Vegetative Propagation of *Bombax costatum* Pellegr. & Vuillet (Malvaceae) by Root Segments Cuttings: Effects of Mother Tree Diameter and Origin of Cuttings

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Abstract: *B. costatum* is a local species of great socio-economique importance whose flowers are consumed and commercialized in local and regional market. Despite its economical and social value in local communities, it is still in the wild stand in the Guinean Savannah Highlands of Cameroon. The objective of the study is to contribute to its domestication by root segments cuttings. Root system of the mother trees was carefully excavated and the root samples (0.5–1kg) for the carbohydrate analysis were collected monthly throughout the year. In addition, a total of 180 root segments cuttings (RSC) were collected from 20 parent trees of different diameters at breast height (dbh) (15 - 30 cm; 30.1 - 45 cm and > 45 cm) in «Gamba» and «Karna manga». The experimental design explored was a split - plot with 03 replicates. The trial was conducted in a non mist polypropagator. The results reveal that among the carbohydrates evaluated, fructose decreased from the dry to the rainy seasons while soluble and total sugars move in inverse order. The first buds appear 08 weeks after planting. At the end of the essay, the highest budding rate (81.11±19.65%) was obtained in root segments cuttings from Karna manga against 68.33±34.88% in RSC from mother trees of dbh 30.1-45 cm. Regarding the origin of root segments cuttings by dbh of parent trees interaction, the budding rate varied significantly from 36.66±5.77% in RSC from mother trees of dbh 15.1-30 cm originated from Gamba to 100±0% in those from parent trees of the same dbh in Karna manga (0.0046 < 0.01). The average height of leafy shoots varied from 8.28±5.23 cm in RSC from Gamba to 11.91±7.35 cm in those from Karna manga. The rooting rate fluctuated from 3.33±2.53% in root segments cuttings from mother trees of dbh 30.1 - 45 cm and dbh > 45 cm originated respectively from Gamba and Karna manga to 50±26.45% in those from parent trees of dbh 15 - 30 cm from Karna manga. These results will help to elaborate appropriate domestication and conservation strategies of the species.

Keywords: *B. costatum*, Carbohydrates, Domestication, Vegetative Propagation, RSC, Root Ability

1. Introduction

Bombax costatum, locally known as ‘‘Djoye’’ in peulh, is among the most socio-economic important tree species of the Guinean Savannah Highlands (GSH) of Adamawa in

Cameroon [1, 2]. Plants that bear edible Non Timber Forest Products are subjected to abuse by the operators. They offer flowers and fruits that are regularly harvested and consumed or sold in local and regional markets [3]. Different parts of the species are utilized in folk medicine, handicraft and

feeding [4, 5]. The wood is valued in the manufacture of kitchen tools. The bark of the tree is used in the treatment of wounds and diarrhea [6] as well as facilitate childbirth [7, 8]. Leaves nurse fever while flowers are used as condiment [3]. Dried immature fruits are used as spices [9]. The great utility of the species can become harmful, although, it can be an asset in favor of its promotion and the acquaintances of the establishment modality of germplasm development. In the GSH, the plant is overexploited and most trees are unable to bear fruits. Lack of seeds limits the possibility to propagate the tree [2, 10]. In the near future, if nothing is done, it could disappear and it would be a great loss for the population of this zone. Therefore its conservation is urgent. Absence of informations related to its conservation is an important handicap for the population. It was reported in Burkina Faso that *B. costatum* can be propagated vegetatively [11, 12]. In the agro-ecological conditions of the GSH of Cameroon, numerous works have been carried out on the propagation by root segments cuttings (RSC) of many species [13-17]. However, none of these works was focused in *B. costatum*. The main purpose of the present study was to fill the gaps and contribute to the domestication of the species. More specifically, to evaluate seasonal fluctuation of carbohydrates and the relationship between carbohydrate contents and ability of root segments cuttings to regenerate; to assess the influence of the origin of parent trees as well as its diameter at breast height on the ability of RSC to form adventitious buds and roots. Our hypothesis was that sprouting and rooting efficiency of root cuttings taken from parent trees are directly related to carbohydrate concentration. In addition to a test of this putative relationship, we wanted to profile the fluctuation of carbohydrates during the seasonal cycle of one year.

2. Materials and Methods

2.1. Study Site

Root segments cuttings (RSC) used in the study were sampled in 20 genotypes from «Gamba» and «Karna manga» located in Guinean Savannah Highlands of Adamawa region. The climate is guinean type with a dry season (November to March) and rainy season (April to October) [18]. For the period of 2008-2017, the average annual rainfall and temperature were respectively 1467.13 mm and 23. 28°C. The area is covered with shrub and / or woody savannah dominated by *Daniellia oliveri* and *Lophira lanceolata* [19]. The evolution of the vegetation is severely hampered by anthropic activities [20]. The soil of the region is constituted in majority of red ferralitic structure developed on old basalt [21]. This region is constituted of multicultural ethnic groups among which the most dominant are Foulani, Mboum, Pere, Niza'a and Dourou [18].

2.2. Sampling of Roots and Carbohydrates Analyses

Due to the importance of carbohydrates as a source of energy and building blocks of macromolecules during the

root development, the first step of this study focus on change of carbohydrate concentrations in mother trees during the year. They are therefore important for root formation. It is often considered exclusively that the availability of carbohydrates as an energetic requirement to drive rooting and together with phytohormones [22]. Root system of mother trees was carefully excavated and root samples (0.5–1kg) of 2cm diameter were collected monthly throughout the year (January-December) for carbohydrate analysis. The samples were dried in ambient temperature (23.28°C), then ground in a mill and stored in airtight plastic bags. Analysis of carbohydrates (soluble sugar, total sugar and fructose) were carried out through Acid 2-hydroxy - 3,5 - dinitrobenzoique method in standard procedures [23].

2.3. Preparation and Cultivation of Root Segments Cuttings

The experiment was conducted in a non mist polypropagator installed at the nursery of the Laboratory of Biodiversity and Sustainable Development of the University of Ngaoundere (Altitude: 1079 m; Latitude North: 7°24'; Longitude East: 13°32'). A polypropagator is raised under a shed with roof in corrugated iron where 06 white transparent sheets were inserted. It is subdivided in three compartments thus corresponding to three replications. This low-cost installations provide conditions conducive to rooting cuttings. The system is simple, inexpensive and easy to operate. The polypropagator is a wooden frame enclosed in a single sheet of polythene such that the base is completely water tight. The frame was covered tightly with single piece of polythene and a closely-fitting lid. It was 3 m long versus 1 m wide and 1 m height at the back and 0.5 m in the main face. The base of polypropagator was covered with a thin layer of sand to protect the polythene and large stones were placed on top of the sand to a depth of 10-15 cm. This was then covered by successive layers of small stones and gravel to a total depth of 20 cm. The saturated layers of stones and gravel were covered by a rooting medium composed with 50/50% homogeneous mixtures of sand/sawdust which was shown performant in the previous studies [16, 17]. The rooting media remained moist by capillarity and could be dampened from the above as necessary. This resulted in a permanently humid environment throughout the propagation period [24]. The relative humidity prevailing inside the polypropagator varied from 80-100% while the temperature varied from 28-30°C.

Preferred characteristics of local populations were considered in the selection of the 20 genotypes used: good sanitary conditions, regular production and low branching. A total of 180 root segments cuttings (RSC) were taken in «Gamba» and «Karna manga» from the same trees as the root samples for the carbohydrate analyses. In each of these sites, the partial excavation was undertaken in root system of mother trees of different diameters at breast height (dbh). The RSC of about 2 cm diameter, 20 cm long [16] were wrapped in a moistened newsprint paper and transported in a cooler to the nursery. Root segments cuttings of the diameter

similar to those typically used in carbohydrate analyses, arrived at the nursery on the same day. They were inserted vertically in rooting medium (homogenous mixture of sand/sawdust 50/50%). A 1 cm notch was incised at the distal end of each root segments cuttings [25]. The essay was watered twice a day (morning and evening) using a hand sprayer. An open cylinder made of PVC pipe was inserted vertically into the medium and stones. This pipe was used as the filling point for the water and allowed a regular check of the water table. A root segments cuttings with at least one leafy shoot is considered budded. A RSC was considered as rooted if the length of the root was greater than 1 cm [26]. Rooted RSC were inserted vertically in large perforated polyethylene bags (27 cm diameter $\times$ 40 cm height) containing the homogenous mixture of arable soil / sawdust which was found performant. These bags were introduced in acclimatization propagators and watered mornings and evenings. During the acclimatization phase, the propagators were left open each night during a month. Watering was then reduced to once a day. After this phase, the plants were transferred to the open field.

2.4. Experimental Design and Data Analysis

Globally, the experimental design exploited for the two trials was a Split-plot each.

For the carbohydrate analysis, the main treatment was season (beginning of the dry season, end of the dry season, beginning of the rainy season and end of the rainy season) and the sub-treatment was represented by sugars (fructose, soluble sugar and total sugar). The number of replications was two.

3.1. Seasonal Fluctuation of Root Carbohydrates in Mother Trees

Figure 1. The seasonal fluctuation of soluble sugar, total sugar and fructose in roots of parent trees in savannah conditions.

Root concentrations of carbohydrates clearly fluctuated under field conditions throughout the year, being highest at

Regarding the vegetative propagation by root segments cuttings, the origin (Gamba, Karna manga) of RSC was the main treatment while the mother trees diameter (15 - 30 cm; 30, 1 - 45 cm and > 45 cm) corresponded to sub-treatment. The number of replications was three while the experimental unit was made up of 10 cuttings. A total of 180 RSC (2x3x3x10) were prepared for the experiment. The following parameters were determined: the rate of budding, the number and the height of leafy shoot and the number of leaves per leafy shoot, the rooting rate, the number and the length of roots. Budding corresponds to bud burst of latent buds that emit one or more leafy shoots on the RSC; the rate of rooting (equated to the success rate of root segments cuttings), the pole on which the leafy shoots were developed (distal or proximal pole) was determined. The distal pole contrarily to proximal corresponds to the extremity of the root segments cuttings which was originally located farthest from the base of the parent tree. For this part, data on rooting were collected at the end of the experiment (24 weeks).

Quantitative data were subjected to an analysis of variance and Post-hoc comparisons were done with the Duncan's Multiple Range Test when significant mean effects were noted. All the analysis were performed using the Statgraphics 5.0 software.

3. Results

In vegetative propagation by root segments cuttings (RSC), adventitious bud and root formation are prerequisite of successful propagation.

the end of rainy season and lowest at the end of dry season except for fructose. The analysis of variance indicated that

the time of sampling condition of the mother trees account significantly for over 99.99% of total variation in fructose, soluble and total sugar concentrations among root samples ($0.0000 < 0.001$). Regarding the seasons, there was also a significant difference ($0.0000 < 0.001$) as well as the carbohydrate by season interaction ($0.0000 < 0.001$). Thus, the seasonal variation of fructose content was different, high in the beginning of dry season (4.71g/100g.MS) and lowest (1.18g/100g.MS) in the beginning of the rainy season. The soluble sugar increased from the beginning of the dry season (5.58g/100g.MS) to the end of rainy season (11.18 g/100g.MS). In the beginning of the dry season (from 18.32 g/100g.MS), the value of total sugar decreased to 12.24 g/100g.MS in the end of dry season then increased up to 27.96 g /100g.MS at the end of rainy season (Figure 1). Analogous variations were reported in hybrid aspen clones [27]. The authors argued that carbohydrate reserves varied during the annual cycle of plants, with root carbohydrate concentrations being highest during the dormant period and reducing in the spring when they are transported to the shoot for bud burst and vegetative growth.

Among the three carbohydrates evaluated, fructose decreased from the dry to the rainy seasons while soluble and total sugars move in inverse order. Many experiments have indicated that the source of carbohydrates is critical for rooting. Therefore, they conclude that a large amount of carbohydrates is essential to initiate and accompany the rooting process [28, 29].

During this process, carbohydrates act as a source of energy and as constitutive elements for the newly formed cells. Based on the trend of fructose, RSC for the present trial were collected on the mother trees during the end of the dry season. For the coming trials, RSC will be collected during the rainy season.

3.2. Effect of Origin of Root Segments Cuttings and Dbh of Mother Trees on Budding

3.2.1. Rate of Budding

The root segments cuttings were put in culture at the beginning of the dry season and the trial lasted 24 weeks. The first buds were observed after 08 weeks in cuttings originated from Gamba in parent trees of dbh 30.1- 45 cm. All RSC have developed leafy shoot without formation of adventitious roots (Figure 2).

Figure 2. Root segments cuttings without adventitious roots.

Concerning the origin of root segments cuttings, at the end of the essay, the rate of budding varied from $44.44 \pm 14.24\%$ in root segments cuttings from Gamba to $81.11 \pm 19.65\%$ in those from Karna manga (Figure 3). A significant difference was registered among the origins of RSC ($0.0000 < 0.001$).

Figure 3. Rate of budding of *B. costatum* according the origin of root segments cuttings.

For the diameter at breast height (dbh) of the parent trees, the rate of budding fluctuated from $51.66 \pm 11.69\%$ in root segments cuttings collected from trees of dbh > 45 cm to $68.33 \pm 34.88\%$ and $68.33 \pm 24.01\%$ in those belonging to dbh

15.1 - 30 cm and 30.1 - 45 cm respectively (Figure 4). A significant difference was registered among the dbh sizes of the parent trees ($0.0339 < 0.05$).

Figure 4. Rate of budding of *Bombax costatum* according to the dbh of parent trees.

The origin of RSC by dbh of mother trees interaction has significantly influenced the budding rate ($0.0046 < 0.01$). The rate of budding varied from $36.66 \pm 5.77\%$ in RSC

originated from Gamba gathered on mother trees of dbh equal to 15.1-30 cm to $100 \pm 0\%$ in those from Karna manga from mother trees of dbh 15.1 - 30 cm (Figure 5).

Figure 5. Budding rate of RSC according the diameter at breast height of parent trees.

3.2.2. Number of Leafy Shoots Per Root Segment Cutting

At the end of the experiment, the average number of leafy shoots varied from 0.84 ± 0.28 in RSC originated from Gamba

to 1.3 ± 0.63 in those from Karna manga (Table 1). The origin of root segments cuttings has significantly influenced the number of leafy shoots ($0.0139 < 0.05$).

Table 1. Effect of the origin of root segments cuttings on growth characteristics of leafy shoots after 24 weeks.

Origin of RSC	Number of leafy shoots	Height of the leafy shoots (cm)	Number of leaves per leafy shoot
Gamba	$0.84 \pm 0.28b$	$8.28 \pm 5.23a$	$2.01 \pm 1.05a$
Karna manga	$1.3 \pm 0.63a$	$11.91 \pm 7.35a$	$2.83 \pm 1.83a$
P. value	0.0139	0.1069	0.0928

Means followed by the same letter are not statistically different at 5%

The average number of leafy shoots varied from 0.08 ± 0.28 in RSC from parent trees of dbh ≥ 45 cm to 1.28 ± 0.59 in those collected from parent trees of dbh equal to 15 - 30 cm

(Table 2). Contrarily to the effect of origin, there was no significant difference between dbh of mother trees ($0.0742 > 0.05$).

Table 2. Effect of dbh of mother trees on growth characteristics of leafy shoots after 24 weeks.

Dbh (cm)	Number of leafy shoots	Height of the leafy shoots (cm)	Number of leaves per leafy shoot
15- 30	1.28±0.59	12.65±8.45	3.18±2.04
30 - 45	1.13±0.61	8.66±4.67	2.08±1.07
> 45	0.8±0.28	8.97±6.08	2±1.16
P. value	0.0742	0.2597	0.0955

Dbh= Diameter at breast height

Regarding the origin by dbh of mother trees interaction, the average number of leafy shoots fluctuated from 0.6±0.1 in root segments cuttings harvested on mother trees of dbh >45 cm from Karna manga to 1.8±0.2 in RCS from

parent trees of dbh 15-30 cm originated from the same locality (Table 3). There was a significant difference for the origin by dbh interaction (0.0074 < 0.01).

Table 3. Number of leafy shoots for origin of root segments cuttings by dbh of mother trees interaction.

Dbh (cm) / Origin	15 - 30	30.1 - 45	≥ 45	Mean
Gamba	0.76±0.21a	0.76±0.38a	1±0.26ab	0.84±0.28
Karna manga	1.8±0.2b	1.5±0.61b	0.6±0.1a	1.3±0.31
Mean	1.28±0.21	1.13±0.49	0.8±0.18	1.07±0.29
P.value		0.0074		

Means followed by the same letter are not statistically different at 5%

3.2.3. Height of Leafy Shoots

The origin of root segments cuttings has not significantly influenced the height of leafy shoots (0.1069 > 0.05). The average height of leafy shoots varied from 8.28±5.23 cm in root segments cuttings originated from Gamba to 11.91±7.35 cm in those from Karna manga (Table 1).

The average height of leafy shoots ranged from 8.66±4.67 cm in RSC collected from mother trees of dbh 30.1 - 45 cm to 12.65±8.45 cm in those gathered on mother trees of dbh

15 - 30 cm (Table 2). Nevertheless, the dbh of mother trees has no significant difference among the height of leafy shoots (0.1069 > 0.05).

The average height of leafy shoots varied from 4.87±1.72 cm in RSC from parent trees of dbh ≥45 cm originated from Karna manga to 19.3±6.04 cm in those from parent trees of dbh 15 - 30 cm of the same locality (Table 4). There was a significant difference for the interaction (0.0038 < 0.01).

Table 4. Height of leafy shoots according to the origin of root segments cuttings by dbh of mother trees interaction.

Dbh (cm) / Origin	15 - 30	30 - 45	> 45	Mean
Gamba	6±3.04a	5.76±3.06a	13.08±6.23ab	8.28±4.11
Karna manga	19.3±6.04b	11.56±4.47ab	4.86±1.72a	11.90±4.07
Mean	12.65±4.54	8.66±3.76	8.97±3.97	10.09±4.09
P.value		0.0038		

Means followed by the same letter are not statistically different at 5%

3.2.4. Number of Leaves Per Leafy Shoot

The average number of leaves per leafy shoot oscillated between 2.01±1.05 in RSC from Gamba and 2.83±1.83 in those from Karna manga (Table 1). However, there was no significant difference among the origin of RSC (0.0928 > 0.05).

The average number of leaves per leafy shoot oscillated between 2±1.16 in RSC collected from mother trees of dbh ≥45 cm and 3.18±2.04 in those collected from parent trees of

dbh 15 - 30 cm (Table 2). However, there was no significant difference between dbh of mother trees (0.0955 > 0.05).

Concerning the origin by dbh of mother trees interaction, the mean number of leaves per leafy shoot fluctuated from 1.13±0.49 in RSC from parent trees of dbh ≥45 cm collected in Karna manga to 4.86±1.17 in those from parent trees of dbh 15 - 30 cm of the same locality (Table 5). Significant difference existed for the interaction origin by dbh of mother trees (0.0021 < 0.01).

Table 5. Number of leaves per leafy shoot according the origin of cuttings by dbh of mother trees interaction.

Dbh (cm) / Origin	15 - 30	30 - 45	> 45	Mean
Gamba	1.5±0.75a	1.66±1.13a	2.86±0.95ab	2.01±0.94
Karna manga	4.86±1.17b	2.5±1.05ab	1.13±0.49a	2.83±0.91
Mean	3.18±0.96	2.08±1.09	1.99±0.72	2.42±0.92
P.value		0.0021		

Means followed by the same letter are not statistically different at 5%

3.3. Effect of Origin of Root Segments Cuttings and DBH of Parent Trees on Root Formation

3.3.1. Rate of Rooting

The rooting rate ranged from $10 \pm 10\%$ in root segments cuttings from Gamba to $27.77 \pm 24.88\%$ for those originated from Karna manga. There was significant difference between origin of cuttings ($0.0128 < 0.05$). The RSC from Karna manga produced more vigorous adventitious roots than those from Gamba (Figure 6).

a

b

Figure 6. RSC rooted: from Karna manga (a); from Gamba (b).

For the diameter at breast height of the parent trees, the rooting rate varied from $11.66 \pm 9.57\%$ in root segments cuttings from trees of dbh ≥ 45 cm to $28.33 \pm 26.45\%$ in those from trees of dbh 15 - 30 cm (Table 6). Nevertheless the dbh of mother trees has not significantly influenced the rate of rooting ($0.1127 > 0.05$).

Table 6. Effect of dbh of mother trees on the rate of rooting.

Dbh (cm)	15 - 30	30.1 - 45	> 45	Mean	P.value
Rooting (%)	28.33 ± 26.45	16.66 ± 16.33	11.66 ± 9.57	18.88 ± 17.45	0.1127

Regarding, the origin of cuttings by dbh parent trees interaction, there was significant difference between the origins of RSC ($0.0048 < 0.01$). The rate of rooting fluctuated from $3.33 \pm 2.53\%$ in root segments cuttings from mother trees of dbh 30.1 - 45 cm and dbh > 45 cm originated

respectively from Gamba and Karna manga to $50 \pm 26.45\%$ in those from parent trees of dbh 15 - 30 cm from Karna manga (Figure 7). The combination between dbh of parent and their origin improved significantly the rate of rooting.

Figure 7. Root rate of RSC according the diameter at breast height of parent trees.

3.3.2. Number of Roots

The average number of roots per root segment cutting varied from 2.61 ± 2.06 in root segments cuttings from Gamba to 2.79 ± 1.65 in those from Karna manga. Nevertheless there was no significant difference between origins of RSC ($0.8128 > 0.05$). The number of roots per root segment cutting was very low.

At the end of the essay, the average number of roots per RSC varied from 2.46 ± 0.55 in RSC from mother trees of dbh 30.1 - 45 cm to 3.07 ± 0.44 in those harvested on parent trees of dbh equal to 15 - 30 cm (Table 7). However, the dbh of parent trees has not significantly impacted the number of roots ($0.7949 > 0.05$).

Table 7. Effect of origin of RSC on growth characteristics of roots.

Origin of RSC	Number of roots	Length of roots (cm)
Gamba	2.61 ± 2.06	2.48 ± 1.87
Karna manga	2.79 ± 1.65	3.41 ± 2.52
Mean	2.7 ± 1.85	2.95 ± 2.19
P.value	0.8128	0.3374

Regarding, the origin by dbh of parent trees interaction, the number of roots varied from 1 ± 0.12 in RSC of mother trees of dbh 30.1 - 45 cm from Gamba to 3.92 ± 0.38 in those of dbh 30.1 - 45 cm from Karna manga (Table 8). However there was significant difference for the interaction ($0.0449 > 0.05$).

Table 8. Number of roots according the interaction origin of RSC *dbh of mother trees.

Origin / Dbh (cm)	15-30	30.1-45	> 45	Mean
Gamba	$3 \pm 2.65b$	$1 \pm 0.12a$	$3.83 \pm 2.31b$	2.61 ± 1.69
Karna manga	$3.15 \pm 0.45b$	$3.92 \pm 0.38b$	$1.33 \pm 0.31a$	2.8 ± 0.38
Mean	3.07 ± 1.55	2.46 ± 0.25	2.58 ± 1.31	2.71 ± 1.04
P.value	0.0449			

3.3.3. Length of Roots

Concerning the length of roots, it oscillated between 2.48 ± 1.87 cm in root segments cuttings from Gamba to 3.41 ± 2.52 cm in those of Karna manga (Table 7). However, there was no significant difference between origins of RSC ($0.3374 > 0.05$).

The length of roots varied from 2.46 ± 1.92 cm in RSC of mother trees of dbh > 45 cm to 3.77 ± 2.74 cm in those from mother trees of dbh 15-30 cm (Table 9). However, the dbh of parent trees has not significantly influenced the length of roots ($0.4729 > 0.05$).

Table 9. Effect of dbh of mother trees on growth characteristics of roots.

Dbh (cm)	Number of roots	Length of roots (cm)
15-30	3.07 ± 0.44	3.77 ± 2.74
30.1-45	2.46 ± 0.55	2.59 ± 2.03
> 45	2.58 ± 0.63	2.46 ± 1.92
Mean	2.71 ± 0.54	2.94 ± 2.23
P.value	0.7949	0.4729

Concerning the origin by dbh of parent trees interaction, the length of roots ranged from 1.22 ± 1.12 cm in RSC of

parent trees of dbh 30.1 - 45 cm from Gamba to 5.02 ± 3.05 cm in those from parent trees of dbh 15 - 30 cm in Karna manga (Table 10). The dbh of parent trees by origin interaction has no significant difference among the length of roots ($0.0740 < 0.05$).

Table 10. Length of roots according the interaction origin of RSC*dbh of mother trees.

Origin / Dbh (cm)	15-30	30.1-45	> 45	Mean
Gamba	2.53 ± 2.21	1.22 ± 1.12	3.68 ± 0.31	2.47 ± 1.22
Karna manga	5.02 ± 3.05	3.96 ± 0.35	1.25 ± 1.16	3.41 ± 1.52
Mean	3.77 ± 2.63	2.59 ± 0.74	2.46 ± 0.74	2.94 ± 1.37
P.value	0.0740			

3.3.4. Polarity of Root Segments Cuttings

The leafy shoots were developed in proximal (86.72%) and median (13.28%) poles. The whole root was formed in distal position (100%).

4. Discussion

4.1. Carbohydrates

The root segments cuttings (RSC) were collected in the beginning of the dry season mainly in December considering the amount of fructose. Among the carbohydrates analysed, only fructose was at its peak in the aforementioned period while soluble and total sugars reached at theirs at the end of rainy season. Fructose is known to facilitate root formation in various species such as *Nauclea diderrichii*, *Stevia rebaudiana* and *Morus alba* [29-31]. Others observed that glucose give the best result in *Rosa rugosa* [32]. Rooting success is associated with low pre-severance starch and water-soluble sugar concentrations and greater total water-soluble carbohydrates [33]. Clearly, roots to be used for regeneration, should be taken at the time of maximum carbohydrate accumulation. The constant sugar levels maintained by all regenerating RSC despite depletion of reserves, indicate a mechanism for maintaining a minimum metabolic sugar level [34].

B. costatum is a deciduous tree which shed off leaves during the dry season. During leaf fall, carbohydrates moved into the roots of mother trees and rapidly converted to polysaccharides. The species analysed in this study displayed contrasting seasonal changes of plant carbohydrates. Indeed, during the rainy season, leaves accumulated soluble and total sugar while fructose greatly declined. Conversely, photosynthetic capacity declined from the beginning to the end of dry season. Thus, as reduced photosynthetic capacity induced leaf senescence, fructose content in leafy Bombax leaves increased. The most likely explanation is that carbohydrate was mobilized during leaf senescence for translocation to roots of the mother trees. However, the same carbohydrate can give very controversial effect according to the species. It is postulated that each step is controlled in different ways by balances of hormones, inhibitors and other biochemicals. Root formation in cuttings is a complex

phenomenon in which there are at least three distinct steps: callus formation; differentiation within callus resulting in root initiation; and root growth [35]. The response of root segments cuttings, collected during the dry season to the different treatments did not form callus but confirm that budding and rooting are correlated to the large amount of soluble carbohydrates which suddenly decreased at the end of the dormancy period. Similar to soluble sugar (11.18g.MS/100g) during the rainy season, the amount of total sugar increased till an optimum (27.96g.MS/100g). Contrarily to our expectations, fructose was the only carbohydrate that had strong correlation with the sprouting efficiency of root segments cuttings. The origin by dbh of parent trees interaction improved the sprouting of RSC up to 100%. Though, clearly, fructose levels do not entirely explain the root sprouting efficiency found in this study despite the fact that soluble sugar and total sugar reached their optimum during the rainy season. Lack of a high rooting response during the dry season may be attributed to the reduction in the endogenous level of auxin or nutritional status of the cuttings [36]. The rooting process is a high demanding carbohydrates process and in our experiment the carbohydrate content in cuttings showed an overall decrease during the rooting process. However, the interaction between origin and dbh of mother trees improved the rooting potential of RSC up to 50%.

4.2. Budding and Rooting

The first buds in RSC of *B. costatum* appeared 08 weeks after planting. This latent period has given conflicting results. It disagreed those obtained in Burkina Faso on the same species [11] as well as those in *Detarium microcarpum* in the aforementioned country [25]. Nevertheless, it corroborated those in *Vitex doniana* in Cameroon [14]. For these authors, the appearance time of the first buds in these species was observed at four, five and eight weeks after planting. In fact, the latency period varied depending on the species, the period of harvesting of the cuttings and even following the application of hormones or not [16, 25, 27]. The sprouting process promotes the start of adventitious rooting. When the RSC sprouted, the carbohydrates supply from the leaves supported the root growth [37].

The provenance significantly influenced the rate of budding, the number of aerial buds, and the rate of rooting. The experiment was conducted in the Guinean Savannah Highlands and this would explain the fact that the budding rate and the rooting rate were high for RSC from Karna manga. Root segments cuttings tend to be more budded and rooted in the agro-ecological conditions of their environment. Indeed, Karna manga is located in the agro-ecological area where the essay took place. Hence a favorable response of root segments cuttings from Karna Manga compared to those of Gamba was remarkable.

The dbh of the parent trees influenced budding. This result is shaped with that obtained in *Vitex doniana* in the same area [13]. These authors obtained like in the present study, a high budding rate from cutting taken from low dbh (5-10

cm). Indeed, when the plant is young, it produced abundantly sugar and hormones responsible of budding. Budding and rooting of cuttings are influenced by the interaction of internal factors of the plant and environmental conditions [37]. A higher level of total carbohydrates was observed in an easy-to-root cultivar of chrysanthemum and it produced a greater number of roots than did the difficult to root cultivar [38].

Despite this slight variation observed, the dbh of the mother trees did not influenced the number of aerial buds, their height and the number of leaves/shoot. Root segments cuttings from parent trees of low dbh showed a slightly superiority on growth parameters, compared to those from parent trees of large dbh. Thus, the mother trees that had the dbh 15-30 cm produced the highest number and length of leafing shoots as well as the roots. This result is in agreement with that reported in *Vitex doniana* [15]. The emission of adventitious roots is low compared to the emission of buds. In fact, the emitted buds elaborated the sugars and hormones responsible for the formation of adventitious roots through the process of photosynthesis [14, 16, 25]. The origin of RSC by mother tree dbh interaction was significant and the rate of rooting was 50% suggesting the ability to produce roots. This is a remarkable result. Efficient rooting is essential, because sprouted RSC will not survive long without roots.

The concentrations of carbohydrate among which total sugar and soluble sugar except fructose were not at their optimum to induce abundant roots in the dry season. Therefore, it can be concluded that the carbohydrates were essential to initiate and accompany the rooting process. Although the effects of origin, season of gathering RSC, and size of the parent tree probably hold in a general way for *B. costatum* in the Guinean Savannah Highlands. The rooting percentage obtained in this work is low compared to that reported in Burkina Faso which was 90% [11]. The season of collection and pedoclimatic conditions may be the explanation which could motivated this variation.

Concerning the position of the buds and roots on the RSC, its polarity was conserved despite the fact that a few buds appeared in median pole.

5. Conclusion

Despite the relatively small influence of carbohydrate noted during the rooting period, the present results with *B. costatum* support the theory that the physiological status of the stock plant at the time that root segments cuttings are excised is of utmost importance for the subsequent rooting process. In addition, this study demonstrated that *B. costatum* could be very easily propagated from RSC. In the present study, observations showed that, the budding and rooting of *B. costatum*'s plantlets are affected by the origin of RSC and the diameter at breast height of the parent trees as well as their endogenous carbohydrate contents.

Further studies are required: to determine, in particular, the effect of genotype, the period of optimal concentration of carbohydrates in mother trees before harvesting RSC, type of

optimal carbohydrate to the development of RSC, different carbohydrates produced of RSC during the propagation process and effect of mycorrhiza inoculums on sprouting and rooting.

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