

Output: RTB-CC2.1.3.2 - Models and decision support systems for understanding and managing seed degeneration

Deliverable: 18037-Equipment and facilities for rapid multiplication innovation

Sustainable rapid production of disease-free cassava planting material

Imran Malik, Jonathan Newby

Alliance of Bioversity International and CIAT (Asia), Lao PDR Office, Dong Dok, Ban Nongviengkham, Vientiane, Lao PDR

Background and Objectives:

Cassava Mosaic disease has spread throughout the main cassava producing regions in mainland Southeast Asia within a very short span of time. SLCMV was first reported in Ratanakiri Province in eastern Cambodia (Wang et al., 2016) and now disease found in main production areas of Vietnam, Cambodia, Thailand. Laos reported the presence of CMD in September 2020 in border region with Cambodia with farmers bring infected stems from family in Cambodia. Subsequently, it was found in Attapeu Province with infected stems introduced by a private sector actor linked to Vietnam.

To inject disease free planting material to the cassava growers and also for rapid dissemination of improved variety(s) CIAT cassava program has established practicable multiplication innovations in Laos and Cambodia during 2020. There are two approaches **a.** In vitro multiplication and **b.** tunnel systems. In the coming season, vitro multiplication will feed the tunnel systems for developing clean seed systems. In vitro multiplication laboratories at NAFRI, Vientiane Laos and at CARDI, Phnom Penh, Cambodia have been upgraded and now operational with both laboratories receiving physical and later virtual training. Tunnel multiplication system has been established in Laos; however, travel restriction (due to COVID-19) slowed down the progress of setting up tunnel multiplication system in Cambodia. The new date for establishing tunnel multiplication system is February 2021.

Status of multiplication in each country

Laos:

In vitro multiplication: Upgraded tissue culture laboratory at Rice Research Centre, NAFRI is operating efficiently. The laboratory has received in vitro KU50 in 100 tubes shipped from CIAT-Colombia (Figure 1). KU50 plantlets were transferred to new culture medium (Figure 2). Initially there were some issues regarding purity of the chemicals that were used for sub-culture and also with bacterial contaminations. After several tele-consultations with Mr. Roosevelt Escobar, CIAT-Colombia, these issues were resolved. The laboratory aims to produce 2,448 plantlets from this batch and keep on maintaining ~200 KU50 plantlets all the time. A test batch (total of 7) of in vitro grown KU50 has been transplanted to the field following hardening in the screen house (Figure 2,3,4). Another batch of 200 in vitro grown KU50 will be transplanted to field mid-February 2021.

The tissue culture laboratory received five IITA varieties, TMEB419, IITA-TMS-IBA980581, IITA-TMS-IBA980505, IITA-TMS-IBA972205, IITA-TMS-IBA920057, that were identified through consultations as suitable for the market segments in Asia and resistant to Cassava Mosaic Disease (CMD) resistant. This material was initially shipped to AGI, Vietnam and subsequently to partners in Laos and Cambodia as in vitro material. These varieties have been successfully multiplied in vitro and a test

batch (a total of 64 plants) of 5 varieties (plant ID-Hanoi 1 to 5) has been transplanted to the field and another 200 plants are at hardening stage in the screen house getting ready to be transplanted to the field at NAFRI.



Figure 1: Variety KU50 arrived at the tissue culture laboratory at RRC, Nhapokh, Laos.



Figure 2: In vitro multiplication (right) and root initiation (left) of KU50 at the tissue culture laboratory at RRC, Nhapokh, Laos.



Figure 3: In vitro plants are transplanted in the soil system in the screen house for hardening.



Figure 4: In vitro plants hardened in the screen house and ready to be transplanted to the field.



Figure 5: IITA clones established in the field at NAFRI, Laos

Tunnel multiplication: CIAT cassava program has established tunnel rapid multiplication system at Naphok research station, Vientiane in collaboration with NAFRI. At this station, 6 tunnels have been established and disease-free planting materials of popular cassava varieties (KU50 and Rayong11) are being multiplied. Different steps of the multiplication described in Figures 6 to 13. KU50 was previously the most widely grown variety in Southeast Asia, however had declined in area in recent years. Experiments in Cambodia showed that it was less susceptible to CMD than other elite Asian varieties¹. However, KU50 has been shown to be more susceptible to Cassava Witches Broom Disease (CWBD). Rayong11 has been shown in participatory studies to be less susceptible to CWBD, however the trials in Cambodia show it is susceptible to CMD. At this stage, maintaining clean planting material of both is seen as a priority until sources of resistance to CWBD can be identified.

Scaling of the rapid multiplication technology has occurred in a partnership with Khouasap Import-Export Company, Bachiang District, Champasak Province another 10 tunnels has been established and, in both places, multiplication is ongoing. A further 6 tunnels will be constructed in early 2020 in partnership with a USDA funded project in southern Laos.

Approximately 35 plant (two long stem each plant) give about 700 two node cuttings were horizontally placed in the sand bed in each tunnel. Moderate level of fertilizer application and adequate soil moisture, optimum temperature (30°C) and high humidity can produce about 5000 plants in a year per tunnels (Table 1).

Table 1. Number of plants produced from tunnel systems-an example of tunnel multiplication in one year from two tunnels.

Variety	Start date in the tunnel	No of viable sprout /cutting	No of days to get new plantlets	No of days to transplant to field	Total Number of plants in the field from 5 cuttings	Success rate of trans planting to the field (%)
KU50	23 rd January, 2020	900	50 ± 4.6	90 ± 12	*3850	100
Rayong11	12 th March 2020	1000	47 ± 3.4	100 ± 5	5040	100

* Lost one batch to mealybugs

Challenges: Keeping the temperature at optimum level is a challenge without automated system in place. Furthermore, access to ground water and electricity for irrigation is essential for the success of this innovation.

¹ Deliverable: 17008-Effect of Cassava Mosaic Disease (CMD) on Cassava Value chain



Figure 6: Aerial view of six multiplication tunnels and screen house at NAFRI research station at Naphok, Vientiane, Laos.



Figure 7: Two node stem-cutting horizontally placed in the sand bed (left) and sprouted nodes after a week of planting (right)



Figure 8: Variety KU50 sprouted and ready to be translated to trays with cocopeat for rooting.



Figure 9: Cassava plantlets are planted in the trays with substrates (rice husk and cocopeat) received from Mekong Timber Plantations. Cocopeat with osmocote slow release fertilizer are used in timber nursery for multiplication.

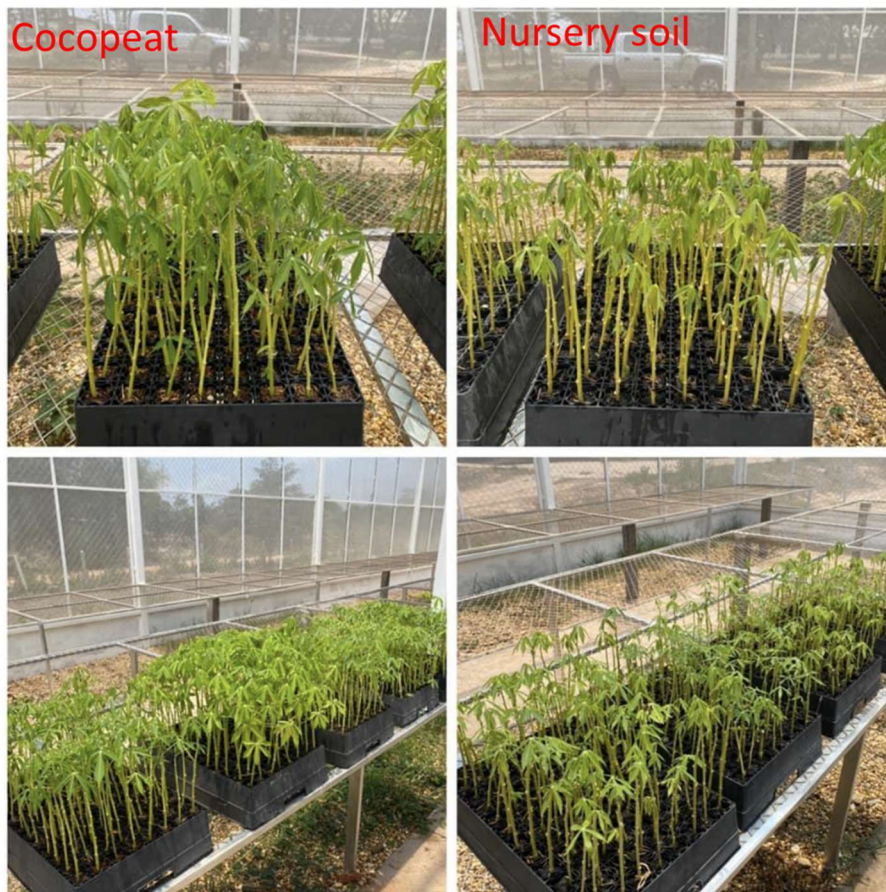


Figure 10: Cassava plantlets are growing in screen house for rooting. Cocopeat with osmocote (slow release fertiliser) are used in timber nursery for multiplication and it also performed better for cassava.



Figure 11: Cassava plantlets are growing in screen house for rooting. Cocopeat with osmocote (slow release fertiliser) are used in timber nursery for multiplication and it also performed better for cassava.



Figure 12: Transplanting in the field with a drip irrigation system in place.



Figure 13: Heavy rain flooded part of the field and some transplanted plantlets exposed to soil waterlogging for couple of days.

Cambodia

In vitro multiplication: Upgraded tissue culture laboratory at CARDI, Phnom Penh, is operating efficiently. The laboratory has received in vitro KU50 in 100 tubes shipped from CIAT-Colombia. KU50 plantlets were transferred to new culture medium. Initially there were some issues regarding purity of the chemical that were used for sub-culture, intensity of lights in the growth room and also with bacterial contaminations. After few tele-consultations with Mr. Roosevelt Escobar, CIAT-Colombia, issues were resolved.

The tissue culture laboratory received five IITA varieties, TMEB419, IITA-TMS-IBA980581, IITA-TMS-IBA980505, IITA-TMS-IBA972205, IITA-TMS-IBA920057, reputed for Cassava Mosaic Disease (CMD) resistant, from AGI, Vietnam as in vitro material. In vitro multiplication is ongoing and about 550 in vitro grown plants are ready to be transplanted. A test batch of these varieties have been already transplanted (see figure). A big flood in Cambodia during 2020 delayed transplantation to the field.



Figure 14: In vitro multiplication of KU50 at CARDI tissue culture laboratory.



Figure 15: In vitro multiplied IITA varieties translated at CARDI field.

Tunnel multiplication: CIAT cassava program plan to establish tunnel rapid multiplication system at CARDI, Phnom Penh and General Directorate of Agriculture (GDA) farm near Kampong Cham, Cambodia. Site selection has been completed. In each site 6 multiplication tunnels will be established.

Acknowledgement: Thanks to Mr. Roosevelt Escobar from CIAT Colombia for spending time late 2019 in Vientiane to establish the first tunnel and tele-consultation with national scientists. Mr. Erik Delaquis (CIAT-Laos) for helping establishing the tunnels.

In Laos, Mr. Laothao Yubee (CIAT-Laos) and Mr. Saythong Oudthachit (NAFRI) for continuously looking after the Tunnel systems for multiplication. Ms Soukphathay Simeuang looking after the in vitro multiplications and hardening of the plants in the screen house. In Cambodia, Dr. Chhourn Orn managing the in vitro multiplication and transplantation of the plants to the field.

Funding from ACIAR, RTB, China-fund and India- fund are gratefully acknowledged.