



Plant quality of *Gmelina arborea* Roxb. produced with different substrate mixtures at the nursery

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Abstract:

The quality of the plant that is produced in the nursery is important to achieve the successful establishment of forest plantations. The aim of the study here described was to assess the growth and quality of *Gmelina arborea* plants, a species that has adapted easily to the humid and subhumid tropical regions of Mexico, with different substrate mixtures through the traditional nursery production system. A completely randomized experimental design was applied, with six treatments, five repetitions and 25 plants per experimental unit. The treatments evaluated were: T1 (50 % soil + 50 % coffee husk), T2 (50 % soil + 50 % sawdust (mixture of *Mangifera indica*, *Tabebuia rosea* and *Tabebuia donnell-smithii*), T3 (50 % soil + 50 % cocoa shell), T4 (50 % soil + 50 % sugarcane bagasse), T5 (50 % soil + 50 % livestock manure) and T6 (100 % forest soil). After 75 days of having carried out the sowing, the statistical analysis showed significant differences ($P \leq 0.05$) among them, in which the T5 generated the highest values in diameter (8.76 mm), height (66.08 cm), area foliar ($1\ 097.42\ cm^2$), total biomass (11.48 g) and Dickson quality index (1.00). It is concluded that this substrate produces quality plants of the tested species.

Key words: Sawdust, sugar cane bagasse, coffee husk, *Gmelina arborea* Roxb., plant propagation, traditional production system.

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Introduction

In recent years, there has been a need to stimulate reforestation programs and commercial forest plantations with fast-growing species to meet the growing demand for forest products and environmental services (Conafor, 2011; Fierros, 2012), in addition to recovering denuded areas with vegetation, as a consequence of the serious problem of deforestation caused mainly by land use change (Semarnat, 2015).

Gmelina arborea Roxb. Is a tropical forest species native to Southeast Asia, of great economic importance for different purposes (Patiño et al, 1982; Hossain, 1999; Dvorak, 2004). Their speed of growth (Juárez and Ramírez, 1985), their diversity of uses and their easy adaptation to the different ecological conditions (Vázquez and Ugalde, 1995, Martínez et al., 2015) have made them include in most of the programs of forest plantations in tropical countries (Murillo, 1991). In Mexico it has adapted easily to humid and subhumid tropical regions in the states of *Yucatán, Veracruz, Jalisco, Oaxaca, Quintana Roo, Campeche, Colima, Nayarit, Michoacán, Tabasco and Chiapas*.

Forest ground is the most common substrate in the Mexican nurseries that work with the traditional production system in forest and fruit species; however, its frequent use causes an undesirable environmental impact due to extraction of large amounts of soil, mainly from coffee and cacao plantations. NOM-003-RECNAT-1996 (Semarnat, 1996) establishes the procedures, criteria and specifications for the exploitation, transport and storage of the forest ground, although it mentions that its irrational use causes serious problems to the ecosystem due to the erosion and loss of soil productivity.

On the other hand, in some regions of the country there are by-products of the wood industry and other natural materials such as the residues of agricultural crops (sugar cane bagasse, coffee husk, cocoa husk, coconut husk, livestock and sheep manure, among others) that can be used as components to improve substrate mixes and substitute the use of forest soil (Reyes *et al.*, 2005; Maldonado *et al.*, 2011; Mateo *et al.*, 2011).

Producing quality nursery plants helps to guarantee a higher percentage of survival in forest plantations and in reforestation programs (Birchler *et al.*, 1998; Rodríguez, 2008; Prieto and Sáenz, 2011). To achieve this, it is important to have an effective control of the production process (Reyes *et al.*, 2005; Basave *et al.*, 2014) and to have an adequate substrate for the selected species (Cabrera, 1999; Pastor, 2000). Given that the volume of a container is limited (Prieto *et al.*, 2007; Sánchez *et al.*, 2016), the substrate and its components must possess physical and chemical characteristics that, combined with a comprehensive management program, allow the growth and optimal development of the seedling (Arteaga *et al.*, 2003; Sánchez *et al.*, 2008; Orozco *et al.*, 2010) and that reduce production costs, but that guarantee the quality of the plant.

The concept of plant quality is mainly based on morphological and physiological characteristics (Thompson, 1985; Birchler *et al.*, 1998; Rodríguez, 2008; Prieto *et al.*, 2009), which allows to know their real status and probable behavior according to the conditions of the place where it will be planted; it is important because it supposes a survival and initial growth starting from the different cultivation practices. In this context, the objective proposed for the research described here was to evaluate the initial growth of *Gmelina arborea* seedlings produced in the traditional system, with different mixes of substrates and to determine the morphological characteristics of the plant that favor its survival in the final place of plantation.

Materials and Methods

Study area

The research work was carried out in the forest nursery of the *Facultad de Agricultura* of the *Universidad Autónoma de Chiapas* (School of Agricultural Sciences Campus IV of the Autonomous University of Chiapas). It is located in *Huehuetán* municipality, *Chiapas*, at an altitude of 44 m, between the parallels 15°00' and 15°30' N and between the meridians 94°30' and 95°00' W.

Weather conditions

The climate that predominates in the study area corresponds to Aw₂ (w") ig which refers to the warm humid with summer rains according to the Köppen classification system modified by García (1973). The average annual rainfall is 2 415 mm from May to November, with an average temperature of 28.5 °C; average maximum of 37.1 °C and minimum average of 19.7 °C and an average evaporation of 1 990 mm.

Vegetal material

The *Gmelina arborea* seed collection was carried out in a seed stand within the experimental station of the School of Agricultural Sciences. The average DBH of the trees is 38 cm and the average total height is 21 m. The seeds to establish such stand come from the *El Chaparral* Ranch, located in the *Juchique de Ferrer* municipality, *Veracruz*.

Treatments and experimental design

A completely at random experimental design was used; six treatments of substrate mixes were tested (Table 1), with five replications and 25 plants per experimental unit, from which 12 were taken for the destructive analysis.

Table 1. Substrate mixes treatments for the initial growth and plant quality of *Gmelina arborea* experiments.

Treatment	Substrate mix
1	50 % forest ground + 50 % coffee husk
2	50 % forest ground + 50% sawdust
3	50 % forest ground + 50 % cocoa husk
4	50 % forest ground + 50 % sugar cane bagasse
5	50 % forest ground + 50 % livestock manure
6	100 % forest ground

Description of substrates

Six substrates were used that are available and easy to get in the region, such as coffee husk (*Coffea* sp.) from a factory one year after being processed; sawdust from *Mangifera indica* L., *Tabebuia rosea* (Bertol.) DC. and *Tabebuia donnell-smithii* Rose, also one year after being processed and dried out in the open; cocoa husk (*Theobroma cacao* L.) with six months of drying under the shadow of a cacao plantation; cane bagasse (*Saccharum officinarum* L.) one year after being processed under shade; livestock manure collected fresh and ground from a cacao plantation. The coffee husk, sawdust, cocoa husk and cane bagasse were ground into a mechanical mill, resulting in particles less than 5 mm in size. The amount of substrate was measured to prepare the mix corresponding to each treatment, which were incorporated later and stirred with a shovel until the mix was homogeneous.

Plant production

The germination of *Gmelina arborea* seeds consisted of immersing them in water for 48 h and 24 h after leaving them in the open air under shade so that they would lose moisture; subsequently, two seeds were sown in the center of each container (black 15 × 20 cm polyethylene bags, 400 µm) at a depth twice their size and covered with the same substrate mix corresponding to the treatment. The material was deposited manually into each container.

Cultivation practices

After the emergence of the seeds, Captan® 500 was applied in a dose of 2 g L⁻¹ of water each week for a period of 30 days, in order to prevent damping-off or seedling disease. Three applications of the Bayfolan® foliar fertilizer were carried out in doses of 2 g L⁻¹ of water; the first application was made 30 days after the emergence and the other two, every 8 days. A single removal was carried out 10 days after the emergence, when it was left to the plants that had favorable characteristics for their growth and development.

Assessed variables

The plants of *Gmelina arborea* were assessed 75 days after sowing in all the variables except for the time of emergence that was quantified when the plant was visible in each bag; the height was measured with a ruler graduated in centimeters, from the base of the stem to the apical bud; the diameter was measured in millimeters at the base of the stem, with a digital vernier (Traceable® 3416 model), the total number of leaves of each

evaluated plant was quantified and the leaf area was determined with the help of the leaf area integrator LI-COR® LI-3000^a model, in cm².

To evaluate biomass, 12 plants were extracted at random from each experimental unit, to which the adhering substrate was eliminated by washing in running water. The samples were placed in paper bags with their respective identifications and dried in an oven (*VWR Sheldon Manufacturing* 1390FM model) at 70 ° C up to their constant weight. After that period, the aerial and radical part was weighed separately on an analytical balance with precision to milligrams (Denver Instrument 310 model).

The aerial part / root ratio was obtained by dividing the aerial dry weight (g) between the radical dry weight (g) and the slenderness index, by dividing the height between the diameter of the stem (Thompson, 1985). The signification index corresponds to the percentage of dry weight in relation to the water content in the plants (Prieto *et al.*, 2004). The Dickson Quality Index (ICD) resulted from integrating the values of total biomass, the slenderness index and the aerial / root part ratio, in which high values indicate plants of better quality (Dickson *et al.*, 1960); its formula is the following:

$$ICD = \frac{\frac{\text{Total dry weight (g)}}{\text{Height (cm)}} + \frac{\text{Aerial dry weight (g)}}{\text{Diameter (mm)}}}{\frac{\text{Root dry weight (g)}}{}}$$

Statistical analysis

The data of each plant corresponding to the evaluated variables were analyzed using the SAS version 9.0 (SAS, 1999) statistical software. The analysis of variance was carried out using the PROC GLM procedure; later, the variables with significant difference were subjected to Tukey's means comparison test ($p \leq 0.05$) to determine the treatment with the most outstanding values.

Results and Discussion

The results of the analysis of variance showed significant differences ($p < 0.05$) for the variables days to emergence, number of leaves and foliar area; and significant ($p < 0.01$) for the following variables: stem diameter, plant height, total biomass, aerial part / root ratio, lignification index and Dickson quality index. There were no significant differences for the slenderness index variable (tables 2 and 3).

Table 2. Results of the analysis of variance for morphological characteristics considered in the initial growth of *Gmelina arborea* Roxb. in the nursery in different substrate mixes.

Source of variation	D.F.	Mean square and significance				
		DMER	DIAM	ALTU	NHOJ	AFOL
Mix	5	16.50*	95.94**	483.48**	950.52*	1719255.29*
Error	354	4.82	0.90	95.08	78.99	32083.25
C.V.		17.93	12.90	17.50	24.40	19.54
r^2		0.04	0.59	0.41	0.14	0.43

DMER = Days of emergence; DIAM = Diameter; ALTU = Height; NHOJ = Number of leaves; AFOL = Foliar area; *Significant with $p \leq 0.05$; ** Significant with $p \leq 0.01$; C.V. = Coefficient of variation (relation between the size of the mean and the variability of the variable); D.F. = Degrees of freedom.



Table 3. Results of the analysis of variance of total biomass, aerial part /root ratio, slenderness index, lignification index and Dickson's quality index in the initial growth of *Gmelina arborea* Roxb. in different substrate mixes.

Variation sources	D.F.	Mean square and significance				
		BIOT	RPAR	IESB	ILIG	ICD
Mix	5	427.08**	0.56**	2.40 ^{ns}	210.02**	3.17**
Error	354	4.63	0.97	1.67	17.07	0.04
C.V.		26.81	0.97	17.00	17.52	30.21
r^2		0.56	0.07	0.01	0.14	0.48

BIOT = Total biomass; RPAR = Aerial part /root ratio; IESB = Slenderness index; ILIG = Lignification index; ICD = Dickson's quality index; *Significant with $p \leq 0.05$; ** Significant with $p \leq 0.01$; C.V. = Coefficient of variation (relation between the size of the mean and the variability of the variable); D.F. =degrees of freedom.

Days to emergency

Tukey's mean comparison test ($p \leq 0.05$) revealed that the seeds of *Gmelina arborea* sown in 100% cacao ground emerged on average at 11.7 days, in contrast to the treatment of the 50 % mix of forest ground + 50 % coffee husk took longer to emerge, on average 13.1 days. This species requires that the substrate is of medium porosity, moisture retention, without mechanical resistance to the emergence, in addition to the depth of the sowing must be twice the thickness of the seed. Orozco *et al.* (2011) point out that the soil that is used to fill the containers and seedbeds must fulfill several functions: letting in and retaining water; be rich in nutrients; soft so that the root can grow and the earth collected in the cacao plantation allowed the seeds of *Gmelina arborea* to have a higher emergence speed (tables 4 and 5).

Table 4. Average values in days to emergence, diameter, height, number of leaves and leaf area in the initial growth of *Gmelina arborea* Roxb. in different substrate mixes.

Treatment	DMER (days)	DIAM (mm)	ALTU (cm)	NHOJ	AFOL (cm ²)
1	13.11 a	7.78 bc	59.47 bc	38.36 a	1012.34 a
2	12.53 ab	5.08 e	39.76 e	28.76 b	774.47 b
3	11.75 b	7.48 cd	54.90 cd	35.36 a	801.33 b
4	12.20 ab	8.13 b	60.37 b	39.26 a	1090.07 ^a
5	12.16 ab	8.75 a	66.08 a	38.20 a	1097.42 a
6	11.71 b	7.08 e	53.56 d	38.53 a	722.10 b

$T_1 = 50\% \text{ forest ground} + 50\% \text{ coffee husk}$; $T_2 = 50\% \text{ forest ground} + 50\% \text{ sawdust}$; $T_3 = 50\% \text{ forest ground} + 50\% \text{ cacao husk}$; $T_4 = 50\% \text{ forest ground} + 50\% \text{ sugarcane bagasse}$; $T_5 = 50\% \text{ forest ground} + 50\% \text{ livestock manure}$; $T_6 = 100\% \text{ cocoa ground}$. t = Average values in the same column followed by the same letter are not statistically different from each other ($p < 0.05$); DMER = Days of emergence; DIAM = Diameter; ALTU = Height; NHOJ = Number of leaves; AFOL = Foliar area.



Table 5. Average values in total biomass, aerial part / root ratio, slenderness index, lignification index and Dickson quality index in the initial growth of *Gmelina arborea* Roxb. in different substrate mixes.

Treatment	BIOT	RPAR	IESB	ILIG	ICD
1	8.59 c	3.96 ab	7.69 a	21.97 bc	0.76 c
2	3.72 e	3.55 bc	7.93 a	20.69 c	0.33 e
3	7.72 cd	3.37 c	7.36 a	25.09 a	0.73 c
4	9.84 b	3.91 ab	7.45 a	23.71 ab	0.88 b
5	11.48 a	4.03 ab	7.58 a	24.64 a	1.00 a
6	6.83 d	4.14 a	7.62 a	25.35 a	0.61 d

$T_1 = 50\% \text{ forest ground} + 50\% \text{ coffee husk}$; $T_2 = 50\% \text{ forest ground} + 50\% \text{ sawdust}$;
 $T_3 = 50\% \text{ forest ground} + 50\% \text{ cacao husk}$; $T_4 = 50\% \text{ forest ground} + 50\% \text{ sugarcane bagasse}$; $T_5 = 50\% \text{ forest ground} + 50\% \text{ livestock manure}$; $T_6 = 100\% \text{ cocoa ground}$. t = Average values in the same column followed by the same letter are not statistically different from each other ($p < 0.05$); BIOT = Total biomass; RPAR = Aerial part /root ratio; IESB = Slenderness index; ILIG= Lignification index; ICD = Dickson's quality index.

Diameter of plants

After 75 days, the *Gmelina arborea* plants sown in the substrate mix of 50 % of forest ground + 50 % of livestock manure had the largest diameter to the base (8.76 mm) while the plants of smaller diameter were the produced in the mix of 50 % forest ground + 50 % sawdust, with average of 5.08 mm. This represents 42 % difference in diameter growth of the plants compared to the best substrate mix. Reyes *et al.* (2015) recorded growth averages in diameter of 4.40 mm in *Gmelina arborea* seedlings produced in the base mix containing 60 % Peat moss® + 30 % agrolite + 10 % vermiculite. It is relevant as plants with diameters greater than 5 mm are more resistant to bending and tolerate damage by pests and noxious fauna, although this

varies according to the species (Prieto *et al.*, 2009); however, larger plants have greater survival in all environments (Rodríguez, 2008). The plants produced in this experiment reached an average diameter of 8.76 mm, so that they satisfy the required diameter measurements to be considered as quality plants and to be taken to the final site of the plantation.

Height of plants

The plants had the highest height (66.08 cm) when they grew in a mix containing 50 % forest soil + 50 % livestock manure. The lowest growth was in seedlings that had as substrate 50 % forest land + 50% sawdust, with a height of 39.76 cm. The best substrate produced 40 % higher seedlings in the 75-day period. It should be mentioned that the height of the plant is used for its qualification, although the initial height does not correlate with survival; that is, by itself it is of limited value, but combined with other variables such as diameter, it has an important influence (Prieto and Sáenz, 2011).

In this context, Rodríguez (2008) states that it is more difficult to plant tall specimens which become more susceptible to mechanical damage, but that the small plant is more prone to be stepped on, to competition and other factors that affect its development; to avoid the above, it is preferable that the selected nursery plant has the height that allows it to compete and grow successfully in the field (Prieto *et al.*, 2003).

Number of leaves per plant and leaf area

The plants that grew in the substrate mix of 50 % forest ground+ 50 % sugarcane bagasse produced the highest number of leaves with an average of 39.27 with respect to the other treatments evaluated. The plants that formed the smallest number of leaves were those of the treatment of 50 % forest ground + 50 % sawdust, with an average of 28.77 leaves. In this regard, Rodríguez (2008) mentions that the morphological characteristics that denote low quality are shortage of foliage, symptoms of mechanical damage (breakage), twisting, yellowish foliage

and smallness, very large size and chlorosis or deformations related to nutritional deficiencies (Ramírez, 2017).

The plants that were produced with the mix of 50 % forest ground +50 % livestock manure had the highest values of leaf area with an average of 1 097.42 cm², with respect to the other assessed treatments. In the plants with the least foliar area, 100 % forest ground was used as a substrate, with an average of 722.10 cm². According to Shibles (1978), leaf area is the usual measure of the photosynthetic tissue of a community of plants; it is important because it determines the amount of solar energy that is absorbed and converted into organic materials.

Total biomass in plants

At 75 days after sowing, *Gmelina arborea* plants that had a mix of 50 % forest land + 50 % livestock manure recorded the highest root and stem dry weight, with an average of 4.14 g, compared to the other treatments evaluated. The plants that had the opposite response come from the substrate composed of 50 % of forest ground + 50 % sawdust, with an average of 3.72 g. Vera (1995) and Prieto *et al.* (2003) indicate that the biomass of the plants has a strong correlation with the survival in the field and suggest that for greater consistency in the results, the values of dry weight should be managed, because green weight has great variation of water in tissues within the same species, which was corroborated in the present work. Dry weight is also an effective indicator when the aerial part is related to the root system.



Plant quality indexes

For the variables considered as indicators of plant quality, the lowest average value of the aerial part / root ratio (3.37) comes from the seedlings that grew in the mix made up by 50 % forest ground + 50 % cocoa husk. The highest value of this relation (4.14) was registered in individuals with 100 % forest ground substrate. In this context, the results of the present work indicate that aerial dry weight is greater than root dry weight. In general, it is recommended that, in places with low rainfall, the quotient should not be greater than 2.5 (Prieto et al., 2003). Santiago et al. (2007) state that the lower the value obtained in the height / diameter relationships for tropical species, the greater the vigor of the plant. Those with a figure > 2.5 should be planted, preferably, in places where favorable environment or where watering can be applied during the establishment phase, which would be ideal for the *Gmelina arborea* seedlings produced in this experiment, since plantations are made during the rainy season in the region.

For the slenderness index, the lowest value appeared in the 50 % forest ground + 50 % cocoa husk mix (7.36). In this variable, the reduced values are important since the opposite indicates imbalance in the growth of the seedlings (Thompson, 1985). With this number and according to the classification of quality of plants of forest species by Prieto and Sáenz (2011), the results obtained in this study carried out would be classified as high quality. The plants that grew on the 50 % forest ground+ 50 % sawdust substrate, reached the highest value (7.94). It is worth mentioning that the optimal values vary according to each species and also depend on the place where it is planted (Cuevas, 1995).

Regarding the index of lignification, which is a parameter that in recent years is essential to assess the plant before it goes out to the field, the plants in which 100 % forest ground were cultivated had the highest values compared to the other treatments evaluated, with an average of 25.36 %. The plants with the lowest percentage were grown with the 50 % forest ground+ 50 % sawdust mix, and 20.70 % average. During the pre-conditioning phase, it is necessary to reduce the supply of water to cause plant stress, which helps to reduce growth in height, promotes the appearance of the apical bud and initiates

mechanisms of resistance to droughts and low temperatures (Prieto *et al.*, 2004), which favors hardening and lignification of the stem. Lignin is a fundamental component of the cell wall and intervenes in the process of linking plant cells (Toral, 1997).

The highest values of the Dickson quality index (1.00) were found in the plants whose substrate was the 50 % forest ground + 50 % livestock manure mix. The lowest (0.34) was recorded in the composite of 50 % forest ground + 50 % sawdust. If in the same lot the specimens record high numbers in this index, or are better balanced in the aerial part and the root, they represent higher quality individuals; in this case, treatment 5 produced this response. In *Gmelina arborea* seedlings produced in the base mix of 60 % Peat moss® + 30 % agrolite + 10 % vermiculite, Reyes *et al.* (2015) obtained an average of 0.67 in this criterion. According to the methodology of Rueda *et al.* (2010), the values calculated in this work indicate that the plants meet the quality standards to be planted in the field.

Conclusions

The forest ground with livestock manure mix favors the production of *Gmelina arborea* plants with morphological characteristics of ideal size in the traditional production system in a production cycle of 2.5 months that will allow high percentages of survival in the field. The environmental impact can be reduced by the extraction of forest ground by using other local substrates for the production of forest plants.



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Conflict of interest

The authors declare no conflict of interest.

Contribution by author

Jorge Reyes Reyes: design and general approach to the experiment; structure and writing of the manuscript; Dorian de Jesús Pimienta de la Torre: review and corrections of the document; Juan Alberto Rodríguez Morales: statistical analysis of the data and review of the document; Mario Alonso Fuentes Pérez: review and corrections of the document; Emilio Palomeque Figueroa: training of the experiment at the nursery and data collection.