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Study of the efficacy of five growing media for nursery production of an endangered species, *Afzelia africana* Smith ex Pers in Côte d'Ivoire

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Abstract

Afzelia africana Smith ex Pers is an endangered plant species listed on the International Union for Conservation of Nature (IUCN) Red List. The low level of natural regeneration of this species requires it to be reproduced in a controlled environment in order to reconstitute its population by providing good quality seedlings. As the growing medium is an important factor in the quality of the seedlings, the influence of the substrates soil + wood chip compost, soil + poultry droppings, soil + rice straw compost, soil + cocoa pod husk compost and soil on the germination and growth of *Afzelia africana* in the nursery was evaluated through a completely randomised design. The soil + wood chips compost performed best, with a high germination rate (76.7%). It enabled the production of taller seedlings (15.6 cm; $p = 0.004$), with larger collar diameters (4.4 mm; $p = 0.001$), higher total dry matter (2.25 $g.plant^{-1}$; $p = 0.005$) and higher root dry matter (2.25 $g.plant^{-1}$; $p = 0.005$). The mixture of three parts soil + one part poultry droppings was toxic for the seedlings, resulting in 60.6% mortality. The use of growing media containing a mixture of soil and wood chips compost is effective for the production of vigorous seedlings. This compost can be recommended for nursery production of *Afzelia africana*.

Key words: *Afzelia africana*; Endangered Plant Species; Growing Media; Efficiency; Vigorous Seedlings

1. Introduction

Afzelia africana Smith ex Pers is a woody species belonging to the Fabaceae family. It grows well in tropical zones, in dense semi-deciduous rainforests [1], gallery forests and mountain vegetation [2]. The tree can reach 35 m in forest areas [3]. It is best suited to deep and well-drained soils [4]. *Afzelia africana* is used for several purposes. The species is widely exploited as timber, service wood and energy wood. Bamigboye *et al* [5] reported that in Ogun State in Nigeria, the primary use of *Afzelia africana* is for spiritual purposes, with all parts of the plant (roots, bark, leaves, stems and seeds) being used. It is also used medicinally to heal wounds, facilitate childbirth, treat eye infections, gonorrhoea, improve male sexual performance, treat inflammation of the knees, cure food poisoning, and to treat headaches, malaria, ulcers, chronic coughs, toothaches, haemorrhoids, stomach upsets, meningitis [5-7]. The leaves are source of food for humans and livestock [8, 9]. As a timber, this species whose average wood density is 716 $kg.m^{-3}$ is highly prized for its resistance to mould, termite attack and many chemicals. In Nigeria, it is considered as first-grade commercial wood [10]. In addition to all these uses, which are exerting strong pressure on the existence of *Afzelia africana*, there is the deforestation of stands for agricultural purposes [11]. The over-exploitation of *Afzelia africana* has made it a vulnerable species on the International Union for Conservation of Nature (IUCN) red list [12]. The low level of natural regeneration of this species due to human pressure means that it needs to be reproduced in large quantities in a controlled environment in order to reconstitute its population by providing good quality seedlings. Assisted reproduction of *Afzelia africana* is, however, limited by several factors, including seed dormancy [11] and the low seed germination rate observed in the wild [13]. Adopting special techniques such as pre-treatment of the seeds can help to improve the germination rate. After the seed germination stage, there is the problem of seedling vigour, which depends, among other things, on the quality of the growing media [8, 14]. One of the most widely used growing media in nurseries is compost,

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as it is free of phytotoxins and a source of carbon dioxide, water, minerals and humus, which are beneficial to plants [15]. These characteristics make compost a complement or substitute for the mineral fertilisers on which intensive agriculture depends. Compost production also makes it possible to recycle waste and clean up the environment. In Côte d'Ivoire, agricultural and forestry residues such as rice straw and wood chips, produced in large quantities, are mostly burnt [16]. The production of composts of known quality from these residues could help nursery growers to improve growing media for *Afzelia africana* and to recycle these unused residues. The aim of this study was to assess the effectiveness of three composts derived from agricultural and forestry residues on the germination and growth of *Afzelia africana* in nurseries.

2. Methodology

2.1. Composting of agricultural and forestry residues

Heap composting was carried out using three main agricultural and forestry residues, to which other raw materials were added. Rice straw compost, cocoa pod husk compost and wood chips compost were formulated from different raw materials. The rice straw compost consisted of rice straw, poultry droppings, wood ash, *Albizia lebbeck* (L.) Benth leaves, eggshells and water. Cocoa pod husk compost was made up of cocoa pod husk, rice straw, poultry droppings, wood ash and water. Finally, wood chip compost was made from non-chemically treated wood chips, rice straw, poultry droppings and wood ash. The quantities of raw materials used to compost the residues are shown in Table 1.

Table 1 Quantity (kg) of raw materials used for composting agricultural and forestry residues

Compost	Rice straw	Cocoa pod husk	Wood chips	Poultry dropping	*Albizia lebbeck*	Egg shell	Wood ache	Water (l)
Rice straw compost	27.6	0	0	27.6	6	11	15.5	180
Cocoa pod husk compost	15	136.5	0	68.2	0	0	15.5	250
Wood chips compost	15	0	129.9	129.9	0	0	15.5	275

Composts made from agricultural and forestry residues were compared with non-composted poultry droppings and soil growing media. The organic substrates consisted of cocoa pod husk compost, rice straw compost, wood chips compost produced on the nursery site and non-composted poultry droppings. The poultry droppings and composts were individually mixed with soil. In this way, one (1) portion organic substrates was mixed with three (3) portions of soil from the topsoil to ensure the survival of seedlings in the nursery [17, 18]. Composting data and the chemical composition of the growing media were determined. Chemical analysis of the growing media was carried out at the soil analysis laboratory of the Institut National Polytechnique Félix Houphouët-Boigny in Yamoussoukro. The pH was determined using a pH meter in a substrate/water suspension of 1/2.5. Total nitrogen was determined by the Kjeldahl method [19]. The soil available phosphorus content was determined by the Olsen 2 method [20] and the soil Ca, Mg and K contents were determined by the ammonium acetate saturation method at pH7 [21]; those of the organic substrates were determined by the dry method [22]. For the determination of Ca, Mg, K and trace elements (Fe, Mn, Zn), the samples were mineralized by the dry method and then the ions were extracted by acid attack. The concentration of Ca, Mg, K and trace elements in solution was determined by atomic absorption spectrometry. The concentration of P was determined by spectrophotometry.

2.2. Plant material for the nursery test

The plant material consisted of *Afzelia africana* Smith ex Pers seeds harvested on 10 March 2023 in Nazinga, Burkina Faso. The seeds had an average weight and diameter of 2.5 ± 0.5 g and 10 ± 2 mm, respectively. They were soaked in water for 24 h, then taken out and left out in the open for at least 30 min before sowing, in order to lift the dormancy.

2.3. Experimental design and trial management

The nursery trial was carried out at the Jean Lorougnon Guédé University (6°54'34''N; 6°26'14''W), in west-central Côte d'Ivoire. The trial was set up on 14 december 2023 and monitored until the third month after sowing, during the long dry season. The average temperature recorded on the experimental site during the trial period was 36°C.

The seedlings were sown directly in polyethylene bags measuring 15 cm x 10 cm. The bags were then laid out in rows, protected from direct sunlight, under a shade canopy, in a completely randomised design. The seedbeds were watered twice a day with an average of 0.66 litres of water per day. Manual weeding was carried out systematically.

2.4. Germination parameters

The number of germinated seeds was recorded daily for 30 days after sowing. Germination parameters such as time to first germination and germination rate were determined. The germination time of the first seed represents the time (in days) between sowing and the first germination. The germination rate (GR) represents the ratio of the number of germinated seeds to the total number of seeds sown. The ratio of the number of dead seedlings to the number of germinated seedlings, or mortality rate was also determined at three months after sowing.

2.5. Seedling growth parameters

Growth parameters were determined at one month and three months after sowing for all seedlings. The diameter at the collar was measured using a digital calliper with an accuracy of 0.2 mm. Seedling size was measured with a tape measure. The number of leaves per seedling was counted. The average leaf area was determined on a sample of leaves taken from 10 plants per treatment at three months after sowing. The biomass of the roots, stems and leaves of this sample was measured using a digital balance with a capacity of 500 g and accuracy of 0.01 g. Dry matter was assessed by weighing the organs of the seedlings after rinsing and drying in an oven at 70°C for 48 hours.

2.6. Data analysis

Data were processed using STATISTICA 7.1 software. When the distribution of the variables was normal, the analysis of variances was performed, the means were compared and the least significant difference was detected at 5%. When the variable distribution was not normal, the Kruskal-Wallis test was used to compare the means. Leaf area was determined using MESURIM (https://acces.ens-lyon.fr/acces/logiciels/applications/mesurim).

3. Results

3.1. Composting data

The average temperature of the first mesophilic phase was 32.8°C for the rice straw compost, 32°C for the cocoa pod husk compost and 32°C for the wood chip compost. The thermophilic phase of composting began in the first week after heaping, in all composts. The average temperatures measured at the start of the thermophilic phase were 43.9°C for the rice straw compost, 46°C for the cocoa pod husk compost and 52.9°C for the wood chips compost. Maximum heap temperatures were reached during the third week after heaping. The lowest maximum temperature was recorded in the cocoa pod husk compost heap (56.7°C). The thermophilic phase lasted 18 weeks in the rice straw compost, with an average temperature of 41.1°C. The thermophilic phase lasted 21 weeks in the cocoa pod husk compost and the wood chips compost.

Table 2 Composting temperatures and times for agricultural and forestry residues

Composting parameters	Rice straw compost	Cocoa pod husk compost	Wood chips compost
Mean temperature of the mesophilic phase (°C)	38.2	32	32
Mean temperature at the start of the thermophilic phase (°C)	43.9	46	52.9
Maximum temperature of the thermophilic phase (°C)	60.1	56.7	61.8
Mean composting temperature (°C)	38.7 ± 7.7	42 ± 6.2	47.2 ± 8.8
Composting duration (week)	18	21	21

3.2. Chemical characteristic of substrates

The topsoil used to make up the growing media was acidic (pH = 5.2). The organic matter (OM = 3.29%) and total nitrogen (N = 0.2%) contents were relatively low (Table 3). The C/N ratio indicates a good capacity for decomposition of the organic matter (C/N = 9.8).

Table 2 Chemical characteristics of the topsoil

pH	C (%)	Nt (%)	C/Nt	OM (%)	Av. P (ppm)	CEC ($cmol.kg^{-1}$)	Ca^{2+} ($cmol.kg^{-1}$)	Mg^{2+} ($cmol.kg^{-1}$)	K^{+} ($cmol.kg^{-1}$)	Na^{+} ($cmol.kg^{-1}$)
5.2	1.91	0.2	9.8	3.29	34	12	1.312	1.084	0.255	0.39

The pH values of all the growing media based on organic matter were slightly acidic to neutral (6.4 ≤ pH ≤ 7.1). The organic matter content was high (OM ≥ 10%). The available phosphorus content of all the growing media was low. The C/N ratio was all below 15 (Table 4).

Table 3 Some chemical characteristics of growing media based on organic matter

Substrats	**pH**	**C (%)**	**Nt (%)**	**OM (%)**	**C/Nt**	**Av. P (ppm)**	**K (%)**	**Ca (%)**	**Mg (%)**
Rice straw compost	6.8	17.41	1.26	30.01	13.82	0.76	0.83	0.93	0.23
Cocoa pod husk compost	6.5	18.77	1.48	32.36	12.65	5.96	1.28	2.25	0.29
Wood chips compost	6.5	19.76	1.68	34.07	11.76	5.71	3.30	2.16	0.31
Soil + Cocoa pod husk compost	6.4	6.04	0.42	10.41	10.40	1.17	0.746	0.621	0.355
Soil + Rice straw compost	7.1	6.45	0.64	11.12	11.09	2.16	0.682	1.158	0.264
Soil + Poultry droppings	6.7	8.11	0.56	13.98	13.96	2.02	0.822	0.836	0.288

3.3. Germination parameters and seedling mortality rate

The germination time for *Afzelia africana* seeds was 19 days for all the growing media. The germination rate assessed at one month after sowing varied according to the growing media (Table 5). The highest germination rate was obtained on the soil + wood chip compost (76.7%), while the lowest was obtained on the soil + cocoa pod husk compost (51.7%). The soil, soil + rice straw compost and soil + poultry droppings growing media had germination rates of 68.3%, 63.3% and 55% respectively.

Table 4 *S*eed germination and seedling mortality rates

Parameters	**Soil + wood chips compost**	**Soil**	**Soil + rice straw compost**	**Soil + cocoa pod husk compost**	**Soil + poultry droppings**
Germination rate (%)	76.7	68.3	63.3	51.7	55.0
Mortality rate (%)	4.3	0.0	2.6	0.0	60.6

At three months after sowing, the soil + poultry droppings resulted in high seedling mortality (60.6%) compared with soil + wood chips compost (4.3%), soil (0%), soil + rice straw compost (2.6%) and soil + cocoa pod husk compost (0%).

3.4. Seedling growth parameters assessed at one month after sowing

The soil + wood chips compost provided the best growth in height and diameter of the seedlings at one month after sowing (Figure 1). The height of the seedlings (15.6 cm; p = 0.004) and the diameter at the collar (4.4 mm; p = 0.001) were significantly higher on the soil + wood chips compost. The lowest values for seedling height and diameter at collar were obtained on the soil + cocoa pod husk compost (13.9 cm) and soil (3.9 mm), respectively. In addition, the soil + poultry droppings favoured higher leaves production (n = 2.5; p = 0.000).

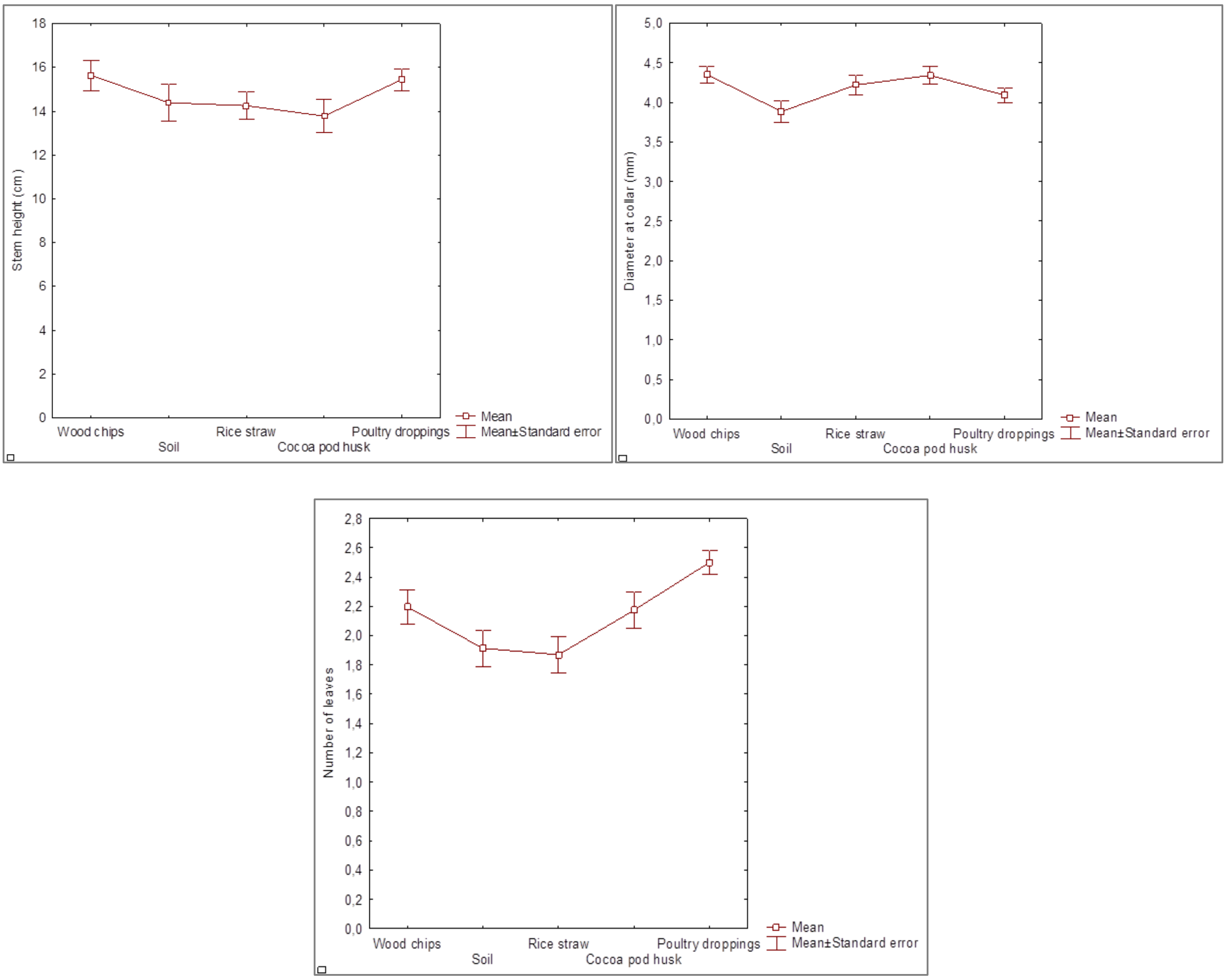

Figure 1 Compared means of the height, diameter at collar and number of leaves of the seedling at one month after sowing

3.5. Seedling growth parameters assessed at three months after sowing

3.5.1. Growth of aerial organs

At three months after sowing, the height and number of leaves of seedlings assessed on all substrates were statistically equal (Figure 2). The diameter at the collar of seedlings grown on the soil + wood chips compost was the highest (4.8 mm; p = 0.002). The values obtained on the soil (3.4 mm), soil + rice straw compost (4.1 mm) and soil + cocoa pod husk compost (4.2 mm) were the lowest.

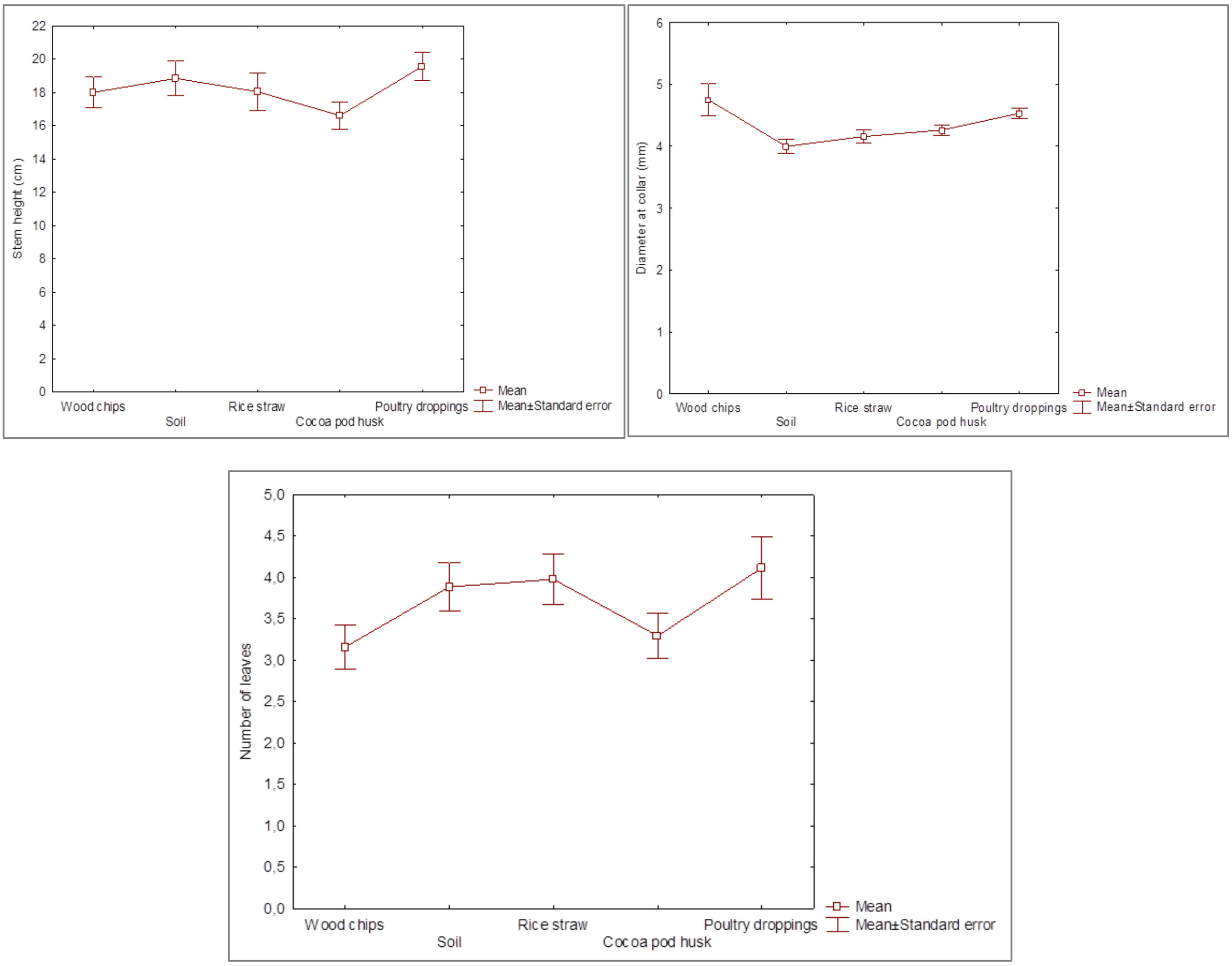

Figure 2 Compared means of the height, diameter at collar and number of leaves of the seedling at three months after sowing

The increase in seedling height, collar diameter and number of leaves from the first to the third month after sowing was assessed (Table 6).

Table 5 Increase in stem height, diameter and number of leaves of seedlings at three month after sowing

Treatments	Increase in height (%)	Increase in diameter at collar (%)	Increase in leaves number (%)
Soil + Wood chips compost	15.2	9.2	43.9
Soil	29.5	2.7	92.7
Soil + Rice straw compost	26.3	2.0	94.6
Soil + Cocoa pod husk	20.9	-0.3	47.8
Soil + Poultry droppings	26.1	11.2	64.4

The increase in height was lower in seedlings growing on the soil + wood chips compost (15.2%), whereas the soil alone had favoured the highest seedling growth (29.5%). In terms of seedling diameter growth, the soil + poultry droppings (11.2%) and soil + wood chips compost (9.2%) favoured greater growth. Seedlings grown on soil + cocoa pod husk compost had a relatively low mean height (20.9%) between the first and third month after sowing. The highest increase in number of leaves was obtained in seedlings sown on soil + rice straw compost (94.6%) and soil (92.7%).

3.6. Leaf area

The mean leaf area of seedlings growing on the soil (38.40 ± 5.84 cm^2), soil + poultry droppings (32.91 ± 2.72 cm^2), soil + wood chips compost (33.44 ± 2.93 cm^2), soil + cocoa pod husk compost (29.71 ± 2.89 cm^2) and soil + rice straw compost (35.79 ± 5.48 cm^2) were statistically similar.

3.7. Roots growth

The average length and diameter of the roots were assessed. The longest roots ($p = 0.024$) were those of seedlings grown on the soil (19.5 cm) and soil + wood chips compost (17.8 cm). Root diameters were statistically similar on all growing media (Table 7).

Table 6 Root mean length and diameter of seedlings at three months after sowing

Treatment	Roots length (cm)	Roots diameter (mm)
Soil + Wood chips compost	$17.75±1.06^{ab}$	$5.73±0.45^{a}$
Soil	$19.5±1.47^{b}$	$5.2±0.36^{a}$
Soil + Rice straw compost	$15.65±1.39^{a}$	$5.91±0.34^{a}$
Soil + Cocoa pod husk compost	$14.2±0.90^{a}$	$5.49±0.33^{a}$
Soil + Poultry droppings	$15.15±1.18^{a}$	$5.81±0.44^{a}$

3.8. Seedlings dry matter

The total dry matter of *Afzelia africana* seedlings and that of the leaves, stems and roots of this species were assessed separately. The discriminating effect of growing media on seedling dry matter was observed for both total dry matter and root dry matter (Table 8). Seedlings grown on the soil + wood chips compost was the most effective in terms of total seedling dry matter (2.25 $g.plant^{-1}$; $p = 0.005$) and root dry matter (2.25 $g.plant^{-1}$; $p = 0.005$). The lowest performing growing medium was soil + cocoa pod husk compost which total seedling and root dry matter was 1.30 $g.plant^{-1}$ and 0.37 $g.plant^{-1}$ respectively. The soil, soil + rice straw compost and soil + poultry droppings had intermediate values for total dry matter (1.89 $g.plant^{-1}$, 1.83 $g.plant^{-1}$, 1.75 $g.plant^{-1}$, respectively), leaf dry matter (0.81 $g.plant^{-1}$, 0. 81 $g.plant^{-1}$, 0.81 $g.plant^{-1}$, respectively), stem dry matter (0.47 $g.plant^{-1}$, 0.47 $g.plant^{-1}$, 0.48 $g.plant^{-1}$, respectively) and root dry matter (0.61 $g.plant^{-1}$, 0.55 $g.plant^{-1}$, 0.47 $g.plant^{-1}$, respectively).

Table 7 Dry matter of leaves, stems and roots of seedlings at three months after sowing

Treatments	Total dry matter weight ($g.plant^{-1}$)	Leaves total dry matter ($g.plant^{-1}$)	Stems total dry matter ($g.plant^{-1}$)	Roots total dry matter ($g.plant^{-1}$)
Soil + Wood chips compost	$2.25±0.26^{b}$	$1.02±0.21^{a}$	$0.52±0.08^{a}$	$0.71±0.15^{b}$
Soil	$1.89±0.15^{ab}$	$0.81±0.12^{a}$	$0.47±0.07^{a}$	$0.61±0.10^{ab}$
Soil + Rice straw compost	$1.83±0.17^{ab}$	$0.81±0.16^{a}$	$0.47±0.08^{a}$	$0.55±0.05^{ab}$
Soil + Cocoa pod husk compost	$1.30±0.13^{a}$	$0.56±0.10^{a}$	$0.37±0.03^{a}$	$0.37±0.04^{a}$
Soil + Poultry droppings	$1.75±0.31^{ab}$	$0.81±0.23^{a}$	$0.48±0.08^{a}$	$0.47±0.11^{ab}$

4. Discussion

4.1. Composting and the chemical quality of compost

The addition of *Albizia lebbeck* leaves and poultry droppings, both rich in nitrogen, to the rice straw compost heap favoured rapid decomposition of the substrates by the microorganisms, whose activity raised the temperature [23]. As a result, the composting time was reduced. On the other hand, the lignin content of rice straw, estimated at 9.8% [24], which is lower than that of wood, which is around 20-30% [25], would have contributed to slowing down the decomposition of the wood chip compost heap and extending its composting time, as it is known that lignin is difficult for microorganisms to decompose [26, 27]. Furthermore, a C/N ratio of less than 15 in composts indicates that they are mature [28] and that organic matter tends to decompose rapidly, releasing nitrogen at high speed. In well-humified soils with a C/N ratio close to 10, the addition of compost with a C/N ratio below 15 can maintain the soil's microbiological balance [29]. The slightly acidic pH of compost is ideal for plant nutrition. The relatively high mineral content of these composts could enable them to be used as organic fertilisers.

4.2. Seed germination and seedlings mortality rate

The germination of plant seeds is influenced by physical and chemical conditions such as acidity, oxygenation, heat and humidity. A good growing medium must promote water drainage and gas exchange (oxygen and carbon dioxide), sufficient to maintain a good water retention capacity and a pH close to neutrality [30-33]. In this study, the soil + wood chips compost gave the highest germination rate (76.7%). Indeed wood chips compost is a fairly porous, low-density medium that is not very compact, providing temperature and water drainage conditions that are suitable for good seed germination [34-35]. This environment also ensures good emergence of germinated seedlings, with a relatively low mortality rate (4.3%). In contrast, the soil + poultry droppings proved not to be suitable growing medium for germination (55% germination) for *Afzelia africana* and was toxic (60% mortality) for young seedlings. Similar results were obtained by Delgado *et al*. [36], working on the toxicity of poultry droppings-based growing substrates for cress (*Lepidium sativum*, L.) seedlings. These authors showed that the application of a non-composted poultry droppings is toxic and leads to a drop in the germination index when the proportion of poultry droppings exceeds 25%. Although less toxic at the 25% dose, the toxicity of poultry droppings at this dose resulted in a lower germination index compared with seeds sown on growing media containing an equal dose of composted manure.

4.3. Seedlings growth

The growing medium is the source of nutrients for the seedling's growth and development. The quality of the seedlings is strongly influenced by the growing media. The nutrients uptake by the seedling depends first and foremost on sufficient quantities of water and oxygen in the growing medium [37]. The characteristics of the growing medium therefore directly affect the development and subsequent maintenance of a functional and extensive root system in plants [38]. The near-neutral pH values of soil + organic matter growing medium ensure good availability of essential nutrients for plants and provide an enhanced ability for plants to take up mineral nutrients. Adding compost to the growing media increased their organic matter content. In addition, when the C/N ratio of composts is below 15, they tend to promote the rapid release of nitrogen into the soil and maintain the soil's microbiological balance [29]. Under these conditions, nutrients are regularly made available to the seedlings [38]. The soil + wood chips compost was the most effective in promoting height growth and enlargement of seedlings at one month after sowing, and then the highest growth and dry matter of roots at three months after sowing. This result is thought to be linked to the fairly porous, slightly compact and relatively low-density nature of this growing medium, which facilitates the gas exchange enough for good root growth [39]. In addition, the abundance of roots contributes to aggregate formation through the secretion of mucilage, which further increases growing media aeration and fluid flow [40]. Furthermore, the chemical composition and the pH close to neutral in this growing medium are favourable conditions for the mineralization of organic matter and the availability of essential nutrients for seedling growth [41]. The performance of the soil + wood chips compost on the seedlings during the first month of growth helped to enhance the physiological functionality and subsequent vigour of the seedlings, as initially high diameter and high seedling size tend to maintain their advantage over time [42,43]. Thus, despite the lower number of leaves produced by seedlings growing on the soil + wood chips compost in the first month after sowing, this substrate resulted in seedlings with a high total dry biomass and long, vigorous roots. This could also be explained by the adequate nutrition of the seedlings provided by the increased intrinsic competitiveness of well-developed roots [44]. In fact, around 95% of plant biomass comes from carbohydrates produced in the green parts by photosynthesis, and the remaining 5% comes from mineral elements absorbed by the roots [45].

Plant dry matter is known to reflect the integration of all the physiological processes undergone throughout the growth stage [46, 47], and the functional aspects of assimilation [48]. The high dry matter value of seedlings grown on the soil

+ wood chips compost would therefore reflect a higher photosynthetic activity [49]. Furthermore, the early vigour of seedlings grown on the soil + wood chips compost is a good indicator of the ability of these seedlings to proliferate better in the field [18, 44, 50, 51]. In addition, the high competitiveness of the roots could enable the seedlings to be more resistant to disturbances caused by transplanting and improve their ability to adapt to new environmental conditions after transplanting [52], root biomass being a more accurate indicator of the seedlings' performance in the field after transplanting [53]. The soil + wood chips compost is a good substrate for nursery propagation of *Afzelia africana*, compared with soil + poultry droppings, soil + rice straw, soil + cocoa pod husk and soil.

5. Conclusion

The production of high-performance seedlings of *Afzelia africana* in nurseries is necessary for the recovery of stands through reforestation. The use of vigorous seedlings is a guarantee of an improved survival rate once the seedlings have been transferred from the nursery to the field. The development of growing substrates to produce good quality seedlings should be encouraged. The soil + wood chips compost promoted good seed germination and the production of seedlings with a high total biomass and vigorous roots. It also produced taller plants with a larger collar diameter. It could help *Afzelia africana* seedlings adapt better when they are transferred to the field. The use of growing media consisting of three (3) parts soil + one (1) part wood chips compost has been beneficial for the production of seedlings in the nursery, and may be recommended for the large-scale production of *Afzelia africana*.

Compliance with ethical standards

Disclosure of conflict of interest

The authors have no relevant financial or non-financial interests to disclose. The authors have no competing interests to declare that are relevant to the content of this article. All authors certify that they have no affiliations with or involvement in any organization or entity with any financial interest or non-financial interest in the subject matter or materials discussed in this manuscript. The authors have no financial or proprietary interests in any material discussed in this article.

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