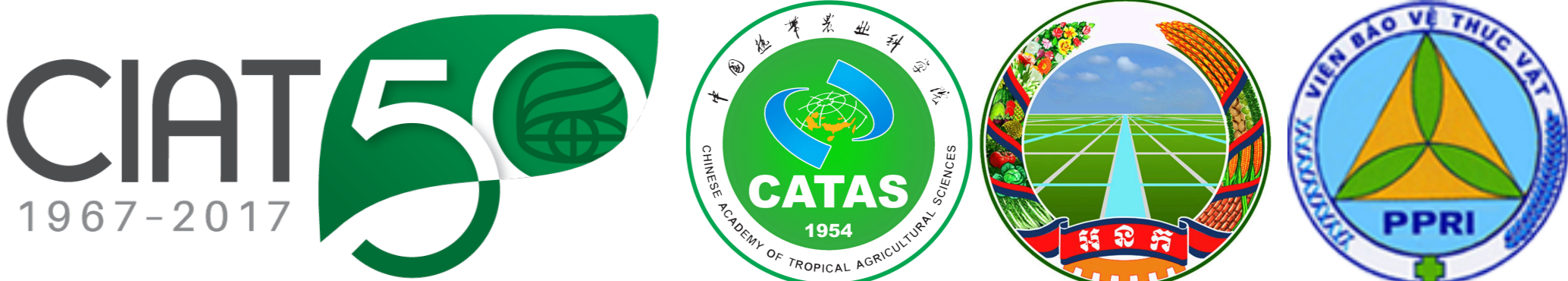


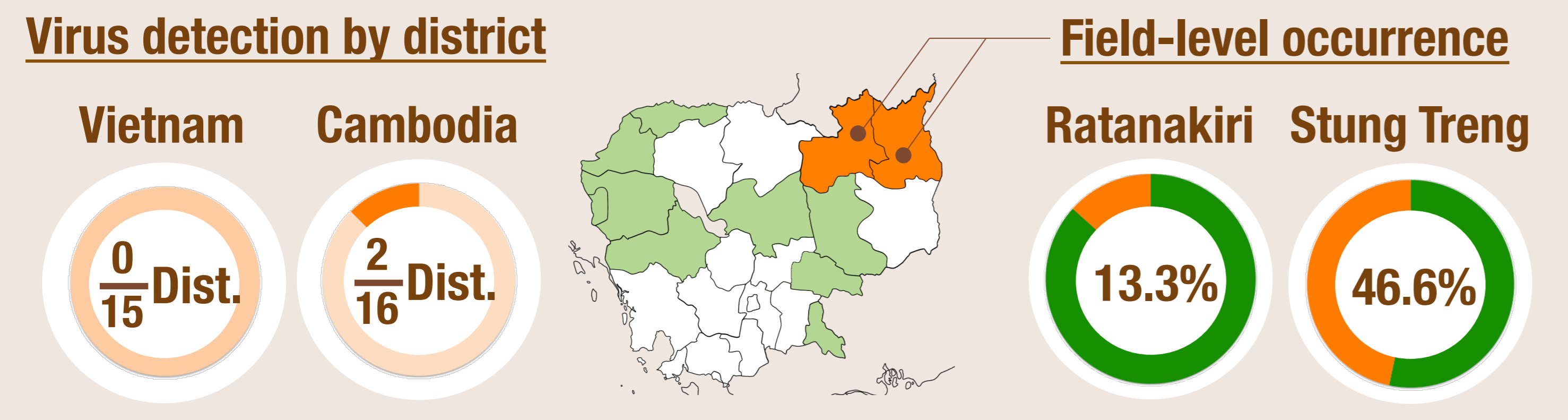
# Incidence and spread of cassava mosaic disease in Cambodia and Vietnam

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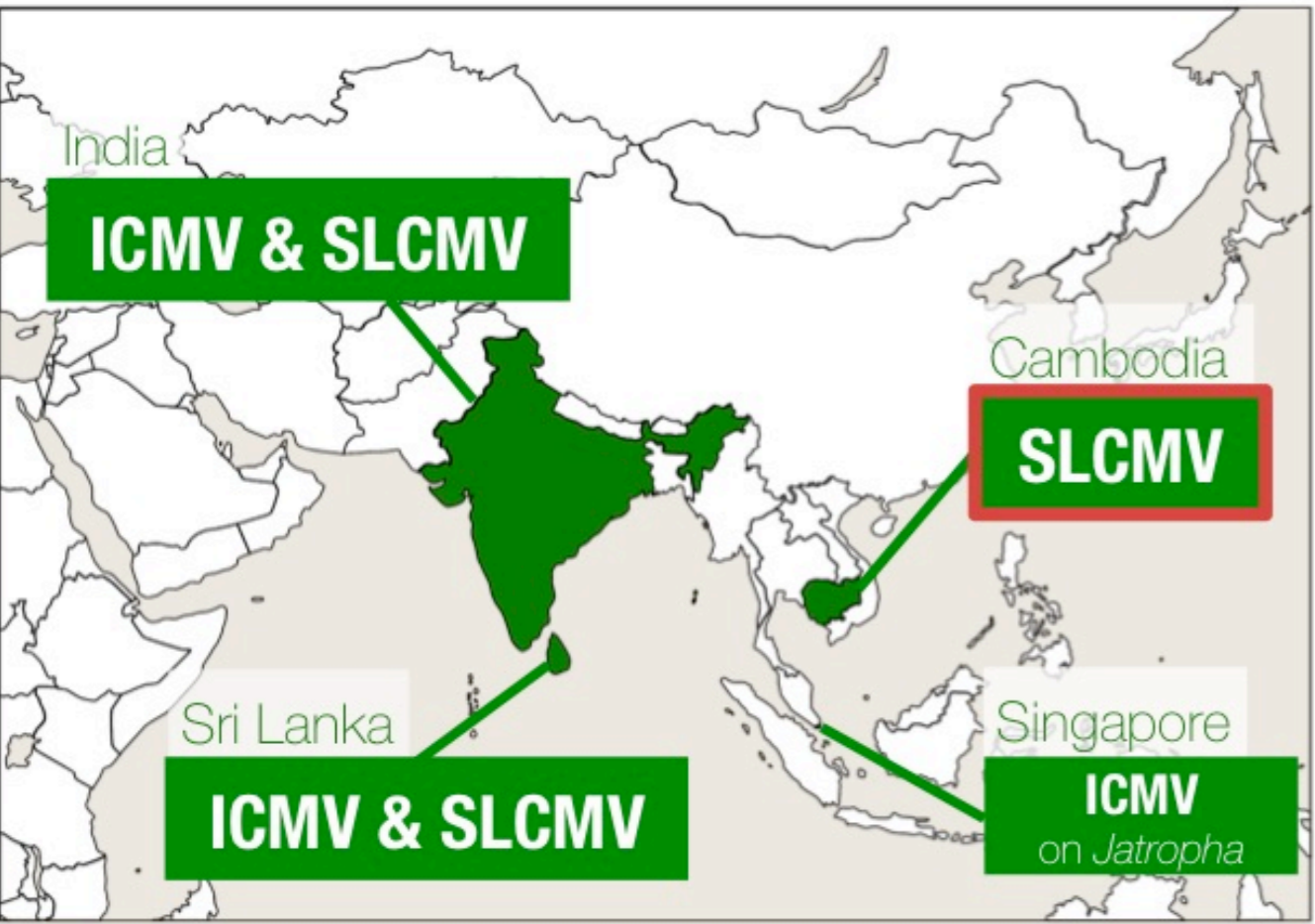
## OVERVIEW



- Cassava production areas of Vietnam and Cambodia in 2014 were 552,760 and 329,781 ha, respectively (FAOSTAT, 2017).
- Sri Lankan cassava mosaic virus (SLCMV) is one of the causal agents of cassava mosaic disease. SLCMV was first detected in Cambodia in the 2015 cropping season.
- Our project conducted binational virus surveillance throughout cassava fields in Cambodia and Vietnam in 2016.
- We found 2 districts with SLCMV occurrence in Cambodia, while no virus infection was found in Vietnam, including Northwest mountainous region (Son La and Yen Bai).
- In addition to the initial historic virus detection site in Ratanakiri province, 46.6% of fields in the neighboring province Stung Treng also contained virus infection, indicating that the virus has spread in 2016.

## 1. Introduction

- In 2016, cassava mosaic disease (CMD) was first reported in Cambodia (**Figure 1**), from a single commercial plantation with Sri Lankan cassava mosaic virus (SLCMV) infection in Ratanakiri province (Ref.1).
- This marked the first confirmation of CMD occurrence in Southeast Asia, with earlier reports confined to South Asia *i.e.* India and Sri Lanka (**Figure 1**; Ref.2&3).



**Figure 1. Distribution of cassava mosaic geminiviruses in Asia in 2016.**



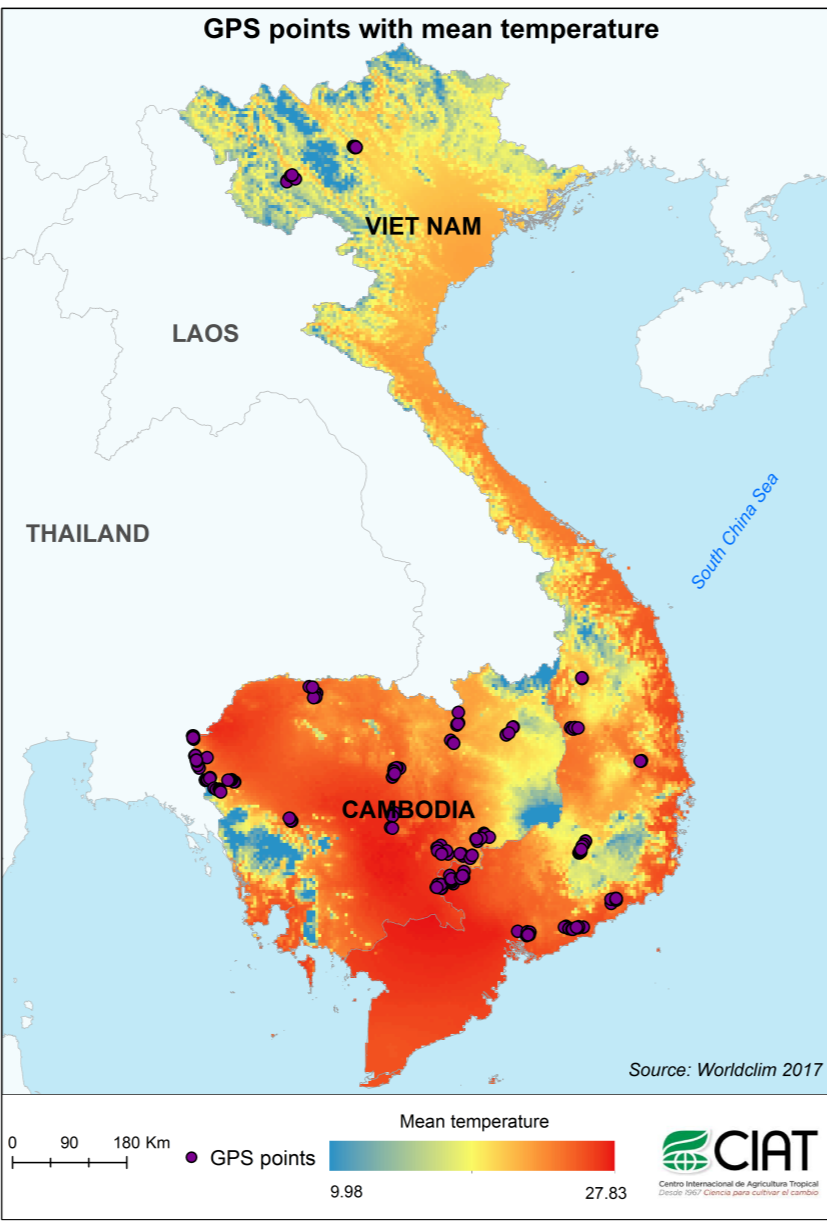
To investigate the current geographical distribution of SLCMV beyond the location of initial detection, we embarked on an extensive binational surveillance program in 2016, pairing virus detection with a related study on movement cassava planting material in Cambodia and Vietnam in the cassava cropping season following the first report of the disease's presence.

## 2. Research Approach

**Table 1. Location of survey sites.**

Country	Surveyed Provinces and Districts
Vietnam	1 Son La Thuan Chau
	2 Yen Bai Van Yen
	3 Gia Lai Chu Prong
	4 Dak Lak Mad Lak
	5 Dak Lak Eaker
	6 Dak Nong Dak Glong
	7 Kon Tum Sa Thay
	8 Gia Lai Krong Pa
	9 Phu Yen Song Hinh
	10 Binh Thuan Bac Binh
	11 Binh Thuan Ham Tan
	12 Tay Ninh Tan Bien
	13 Tay Ninh Tan Chau
	14 Binh Thuan Ham Thuan Nam
	15 Dong Nai Long Thanh
Cambodia	1 Oddar Meanchey Anlong Veaeng
	2 Banteay Meanchey Malai
	3 Pailin Sala Krau
	4 Pailin Pailin
	5 Battambang Kamrieng
	6 Battambang Phnum Proek
	7 Battambang Rattanak mondul
	8 Pursat Kravanh
	9 Ratanakiri Koun Mom
	10 Steung Treng Steung Treng
	11 Kratie Snoul
	12 Tbong Khmum Dambae
	13 Tbong Khmum Memot
	14 Svay rieng Romeas Haek
	15 Kampong Thom Sandan
	16 Kampong Thom Baray

- Surveillance was conducted in 15 districts with the largest cassava production. Koun Mom, Ratanakiri was additionally sampled due to its status as the first reported infection site (**Table 1**).
- For virus diagnosis randomly selected 15 fields/district, and 16 plants/field were surveyed. Plant photographs, young leaf tissues, GPS coordinates, and planting material trading data were collected in a total of 419 fields/households for a total of 6,480 plants (**Figure 2**).
- Total DNA was extracted using the CTAB method, and then used for SLCMV detection by PCR amplifying viral *AC1* (*Replicase*) gene.
- Sequences obtained from PCR amplicons were compared with available SLCMV and ICMV sequences in GenBank.
- The phylogenetic tree was constructed using a maximum-likelihood method with 1,000 bootstrap trials in MEGA7.



**Figure 2. Location of 419 field sampling sites in Cambodia & Vietnam.**

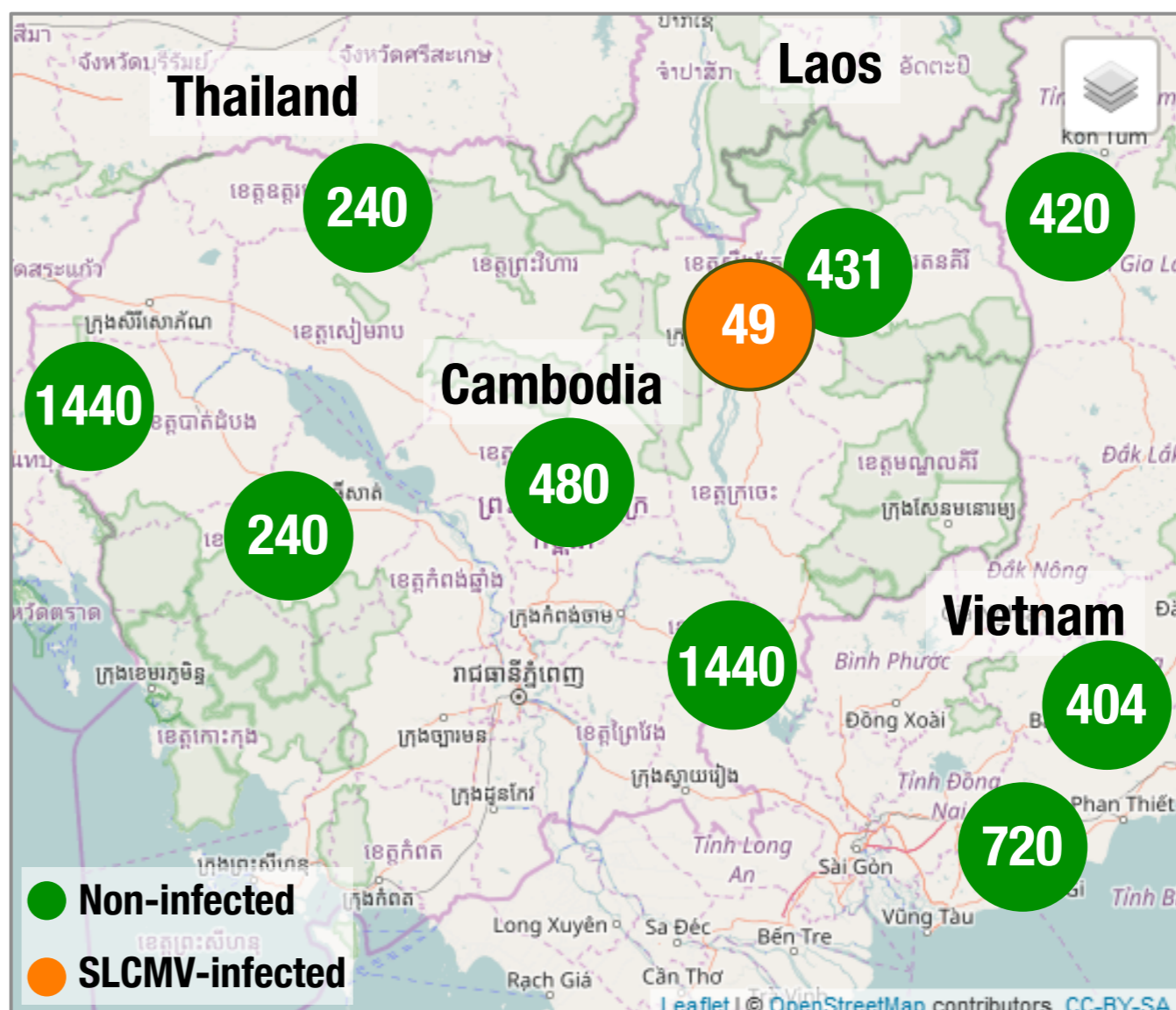
## 3. Results

### KEY FINDING 1:

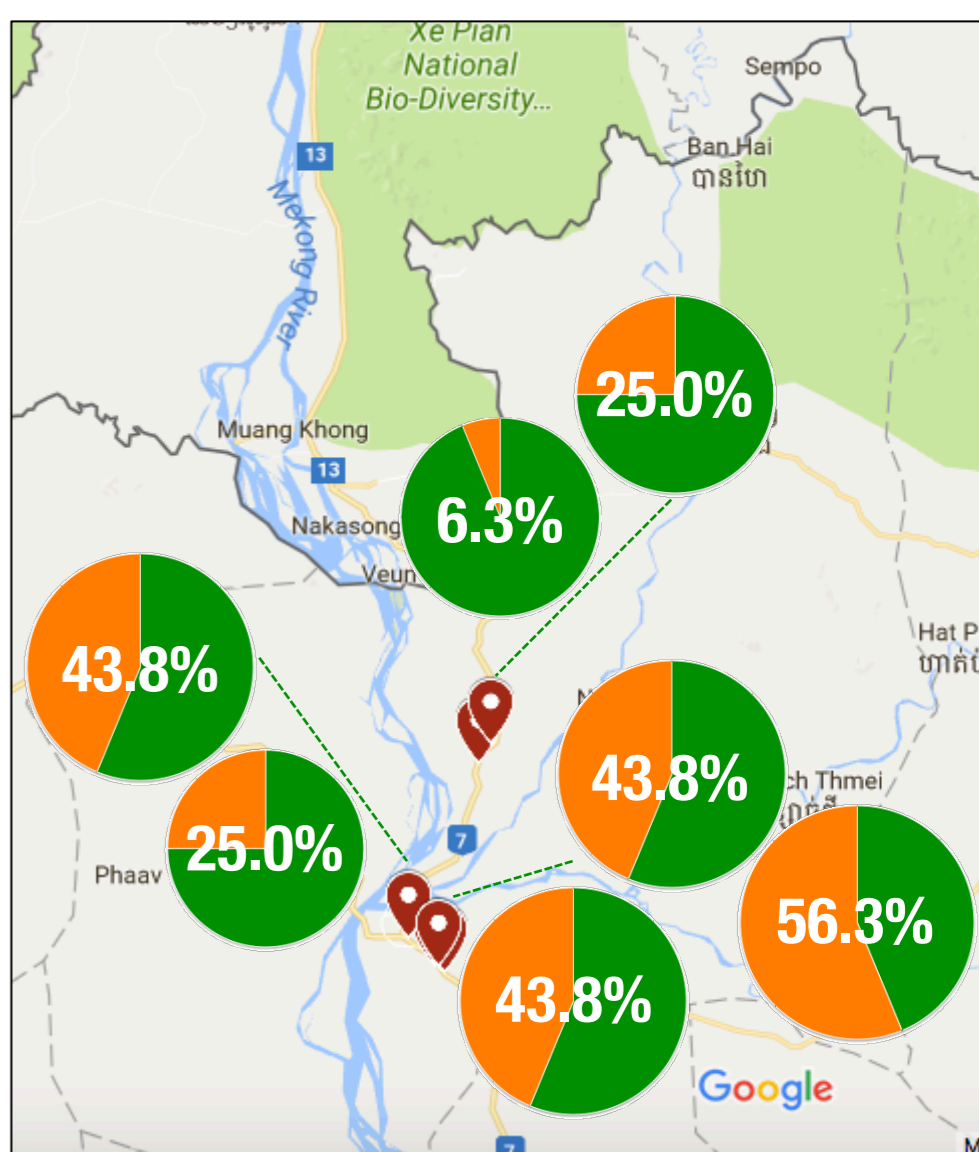
- From the 419 fields surveyed in Vietnam and Cambodia, 9 SLCMV-infected fields (49 total infected plants) were found. Infections were restricted to Ratanakiri and Stung Treng provinces of Eastern Cambodia, with 13.3% and 46.6% of the occurrence, respectively (**Figure 3 & 4**).
- In Stung Treng province, 4 of 7 infected fields showed within-field SLCMV incidences higher than 40% (**Figure 5**). The most remote virus-infected field was approx. 70 km away from the initial reporting site of 2015.
- Over 25% of SLCMV-infected plants did not exhibit clear mosaic symptom in the field.



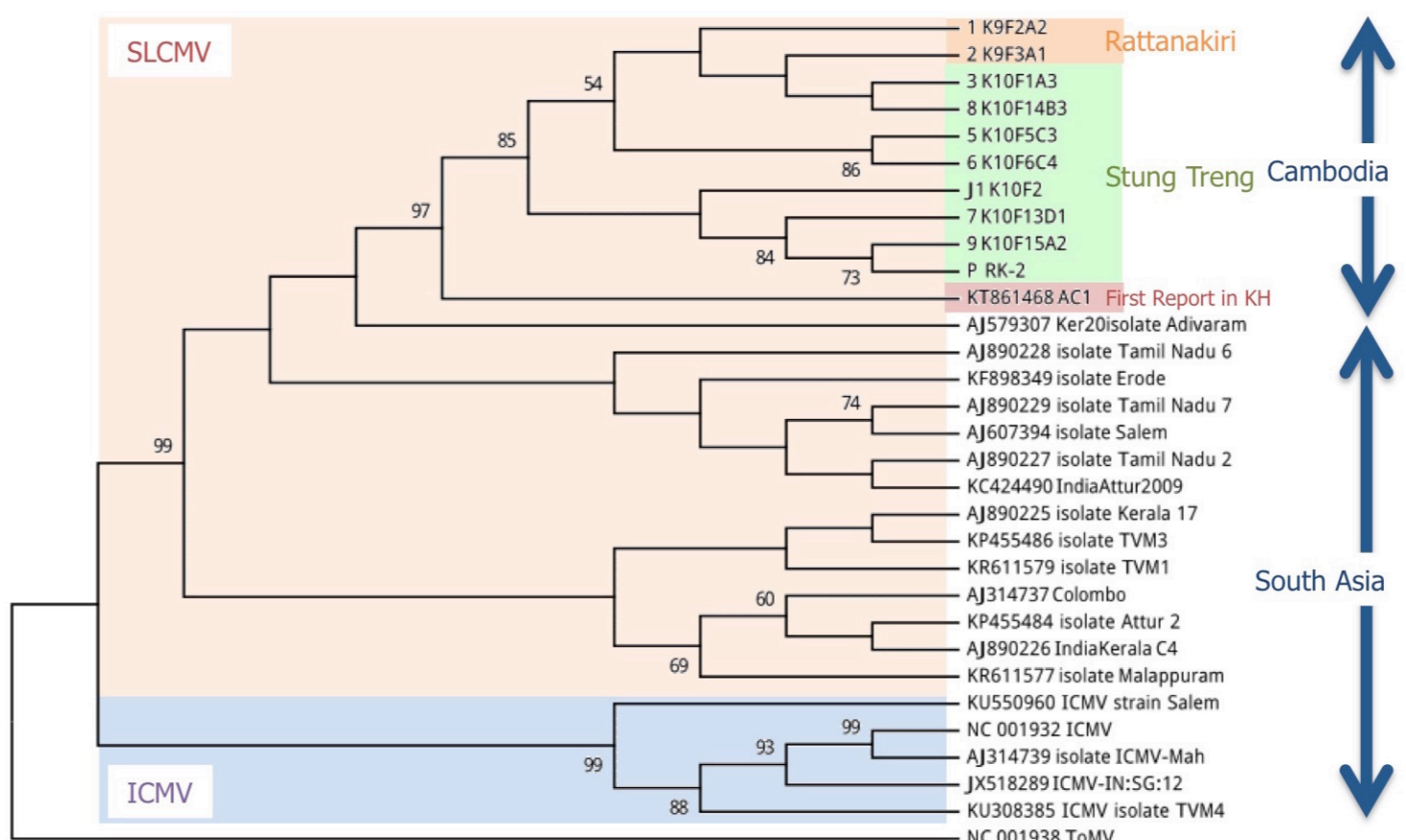
**Figure 3. Symptoms on cassava.**



**Figure 4. Distribution of SLCMV-infected plants in Cambodia & Southern Vietnam.**



**Figure 5. SLCMV incidence in infected fields of Stung Treng province.**



**Figure 6. Phylogenetic tree of *AC1* gene of available SLCMV and ICMV sequences.**

### KEY FINDING 2:

- The resulting topology of phylogenetic tree showed that:
  - (1) SLCMV and ICMV were clearly differentiated into two different groups (**Figure 6**).
  - (2) all SLCMV isolates from our study (10 top seq in Figure 6) were likely derived from the first isolate of Cambodia (**Figure 6**).

## 4. Discussion & Conclusion

Our study confirms the presence of CMD in Ratanakiri province, and that the disease has spread at least 70 km away from the first reported location. Overall, both field-level incidence and infection rates were higher in neighboring Stung Treng than in Ratanakiri, suggesting that SLCMV is undergoing expansion to further production areas. It is likely that such long distance spread is mediated through stake movement as farmers obtain new planting materials. Interestingly, our seed trading survey revealed that most virus infected fields acquired planting materials from their own stock in 2015 (data not shown). Our surveillance found infected plants showing both systemic and non-systemic (limited to upper leaves) symptoms, indicating that both planting material exchange and whiteflies might be contributing to vectoring SLCMV in Cambodia in 2016. The geographical distribution found in this study suggests that regulation of stake movement may still be options to contain the disease. Although our study did not detect SLCMV in Northwestern Vietnam, once CMD occurs it may spread rapidly to smallholder farms with limited access to phytosanitary information. Clean and relatively isolated area like Northwest highlands could take advantage and specialize in clean stake business development. The development of short- and long-term management strategies is urgently needed to mitigate the spread and impact of this disease in Vietnam and Cambodia.

## Acknowledgement

We are especially grateful to Dr. Bui Xuan Phong (PPD), Ms. Hanh TH Luu (PPD), Dr. Nhung Thi Tuyet Le (PPRI), Mr. Lam Van Nguyen (SubPPD), and Ms. Diem Ngoc Huynh (SubPPD) for their contribution to surveillance in VN, and also to Ms. Sreymom Tep (GDA), Ms. Sereyath Sorn (GDA), Mr. Somhoum Not (GDA), Mr. Savoeun Dok (PDAFF-Tboung Khmum), Mr. Thorngum Ieng (PDAFF-Tboung Khmum), and Mr. Chhorvorn Phal (independent consultant) for surveillance in KH. We would like to thank Dr. Jingjing Xue, Ms. Xiuqin Luo, Dr. Yuling Qin, Ms. Feifei An, Ms. Wenli Zhu (CATAS) for their great support in diagnostics. We also thank Than Nguyen (GIS team, CIAT) for her help on GIS mapping. We appreciate the assistance of Le Nghiem and Huyen Tran (CIAT) in the survey training. This project was funded by the Australian Centre for International Agricultural Research (ACIAR). We sincerely appreciate the support from the CGIAR Research Program on Roots, Tubers and Bananas (RTB) and JSPS.



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