirmed by sequencing

Restriction

qPCR

39,890

104,110

Laos

Philippines

Malaysia

Indonesia

Papua New Guinea

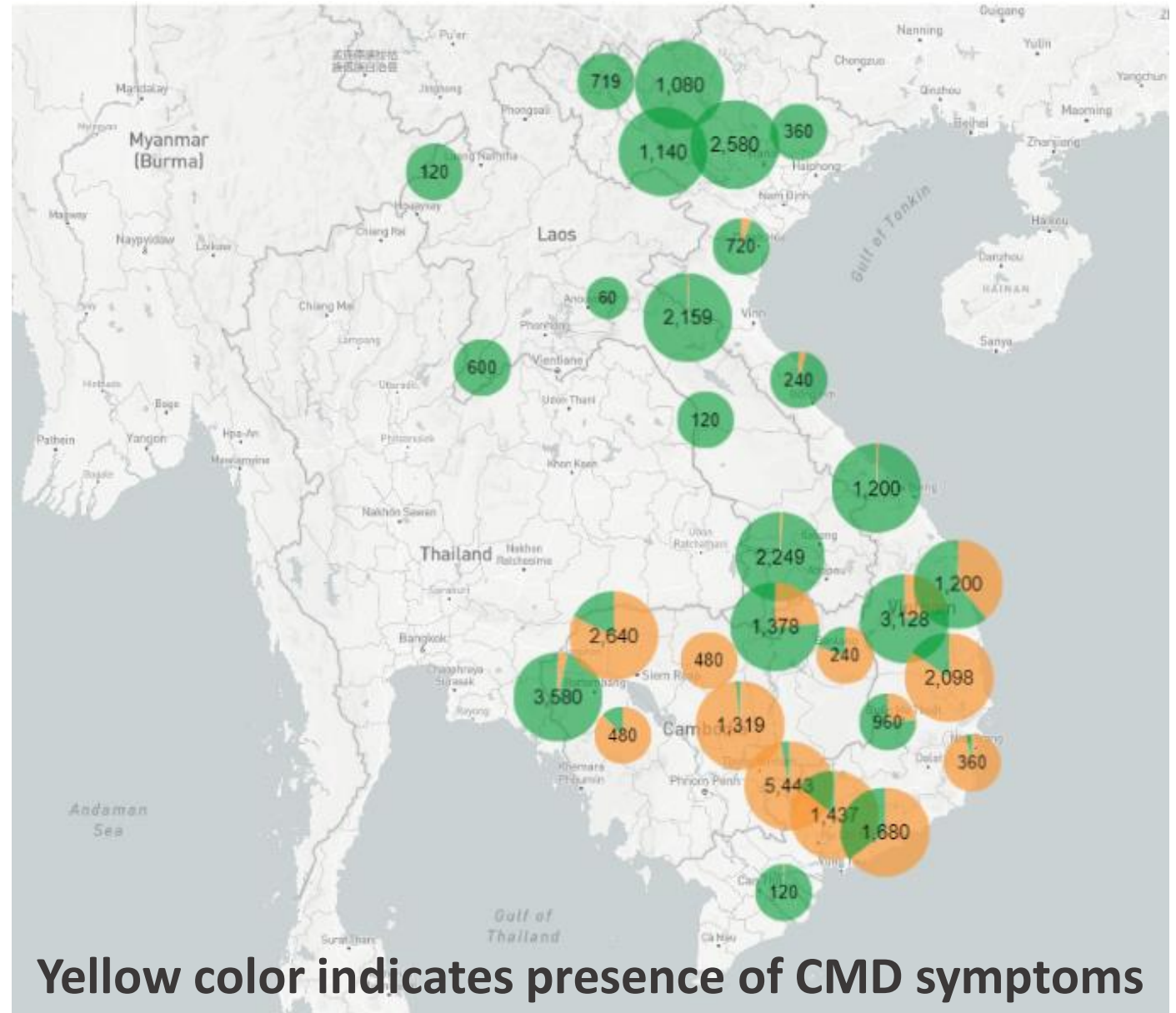
CIAT

International Center for Tropical Agriculture

Since 1967 dedicated to sustainable change

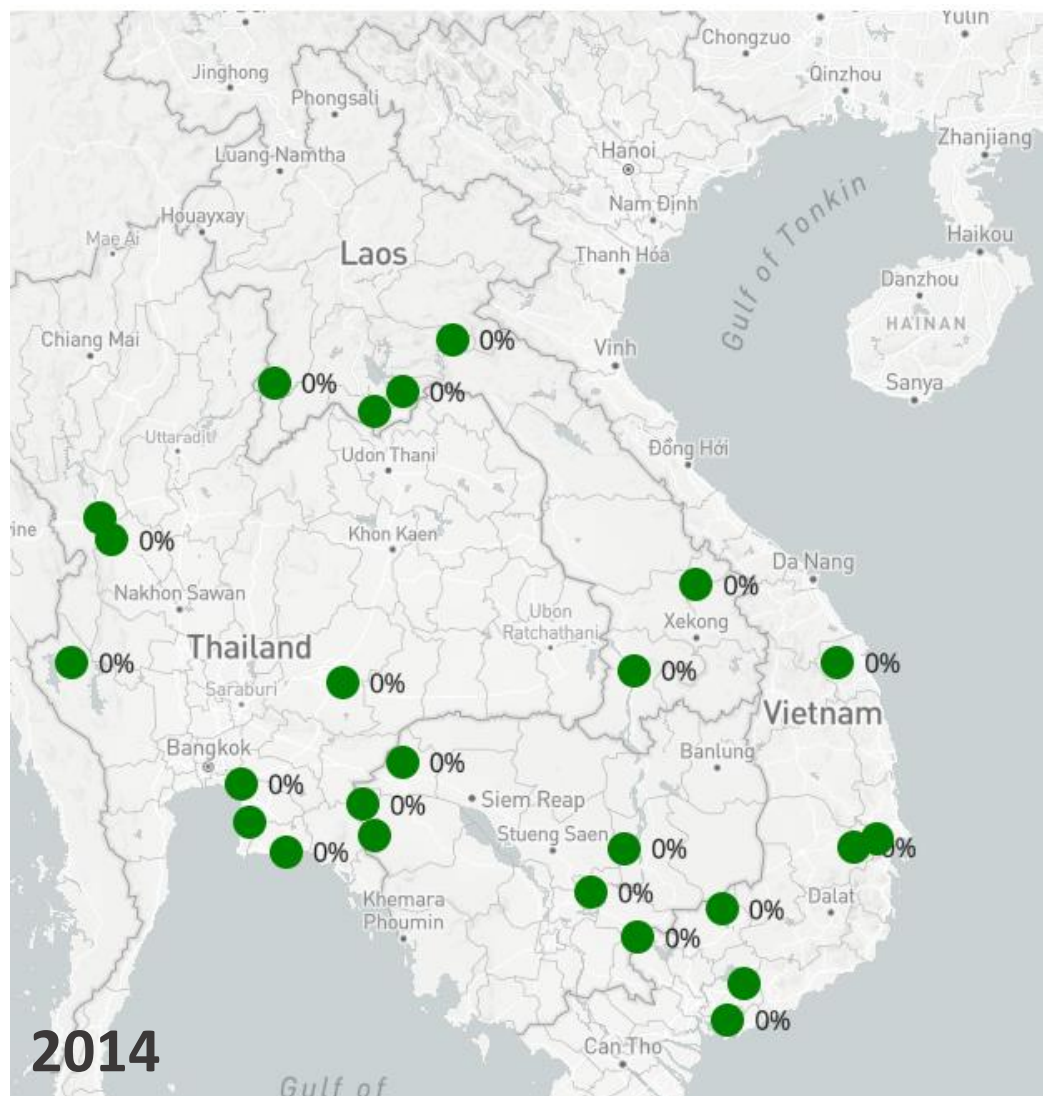
Surveillance

- Two consecutive years. Using a standard protocol in 3 countries (5% symptoms, 95% prob/Ha).
- 28,920 'observations' in 2020 and 10,970 'observations' in 2021 (COVID impact)
- Raw data maps updated in 'real-time'. Analysis>curation. All data is shared through the platform.
- Photo records 'observations' allow the identification of CMD, CWBD and other symptoms.

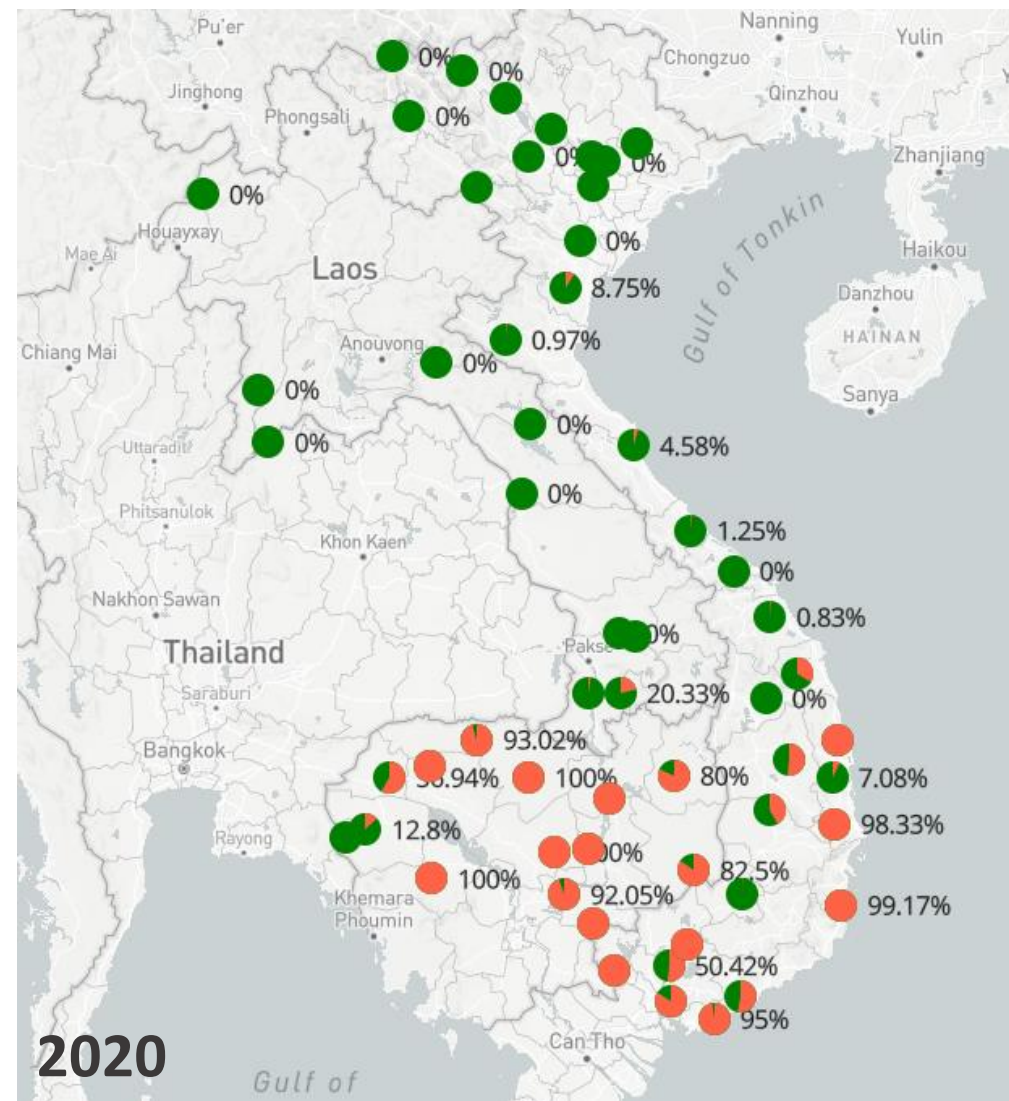


Incidence maps (Province level): CMD

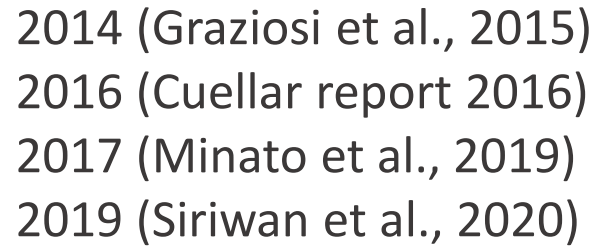
https://pestdisplace.org/projects/PDP_00063/provinces/map#3.44/18.47/103.34



Transect sampling, 50 observations per field; 3-5 months plants

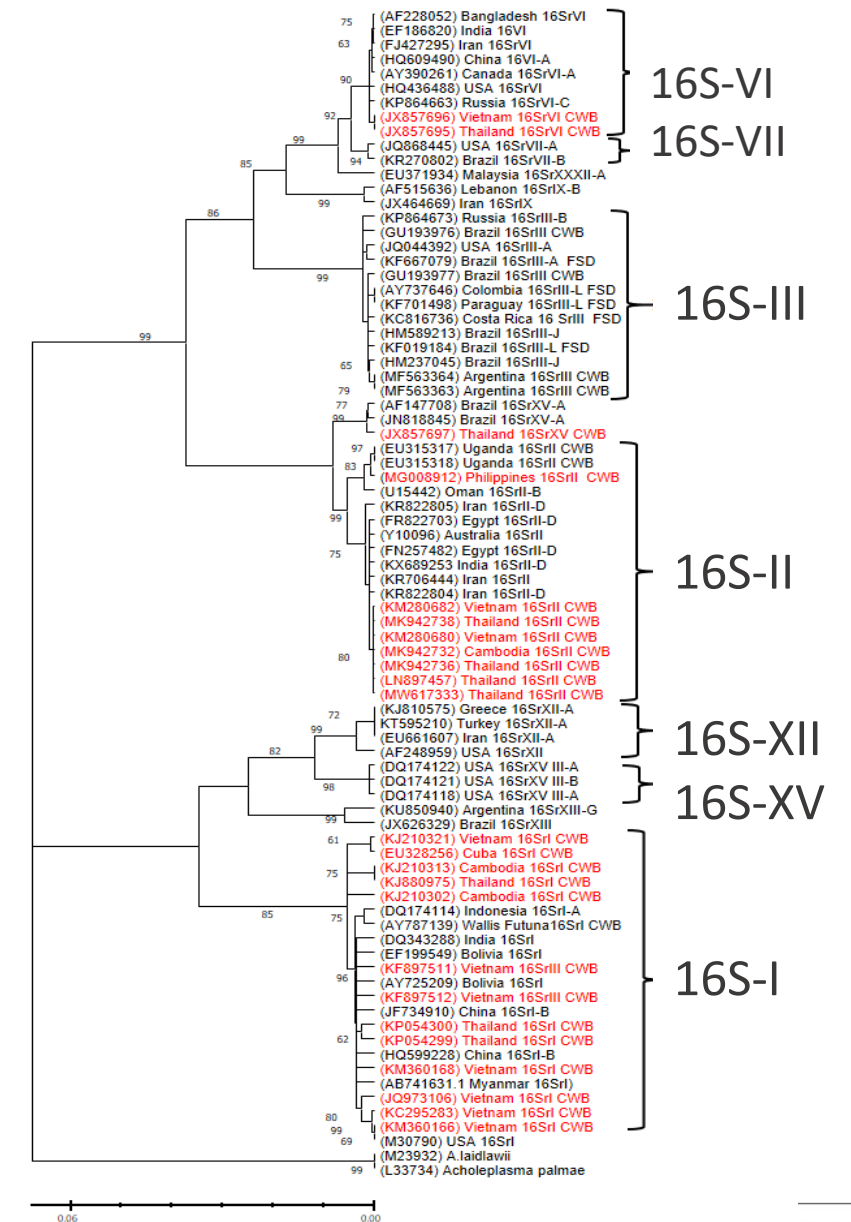


Transect sampling, 60 observations per field; 4-6 months plants

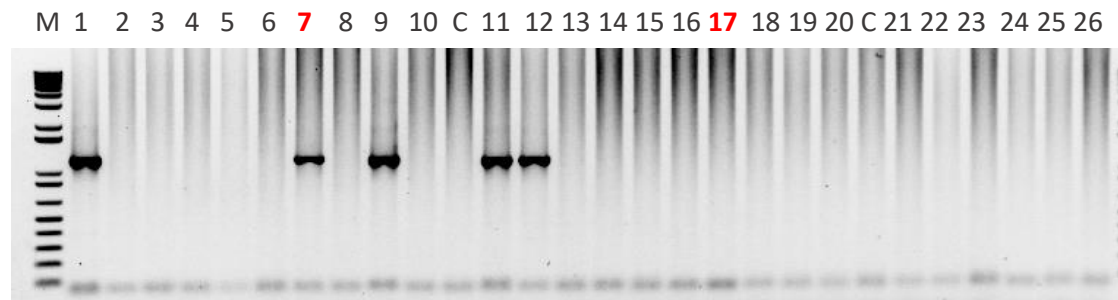


Diversity of phytoplasmas sequences in SEA

- Phytoplasma can be classified based on the analysis of their 16S rRNA into ribosomal groups and subgroups
- Sequences of CWBD-associated phytoplasmas belong to groups I, II, and VI (in red).
- Phytoplasmas infecting cassava in LAC belong to a separate group (III).
- Current detection methods target the 16S sequences but are not efficient



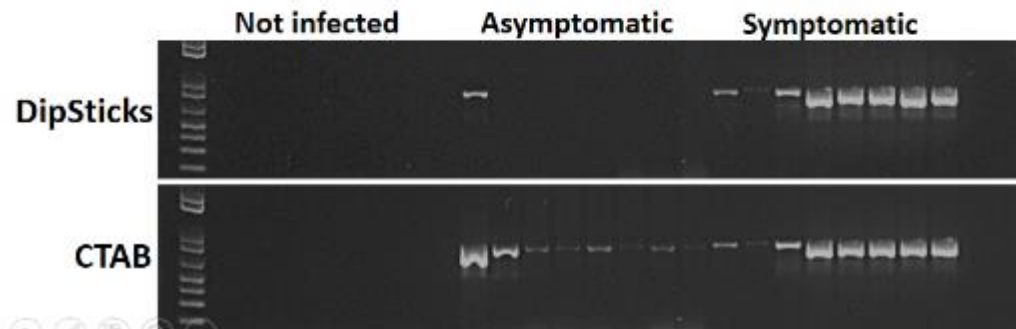
Testing current nested PCR protocols for detection of CWBD-associated phytoplasma



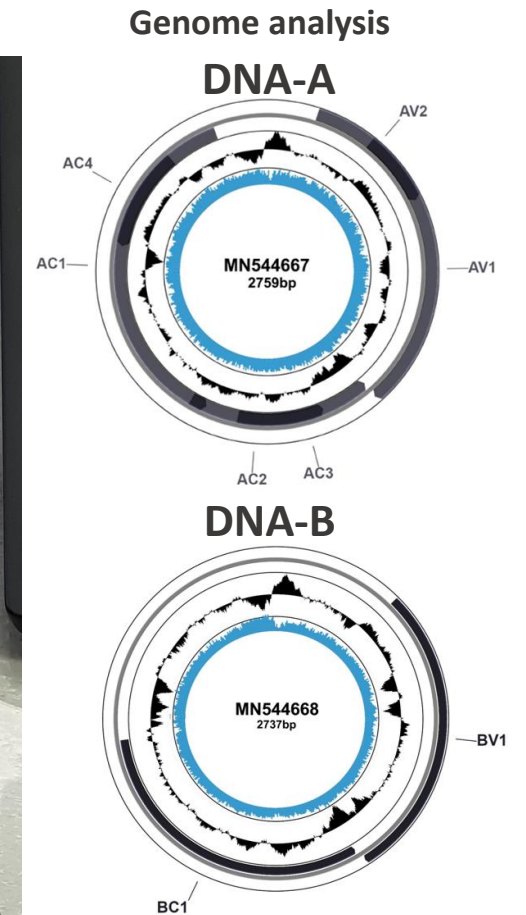
N=120	CWBD symptoms	No CWBD symptoms
PCR(+)	4	23
PCR (-)	13	80

Less than 50% of the PCR bands correspond to phytoplasma sequences

Diagnostics CMD: PCR, LAMP (droid), genome sequencing

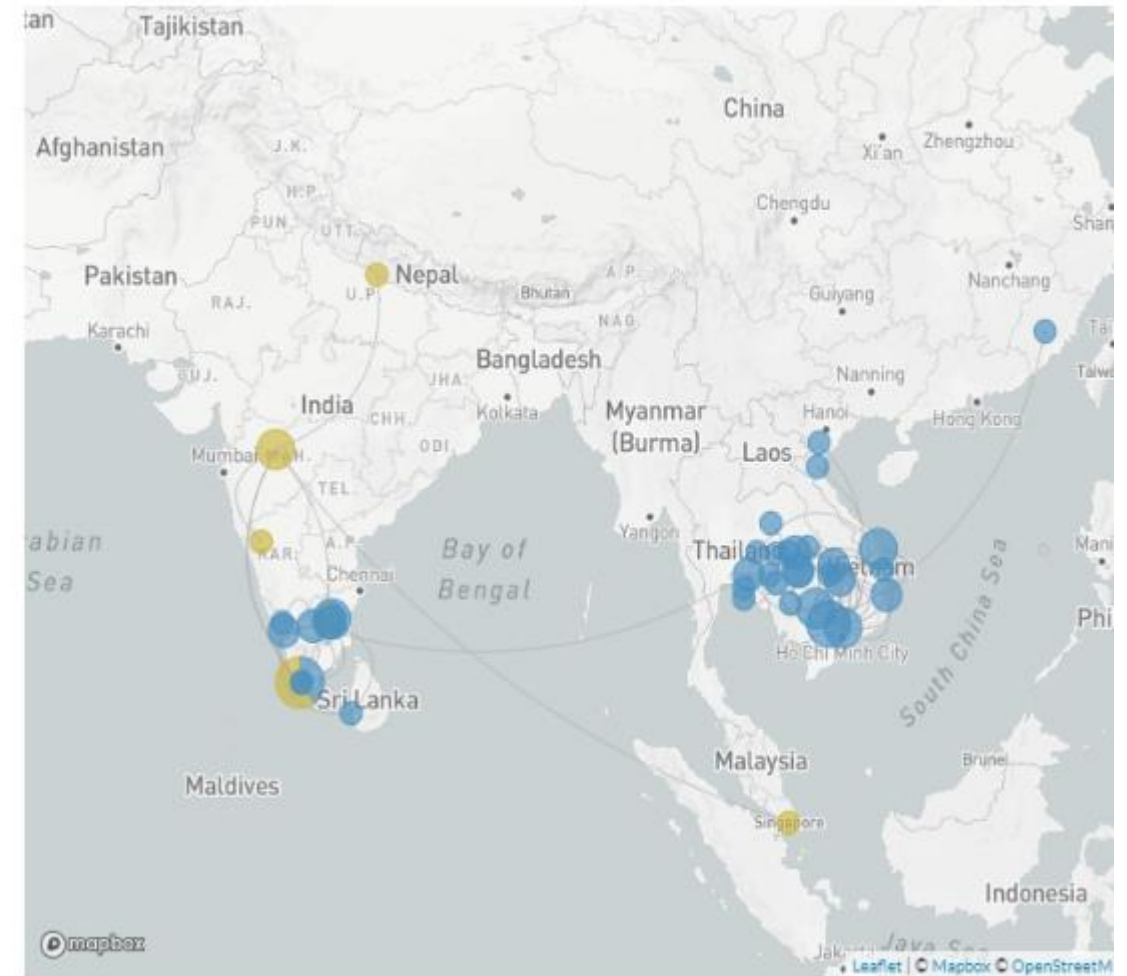
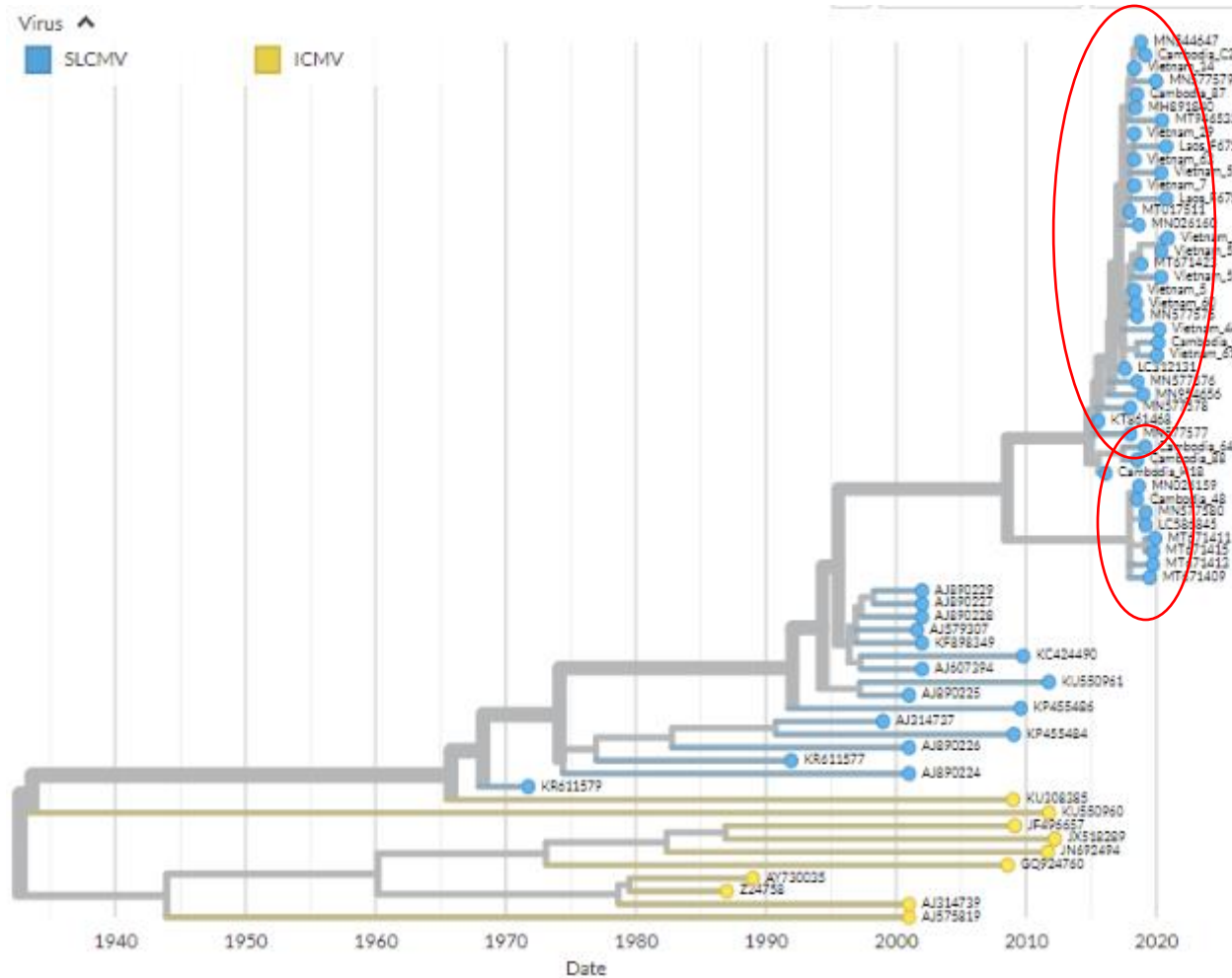


UQueensland

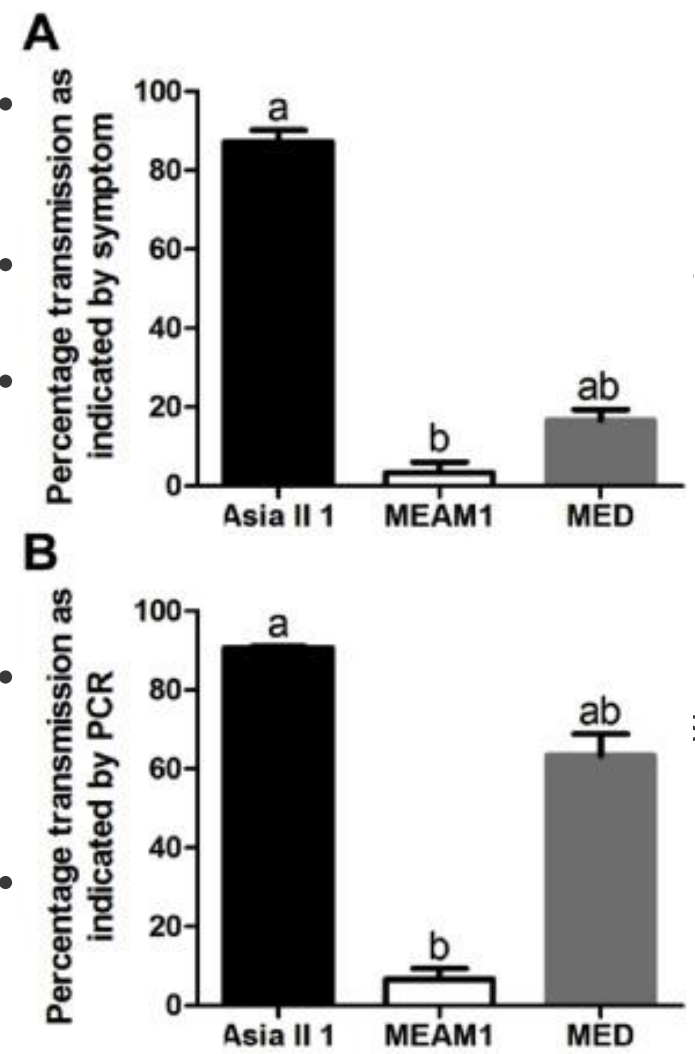


Genome analysis of SLCMV

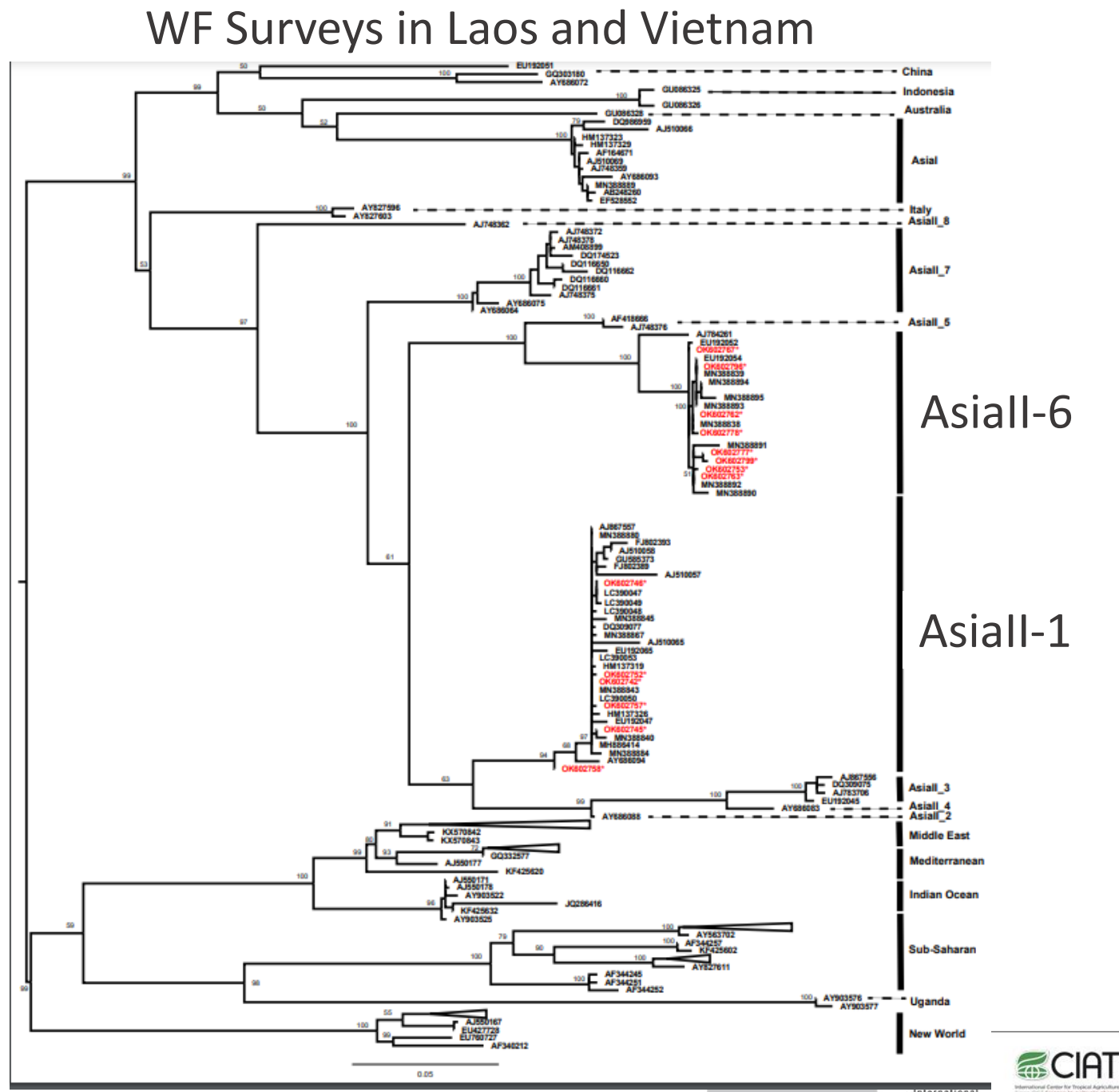
<https://nextstrain.org/community/pestdisplace/CMDASIA1?c=virus&r=location>



Whiteflies (WF)



Chi et al., 2020



Awareness raising leads to early detection and rapid response



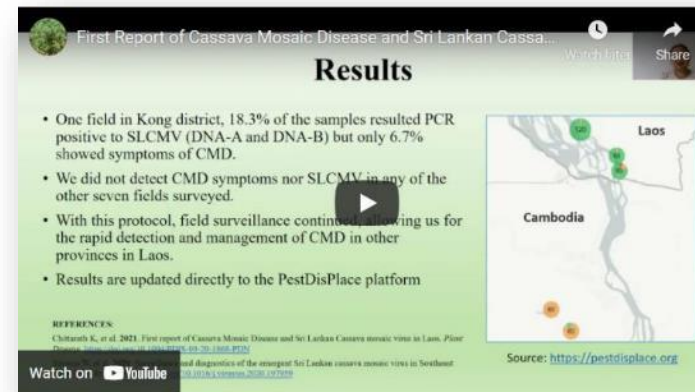
Capacity building: participation in Intl. conferences and scientific publications

Australasian Plant Pathology Society Conference - 2021

- Khonesavanne Chittarath and Pinkham Vongphachanh
PPC – Lao PDR
- Le Thi Hang: PPRI - Vietnam

PUBLICATIONS

- Siriwan W, Jimenez J, et al. **2020**. Surveillance and diagnostics of the emergent Sri Lankan cassava mosaic virus in Southeast Asia. *Virus Research*. <https://doi.org/10.1016/j.virusres.2020.197959>
- Leiva AM; Siriwan W, et al. **2020**. Nanopore-based complete genome sequence of a Sri Lankan cassava mosaic virus strain from Thailand. *Microbiology Resource Announcements*. <https://doi.org/10.1128/MRA.01274-19>
- Chittarath K, Jimenez J, et al. **2021**. First report of Cassava Mosaic Disease and Sri Lankan Cassava mosaic virus in Laos. *Plant Disease*. <https://doi.org/10.1094/PDIS-09-20-1868-PDN>
- Jimenez et al., **2021**. An optimized nucleic acid isolation protocol for virus detection in cassava (*Manihot esculenta* Crantz.). *MethodsX*. <https://doi.org/10.1016/j.mex.2021.101496>



In summary

- Diagnostic protocols for CMD have been validated >> implement portable LAMP and sequencing in the region (COVID permitting).
- Surveillance for CMD has mapped regions of high and low disease incidence in each country >> clean seed propagation, resistance trials, variety identification
- Validate CWBD decreased incidence >> consider cassava genotyping data
- Improve molecular identification of CWBD >> genome sequencing
- Two biotypes of WF >> Contribution to transmission & dissemination of CMD

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Thank you!

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<https://cassavadiseasesolutionsasia.net/> -



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