American Journal of Agricultural and Biological Sciences

Original Research Paper

Vegetative Propagation by Root Segments Cuttings of *Sclerocarya Birrea* (A. Rich.) Hochst: Effects of Substrate and Mycorrhizal Inocula on the Ability to Root Cuttings

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Article history
Received: 07-11-2021
Revised: 03-06-2022
Accepted: 16-06-2022

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Abstract: *Sclerocarya birrea* (A. Rich.) Hochst is one of the woody plants of the soudano-Sahelian zones, with great socioeconomic importance. The local populations over-exploit this species for their daily socio-economic needs and this endangered its durability. The objective of this study was to contribute to the domestication of this species by the low-cost technique of Root Segments Cuttings (RSC). The substrates sand/sawdust and dark soil/Sawdust have been inoculated with 0, 10, and 20 g of mycorrhizae. The experimental design was a split plot with 4 repetitions. The experimental unit is of 10 cuttings. The results show that the best substrate for the budding of the RSC is the mixture of Sand/Sawdust (37.77±17.15%). The appropriate dose of mycorrhizae for the budding of the RSC is 10 g (33.33±21.6%). For the height of the aerial axes, a satisfactory result was obtained in the dose of mycorrhizae of 10 g (3.43±2.40 cm). Concerning the number of the leaves, the maximum is recorded in the RSC inoculated with 20 g of mycorrhizae (3.60±1.36). The analysis of variance did not show a meaningful effect for the treatments (0.54>0.05), as well as for the substrates (0.76>0.05). Regarding the rooting of the cuttings, one must notice that the best substrate for the number of newly formed roots is the mixture of Sand/Sawdust (2.22±1.01) while the dose of 20 g has much allowed the appearance of roots (3.41±1.24). The best substrate for the length of the newly formed roots is the mixture of Sand/Sawdust (2.9±1.3 cm), whiles the proportion of 10 g has permitted a lot of the growth of roots (4.24±1.69 cm) as far the plum tree of Africa is concerned. These results show that improving some parameters at *S. birrea* is possible by vegetative propagation.

Keywords: *Sclerocarya Birrea*, Domestication, Root Segments Cuttings, Inoculum Mycorrhiza, Guinean Savannah Highlands

Introduction

Tropical forest constitutes an immense reservoir of biological diversity; their ecological functions are essential for humanity (Matig *et al*., 2006). As elsewhere, the peoples of northern Cameroon have been exploiting and using plant species for centuries like raw materials for the manufacture of spatula, chairs, beds, sources of energy, etc. Despite a sparsely forested environment, frequently overmastered by bush fires and land clearing, these populations have made a great profit from the different potentialities of the local flora (Gormo and Nizesete, 2013). Plant species also provide farmers with non-timbers forest products to improve their standard of living in local and regional markets through the diversification of their source of income (Leakey *et al*., 2000). The ecosystems of the Guinean Savannah Highland of Cameroon are diverse and rich in species of socioeconomic interest (Mapongmetsem *et al*., 2008; 2010). These species thus play a significant role in the nutrition and the improvement of socio-economic conditions of rural populations, particularly in Africa (Larwanou *et al*., 2010; Mapongmetsem *et al.*, 2012a). However, over time, the population of many woody species has declined in the Guinean zone and the most important causes are natural

disasters and anthropic pressures, which are increasing (Ouedraogo *et al*., 2005). These lead to the deforestation of vast areas (Matig *et al.,* 2006). Thus, to maintain this biodiversity, domestication of these wild woody plants could be an alternative and contribute to food security (Tchiagam *et al.*, 2011).

Natural regeneration by the seed of some of these species is often difficult because of the non-availability of seeds and the difficulty of conserving their germination viability (Thiombiano *et al.,* 2010; Muok *et al.,* 2011). Vegetative propagation, which is faster and less costly (Bellefontaine and Monteuuis, 2002), appears to be an alternative (Mapongmetsem *et al.,* 2012a), an adaptive strategy for these species to environmental disturbances and climatic hazards (Ouedraogo *et al.,* 2005). The domestication of these species and the fight against the desertification of the environments they are attached to will be facilitated by controlling their regeneration methods (Bellefontaine and Monteuuis, 2002 Blanc, 2003).

Woody species of high socioeconomic interest, such as *Sclerocarya birrea* (A. Rich.) Hochst requires special attention (Mapongmetsem *et al.*, 2012a). It is one of the sixteen priority species in the Guinean high savannahs of Cameroon (Mapongmetsem *et al.,* 2012a). The fruits are consumed and marketed. When ripe, the pulp can be made into a fermented drink or not. The bark of the trunk and roots is used against diabetes (Mapongmetsem *et al*., 2015). It is rich in various nutrients, the most abundant being vitamin C. In South Africa, the pulp is used to manufacture a local drink named "Marula", which is enjoyed and traded in local, regional, and international markets (Leakey *et al*., 1996). The fruit kernel is consumed as a nut by many tribes in Cameroon (Tupuri, Guiziga, Mundang, etc.,) (Mapongmetsem *et al.,* 2012a). It is one of the utilitarian species recorded in the savannahs of Togo where it is used for food and medicinal purposes (Atato *et al.,* 2010). In Mali, the leaves of the tree are used as fodder (Hamer *et al.,* 2007). South Africa is among the species selected for agroforestry development and domestication (Maghembe *et al.*, 1998). In Botswana, the tree provides farmers with 39 000 CFA francs annually (Taylor *et al.,* 1996). All parts of the species are used in the treatment of several diseases: Schistosomiasis, snake bite, etc. (Arbonnier, 2008); diabetes (Apema *et al.,* 2009); malaria, HIV (Brueton *et al.*, 2009).

Despite the importance of this species in the rural environment, it is still exploited in the wild. In Togo, a study of the status of the stand of this species revealed a regression trend due to its low regeneration rate (Agbogan, 2007). The difficulties of regeneration of this species by Root Segment Cutting (RSC) were reported by Mapongmetsem *et al.* (2016a). The mycorrhizal association is recommended in the nursery (Nelson, 1987). The use of endomycorrhizal inoculums during cuttings improves rooting. Mycorrhizal inoculums have increased rooting, callus size, and survival of cuttings of some species such as *Viburnum dentarum* (Verkade and Hamilton, 1987), *Sciadopitys verticillata* (Douds *et al.*, 1995). The general objective of this study is to contribute to the domestication of *S. birrea* by RSC. Specifically, it aims to evaluate the influence of some external factors on the ability of RSC to neoformed buds and roots; to evaluate the effect of substrate on RSC in the neoformation of adventitious buds and roots; to study the influence of mycorrhizae inoculum on rhizogenesis and calogenesis of root segment cuttings; to determine the best dose of mycorrhizae for the neoformation of buds and roots by RSC.

Materials

Description of the Study Site

The investigations on the cutting of *Sclerocarya birrea* took place in the Guinean savannah highlands, which are periodically burnt Fig. 1 and grazed, notably in the localities of Dang (altitude: 1079 m; latitude: 7°24' North; longitude: 13°32' East) and Gamba (altitude: 2872 m; latitude: 7°48'47.78" North; longitude: 13°34'47.02" East). This area is subjected to a Guinean climate characterized by two seasons: A dry season from November to March and a rainy season that begins in April and ends in October. The human population of the locality is mostly made up of herders (Bororo and Fulbe) and farmers (Mboum, Dii, and Gbaya) (Fawa *et al.,* 2015). This area is covered by shrubs trees savannahs dominated by *Daniella Oliveri* and *Lophira lanceolata* (Letouzey, 1968).

Description of the Site where the Root Segments were Collected

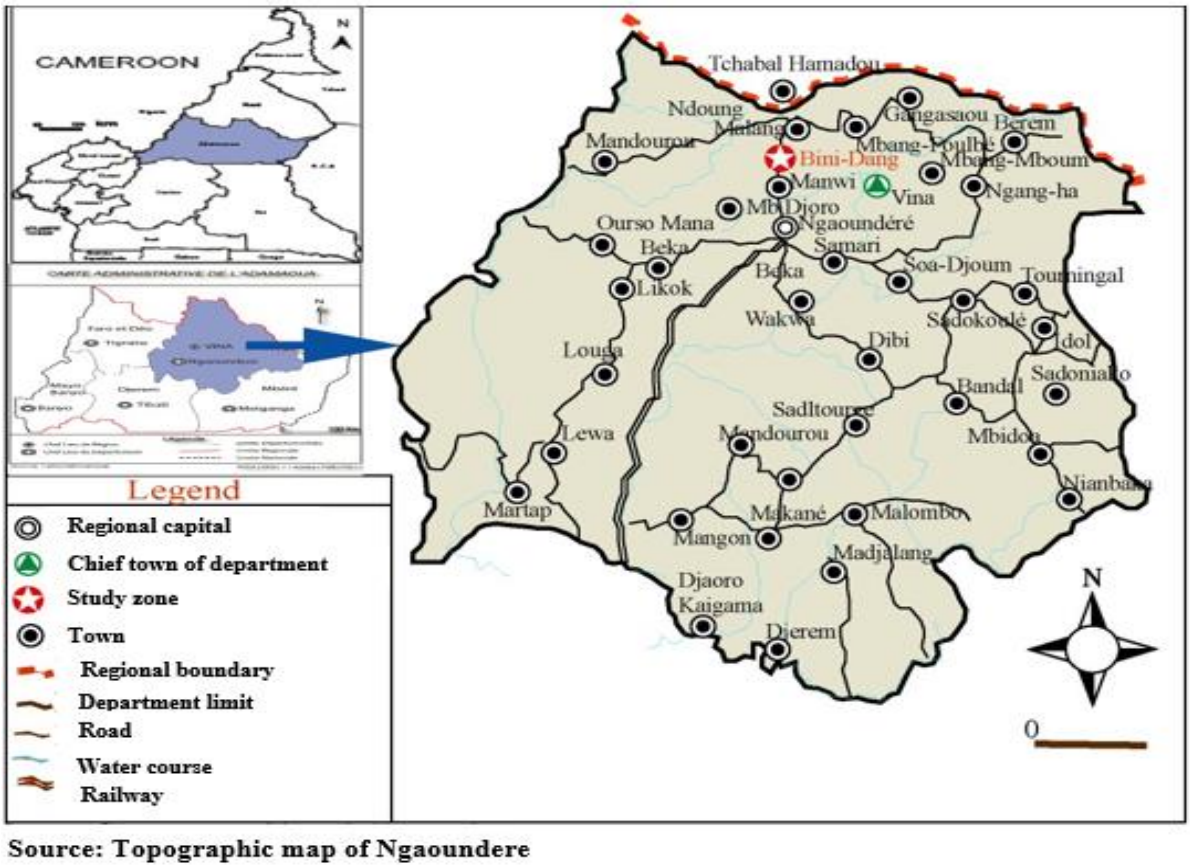
The root segments used in this study were taken from mature trees in the savannah of Poumdjéré. The root fragments were taken from the roots of mature trees after careful excavation of their root systems. Root fragments collected from the savannah with a diameter of 1 to 3 cm were transported in an icebox containing ice blocks to avoid dehydration during excavation and transported from the savannah to the nursery. This precaution keeps the cells turgid. The fragments were cut into Root Segment Cuttings (RSC) on arrival at the nursery.

Presentation of the Nursery and the Polypropagator

The cutting trials were carried out in the nursery of the Laboratory of Biodiversity and Sustainable Development of the University of Ngaoundéré located in Manwi, near the Bini river. Shading is provided by a shed covered with straw that filters out the outside light, the temperature inside the frame was 23-27°C. The propagation device is made from local material and

DOI: 10.3844/ajabssp.2022.40.50

subdivided into 4 compartments. Its shape is like a parallelepiped. This wooden box is covered with a 1 mm thick transparent polyethylene film to maintain a moderate temperature, humidity, and light intensity favorable for the best development of the cuttings (Leakey *et al.*, 2003). From bottom to top, the following layers are arranged: A thin layer of fine sand, large pebbles, medium pebbles, gravel, sand, and finally, the rooting substrate (Fig. 2) (Mapongmetsem *et al*., 2012b). All this material is immersed in water, the height of which is limited to the second layer of sand. The different substrates mixed occupy the upper part of the slick and the cuttings will absorb the water by capillary action. A PVC pipe is inserted at the corner of each compartment and allows the water level in the frame to be gauged regularly (Mapongmetsem *et al.*, 2012b)



Source: Topographic map of Ngaoundere

Fig. 1: Map of the location of the study area

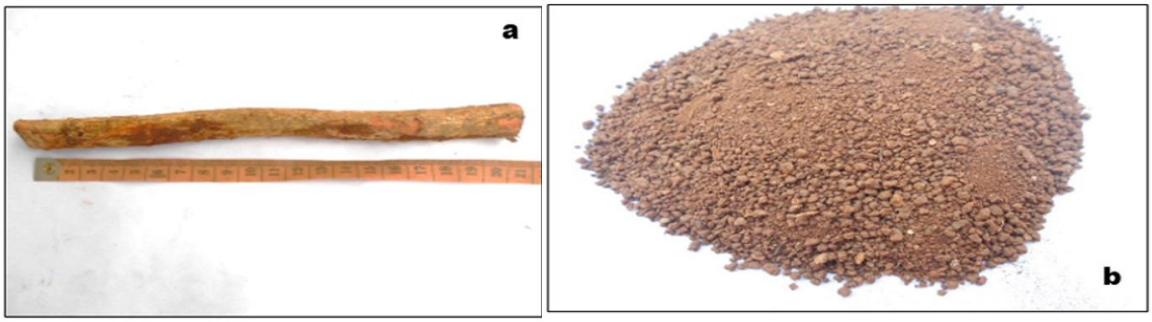


Fig. 2: *Sclerocarya birrea* cuttings (a) and mycorrhizal inoculum (b)

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Methods

Description of the Trials

When returned to the shed, the segments taken from the field were cut into 20 cm cuttings with a pruner which was introduced horizontally into the substrate consisting of the mixture of black soil/sawdust and sand/sawdust. Each location of the root segment cuttings was seeded with a dose of mycorrhizae (10, 20 g) or not (no mycorrhizae or control). The cuttings were watered twice a day, in the morning and evening, using a sprayer that delivers fine drops of water. Evaluations were carried out every week until the end of the trial. A cutting was said to be rooted if the length of the root is equal to or greater than 1 cm, otherwise, it was reinserted into the substrate (Mapongmetsem, 1994). Rooted segment cuttings were placed in black perforated polyethylene bags for acclimatization trials. Cuttings and dead leaves were systematically removed.

Experimental Device

The experimental design used was a split-plot with 4 replicates. The main treatment consisted of the substrate: The black soil/sawdust mixture and the sand/sawdust mixture, while the second treatment was the mycorrhizal inoculum (0, 10, and 20 g). The cuttings that did not receive the mycorrhizal inoculum (0 g) were the control. The different compartments of the polypropagator represented the replicates. The experimental unit was set at 10 cuttings because of the rarity of the species in the area. A total of 240 cuttings ($10 \times 4 \times 3 \times 2 \times 1$) were handled.

Data Collection and Processing

Data collected in each evaluation included the number of cuttings that budded, the number of aerial axes formed, the number of leaves per aerial axis, the height of the leafy axes, the number of rooted cuttings, the number of roots per cutting, and the size of the roots per cutting. Statistical analyses were performed on the variance. The separation of effective means was done using the Duncan multiple range test. The statistical analysis program used was Statgraphics Plus 5.0.

Results

Budding of Root Segment Cuttings

The Root Segment Cuttings (RSC) were implanted on 28 May 2016 and the first budding took place on 22 June 2017, which is 26 days (4 weeks) after planting. Root segment cuttings started budding Fig. 3 on the two substrates used at different times.

Substrate Effect

The first aerial shoots appeared in the sand/sawdust mixture in the 4th week after the RSC trial and in the 5th week in the black soil/sawdust mixture. At the 18th week after cultivation, the percentage of budding ranged from 15.55±4.77% in the black soil/sawdust mixture substrate to 37.77±17.15% in the sand/sawdust mixture Fig. 4. There was a significant difference between substrates (0.006<0.01). Substrates influenced the budding of root segment cuttings Sa/Sc = Sand/Sawdust; Tn/Sc = Dark soil/Sawdust.

Mycorrhizae Effect

Root segment cuttings started to bud at the 4th week after being grown in substrates infested with 20 g of mycorrhiza, whereas at the 10 g dose and in the control substrate, budding started at the 5th week Fig. 5. From the 5th week onwards, the cuttings grown in the 10 g mycorrhizal substrate took over from the others until the end of the experiment.

The percentage of budding until the end of the experiment varied from 18.33±5.85% in the control cuttings to 33.33±21.6% in the cuttings inoculated with 10 g mycorrhizae. Despite this fluctuation, no significant difference (0.22>0.05) is noticeable. In Senegal, T0 = Control, M1 = M1 = Mycorrhizae 10 g; M2 = Mycorrhizae 20 g.

Substrate Mycorrhizal Inoculum Interaction Effect

Regarding the effect of substrate mycorrhizal inoculum interaction, the percentage of budding varied from 13.33±8.27% in root segment cuttings mycorrhized with 10 g grown in the black soil/sawdust mixture to 50±10% in those also mycorrhized with 10 g in the sand/sawdust substrate Fig. 6. Analysis of variance revealed that the substrate*inoculum interaction was not significant (0.39>0.05).

Number of Aerial Axes Per Cutting

Substrate Effect

Up to the 18th week of the experiment, the number of aerial axes ranged from 2.98±0.79 in the black soil/sawdust mixture substrate to 4.7±0.72 in the sand/sawdust mixture Table 1. The analysis of variance did not show a significant difference between substrates (0.15>0.05). This result indicates that substrates did not influence the number of aerial axes.

Influence of Mycorrhizae

Regarding the treatment of substrates with mycorrhizal inoculum, the number of aerial axes varied from 1.58±0.97 in cuttings that received 0 g of mycorrhizae to 5.13±2.98 in those inoculated with 20 g of mycorrhizae Fig. 7. This disparity is confirmed by the analysis of variance, which shows a significant difference between cuttings (0.04<0.05). Thus inoculation of cuttings with mycorrhizae truly influenced the number of aerial axes.

DOI: 10.3844/ajabssp.2022.40.50

Fig. 3: Leafy cutting of *Sclerocarya birrea*

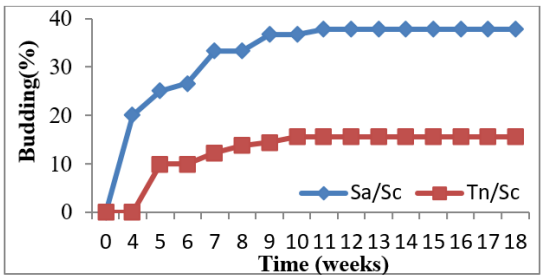

Fig. 4: Budding rate over time

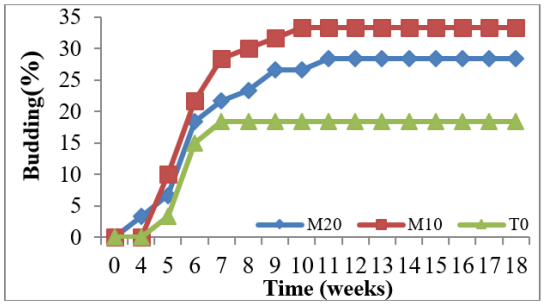

Fig. 5: Budding rate over time

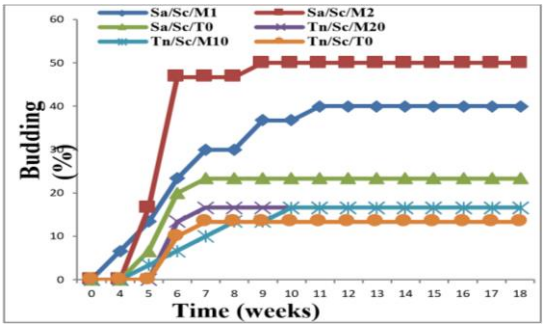

Fig. 6: Budding rate of cuttings according to the substrate by mycorrhizal inoculum interaction

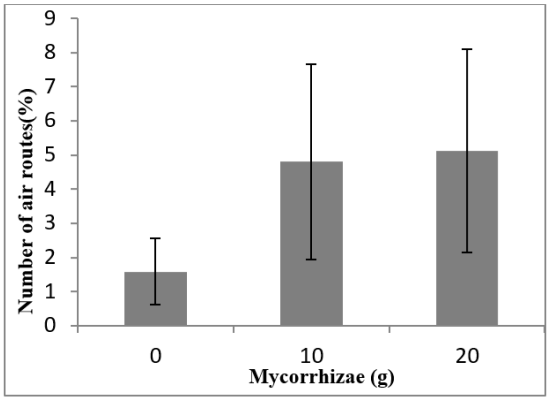

Fig. 7: Number of aerial axes as a function of mycorrhizae

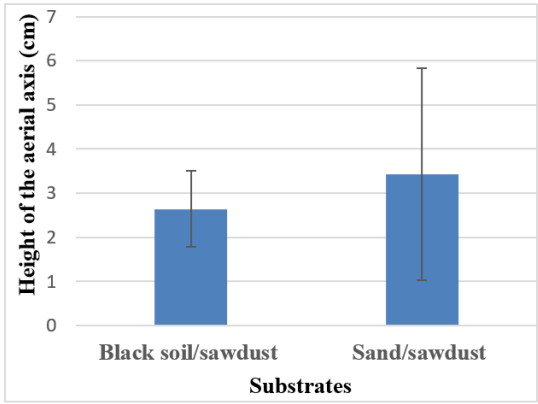

Fig. 8: Height of the RSC according to the substrates

Fig. 9: Cuttings having emitted neo-root

Effect of Substrate Mycorrhizae Interaction

The number of aerial axes varied from 1.5±1.38 in inoculated cuttings grown in the black soil/sawdust mixture to 6.77±1.54 in those inoculated with 10 g grown in sand/sawdust Table 2. Analysis of variance did not indicate a significant difference (0.35>0.05), despite the variation observed.

At the end of the experiment, an average of 5.29 leafy shoots was obtained.

Height of the Aerial Axes

Influence of Substrate

The mean height of aerial axes per BSR fluctuated between 2.64±0.86 cm in the black soil/sawdust mixture and 3.43±2.40 cm in the sand/sawdust mixture Fig. 8. This disparity is only apparent since the analysis of variance does not reveal any significant difference between substrates (0.53>0.05).

Influence of Mycorrhizal Inoculum

Regarding the injection of cuttings with mycorrhizae, the height of the aerial axes ranged from 2.97±1.05 cm in control cuttings to 3.12±2.80 cm in those inoculated with 10 g of mycorrhizae. Despite this disparity, the analysis of variance did not show a significant difference between the doses of inoculum received by the cuttings (0.99>0.05). An increase in height of the aerial axes may be obtained from an average dose of mycorrhizae since it is a symbiotic association between tree roots and fungi.

Substrate Mycorrhizae Interaction Effect

For the interaction between substrate and mycorrhizal inoculum, the height of the cuttings fluctuated between 1.79±1.49 cm in cuttings inoculated with 10 g of mycorrhizae grown in the black soil/sawdust mixture and 4.45±3.39 cm in cuttings with 10 g of mycorrhizae grown in the sand/sawdust mixture Table 3. Despite the fluctuation, analysis of variance shows that the substrate mycorrhizal inoculum interaction is not significant (0.49>0.05). This result suggests that combining these two factors is not beneficial to aerial shoot height growth.

Number of Leaves Per Aerial Axis

Substrate Effect

Up to week 18, the number of leaves per aerial axis varied from 2.92±2.63 in the black soil/sawdust mixture to 3.00±1.11 in the sand/sawdust mixture. Despite this variation, analysis of variance reveals no significant difference between substrates (0.95>0.05). Therefore, the substrate did not influence the number of leaves per aerial axis.

Mycorrhizal Effect

For mycorrhizal cuttings, the number of leaves varied from 2.49±1.80 in cuttings inoculated with 20 g to 3.60±1.36 in those inoculated with 10 g. Despite this variation, the analysis of variance did not show a significant difference (0.84>0.05). These results suggest that the amount of mycorrhizal inoculum determines the growth and development of an explant of *S. birrea*.

Effect of Substrate Mycorrhizae Interaction

For the substratemycorrhizae interaction, the number of leaves per aerial axis varied from 1.61±1.56 in control cuttings grown in the sand/sawdust mixture to 5.26±1.92 in those mycorrhized with 10 g and grown in the sand/sawdust mixture Table 4. The analysis of variance shows the absence of a significant difference (0.35>0.05).

Rooting

After the cultivation of the RSC, the first roots appear around the 4th month. After 18 weeks of cultivation, 7.77% of the cuttings with aerial axes were rooted Fig. 9.

Substrate Effect

At the 18th week after planting the RSC, the rooting rate varied from 3.33±2.05% in the sand/sawdust substrate to 4.44±0.07% in the black soil/sawdust substrate. The analysis of variance did not show a significant difference between the rooting substrates (0.75>0.05).

Mycorrhizae Effect

The root segment cuttings started to root in the 4th month after being grown in the substrates inoculated with 10 g mycorrhiza, whereas in the 20 g dose and the control substrate, rooting started in the 5th month. From this date onwards, the cuttings grown in the 20 g mycorrhizal substrate took over from the others until the end of the experiment.

The rooting percentage until the end of the experiment varied from 1.67±0.27% in the control cuttings to 6.67±3.04% in the cuttings inoculated with 20 g of mycorrhizae. Despite this disparity, analysis of variance revealed that the mycorrhiza inoculum was not significant (0.51>0.05).

Effect of Substrate Mycorrhizal Inoculum Interaction

Regarding the substrate mycorrhizal inoculum interaction, the rooting percentage varied from 0±0% in control cuttings grown in the black soil/sawdust mixture to 6.67±4.3% in cuttings mycorrhized with 10 and 20 g in the sand/sawdust and black soil/sawdust substrate respectively. Analysis of variance revealed that the substrate*inoculum interaction was not significant (0.51>0.05).

Table 1: Variation in the number of overhead axes as a function of substrates

Treatments	Sand/sawdust	Black soil/sawdust	Mean
Number of aerial axes	4.7±0.72	2.98±0.79	3.74±0.75

Table 2: Number of aerial axes according to the substrate mycorrhizal interaction

Substrate/Dose (g)	0	10	20	Mean
Sand/sawdust	1.5±0.830	6.77±1.54	5.83±2.36	4.7±1.5700
Black soil/sawdustsec	1.66±1.52	2.83±2.56	4.44±3.90	2.97±2.660
Mean	1.58±1.17	4.8±2.050	5.13±3.13	3.83±2.110

Table 3: Height of the air axes (in cm) according to the substrates and the mycorrhizal inoculum

Substrate/Dose (g)	0	10	20	Mean
Sand/sawdust	2.38±1.49	4.45±3.39	3.47±0.66	3.19±0.59
Black soil/sawdust	3.55±3.50	1.79±1.49	2.60±2.29	2.39±0.38
Mean	3.91±0.76	2.82±0.23	1.65±0.46	2.79±0.48

Table 4: Number of leaves of cuttings per aerial axis according to substrates and mycorrhizal inoculum

Substrate/Dose (g)	0	10	20	Mean
Sand/sawdust	1.61±1.56	5.26±1.92	2.15±0.44	3.19±0.59
Black soil/sawdust	4.00±3.60	1.94±1.92	2.83±2.75	2.39±0.38
Mean	3.91±0.76	2.82±0.23	1.65±0.46	2.79±0.48

Table 5: Number of roots according to the substrate mycorrhiza interaction

Substrate/Dose (g)	0	10	20	Mean
Sand/sawdust	0.00±0.00	1.76±1.31	5.00±1.75	2.25±1.02
Black soil/sawdust	0.00±0.00	0.00±0.00	3.73±0.31	1.23±0.10
Mean	0.00±0.00	0.88±0.65	4.36±1.03	1.74±0.56

Table 6: Height of the air axes (in cm) depending on the substrates and the mycorrhizal inoculum

Substrate/Dose (g)	0	10	20	Mean
Sand/sawdust	0.00±0.00	4.39±1.79	4.86±2.39	3.08±1.39
Black soil/sawdust	0.00±0.00	0.00±0.00	4.78±1.79	1.59±0.59
Mean	0.00±0.00	2.19±0.89	4.82±2.09	2.33±0.99

Number of New Roots Per Cutting

Substrate Effect

Until the 18th week of the experiment, the number of new roots oscillated between 1.27±1.01 in the substrate based on the black soil/sawdust mixture and 2.22±1.01 in the sand/sawdust mixture. The analysis of variance did not show a significant difference between the substrates (0.52>0.05). This result indicates that the substrates did not influence the number of reformed roots. These observations disagree with those made on *V. doniana* and *Lophira lanceolata* where the best substrate was the black soil/sawdust mixture (Fawa, 2015).

Influence of Mycorrhizae

Concerning the treatment of substrates with mycorrhizal inoculum, the number of neoformed roots varied from 1.0±0.0 in cuttings that received 0 g of mycorrhizae to 3.41±1.24 in those inoculated with 20 g of mycorrhiza Fig. 10. The analysis of variance did not show a significant difference between the cuttings (0.29>0.05). Therefore, inoculation of the cuttings with mycorrhizae did not influence the number of neoformed roots.

In the same vein, Abbott and Robson (1984) and Nelson (1987) reported that even if the needs are low, the fungus acts rather like a parasite by drawing from the plant's reserves of the sugars it needs for its establishment. However, these same authors specify that the delay is compensated for as soon as the symbiosis is established. In the same range, Trépanier (1998) reports on *Juniperus sabina*, *Cornus stolonifera* var. *coloradensis*, and *Prunus cistena* that mycorrhizal inoculums delay the rooting of cuttings because these mycorrhizae fungi use the carbohydrates of the cutting for 3 to 5 months for their development before engaging in symbiosis during root formation. Thus the drying out of the leafy shoots during the experiment would be due to the low carbohydrate content of the RSC being compensated for as soon as the establishment of the symbiosis is effective.

Effect of Substrate Mycorrhiza Interaction

The number of adventitious roots varied from 0.0±0.0 in cuttings inoculated with 0 and 10 g of mycorrhizae grown in the black soil/sawdust mixture to 5.00±1.75 in those inoculated with 20 g of mycorrhizae grown in sand/sawdust Table 5. The analysis of variance does not indicate a significant difference (0.35>0.05), despite the variation observed.

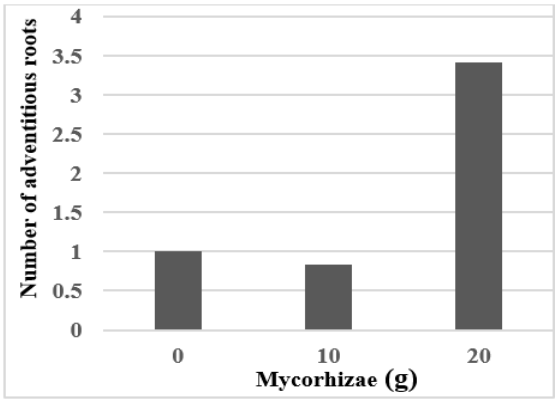

Fig. 10: Number of roots as a function of mycorrhizae

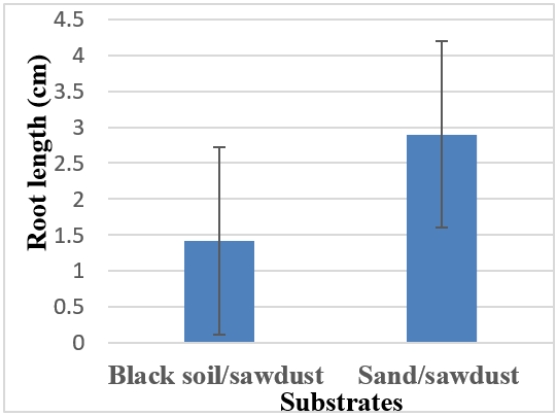

Fig. 11: Length of newly formed roots depending on the substrates

Length of Neoformed Roots
Influence of Substrate

The length of neoformed roots per RSC fluctuated between 1.42±1.3 cm in the black soil/sawdust mixture and 2.9±1.3 cm in the sand/sawdust mixture Fig. 11. This disparity is only apparent since the analysis of variance does not reveal any significant difference between the substrates (0.47>0.05). These results are in disagreement with those obtained on *Vitex doniana* and *Lophira lanceolata* where the best substrate was sawdust (Fawa, 2015).

Influence of Mycorrhizal Inoculum

About the inoculation of cuttings with mycorrhizae, the length of neoformed roots ranged from 0.34±0.8 cm in cuttings inoculated with 0 g to 4.24±1.69 cm in cuttings inoculated with 10 g of mycorrhizae. Despite this disparity, the analysis of variance does not show a significant difference between the doses of inoculum received by the cuttings (0.29>0.05). It is possible that an average dose of mycorrhiza results in high root growth, as it is a symbiotic association between tree roots and fungi.

Effect of the Substrate Interaction Mycorrhizae

For the interaction between substrate and mycorrhizal inoculum, the length of re-formed roots fluctuated between 0.00±0.00 cm in cuttings inoculated with 0 and 10 g of mycorrhiza grown in the black soil/sawdust mixture and 4.86±2.39 cm in cuttings that received 20 g grown in the sand/sawdust mixture Table 6. Despite the fluctuation, the analysis of variance shows that the substrate mycorrhizal inoculum interaction is not significant (0.65>0.05). This result suggests that combining these two factors is not beneficial for the growth of re-formed roots.

Discussion

Budding

The duration of the budding of root segment cuttings of *Sclerocarya birrea* obtained in this study agrees with the results reported by Mapongmetsem *et al.* (2016b) on the same species in Cameroon as well as those of Ky-Dembele *et al.* (2010) in Burkina Faso on *Detarium microcarpum*. These authors obtained the first buds in the 4th week. In Burkina Faso, in *Acacia albida*, it took 8 weeks (Harivel *et al.*, 2006). Similarly, in *Vitex doniana* in Cameroon, they were observed after 8 weeks (Mapongmetsem *et al.*, 2012b). These observations are in disagreement with those obtained in *Brosimum gaudichaudii* (Silva *et al.*, 2011). The budding delay of a species could be explained by different environmental factors such as Pedo-climatic conditions of the habitat, species, genotype, and harvesting period of RSC. In *Sclerocarya birrea*, it could be controlled by these factors. Nevertheless, Duponnois *et al.* (2005; 2007) showed that mycorrhizal inoculums improve the budding of RSC. The authors reported that *Acacia holosericea* associated with ectomycorrhizal fungi, of the genus *Pisolithus* and *Sleroderma*, shows a much higher growth in a controlled environment (greenhouse) than *A. holosericea* without inoculation in the juvenile phase. These results corroborate those of Le Tacon (1997) who reported that on substrates generally lacking natural inoculum, the level of fertility is very high but plant growth is rapid, while root colonization by mycorrhizae fungi is low or nil. Concerning the growth parameters of root segment cuttings, the result was slightly low compared to that obtained on the same species by Mapongmetsem *et al.* (2016a) concerning the number of shoots/RSC (6.75 shoots) and the height (6.59 cm) on average. Similarly, Harivel *et al.* (2006) reported in Burkina Faso an average of 2 shoots per RSC in *Acacia albida*. These authors reported that the substrates that give satisfactory results are sand and sawdust. The result obtained in these substrates would be due to their porosity, which allows water circulation between the cuttings. These observations are analogous to those of Trépanier (1998) who reported that *Cornus stolunifera* var. *coloradensis* seedlings inoculated with 20 and 40% mycorrhizal after the growing period

inoculum showed an improvement in aerial growth up to more than 25% compared to the controls. These observations lead us to believe that the sand/sawdust mixture would be an appropriate substrate for leaf growth in this species. These results are consistent with those reported by Duponnois *et al*. (2005; 2007) in Senegal where *Acacia holosericea* is associated with ectomycorrhizal fungi of the genus *Pisolithus* and *Sleroderma*, showing in a controlled environment a higher juvenile growth than uninoculated *A. holosericea*.

Rooting

The separation of budding and rooting in time shown in this study is justified because the leafy shoots must supply the cuttings with carbohydrates which will stimulate new roots (Mapongmetsem *et al.,* 2016b). However, these authors reported on the same species that sawdust was the best rooting media. These observations are also in disagreement with those obtained on *V. doniana* where the best substrate was sand, sawdust, and black soil/sawdust mixture (Fawa, 2015). These results do not corroborate those of Le Tacon (1997) who reported that on substrates generally lacking natural inoculum, the level of fertility is higher and plant growth is rapid. Still, there is little or no colonization of the roots by mycorrhizal fungi.

In the same vein, Abbott and Robson (1984) and Nelson (1987) argue that even if the needs are low, the fungus acts rather like a parasite by drawing the sugars it needs for its establishment from the plant's reserves. However, these same authors specify that the delay is compensated for as soon as the symbiosis is established. In the same range, Trépanier (1998) reported on *Juniperus sabina*, *Cornus stolonifera* var. *coloradensis*, and *Prunus cistena* that mycorrhizal inoculums delay the rooting of cuttings because these mycorrhizae fungi use the carbohydrates of the cutting for 3 to 5 months for their development before engaging in symbiosis during root formation. Thus the drying out of the leafy shoots during the experiment would be due to the low carbohydrate content of the RSC being compensated for as soon as the establishment of the symbiosis is effective.

Conclusion

This study found that root segment cuttings of *Sclerocarya birrea* showed a good ability to form leafy shoots and rooting. The best substrate for budding was the sand/sawdust mixture, while the 10 g dose favored budding. The highest number of aerial shoots was recorded in the sand/sawdust mixture and the cuttings mycorrhized with 20 g inoculum. The greatest heights were obtained in the sand/sawdust mixture. The highest number of leaves per aerial axis was observed in the mixture of sand/sawdust substrate, and regarding inoculation with mycorrhizae, the 10 g dose was again the best.

It should also be noted that the best substrate for the number of reformed roots was the sand/sawdust mixture, while the 20 g dose was more favorable for the appearance of roots. The best substrate for reformed root length was the sand/sawdust mixture, while the 10 g dose favored the root growth for the African plum. Vegetative propagation by RSC is an alternative to the cultivation of *S. birrea*. The domestication of this species by root segments cuttings associated with mycorrhizal inoculum is to be advised to farmers concerned about a good recovery of their cuttings in the field, given the multiple socioeconomic interests that this species abounds. *Sclrocarya birrea* can be propagated by vegetative propagation by root segment cuttings.

Thus, it would be desirable to carry out an anatomical and histological study of the organs involved in the vegetative propagation of this species to determine the appropriate period for its cutting: To test other modes of vegetative propagation with mycorrhizal inoculum in this species and then to extend it to other woody plants: To carry out cutting tests of this species in the medium of collection of the RSC; to follow the explants produced until fructification to appreciate the effect of the mycorrhizal inoculums.

Acknowledgment

We would like to thank all the referees whose contributions have been very fruitful for the perfection of this manuscript.

Author's Contributions

Herbert Abdoulaye: Carrying out the experiment, collecting and verifying the analyzed data; prepared the draft of the manuscript and approved the final manuscript.

Tsobou Roger, Dona Adoum and Oumarou Haman Zephirin: Member of the Laboratory, experimental monitoring and approved the field data.

Wangbiching Jean De-Dieu: Correction of the translation of the manuscript in English, experimental monitoring, member of the Laboratory and approved the field data.

Binwe Jean-Baptiste: Member of the Laboratory, preparation of the nursery, experimental monitoring, collection of field data and approved the field data.

Megueni Clautilde: Design the research plan, and supervised this study.

Mapongmetsem Pierre Marie: Design the research plan, supervised this study, and approved the final manuscript.

Ethics

This article is original and contains unpublished material. The corresponding author confirms that all of the other authors have read and approved the manuscript and that no ethical issues are involved.

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