

A RAPID METHOD FOR THE PROPAGATION OF CASSAVA (*MANIHOT ESCULENTA* Crantz)

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SUMMARY

A study of methods to propagate cassava rapidly is under way at CIAT. Results so far show that un-lignified shoot tips when planted under mist produce roots during the second week after planting. These rooted cuttings may be transplanted into the field after a ten day hardening off period.

Shoots are produced from stem cuttings planted in an environment of high humidity. More rapid production of shoots occurs in humid chambers than under mist. Propagation by this method is twenty times more rapid than by conventional methods.

RESUME

Des méthodes de propagation rapide du manioc sont en cours d'étude au CIAT. Les résultats déjà obtenus montrent que les boutures de pousse non aoutées produisent des racines en deux semaines lorsqu'elles sont plantées en condition brumeuse. Les boutures enracinées peuvent être repiquées au champ après une période fortification de 10 jours.

Les pousses sont obtenues à partir des boutures de tige plantées en milieu d'humidité élevée. On peut obtenir une production de pousses plus rapidement en chambre humide qu'en condition de brume. Cette méthode de propagation est vingt fois plus rapide que par les procédés conventionnels.

RESUMEN

Se lleva a cabo en el CIAT, un estudio sobre métodos rápidos de propagación de yuca. Hasta el momento, los resultados muestran que cuando se siembran brotes terminales, bajo nebulización, producen raíces durante la segunda semana después de la siembra. Estas estacas, ya enraizadas, se pueden transplantar al campo después de un período de acondicionamiento de diez días.

Los vástagos se producen a partir de estacas sembradas en un ambiente de elevada humedad. La producción de vástagos es más rápida en cámaras húmedas que bajo nebulización. La propagación por este método es veinte veces más rápida que por los métodos convencionales.

INTRODUCTION

Crop improvement programmes frequently encounter problems associated with the rate of multiplying planting material. A low multiplication rate impedes agronomic testing of a new variety, and the scarcity of planting material delays distribution to farmers.

This problem is more acute with vegetatively propagated crops. Table 1 compares the multiplication rate of cassava with that of some sexually propagated crops such as rice and maize.

It is important therefore that a cassava improvement programme should have an organized procedure for rapidly multiplying improved cultivars. A successful propagation programme must be able to multiply rapidly from an initial stock of a small number of plants. For example, the CIAT cassava germ plasm bank with more than 2,000 cultivars is maintained with four plants per cultivar because of the problems associated with growing large numbers of plants. In extreme cases, one plant may be the sole foundation stock.

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PROPAGATION METHODS

Stem cuttings, 15-30 cm in length, are used as planting material to establish commercial crops of cassava. These cuttings are selected from the leafless woody portion of the stem of plants normally older than nine months. Results from experiments comparing the size and origin of cuttings generally agree that longer cuttings from the basal portion of the stem germinate and establish better than apical cuttings.^{4,5,7,8,10} However, it has been shown that under optimum conditions for establishment and with the use of good quality planting material, the length of cutting has no effect on establishment.¹²

Single node, woody cuttings have been successfully used for rapid propagation in Venezuela and Brazil (Alvarez-Luna, personal communication). Where internodes are short, preventing the preparation of single node cuttings, eye cuttings (a bud together with a segment of stem split longitudinally) may be used. This method was developed initially in an attempt to produce plants free of cassava bacterial blight from infected stems¹¹.

Techniques have been developed using shoots produced from stem cuttings grown under conditions of high humidity. The first report recommended the use of shoots, excised together with a heel of lignified tissue². These shoots produced roots when placed in a propagating case.

It has recently been shown, however, that non-lignified shoots without the lignified heel also produce roots^{3,13}. These shoots have been used to prepare single node cuttings with the leaf attached which root under mist¹³. Leaves also have been rooted^{10,13}, but so far no technique has been found to stimulate shoot production from rooted leaves. A programme was initiated at CIAT in 1971 to develop simple, efficient methods for rapid propagation of cassava.

MATERIAL AND METHODS

Initially, a bamboo lathhouse was used for preliminary studies of double and single node stem cuttings. Later, a mist propagation unit was used to investigate the potential of shoots as propagules for rapid propagation. The system incorporated an electric leaf (1) that controlled the intermittent mist according to the evaporation rate of water. Subsequently, a propagating frame using a wood and polyethylene roof over a bed of sterile soil was constructed (Fig. 1). Hollow concrete blocks, forming the retaining wall of the soil bed, were cemented on one open side, placed hollow surface uppermost and filled with water. The roof of the frame partially covered the water which evaporated, thus increasing the humidity within the frame.

RESULTS

During the initial phase, observations of the rooting of single and double node cuttings in the lathhouse showed that roots were produced from three distinct sites:

1. Initially from discreet zones associated with the leaf scar and internode bud.
2. Later from the basal end of the cutting associated with callus produced from the cambium.
3. From the etiolated base of the new shoot that developed from the cuttings.

The ability of young shoots to produce roots under such conditions suggested that the excised shoot demonstrated a potential as the propagule upon which to base a rapid propagation system. The efficiency of this system depends upon the success of two phases, shoot production and the rooting and establishment of shoots.

Shoot production

Shoot production under mist was evaluated using 25 cm stem cuttings, planted horizontally immediately below the surface of the medium. Shoots were removed when they had attained a length of 8 cm. Woody stem cuttings produced more shoots after three months than non-lignified cuttings. Most of the latter, despite some rooting, were dead at the end of the experiment. An efficiency of two shoots per node was achieved from woody cuttings of cultivars M Col 75 and 1438 (Table 2).

It was observed on termination of the experiment that, despite a shoot production efficiency of two shoots per node in two cultivars, these shoots were produced from a comparatively small number of nodes, the majority of the nodal buds remaining dormant. This capability for multiple shoot production (Fig. 2) from one bud demonstrated a potentially efficient shoot production system.

A subsequent experiment showed that single and two node woody cuttings produce more shoots per node than longer cuttings, (Table 3). In these minimal size cuttings, all nodal buds were stimulated into shoot production.

To develop a system more adaptable to developing countries, the efficiency of shoot production under mist was compared with production in a propagation frame.

Cuttings grown in the propagation frame produced more shoots than the cuttings under mist (Table

3). The relative humidity under both regimes was similar, but large temperature differences were noted (Table 4), the air in propagation frames reaching 45° at midday. This high temperature had no adverse effects on growth and may be responsible for the increase in shoot production. Other experiments to investigate the effect of environment on shoot production are under way.

Observations were made on the rooting of excised shoots placed under intermittent mist. During the first two days under mist, the shoots wilted but regained turgor after the third or fourth day. Most expanded leaves fell during the first week. The formation of callus at the shoot base began ten days after planting, and roots developed from the callus region during the following ten days (Fig. 3). Twenty days after planting, the root system was sufficiently developed to enable transplanting.

There were no differences in rooting of shoots from woody or non-lignified stem cuttings of different cultivars. Root formation was excellent in both 2 mm gravel and perlite media.

CIAT propagation system

A routine system for the propagation of promising selections has been developed based on the above observations. Two node stem cuttings are planted horizontally in a propagation frame with sterile well-structured soil. Shoots are removed when they are 8 cm long using a sterile blade. A 5 mm section of stem is left from which new shoots are produced. Before planting, all leaves except the unopened leaves and the newly opened leaf are removed when they are 8 cm long using a sterile blade. A 5 mm section of stem is left from which new shoots are produced. Before planting, all leaves except the unopened leaves and the newly opened leaf are removed. The shoots are planted in 2 mm gravel under mist 5 cm apart and left to root for three weeks. After this period the rooted shoots are carefully removed from the medium and planted into peat pots with sterile soil. After a further ten days under reduced mist, during which time the roots break through the peat pot, the rooted shoots are transferred to the field. Fig. 4 shows a typical rooted shoot ready for transplanting. More than 95 percent survival of shoots is regularly achieved using this system.

Recent studies show that shoots can be rooted in propagating frames using IBA hormone. Further work is under way to refine this technique.

DISCUSSION

The rapid propagation programme is developing the entire system of shoot production and rooting using techniques applicable to the developing countries. Although mist propagation which maintains low tissue temperature under high light intensity⁶ offers more advantages than any other system of propagation, the success of an intermittent mist system depends on a reliable supply of both water and electricity. The water supply must be low in salt content; the accumulation of salt on the leaves through evaporation results in leaf loss. Water pressure greater than 2.5 kg/cm² is needed to form mist at the jets. In the tropics many experimental stations lack a reliable, low salt, high pressure water supply. As the salt concentration of the well-water at CIAT is too high for mist propagation, rain water is used.

An intermittent mist system entirely depends on a reliable electricity supply. Similarly, many water supplies rely on electricity as a source of energy for pumps. Unfortunately, electricity supplies in the developing nations of the tropics frequently fail. Therefore, a successful propagation system for cassava in the tropics should not depend upon mist propagation.

The rapid propagation system described above is capable of producing 18,000 stem cuttings after one year, starting with one mature plant (Fig. 5). The efficiency of this method is due to the exploitation of multiple shoot production. Chant and Marden's method², producing 500 stem cuttings from a seed after two years did not allow further shoot production from each node because of the removal of the lignified heel with each shoot.

Using a standard field multiplication procedure, starting from one plant with 15 cm stem cuttings as planting material, it is possible to produce thirty plants after one year. Each plant provides thirty more cuttings, a total production of 900 15 cm stem cuttings. By reducing the cutting size to two nodes which establish under good management³, this output can be increased to 1,800 stem cuttings. Therefore the above system is much more rapid than previously used methods.

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TABLE I

Comparison between sexually and asexually propagated crops.

	Rice	Maize	Cassava
Crops per year	2	2	1
Multiplication rate/year certified seed production	160,000	640,000	30
Breeders' seed production	25 million	1 million	90

TABLE 2

Shoot production from woody and non-lignified 25 cm stem cuttings of four cassava cultivars after three months.

Mean of twelve cuttings.

Cultivar	Type of cutting				mean	
	non-lignified		woody		Shoots/ cutting	Shoots/ node
	Shoots/ cutting	Shoots/ node	Shoots/ cutting	Shoots/ node		
M Co1 22	4.3	0.46	13.7	1.07	9.0	0.77
M Co1 75	6.7	0.95	11.6	1.99	9.2	1.46
M Co1 76	3.2	0.32	7.9	0.89	5.6	0.61
M Co1 1438	2.2	0.34	10.2	1.97	6.2	1.16
Mean	4.1	0.52	10.9	1.48	7.5	1.00

L.S.D. (P=0.05) no/cutting between ages = 1.13: between vars.=1.60
no/node " " = 0.12: between vars.=0.17

TABLE 3

The effect of environment and size of cutting on shoot production after 40 days (M Col 1438)

Cutting size	Intermittent mist		Propagation frame		Mean of environments	
	Shoots/cutting	Shoots/node	Shoots/cutting	Shoots/node	Shoots/cutting	Shoots/node
1 node	0.9	0.9	1.1	1.1	0.9	1.1
2 node	1.4	0.7	2.9	1.4	2.2	1.1
10 cm	1.8	0.5	3.1	0.9	2.5	0.7
20 cm	3.0	0.5	4.2	0.9	3.6	0.7
Mean	1.8	0.6	2.8	1.1	2.3	0.9

L.S.D. (P=0.05) Between cutting size = 0.22.
 Shoots/node Between environment = 0.15

TABLE 4

A comparison of air temperature and relative humidity under mist and in a propagation frame

	Air temp. °C		Relative Humidity(%)		
	Max.	Min.	Max.	Min.	Hours/day RH at 80%
Mist	32.3	20.2	96.0	42.5	14.0
Propn. frame	45.1	22.8	99.0	39.5	15.5



Figure 1. *Propagation frame showing simple inexpensive construction.*



Figure 2. *Multiple shoot production from one nodal bud of cassava.*



Figure 3. Shoots removed from stem cuttings (A), and after three weeks under mist (B).

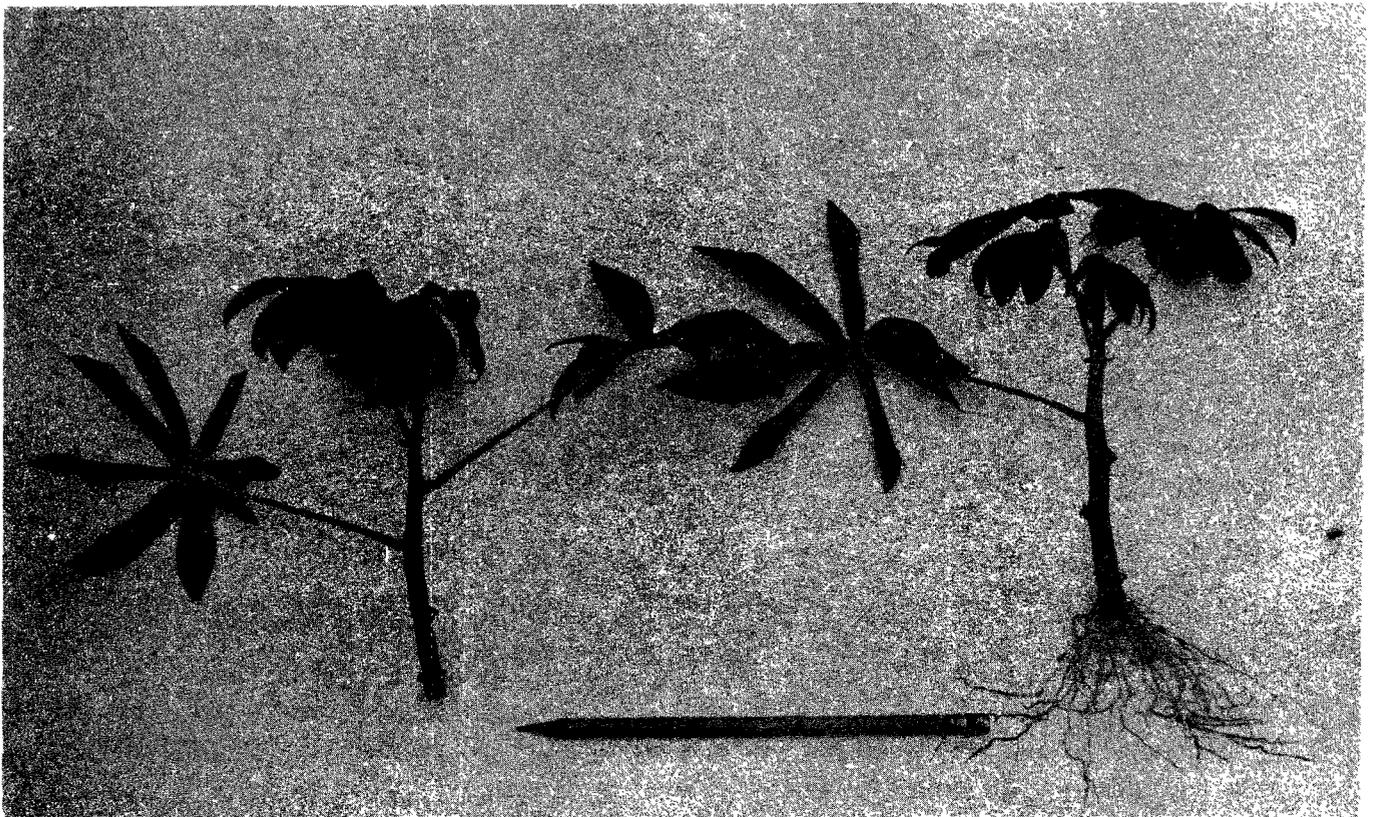
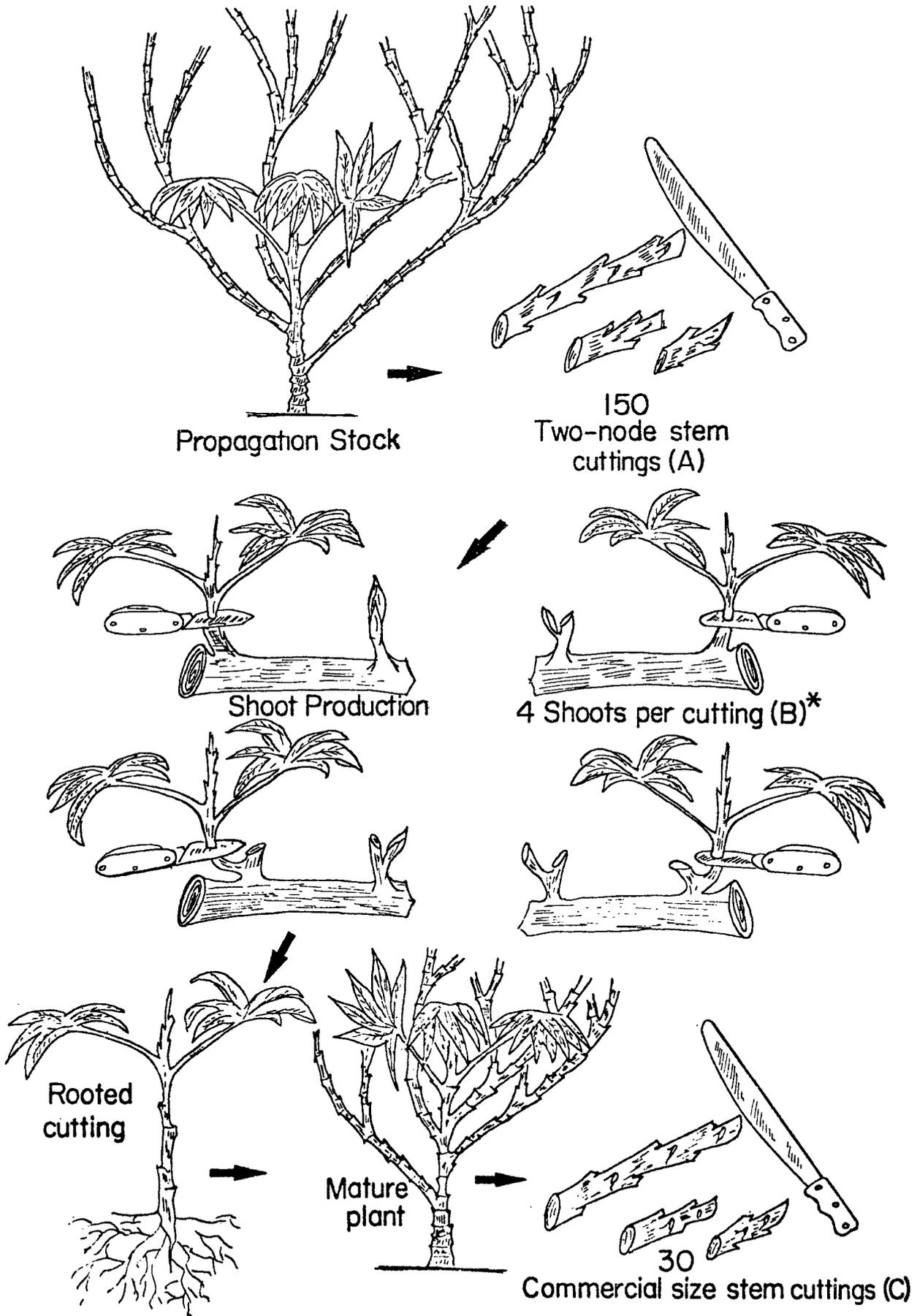


Figure 4. Rooted shoot of variety Llanera ready for planting out in the field.

Fig. 5 Diagram of CIAT'S Propagation System



Multiplication rate, $A \times B \times C = 150 \times 4 \times 30 = 1800$

* Two shoots per node (data from current experiment)