# CHARACTERISTICS AND PRODUCTION OF CASSAVA STEM CUTTINGS FOR RAPID **MULTIPLICATION METHOD**

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#### ABSTRACT

Selection of stem cuttings is an important step for production of high-quality seedlings by rapid multiplication method. This study aimed to assess the sprouting capacity of cassava stem cuttings of different diameters. Experiments were conducted at five planting seasons (June, July, August, September, and October) under a completely randomized design in a factorial scheme  $(5 \times 2)$ , with ten treatments and 20 replications. Treatments consisted of combinations between planting

seasons and stem cuttings with diameters lower (from 15 to 19.99 mm) and higher than 20 mm ( from 20 to 25 mm). Each stem cutting was measured regarding length, diameter, and weight. The mean number of days for emergence, percentage of survival, and sprouting index were assessed at each plot. Planting of stem cuttings with diameter between 20 and 25 mm in September was adequate for obtaining cassava cuttings for the rapid multiplication method.

PALAVRAS-CHAVE: Manihot esculenta Crantz, multiplication rate, vegetative propagation, seedlings.

# CARACTERÍSTICAS DA MANIVA E A PRODUÇÃO DE ESTACAS DE MANDIOCA PELO MÉTODO DE MULTIPLICAÇÃO RÁPIDA

#### RESUMO

A seleção de manivas é uma etapa importante para produção de mudas de alta qualidade pelo método de multiplicação rápida. O objetivo desta pesquisa foi avaliar a capacidade de brotação de manivas de mandioca de diferentes diâmetros. O experimento foi conduzido em cinco épocas de plantio de manivas com duas gemas da cultivar Apronta Mesa (junho, julho, agosto, setembro e outubro). O delineamento experimental foi o inteiramente casualizado, em fatorial (5x2), com 10 tratamentos e 20 repetições. Os

tratamentos foram as combinações entre as épocas de plantio e de manivas com diâmetro menor do que 20 mm (15 a 19,99 mm) e com diâmetro maior do que 20 mm (20 a 25 mm). Foram mensurados em cada maniva o comprimento, diâmetro e peso de maniva e em cada parcela o número médio de dias para a emergência, percentual de sobrevivência e o índice de brotação. O plantio de manivas com diâmetro entre 20 e 25 mm no mês de setembro é o mais indicado para a obtenção de estacas pelo método de multiplicação rápida.

KEYWORDS: Manihot esculenta Crantz, taxa de multiplicação, propagação vegetativa, mudas.

### **1 INTRODUCTION**

Cassava (Manihot esculenta Crantz) is an important species cultivated mainly for commercialization of roots, as it is a highly energetic starch-rich food (El-Sharkawy, 2012). Among the different forms of consumption, the most outstanding is in natura and as flour or chips (Aguiar, 2013), besides being a constituent of several industrialized products, such as sausages, glues, papers, packaging, textiles, paints, and drugs (Mattos & Cardoso, 2003, Morais et al., 2014). Despite its importance for food security, mean root yield in family farms is low in Brazil, with values of approximately 5,770 kg ha<sup>-1</sup> (Brasil, 2009).

Cassava propagation is carried out vegetatively by planting stem sections, called stem cuttings. Traditional planting is usually carried out in non-fertilized furrows and at spacings that vary according to the region of cultivation, cultivar, and technological level. One of the limitations of a traditional planting is low multiplication rate, which varies from 1:7 to 1:10, i.e., up to 10 stem cuttings are produced from each stem (Ceballos et al., 2011). The quality of propagation material is important and influences plant growth and development and, consequently, has a direct effect on yield. However, the low multiplication rate in high-quality propagation materials is a problem already observed by researchers and producers (Ceballos et al., 2015).

Cassava is considered a species of high rusticity, as it may be cultivated in marginal areas of properties, where fertility is suppressed or reduced to small amounts of organic fertilization and yet provides a variable amount of root production. Despite this limitation, this crop has high agronomic and broad production potentials if grown under appropriate conditions. Ishida et al. (2016) pointed out that despite high rusticity, annual succession of crops within the same area leads to onset of bacterial diseases on crops, precluding the use of stems in the following seasons. According to Shiji et al. (2014), micropropagation techniques for rapid multiplication of cassava have been promising to obtain seedlings with assured sanitary quality and viability for transplanting to the field two months after planting.

The rapid multiplication method, which is developed by the International Center for Tropical Agriculture (CIAT) and adapted to northeastern (Santos et al., 2009) and southern (Koefender et al., 2015) Brazil growing conditions, is based on the use of stem cuttings with two buds planted in beds covered with transparent plastic film, where rooting and emission of sprouts are favored. Subsequently, stem cuttings are collected, placed in containers with water for rooting, and then planted in substrate packaging for seedling production. Cassava seedlings are obtained at the end of this process, which will be able to be transplanted to production fields after acclimatization period. This method has a potential to increase rate of use of propagating material, especially in regions where storage of stems is necessary due to frosts during winter, when high mortality rates occur mainly due to bad storage conditions (Piza & Pinho, 2002).

Besides edaphoclimatic conditions, which interfere with rooting and sprouting of cassava stem cuttings, visual assessment of stems is essential for rapid multiplication method as it shows maturity level and presence of pathogens. In addition, length, diameter, and weight of stem

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cuttings indicate accumulation of reserves and water content, which vary with stem diameter and interfere with rooting and sprouting potential (Bezerra, 2012). The balance between water content and reserves in stem cuttings is important for root emission and subsequent sprouting (Alves, 2006), resulting in vigorous stem cuttings and hence cassava seedlings with good rooting capacity.

Due to stagnation of production and reduction of areas planted with cassava crop in the State of Rio Grande do Sul (IBGE, 2019), studying alternative propagation methods is important to enhance production of propagation materials with physiological quality and sanitary, and consequently root yield. This study aimed to assess the sprouting capacity of cassava stem cuttings of different diameter intervals at five planting seasons.

### **2 MATERIAL AND METHODS**

Multiplication of propagation material (cultivar Apronta Mesa) was carried out in the 2016/2017 season for the setting up of experiments in the following season. The multiplication area was planted in the second half of October, and conventional soil tillage was carried with plowing and harrowing. Planting was carried out at a spacing of 1.0 × 0.6 m with six-bud stem cuttings, without additional fertilization. Weeds were controlled through weeding until inter-row closure, and cassava stems were collected in the second fortnight of May 2017.

The experiment was conducted in Rio Grande do Sul (Brazil), at the geographical coordinates 28°38'19" S and 53°36'23" W, with an average altitude of 452 m, at five planting seasons: June, July, August, September, and October. The experimental design was a completely randomized design in a factorial scheme (5  $\times$  2), with ten treatments and 20 replications. Treatments consisted of combinations between planting seasons and stem cuttings of the cultivar Apronta Mesa, with diameters lower (15 to 19.99 mm) and higher (20 to 25 mm) than 20 mm. Cassava stems were cut with a manual saw, and each cutting had 2 buds.

Planting of stem cuttings was carried out in 15-cell black plastic trays, with dimensions of 34 cm long  $\times$  21 cm wide  $\times$  7.8 cm high. Each cell measured 6.2 cm in the top  $\times$  5.0 cm in the bottom × 7.8 cm in height, with five 6-mm basal openings for excess water draining. The trays were filled with commercial substrate (Mec Plant<sup>®</sup>) (Table 1).

Characteristic	Unit	Value	
рН	-	5.36	-
Electric conductivity	μS cm⁻¹	459.00	
Salt index	-	0.06	
Density	g cm⁻³	0.59	
Organic carbon	%	13.25	
Total carbon	%	17.62	
Moisture lost at 65 °C	%	55.85	
Moisture lost at 110 °C	%	6.29	
Total fixed solids at 110 °C	%	93.71	
Total fixed solids at 550 °C	%	21.88	

Table 1. Physicochemical characteristics of Mec Plant <sup>®</sup> substra
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Total volatile solids	%	71.83
Water retention capacity	%	15.75
Cation exchange capacity	mmolc kg <sup>-1</sup>	76.00
Phosphorus	mg kg⁻¹	166.61
Potassium	mg kg⁻¹	123.01
Calcium	mg kg⁻¹	240.55
Magnesium	mg kg⁻¹	211.50
Sulfur	mg kg⁻¹	77.75
Boron	mg kg⁻¹	0.06
Copper	mg kg⁻¹	0.01
Iron	mg kg⁻¹	0.09
Manganese	mg kg⁻¹	1.97
Molybdenum	mg kg⁻¹	0.00
Sodium	mg kg⁻¹	25.05
Zinc	mg kg⁻¹	0.03
Cadmium	mg kg⁻¹	0.00
Lead	mg kg⁻¹	0.00
Chromium	mg kg⁻¹	0.00
Nickel	mg kg⁻¹	0.00
Cobalt	mg kg <sup>-1</sup>	0.00

One stem cutting was planted horizontally per tray cell to a depth of 2 cm. Each experimental unit was composed of a planting tray, totaling 15 stem cuttings per experimental unit. The trays were placed on benches located in a Van Der Hoeven agricultural greenhouse, with a sprinkler irrigation system. Irrigation was carried out daily at the end of the afternoon for substrate moisture maintenance and stem cutting rooting. Irrigation was applied to a water of between 4 and 8 mm per irrigation shift.

Daily readings of minimum and maximum air and substrate temperatures and relative air humidity were carried out during experimental period using two digital thermometers and a digital thermo-hygrometer randomly distributed within the experimental area. Mean daily air and substrate temperatures (Tmean) were obtained from the mean of the three devices. Daily thermal summation (TSd, °C day) was calculated by three methods (Equations 1, 2 and 3) (Gilmore Jr & Rogers, 1958, Arnold, 1960):

Method 1: TSd = (Tmean – Tb) × 1 day if Tmean < Tb then Tmean = Tb

Method 2: TSd = (Tmean – Tb)  $\times$  1 day if Tmean < Tb then Tmean = Tb and if Tmean > Topt then Tmean = Topt

Method 3: TSd = (Tmean – Tb) × 1 day when Tb < Tmean  $\leq$  Topt and TSd = (Topt – Tb) × (Tmax × Tmean)/(Tmax – Topt) when Topt < Tmean  $\leq$  Tmax;

Where: Tb is the base temperature, Topt is the optimum temperature, and Tmax is the maximum temperature for the cassava crop development, and Tmean is the mean daily air temperature. The base temperature was 14 °C (Schons et al., 2007), while optimum and maximum temperatures were 30 and 42 °C, respectively (Matthews & Hunt, 1994). Accumulated thermal summation (TSa, °C day) from stem cutting planting to emergence (SCP–EM) was calculated by summing TSd values.

Length (SL), diameter (SD), and weight (SW) were measured at each stem cutting before planting. The mean number of days for emergence (DFE), percentage of survival (S%), and sprouting index (SI%), expressed as SI (%) = (Ns/Nb) × 100, where (Ns) is the number of sprouts and (Nb) is the number of buds, were calculated at each plot. The linear relationships were verified using Pearson's correlation coefficient. All plants of each experimental unit were assessed for the assessed traits.

Assumptions of mathematical model were verified before analysis of variance by homogeneity of treatment variances and normality of errors by Bartlett's and Shapiro-Wilk's tests (p<0.05), respectively. If the assumptions were not met, a Box-Cox procedure was used to verify an appropriate transformation for data using Action software (Equipe Estatcamp, 2014). Results underwent analysis of variance, and means were compared by the Scott-Knott's test (p<0.05) using SISVAR 5.6 statistical software (Ferreira, 2011).

### **3 RESULTS AND DISCUSSION**

A significant interaction was observed between planting seasons and stem cutting diameter ranges for stem cutting length, diameter, and weight, days for emergence, and percentage of survival. A breakdown of the two-way interaction, i.e., planting seasons within stem cutting diameter and vice versa, was carried out for these traits. Sprouting index showed no significant interaction between factors, and an analysis of the main effect of each factor was performed.

Stem cuttings with a diameter higher than 20 mm had a higher length and weight at all planting seasons (Table 2). Despite having a higher weight at all planting seasons, stem cuttings with a diameter higher than 20 mm did not differ statistically from those with a diameter lower than 20 mm for days for emergence in July, August, September, and October. Thus, the trait days for emergence are possibly influenced mainly by air temperature despite the higher weight and concentration of water, carbohydrates, and minerals for sprout induction and emission (Figure 1). Planting carried out under conditions of lower temperatures leads to an increase in the number of days for cassava stem cutting sprouting (Alves, 2006).

Table 2: Stem cutting length (SL), diameter (SD), and weight (SW), days for emergence (DFE), percentage of survival(S%), and sprouting index (SI%) of cassava stem cuttings with a diameter lower (< 20 mm) and higher (> 20 mm)than 20 mm planted in June, July, August, September, and October.

			Tra	its		
	SL (cm)		SD (mm)		SW (g)	
Season	< 20 mm	> 20 mm	< 20 mm	> 20 mm	< 20 mm	> 20 mm
June	4.03* cB	4.22 bA	17.96 bB	22.56 aA	11.03 aB	18.91 aA
July	4.32 bA	4.26 bA	17.84 bB	22.01 bA	10.64 aB	16.89 bA
August	5.54 aA	4.42 aA	18.39 aB	22.23 aA	10.45 aB	15.77 cA
September	4.29 bB	4.54 aA	17.93 bB	22.38 aA	8.66 bB	15.34 cA
October	4.41 aA	4.43 bA	18.31 aB	21.85 bA	9.06 bB	13.25 dA
CV (%)	6.2	6.20 2.00		00	8.	24
				Traits		

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	DFE		S (%)		SI (%)		
Season	< 20 mm	> 20 mm	< 20 mm	> 20 mm	Época	Dian	neter
June	17.28 bA	11.95 cB	83.74 aA	61.15 cB	15.66 e	< 20 mm	> 20 mm
July	19.98 aA	20.97 aA	87.51 aA	89.01 aA	43.08 d	46.60 A	41.37 B
August	14.36 cA	13.84 bA	70.93 bA	80.25 bA	53.33 b		
September	8.41 eA	8.55 dA	63.63 bB	92.92 aA	60.00 a		
October	10.82 dA	11.14 cA	72.03 bA	78.95 bA	48.83 c		
CV (%)	20.	59	24	.94		25.54	

\*Means followed by different lowercase letters in the column and uppercase letters in the row on each trait differ from each other by the Scott-Knott test at 0.05 probability.



Figure 1: Mean daily air temperature and relative humidity (RH%) during the emergence of sprouts in cassava stem cuttings planted in July, August, September, and October.

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A decrease in stem cutting weight was observed in both assessed diameter ranges as planting seasons advanced, which did not negatively influence the traits related to emergence (DFE) and sprout survival (S%). Planting can be carried out shortly after collection of cassava stems in places where there are no frosts. In the state of Rio Grande do Sul, stems should be stored to remain viable for planting under conditions where the risk of frost is minimal or zero. Nevertheless, according to Santos et al. (2009), the quality of cassava propagation material is drastically reduced after 90 days of storage. IITA (2014) recommends that storage should not exceed 56 days to prevent dehydration of tissues and loss of vigor of cassava stems.

Storage of stems for this study was performed in the shade of trees, with early morning solar incidence, as recommended by IITA (2014). Stems started sprouting when the mean air temperature increased during storage, which was initially observed in September and more intensely in October. This fact may be responsible for reducing the content of water and reserve substances used by sprouts and caused a reduction in stem cutting weight as storage time increased.

Planting carried out in September presented lower values of days for the emergence and higher sprouting rate, which was 60%. The mean temperature was 27.46 °C during the period of sprout emergence, the highest between the assessed planting seasons. In addition, this planting season showed the lowest maximum values of days for emergence among the seasons, which were 22 and 26 days for stem cuttings smaller and larger than 20 mm in diameter, respectively. This result confirms that September, in the absence of late frost forecasts, is the starting point of the appropriate period for cassava planting in the field by the traditional planting method. According to the Ordinance 130/2011 of the Ministry of Agriculture, Livestock and Supply of May 2, 2011, the climate risk zoning was approved for cassava in the state of Rio Grande do Sul, being established that planting in the municipality of Cruz Alta should occur between September 11 and November 30 (MAPA, 2011). September was adequate for stem cutting planting because it is carried out predominantly under a protected environment in the rapid multiplication method.

In general, at planting seasons, stem cuttings with a diameter lower than 20 mm had higher sprouting index (46.60%) in relation to those with diameter higher than 20 mm (41.37%). It is an indication that stem cuttings with a higher diameter possibly present a higher degree of lignification, which reduced the sprouting index. The pith is located in the central portion of stem cuttings, which is a region rich in water. Pith is surrounded by the cortex, a region with the highest reserve concentration. Thus, stem cuttings present adequate physiological conditions for planting when it has less than half of the internal area occupied by the pith (Bezerra, 2012). Farmers can use the pith to cortex ratio as a characteristic of selecting stem cuttings for planting. Nevertheless, Remison et al. (2015) verified that bud emission capacity and vegetative growth were related to the number of buds per stem cutting.

The percentage of survival in the assessed diameter limits showed no significant difference in July, August, September, and October, except for stem cuttings smaller than 20 mm planted in September, in which the percentage of survival was 63.63%, while stem cuttings larger than 20 mm had a 92.92% survival. Although sprout emergence is supplied by stem cutting reserves,

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composed mainly of carbohydrates, part of them showed no root emission to supply the requirements of sprout development by the absorption of water and nutrients. Storage of stems leads to water loss from the propagation material, which causes a reduction in rooting and sprouting (Bezerra, 2012). Initially, adventitious roots should emerge and then sprouts, favoring the shoot development (Alves, 2006; IITA, 2014).

The percentage of survival for stem cuttings smaller than 20 mm presented values of 83.74 and 87.51% in plantings carried out in June and July, respectively. On the other hand, a low sprouting index (15.66%) stood out in June. Therefore, among the 600 stem cuttings planted in June, which corresponded to 1200 buds, only 182 emitted sprouts and 152 survived. In general, under the climate conditions of Rio Grande do Sul, June is not appropriate for cassava planting, which would explain the slow development of surviving sprouts, which made the production of cassava seedling unfeasible at this planting season. Although the planting season carried out in July presented a sprouting index of 43.08%, the percentage of survival was high in both assessed diameter limits, which accredits this season as an alternative for obtaining stem cuttings and producing seedlings suitable for transplanting in early spring. According to Zuffellato-Ribas & Rodrigues (2001), the success of vegetative propagation depends on the levels of natural growth-promoting and inhibiting substances present in plants.

According to Figure 1, the mean air temperatures from stem cutting planting to sprout emergence (SCP–EM) in July, August, September, and October were 21.18, 23.27, 27.46, and 25.37 °C, respectively. The minimum temperatures were 17.43, 13.92, 20.85, and 21.70, while maximum temperatures were 24.05, 27.38, 29.65, and 29.00 °C. This stage showed a variation in the accumulated thermal summation between planting seasons, but its duration did not differ between the methods for estimating the accumulated thermal summation. It occurred because during the stage from stem cutting planting to sprout emergence, the mean daily air temperature did not exceed the optimum temperature for cassava, which was 30 °C.

When the planting was carried out in July, the longest period in days was required to complete the stage from stem cutting planting to emergence (20 days), and the highest value was observed for the accumulated thermal summation (143.53 °C day). On the other hand, the planting season of September had the shortest duration in days (8 days) for this stage and the lowest accumulated thermal summation (107.65 °C day). Planting seasons of August and October were similar, as planting required 14 and 11 days to complete the stage from stem cutting planting to emergence, respectively. A difference of 4.65 °C was found between plantings carried out in August and October for the accumulated thermal summation (Table 3). The cultivar Maus, 10 planted at a depth of 10 cm in incubators and considering as base temperature 13 °C, required about 210 degrees day for the emergence of 50% of sprouts (Keating & Evenson, 1979).

Table 3: Duration of the stage from stem cutting planting to sprout emergence (SCP - EM) in days and cumulative thermal summation (TSa); substrate temperature of SCP - EM (Ts, °C) at the planting seasons of July, August, September, and October; and Pearson's linear correlation coefficients between SCP - EM, substrate temperature for emergence (Ts, °C), and accumulated thermal summation for emergence (TSa, °C day).

Planting season	SCP - EM (days)	SCP - EM (Tsa)	Substrate temp. (ºC) SCP - EM
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July	20	143.53	18.89	
August	14	129.73	20.75	
September	8	107.65	24.75	
October	11	125.08	22.52	
	SCP - EM	Substrate temp. ( <sup>o</sup> C)	Tsa (⁰C day)	
SCP - EM		-0.97*	0.96*	
Substrate temp. (ºC)			-0.99*	
Tsa (ºC dav)				

\*Significant at 0.05 probability by the t-test.

n = 4.

The lowest mean substrate temperature (18.89 °C) was verified in July when the highest accumulation of accumulated thermal summation was required. On the other hand, the highest mean substrate temperature (24.75 °C) was verified in September, when the lowest accumulation of accumulated thermal summation was required for sprout emergence, i.e., a relationship was observed between the accumulated thermal summation necessary for sprout emergence and the mean substrate temperature. Therefore, a higher accumulated thermal summation was required when the mean substrate temperature was lower. In this case, substrate temperature had a direct effect on sprout emergence of the cassava crop. According to Keating & Evenson (1979), soil temperatures below 17 °C decrease the sprout emission rate, and temperatures close to 30 °C accelerate it.

The relationship between the accumulated thermal summation and mean substrate temperature was confirmed by Pearson's linear correlation coefficients (Table 3). A significant negative linear correlation was observed between the stage from stem cutting planting to emergence and substrate temperature (Ts) (-0.97), i.e., an increase in substrate temperature led to a decrease in the stage. A positive linear correlation was found between the stage from stem cutting planting to emergence and accumulated thermal summation (TSa) (0.96), indicating a higher accumulation of thermal summation when the stage was longer. A negative correlation was observed between the accumulated thermal summation and substrate temperature (-0.99), confirming the inversely proportional relationship between the accumulated thermal summation and substrate temperature for the emergence of cassava sprouts.

## **4** CONCLUSION

Planting of stem cuttings with diameter between 20 and 25 mm in September was adequate for obtaining cassava cuttings for the rapid multiplication method.

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