



**Full Length Article**

## Early Growth Comparison of the Fast-Growing Tree Species in Mixed-Species Plantation Planted with Agroforestry and Biofertilization

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### Abstract

This field study was conducted to evaluate the growth of mixed-tree species plantation (*Neolamareckia macrophylla*, *N. cadamba* and *Nauclea orientalis*) grown with different competitive ratio and land equivalent ratio (LER) under different agroforestry and biofertilization at Parungpanjang, Bogor, West Java and Indonesia. The establishment of the mixed-tree species plantation used a randomized factorial design with two factors, *i.e.*, tree species and agroforestry with biofertilization planted in 4 blocks. The results showed the agroforestry using arbuscular mycorrhizal fungi (AMF) and compost fertilization resulted the best tree growth of all tested three species. However, the *N. cadamba* observed the highest plant height, diameter, volume and crown width followed by *N. orientalis* and *N. macrophylla*. The height of *N. cadamba* at the age of 3 years reached 10.87 m (84% higher than the control) with the diameter of 13.43 cm (45% higher than the control), followed by *N. orientalis* with the height of 9.27 m (69% higher than the control) with the diameter of 12.71 cm (67% higher than the control) and *N. macrophylla* with the height of 8.15 m (103% higher than the control) with the diameter of 10.31 cm (100% higher than control). The competitive ability of *N. cadamba* was higher than *N. orientalis* and *N. macrophylla*. The sum of the partial LER of mixed-tree species plantation and partial LER of *A. galanga*, had a value of >1 in all agroforestry and biofertilization. This LER value showed that the mixed-tree species plantations with agroforestry seemed a viable option to get higher profit. © 2024 Friends Science Publishers

**Keywords:** Agroforestry; Arbuscular mycorrhizal fungi; Inoculation; Competitive ratio; Tree growth

### Introduction

Balancing demand for forest products and managing sustainable forest ecosystems is one of the main problems currently faced by the forest sector worldwide. Efficient plantation forest management and intensive silvicultural practices are urgently needed to increase the success and productivity of plantation forests (Alan 2020). In addition, forests must also have ecological and social functions that allow them to go hand in hand. The development of a mixed-tree species plantation with an agroforestry pattern makes it possible to create tree stands with high diversity and productivity and social functions for communities around the forest area. Several studies showed the advantages of mixed-tree species plantation compared to pure (monoculture) stands in terms of biomass production (Marron and Epron 2019), soil fertility (Danise *et al.* 2020), nutrient cycling (Pardos *et al.* 2020) and carbon

sequestration (Molina-Valero *et al.* 2021). Mixed-tree species plantations are thus considered to be better than monocultures for maintaining timber production while improving soil quality (Danise *et al.* 2021). Forests that are developed by involving high genetic variation both within and among species can improve the stability of forest ecosystems and increase their potential adaptation to global climate change (Hamrick 2004).

Establishment of a mixed-tree species plantation is certainly more difficult than monoculture, especially in site matching, species selection, and inter-specific competition (Manson *et al.* 2013; Trogisch *et al.* 2021; Vanclay *et al.* 2022) with management which are more complex (Dickinson *et al.* 2008), so that operationally in the development of industrial plantation forests it is still rarely carried out (Nichols *et al.* 2006; Vanclay *et al.* 2022). Some experts recommend mixed-tree species plantations to be applied in the development of community forestry

(Chechina and Hamann 2015) and forest and landscape restoration (Temperton *et al.* 2019). In this study, three fast growing species from the Rubiaceae family were selected, *i.e.*, *Neolamarckia cadamba* (Roxb.) Bosser, *Neolamarckia macrophylla* (Roxb.) Bosser and *Nauclea orientalis* L. These three species are native Indonesian species which are widely planted both on a large scale (plantation) and on a small scale in the form of community forests (Sarjono *et al.* 2017; Riany *et al.* 2018; Yuniarti *et al.* 2023) and is suitable for development with agroforestry patterns (Indrajaya and Siarudin 2015; Saravanan 2019). However, mixed plantation forests with more intensive silviculture between these three species have never been studied, including their inter-specific competition.

In addition, intensive silviculture has been applied to the establishment of industrial plantation forests through soil preparation, use of high-quality seedlings, fertilization, thinning, and pest and disease management (Alan 2020). Most of the fertilizers used are chemical fertilizers. The application of biofertilizers such as mycorrhiza on an operational scale for nurseries and silviculture for forest tree species in Indonesia is still limited. In fact, mycorrhizae such as arbuscular mycorrhizal fungi (AMF) have the ability to increase nutrient uptake, produce phytohormones, and resistance to abiotic stresses, such as drought, extreme temperatures, metals and salinity (Boyer *et al.* 2014; Amiri *et al.* 2017; Ahammed and Hajiboland 2024). AMF inoculation on the tree seedlings in the nursery can improve seedling vigor so that they are able to adapt when planted in the field (Navarro-Garcia *et al.* 2011; Kumar *et al.* 2017).

Thus, the development of mixed-tree species plantations with agroforestry and biofertilization treatments is expected to provide more effective land use values. One measure to assess the land use value is the land equivalent ratio (LER) which is an important tool used for evaluating agroforestry or polyculture systems (Dariush *et al.* 2006; Mohammed 2012). LER is defined as the relative area of land required by a single crop (monoculture) to produce the same yield when planted with an agroforestry or polyculture system (Mohammed 2012). The hypothesis in the research seeks to ensure that mixed-tree species plantations with agroforestry using biofertilizer can be implemented and is profitable, so the purpose of this study was to: (1) examine the growth of mixed-tree species plantation (*N. cadamba*, *N. macrophylla* and *N. orientalis*), (2) assess the competitive ratio of the plantation and (3) calculate the LER in different agroforestry with biofertilization (AMF and compost) at Parungpanjang, Bogor, West Java and Indonesia.

## Materials and Methods

### Materials

The study was carried out at the mixed-tree species plantation stand at Parungpanjang, Bogor, West Java, Indonesia. The stand was established using three species

from Rubiaceae family, *i.e.*, *N. macrophylla*, *N. cadamba* and *N. orientalis*. The seeds were collected from Luwu Timur-South Sulawesi (3°15'22" S, 120°12'48" E) for *N. macrophylla*, Pomalaa-Southeast Sulawesi (4°00'94" S, 121°30'09" E) for *N. cadamba*, and Parungpanjang-Bogor (6°23'09" S, 106°31'14" E) for *N. orientalis*. The seeds were processed at the Seed Testing Laboratory, Center for Standard and Instrument Implementation of Forestry and Environment, Bogor.

### AMF preparation

The AMF strains were explored from nature forest in PT. Tanjung Redeb Hutani, East Kalimantan (02°10'20" N, 117°28'15" E) and isolated in Laboratory of PT. Inagro, Bogor, West Java (<https://inagro.co.id>). AMF spores were extracted from 100 g of rhizosphere soil of trees with the best and healthy phenotypes using the wet filter pour and centrifugation method (Brundrett *et al.* 2008). The number of each type of spore that was successfully extracted from each type of host was calculated. The spores chosen for identification are spores with complete structures. Identification was carried out based on the identification key of Schenck and Perez (1990) in genus level. Making a single spore culture refers to the method used by Mansur (2000), *i.e.*, the Petridish Observation Chamber (PDOC) with modifications. Culture propagation is carried out by planting a propagation medium (zeolite) with a cover crop (*Pueraria javanica*) so that sufficient pure culture is obtained to test the effectiveness of several types of desired plants. If the results obtained show a positive trend, the isolate will be multiplied for production. The AMF used in this study was a mixture of several AMF strains (*Glomus* sp.-1, *Glomus* sp.-2, *Acaulospora* sp. and *Gigaspora* sp. number of spores 626 per gram).

### Seedling preparation and planting design

Seedlings were raised in the nursery at Nagrak Research Station, Bogor (06°6'74" S, 106°51'27" E). Seeds were sown in mixed media of compost, sand, and charcoal (3: 5: 1, by volume). After two months of age, seedlings with a pair of leaves that grow normally are transferred to polybags with mixed media of topsoil, rice husks, and compost (3:2:1, by volume) (Sudrajat *et al.* 2016; Sudrajat *et al.* 2019). AMF inoculation was carried out when transplanting seedlings from the germination box into polybags containing the media by adding 5 g of AMF to each seedling. Seedlings are maintained in the nursery for 4 months and are ready to be planted in the field test. The results of the analysis of the percentage of inoculation on the seedlings showed that the inoculation level on *N. macrophylla* seedlings ranged from 36.7–43.3%, *N. cadamba* ranged from 40.0–50.0% and *N. orientalis* ranged from 46.7–63.3%.

Planting test was carried out at the Parungpanjang, Bogor, West Java and Indonesia (06°20'42" S, 106°06'15"

E, altitude 52 m asl.). Soil was categorized as podzolic haplic with low pH and low nutrient content (C-Organic = 1.20–2.31%, N-total = 0.22–0.24%, P = 0.88–13.75 ppm, K = 0.10–0.15 cmol<sup>(+)</sup> kg<sup>-1</sup>, Ca = 2.30–4.98 cmol<sup>(+)</sup> kg<sup>-1</sup>, Mg = 1.80–3.47 cmol<sup>(+)</sup> kg<sup>-1</sup>) and pH 4.46–4.61 (Anna *et al.* 2020). Planting of mixed-tree species (*N. macrophylla*, *N. cadamba* and *N. orientalis*) was conducted by spacing of 4 m x 2 m at the planting hole size of 30 cm x 30 cm x 30 cm. The tree planting path for each species runs from east to west (Fig. 1). The design used to develop mixed-tree species plantation was a factorial randomized design with two treatment factors, *i.e.*, tree species (three species) and agroforestry with biofertilization planted in 4 blocks (replications). The treatments were: (1) control (no agroforestry and no biofertilization), (2) agroforestry, (3) agroforestry using the inoculated AMF inoculated seedling, (4) agroforestry pattern with 3 kg compost fertilization, and (5) agroforestry using the AMF inoculated seedling and 3 kg compost fertilization.

The compost (manure and rice husk compost) has the following characteristics and chemical content: pH 8.09, C organic 22.49%, N 0.84%, P 0.26 ppm, K 0.53 cmol(+) kg<sup>-1</sup> and C/N ratio 26.77. Compost fertilization was carried out 3 times, *i.e.*, at the time of out planting as a basic fertilizer (3 kg), at the age of the trees 12 months after out-planting (3 kg), and 24 months after out-planting (3 kg). The second and third fertilization has been done by loosening the soil around the tree with a diameter of 80–100 cm and adding compost to the area. The annual crop chosen in this agroforestry pattern was *Alpinia galanga* with a spacing of 100 cm x 50 cm (Fig. 1). Planting of *A. galanga* was carried out after the tree species. The planting of the three tree and crop species was also carried out using a monoculture pattern as a comparison for calculating the competition ratio and land equivalent ratio.

### Measurement of growth and competitive ratio

The growth was measured on the tree height and diameter carried out at the age of trees 1, 2 and 3 years. Height measurement was carried out using a scaled pole, while diameter measurement (diameter at breast height) was carried out using a diameter measuring tape. In addition, measurements of crown width and tree volume were also carried out. Crown width was measured by considering the widest and narrowest crowns and averaged to obtain the crown width. Tree volume ( $V_t$ ) is only measured at the age of 3 years using the following formula (Saputra *et al.* 2019):

$$V_t = 0.25 \pi (D)^2 H f \quad (1)$$

Where:  $V_t$  = tree volume,  $\pi$  = 3.14;  $D$  = diameter at breast height (m);  $H$  = total height (m);  $f$  = form factor (0.7).

Competitive ratio between tree species in mix-species plantations based on tree volume at 3 years of age. Competitive ratio calculation is done by comparing two tree species and each tree species is compared with

a different tree species so that 6 pairs are formed in comparison to the competitive ratio of the three tree species. The formula used in calculating the competitive ratio adopts Ceunfin *et al.* (2017):

$$CR = \frac{Y_a/Y_{aa}}{Y_b/Y_{bb}} \frac{Z_b}{Z_a} \quad (2)$$

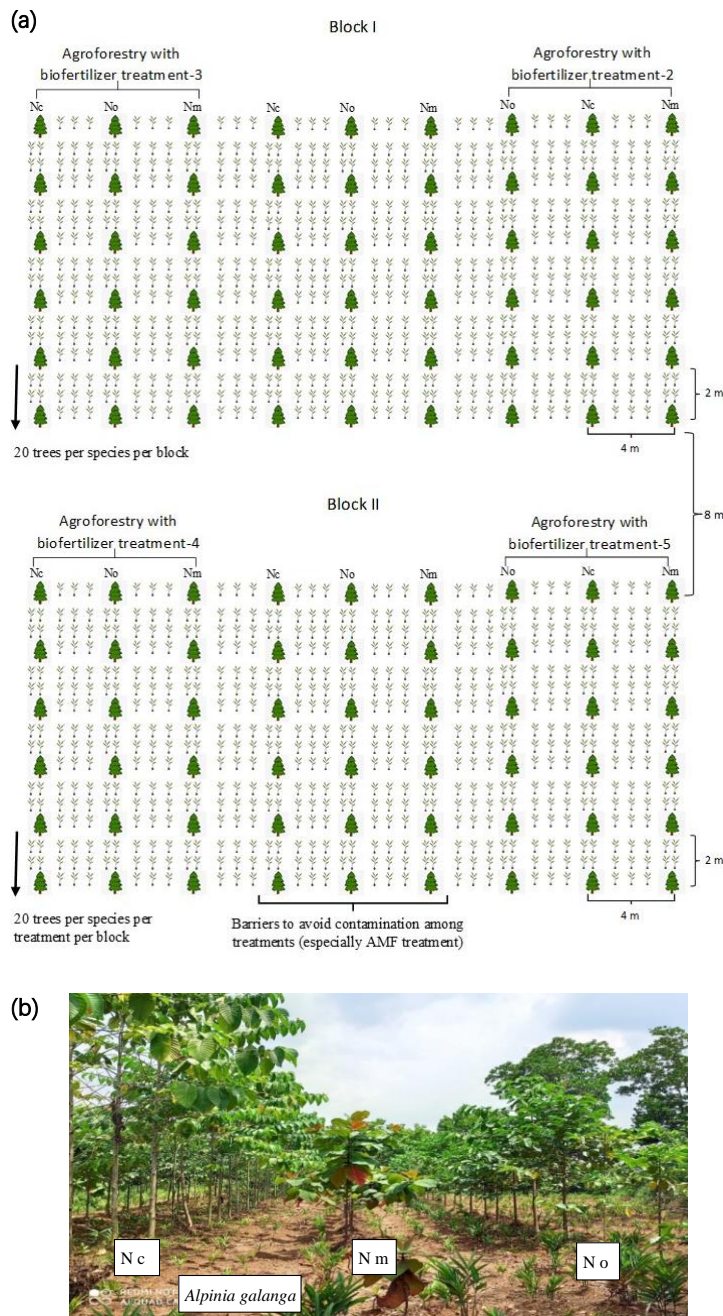
Dimana:  $Y_a$  = tree volume of species a in the mix-species plantation,  $Y_b$  = tree volume of species b in the mix-species plantation,  $Y_{aa}$  = tree volume of species a in monoculture plantation,  $Y_{bb}$  = tree volume of species b in monoculture plantation,  $Z_b$  = proportion of species b in the mix-species plantation,  $Z_a$  = proportion of species a in the mix-species plantation.

### Calculation of LER

Measurement of agroforestry land productivity between mixed tree (*N. macrophylla*, *N. cadamba* and *N. orientalis*) stands and crop species (*A. galanga*) was carried out by calculating tree volume and crop production data in planting test demonstration plots and compared with tree volume and crop production data planted in monoculture. Measurement of *A. galanga* production was carried out by weighing the weight of fresh tubers in each year of production with a total of 25 plants taken at random which were converted into production per unit of land (ton ha<sup>-1</sup>). Determination of tuber production per unit of land (hectare) was done by calculating the average production per clump multiplied by the number of *A. galanga* plant populations for each hectare with a spacing of 100 cm x 50 cm. Meanwhile, the measurement of the productivity of tree stands was carried out for all plants (census) in the planting test area. Timber production per unit of land (hectare) was determined by calculating the average volume per tree multiplied by the number of trees per hectare and the percentage of survival in each year of observation. The increase in timber production from tree species was the difference in the volume of wood per hectare measured at the beginning of the year and the end of the year (annual volume growth). The LER calculation uses a formula adopted from Dariush *et al.* (2006) and Figyantika *et al.* (2020) as follows:

$$LER = \frac{Pa_n}{Ma_n} + \left( \frac{Pb_n - Pb_{(n-1)}}{Mb_n - Mb_{(n-1)}} + \frac{Pc_n - Pc_{(n-1)}}{Mc_n - Mc_{(n-1)}} + \frac{Pd_n - Pd_{(n-1)}}{Md_n - Md_{(n-1)}} \right) \quad (3)$$

Notes:  $Pa$  = yield of galangal (*A. galanga*) tubers cultivated by agroforestry pattern,  $Pb$  = production (tree volume) of *N. macrophylla* grown by polyculture,  $Pc$  = production (tree volume) of *N. cadamba* grown by polyculture,  $Pd$  = production (tree volume) of *N. orientalis* grown by polyculture,  $Ma$  = yield of galangal (*A. galanga*) tubers cultivated by monoculture,  $Mb$  = production (tree volume) of *N. macrophylla* grown by monoculture,  $Mc$  = production (tree volume) of *N. cadamba* grown by monoculture,  $Md$  = production (tree volume) of *N. orientalis* grown by monoculture,  $n$  = plant or tree age.



**Fig. 1:** Planting design of mixed-tree species plantation with agroforestry *Alpinia galanga* and biofertilizer treatments (a), and tree growth performance at 1 year old age (b). Notes: N c = *Neomarcia cadamba*, N o = *Nauclea orientalis*, N m = *Neolamarckia macrophylla*

### Data analysis

Data were analyzed using analysis of variance to examine the effect of different treatments (tree species and agroforestry with biofertilization) on the growth of tree height, diameter and volume. Duncan's multiple range test was carried out if the results of the analysis of variance of a treatment or its interaction have a significant effect on an observed parameter. Competitive ratio and land equivalent

ratio data are tabulated and described to obtain an overview of the treatment with the highest ratio.

### Results

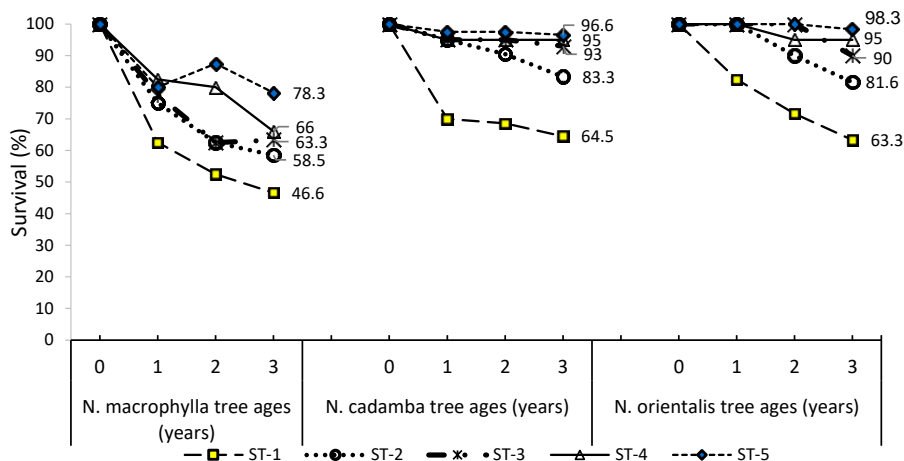
#### Growth performance

Species, agroforestry with biofertilization, and their interactions had a significant effect on tree survival up to 3

**Table 1:** Recapitulation of analysis of variance of influences of species and silvicultural treatment on survival, height, diameter at breast height, volume, and crown width of *N. macrophylla*, *N. cadamba* and *N. orientalis* at 3 years of age

Sources of variation	DF	Survival (%)	Height (m)	Diameter (cm)	Volume (m <sup>3</sup> )	Crown width (m)
Block (B)	2	-	2.38 <sup>ns</sup>	2.77 <sup>ns</sup>	1.49 <sup>ns</sup>	19.31**
Species (S)	2	65.51**	135.86**	110.03**	86.67**	70.68**
Block (B)*Species (S)	4	-	0.84 <sup>ns</sup>	1.68 <sup>ns</sup>	0.89 <sup>ns</sup>	8.75**
Silviculture treatment (ST)	4	30.20**	204.46**	89.16**	68.44**	275.89**
B*ST	8	-	1.24 <sup>ns</sup>	0.15 <sup>ns</sup>	0.12 <sup>ns</sup>	5.79**
S*ST	8	2.71 *	8.86**	6.44**	5.22**	29.83**
B*S*ST	16	-	0.60 <sup>ns</sup>	0.76 <sup>ns</sup>	0.44 <sup>ns</sup>	4.07**

Notes: \*\* = significant at 99% confidence level, \* = significant at 95% confidence level, ns = not significant at 95% confidence level



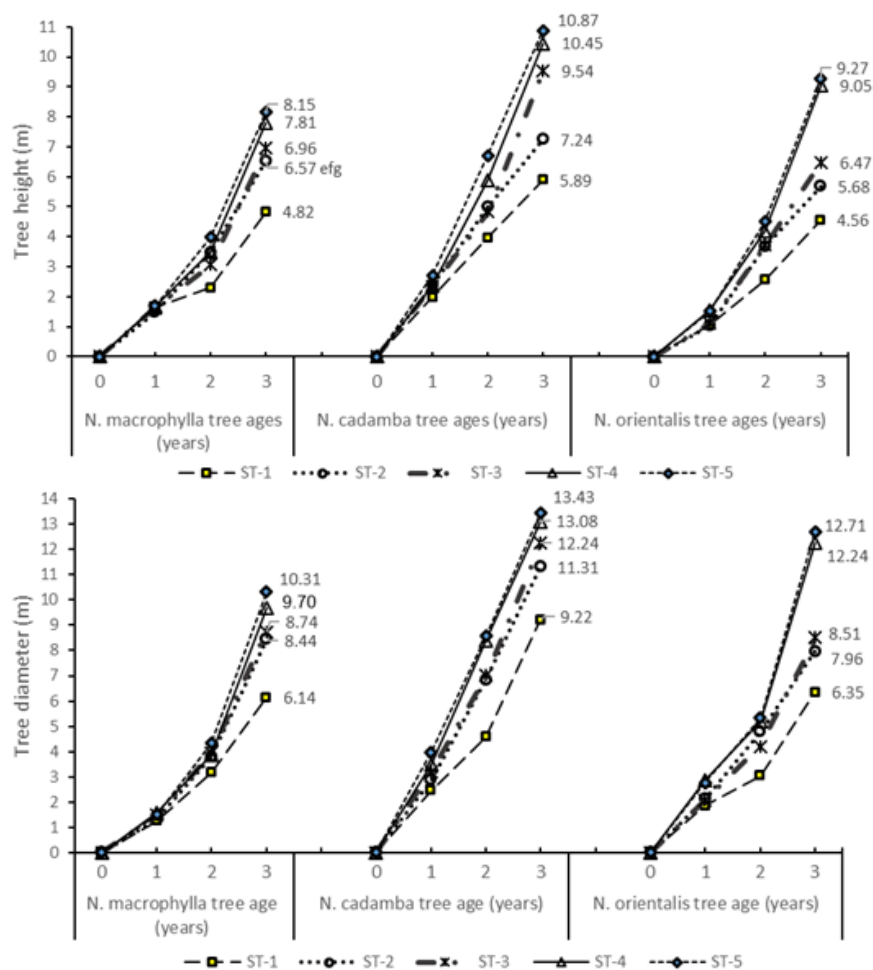
**Fig. 2:** Tree survival of *N. cadamba*, *N. macrophylla* and *N. orientalis* in the agroforestry using AMF inoculated seedling and compost fertilization treatment. Notes: ST-1 = non agroforestry, ST-2 = agroforestry, ST-3 = agroforestry using AMF inoculated seedling, ST-4 = agroforestry with 3 kg of compost per year, ST-5 = agroforestry using AMF inoculated seedling and fertilization of 3 kg of compost per year

years old. The interaction between species and agroforestry with biofertilization had a significant effect on tree height, diameter, volume, and crown width. Most of the block effects had no significant effect on the growth of the three tree species tested, except for the crown width parameter (Table 1). This shows that the planting test blocks are relatively uniform.

The agroforestry and AMF inoculated seedling and 3 kg compost per year until 2 years of tree age gave the highest survival rate for the three tree species tested (Fig. 2). In general, *N. orientalis* had the highest survival. In the best treatment, agroforestry and AMF inoculated seedling and fertilization with 3 kg compost per year, *N. orientalis* had the highest survival (98.3%), followed by *N. cadamba* (96.6%), and *N. macrophylla* had the lowest survival (78.3%). In areas that did not apply an agroforestry system, the survival of *N. macrophylla* also gave the lowest value (46.6%) compared to the other two species. The highest mortality rate occurred in the first year or during the adaptation phase of the seedlings. The treatment of agroforestry and biofertilization with AMF inoculated seedling and 3 kg of compost per year also gave the best height and diameter growth for all species. *N. cadamba* had

higher growth in height and diameter compared to other species, while *N. macrophylla* has the lowest growth. The *N. cadamba* height of a 3-year-old planted with an agroforestry using AMF inoculated seedling and 3 kg of compost per year reached the tree height of 10.87 m (84% higher than control) with the tree diameter of 13.43 cm (45% higher than control), followed by *N. orientalis* with the height of 9.27 m (69% higher than control) with the tree diameter of 12.71 cm (67% higher than control), and lastly *N. macrophylla* with the tree height of 8.15 m (103% higher than control) with the diameter of 10.31 cm (100% higher than control) (Fig. 3).

The trend of tree volume at 3 years of age followed the growth trend in height and diameter with the treatment that produced the highest tree volume obtained from the agroforestry pattern using AMF inoculated seedling and 3 kg compost per year. In this treatment, *N. cadamba* produced a tree volume of 0.1126 m<sup>3</sup>, followed by *N. orientalis* with a tree volume of 0.0881 m<sup>3</sup>, and finally *N. macrophylla* with a tree volume of 0.0501 m<sup>3</sup>. Statistically, for several parameters such as tree height, diameter, and volume of *N. cadamba* and *N. orientalis*, the treatment of agroforestry using AMF inoculated seedling and 3 kg



**Fig. 3:** Tree height and diameter at breast height of *N. cadamba*, *N. macrophylla* and *N. orientalis* on the treatment of agroforestry using AMF inoculated seedling and compost fertilization treatment. Notes: ST-1 = non agroforestry, ST-2 = agroforestry, ST-3 = agroforestry using AMF inoculated seedling, ST-4 = agroforestry with fertilization of compost 3 kg per year, ST-5 = agroforestry using AMF inoculated seedling and compost fertilization

compost per year using the agroforestry pattern was not significantly different from the treatment of agroforestry with fertilization of 3 kg compost per tree per year. For crown width, the best treatment for each species was obtained by agroforestry pattern using AMF inoculated seedling and 3 kg of compost per year using an agroforestry pattern. In this treatment, the crown width of *N. cadamba*, *N. macrophylla* and *N. orientalis* were 10.52 m, 7.98 m and 6.89 m, respectively (Table 2). Visually, the crowns of *N. orientalis* tend to be heavier and denser.

In mix-tree species plantations, there is inter-specific competition which is planted in intermittent row patterns. Based on the competitive ratio, *N. cadamba* has a better competition value than *N. macrophylla*. The same thing was shown by *N. orientalis* which had a higher competition ratio than *N. macrophylla*. When comparing *N. cadamba* and *N. orientalis*, *N. cadamba* has competitive advantages in control, agroforestry, and agroforestry using AMF

inoculated seedling. In more intensive conditions, the treatment of agroforestry with the fertilization of compost 3 kg, and agroforestry using AMF inoculated seedling and fertilization of compost 3 kg, *N. orientalis* has a higher competitive ratio than *N. cadamba* (Table 3).

### Land equivalent ratio (LER)

Total LER, which is the sum of the partial LER of mix-tree species plantation and partial LER of *A. galanga*, had a value of >1 in all agroforestry and biofertilization (Table 4). The range of LER values for trees aged 1, 2, and 3 years were 1.48–2.68, 1.92–2.98 and 1.87–4.41, respectively. At the age of 1 year, the partial LER of mix-tree species had a value of >1 in the compost fertilization (1.34) and the AMF inoculated seedling and compost fertilization treatment (1.82), while at the age of 2 and 3 years, the partial LER of mix-tree species plantation had LER > 1 in all agroforestry

**Table 2:** Tree volume and crown width of *N. cadamba*, *N. macrophylla* and *N. orientalis* on the agroforestry using AMF inoculated seedling and compost fertilization treatment at Parungpanjang, Bogor

Species	Agroforestry and biofertilization treatments	Tree volume (m <sup>3</sup> )	Crown width (m)
<i>N. macrophylla</i>	ST-1	0.0115 g	3.20 g
	ST-2	0.0230 fg	4.22 ef
	ST-3	0.0337 def	4.98 de
	ST-4	0.0473 cde	4.37 e
	ST-5	0.0501 cd	7.98 b
<i>N. cadamba</i>	ST-1	0.0313 defg	4.38 e
	ST-2	0.0611 c	4.83 de
	ST-3	0.0858 b	6.61 c
	ST-4	0.1108 a	8.44 b
	ST-5	0.1126 a	10.52 a
<i>N. orientalis</i>	ST-1	0.0113 g	3.57 fg
	ST-2	0.0215 fg	6.35 c
	ST-3	0.0219 efg	5.16 d
	ST-4	0.0820 b	5.34 d
	ST-5	0.0881 b	6.89 c

Notes: ST-1 = non agroforestry, ST-2 = agroforestry, ST-3 = Agroforestry using AMF inoculated seedling, ST-4 = agroforestry with fertilization of compost 3 kg per year, ST-5 = agroforestry using AMF inoculated seedling and compost fertilization treatment

**Table 3:** Competitive ratio between *N. cadamba*, *N. macrophylla* and *N. orientalis* on the mix-tree species plantation at the 3 years old age at Parungpanjang, Bogor

Treatments	Nm to Nc	Nm to No	Nc to Nm	Nc to No	No to Nm	No to Nc
Control	0.57	0.71	1.75	1.24	1.41	0.81
Agroforestry	0.82	1.00	1.22	1.22	1.00	0.82
Agroforestry using AMF inoculated seedling	0.53	0.76	1.87	1.43	1.31	0.70
Agroforestry with compost fertilization treatment	0.65	0.51	1.53	0.78	1.97	1.29
Agroforestry using AMF inoculated seedling and compost fertilization treatment	0.83	0.64	1.20	0.77	1.56	1.30

Notes: Nm = *Neolamarckia macrophylla*, Nc = *Neolamarckia cadamba*, No = *Nauclea orientalis*

**Table 4:** Land equivalent ration (LER) of mixed-tree species plantation with agroforestry using *Alpinia galanga* at the tree age of 1, 2 and 3 years

Tree age	Treatments	Partial LER of mix-tree species plantation	Partial LER of <i>Alpinia galanga</i>	LER total
1 year	Agroforestry	0.62	0.86	1.48
	Agroforestry using AMF inoculated seedling	0.72		1.58
	Agroforestry with compost fertilization treatment	1.34		2.20
	Agroforestry using AMF inoculated seedling and compost fertilization treatment	1.82		2.68
2 years	Agroforestry	1.09	0.75	1.92
	Agroforestry using AMF inoculated seedling	1.03		1.85
	Agroforestry with compost fertilization treatment	1.66		2.53
	Agroforestry using AMF inoculated seedling and compost fertilization treatment	2.10		2.98
3 years	Agroforestry	1.29	0.59	1.87
	Agroforestry using AMF inoculated seedling	2.15		2.72
	Agroforestry with compost fertilization treatment	3.35		3.90
	Agroforestry using AMF inoculated seedling and compost fertilization treatment	3.87		4.41

and biofertilization, with a range of 1.03–3.87. The partial LER of *A. galanga* tends to decrease with increasing tree age. The highest total LER value was obtained by treatment with AMF inoculated seedling and compost fertilization (3 kg per year) with LER values in years 1, 2 and 3 were 2.68, 2.98 and 4.41, respectively.

## Discussion

Growth performance (tree survival, height, diameter, stem volume, and crown width) of the three species was strongly influenced by agroforestry and biofertilization treatments. The application of agroforestry and biofertilization can

improve tree survival and growth. The growth of the three species, i.e., *N. macrophylla*, *N. cadamba* and *N. orientalis*, increased with increasing biofertilization. The agroforestry using AMF inoculated seedling and 3 kg compost fertilization gave the highest of tree survival, height, diameter and the biggest of tree volume and crown width. In several previous studies, the application of agroforestry was able to improve tree growth, as reported by Supriyono and Setyaningsih (2012) on *T. grandis*, Lucena et al. (2023) on *Khaya ivorensis* and Souza et al. (2023) on *Bertholletia excelsa*. Studies on the effectiveness of AMF inoculation on increasing growth and adaptation of seedlings have been reported in several plant species,

especially agricultural plants. AMF can increase nutrient and water absorption, as well as produce phytohormones, such as auxin, which can stimulate plant growth and development (Ahammed and Hajiboland 2024). AMF can assist host plants in improving tolerance mechanisms and preventing downregulation of key metabolic systems (Begum *et al.* 2019). In this study, the use of a mixture of AMF strains was quite suitable and effective in associating with the root systems of the three species tested, as indicated by the high average percentage of colonization, *i.e.*, *N. macrophylla* 41.1%, *N. orientalis* 54.4% and *N. cadamba* 44.4%. O'Connor *et al.* (2001) stated that the colonization percentage reaches a high level if damage is > 30%. Likewise, the adaptability (survival) and growth of the three trees showed that the trees originating from AMF inoculated seedlings were relatively better than the control. The use of mixed AMF species has also been reported by Jansa *et al.* (2008); Wehner *et al.* (2010) and Shukla *et al.* (2014) is more effective compared to applications using individual AMF species.

Some study also reported the positive effect of fertilization on the tree growth (Subedi *et al.* 2015; Tumushime *et al.* 2019; Anwar *et al.* 2020). Jaquetti and Gonçalves (2021) stated that fertilization is fundamental during the early growth (stage) of forest plantation development. According to Lucena *et al.* (2023), a combination of agroforestry systems with silvicultural interventions (biofertilizer treatment) can increase the ecological and economic values of the tree plantation.

In this research, a mix-tree species plantation with three fast growing tree species from the Rubiaceae family had different adaptations and growth. In the best treatment (agroforestry using AMF inoculated seedling and 3 kg compost fertilization), *N. orientalis* had the highest tree survival, followed by *N. cadamba* and *N. macrophylla* with the lowest tree survival. For its growth, *N. cadamba* had the highest height, diameter, stem volume, and crown width. When viewed from its natural distribution pattern, *N. orientalis* has a wide natural distribution (Riany *et al.* 2018), as well as *N. cadamba* which has a wide distribution and is found throughout the major islands of Indonesia (Sudrajat 2016). The broad description of natural seismicity is thought to be related to its adaptability. In contrast to *N. macrophylla* which is only found in eastern Indonesia, *i.e.*, Sulawesi, the Maluku Islands and Papua (Halawane *et al.* 2011; Yuniarti *et al.* 2023), it is thought that its adaptability is relatively lower. In addition, the diversity of *N. cadamba* is thought to be wider than that of *N. macrophylla*. Several studies showed that *N. macrophylla* had low to moderate genetic diversity, with values of  $He = 0.05-0.42$  and  $Gst = 0.083$  (Andriani 2017; Larekeng *et al.* 2018; Arif *et al.* 2019), while *N. cadamba* showed relatively wider genetic diversity, with  $He = 0.2756$  (0.1489–0.3339) and  $Gst = 0.2707$  (Sudrajat *et al.* 2015). The wider natural distribution and genetic diversity is thought to be related to the

adaptability of each species, especially on marginal lands such as in this research location (Parungpanjang, Bogor). This adaptability also affected its performance in the form of mixed-tree species plantations.

Mixed-tree species plantations planted using an agroforestry pattern have several advantages such as providing a variety of products (biodiversity, timber, and non-timber) compared to pure-species plantations (Montagnini *et al.* 2003), being more adaptive to local site conditions and being more beneficial for ecological balance, and can mitigate the risk of natural hazards (Griess and Knoke 2011), has higher market and social value for local communities (Ball *et al.* 1995). However, on an operational scale for commercial or industry, mixed-tree species plantations are still rarely practiced because management is more difficult (Nichols *et al.* 2006), potential production benefits are not always realized (Kelty 2006), and little is known about interactions and competition between species, especially for tropical native tree species.

In this study, the competition ratio (CR) for each species has different values. CR is an estimate of the relative competitive ability of a tree species against other tree species. CR is related to the ability of each species to obtain resources both vertically and horizontally (Paulus 2005; Ceunfin *et al.* 2017). In this study, the CR of *N. cadamba* against *N. macrophylla* had a higher value compared to the opposite CR value (*N. macrophylla* against *N. cadamba*). This indicated that the competitiveness of *N. cadamba* was higher than that of *N. macrophylla*. Likewise with *N. orientalis* against *N. macrophylla* which has a high CR. *N. cadamba* also had a higher CR than *N. macrophylla* in the control treatment, agroforestry patterns, and agroforestry patterns with NPK fertilization, so that the highest adaptability was *N. cadamba*, *N. orientalis*, and *N. macrophylla*, respectively. However, in the treatment of the agroforestry with compost fertilization and the agroforestry using AMF inoculated seedling and compost fertilization, the CR values for *N. orientalis* showed higher results, so that the order of competence in these conditions was *N. orientalis*, *N. cadamba* and *N. macrophylla*.

To compare the effectiveness of land use in an agroforestry (mixed cropping pattern) with a monoculture pattern, the LER was calculated based on the productivity of each commodity species (Dariush *et al.* 2006; Figyantika *et al.* 2020). The LER value is equal to 1 (one) indicating that there is no difference in results between the intercropping or polyculture system and the monoculture system. Meanwhile, every LER more than 1 (one) will show the benefits of an intercropping/polyculture management system and every LER below 1 (one) shows the benefits of monoculture system management (Lehmann *et al.* 2020). The interaction between plants planted using an agroforestry pattern on a land has a positive impact on each of its components, and this will affect plant productivity. Productivity measurement for mix-tree species plantations was based on growth shown in the tree stand volume value, while for crop species it was



calculated based on the production of tubers. The total LER values for mixed-tree species plantation aged 1, 2 and 3 years showed a range of 1.48–2.68, 1.92–2.98, and 1.87–4.41, respectively, with the highest total LER values produced by the agroforestry using AMF inoculated seedling and 3 kg compost fertilization. The data showed that a mixed crop pattern combined with crop species provides higher productivity than if planted in monoculture. Similar research result was reported by Narendra and Nandini (2013), who stated that the productivity of a species will increase if it is planted in polyculture or agroforestry with fertilizer treatment, such as teak (*T. grandis*) which is planted in agroforestry with corn and peanuts. In this study, LER partial mix-tree plantation species tended to increase with fertilizer treatment and increasing tree age. Studies on other species planted using agroforestry show that the combination of orange, patchouli and papaya in an agroforestry pattern has a total LER value of 1.783 (Setiawan and Himawan 2018). This illustrates that the application of agroforestry provides better growth in good growth, especially for woody plants or trees and provides higher land use efficiency.

*A. galanga* cultivation planted in stands of mixed-tree species from the Rubiaceae family (*N. cadamba*, *N. macrophylla* and *N. orientalis*) showed a decrease in LER from 1 year to 3 years of age. The partial LER values of *A. galanga* at 1, 2, and 3 years of age were 0.86, 0.75, and 0.59, respectively. This value shows that the productivity of *A. galanga* tubers decreased after the main plants (*N. cadamba*, *N. macrophylla* and *N. orientalis*) increased in size. The decrease in productivity of *A. galanga* tubers was not optimal because the shade from the tree crown or canopy widens with age. The range of crown widths for the three tree species in all treatments at 3 years of age was 3.20–7.98 m (*N. macrophylla*), 4.38–10.52 m (*N. cadamba*), and 3.57–6.89 m (*N. orientalis*). The wide canopy causes obstruction of sunlight received by *A. galanga*, so that the production of *A. galanga* tubers was less than optimal. Similar results were reported by Sudrajat *et al.* (2023) on arrowroot (*Maranta arundinacea*) planted under tree stands of *Falcataria molucana* aged 1, 2 and 3 years, the biomass of the tubers tended to decrease as the age of the trees increased. Figyantika *et al.* (2020) also reported on agroforestry patterns between *Acacia auriculiformis* with soybeans and corn in the Gunung Kidul (Indonesia) which stated that the larger the *A. auriculiformis*, the LER value of the crop species (soybeans and corn) decreased. The decrease in light received by crop species as well as competition for water and nutrient absorption is the cause of the decline in crop plant productivity (Figyantika *et al.* 2020). In this study, the decrease in light intensity under mixed-tree species plantation at 1, 2 and 3 years was 59.61, 34.53 and 25.17%, respectively. In general, the agroforestry pattern between mixed-tree species and *A. galanga* in this study showed an LER value of > 1. This shows that the agroforestry system is very suitable and applicable. According to Desmiwati *et al.* (2021), the contribution of

crop species (*A. galanga*) to farmers' income in the same location, *i.e.*, Parungpanjang, Bogor reached 15.8% of their total income per year. In addition, LER > 1 indicates that a monoculture crop system requires a larger area of land compared to an agroforestry pattern, so that the agroforestry system can be declared more efficient in land use (Ceunfin *et al.* 2017).

## Conclusion

The growth of the three species, *i.e.*, *N. macrophylla*, *N. cadamba* and *N. orientalis* increased with increasing fertilization intensity. The agroforestry using AMF inoculated seedling and 3 kg compost fertilization gave the highest of tree survival, height, diameter, and the biggest tree volume and crown width. The competitive ratio of the three species in plantation conditions without agroforestry, with agroforestry, and agroforestry using AMF inoculated seedling was *N. cadamba* > *N. macrophylla* > *N. orientalis*. In more intensive conditions, *i.e.*, agroforestry with compost fertilization and agroforestry using AMF inoculated seedling and 3 kg compost fertilization, the competition ability of the three species was *N. orientalis* > *N. cadamba* > *N. macrophylla*. The range of LER values (sum of the partial LER of mixed-tree species plantation and partial LER of *A. galanga*) for trees aged 1, 2 and 3 years were 1.48–2.68, 1.92–2.98 and 1.87–4.41, respectively. This LER value >1 showed that the agroforestry system in mixed-tree species plantations was more effective in land use.

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## Author Contributions

Each author (DJS, Y, ER, NW, KPP, RUD, N, ES, D, YMMAN, and VY) equally contributed as main contributors to the design and conceptualization of the manuscript, performed the analysis, prepared the initial draft, and revised and finalized the manuscript. All authors have read and agreed to the published version of the manuscript.

## Conflicts of Interest

All authors declare no conflict of interest.

## Data Availability

Data presented in this study will be available on a fair request to the corresponding author.

## Ethics Approval

Not applicable to this paper.

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