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Effect of inorganic fertilizers on initial growth performance of *Suregada multiflora* and *Firmiana colorata* seedlings in the nursery

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Abstract
Before going to large scale plantation of native tree species, optimal fertilization is required for production of quality tree seedlings in the nursery. This study explores the initial growth performance with different levels of inorganic fertilizers (Urea) applied on 9 months seedlings of *Suregada multiflora* and *Firmiana colorata* in the nursery of institute of Forestry and Environmental Sciences, University of Chittagong, Bangladesh during May 2017 to April 2018. Four treatments were applied for *S. multiflora* and five treatments for *F. colorata* in the nursery. The study showed that seedlings applied with urea (N) varied significantly ($P<0.05$) in comparison to control (i.e. unfertilized soil). Total length, number of leafs, sturdiness of *S. multiflora* was highest for 100 kg N ha^{-1} application whereas the highest values for *F. colorata* were found when the seedlings treated with 300 kg N ha^{-1}. Similar trends of result found in shoot, root, fresh and dry biomass production for *S. multiflora* but for *F. colorata,* the values found highest in case of 400 kg N ha^{-1} application. Maximum volume index and height increment of *S. multiflora* were recorded when the seedlings treated with 100 kg N ha^{-1} but for *F. colorata,* the values found highest while 400 kg N ha^{-1} applied in the seedlings. Therefore, the application of N at the rate of 400 kg ha^{-1} for *F. colorata* (Udal) and 100 kg ha^{-1} for *S. multiflora (*ban-naringa) may be recommended to boost up the height, diameter and seedling dry matter production in the nursery.

Keywords: *S. multiflora; F. colorata;* Urea; quality index; height increment

Introduction

World biodiversity is depleting at an alarming rate due to human interferences and environmental degradation, causing high risk of extinction (Rashid *et al*., 2014) [37]. IUCN's Threatened Plants Unit estimated that about 60,000 plant species (25%) would become either extinct or rare by the year of 2050 (Uberoi, 2010) [40]. However, in Bangladesh, the total numbers of angiosperm species reach over 5700 where, 13% are facing threats to extinction due to habitat fragmentation, population pressure and over-exploitation of natural resources (Khan, 1991; Rahman *et al*., 2010; Irfanullah, 2011; Rahman, 2015; Hossain *et al*., 2017) [25, 36, 42].

Suregada multiflora (A. Juss.) Baill. Sometimes called the "false lime tree" is a species in the family Euphorbiaceae (Roskov *et al*., 2014) [38], locally known as ban-naringa (Das and Alam, 2001) [12], a native lesser known tree species in Bangladesh. This plant is used in the treatment of fever, pneumonia, poisonous effect, squint eye and stomach disorder. The timber is used for making tool handles and seeds are edible (Uddin and Rahman, 2006). In Thailand, people commonly used this species to treat inflammation and skin diseases (Wutthithamavet, 1997) [49]. In addition, the bark of *S. multiflora* is appreciable anti- allergic activities (Cheenpracha *et al*., 2006). However, in some regions, the granule products can be made of this species which acts as a powerful organic herbicide (Laosinwattana *et al*., 2010) [26]. Similarly,

Firmiana colorata (Roxb.) R. Br. locally known as Udal is a native, deciduous tree species naturally grows in Bangladesh, Sri Lanka, India, Nepal, Bhutan, Myanmar, Thailand, China and Indonesia (Hossain *et al*., 2017) [42]. In Bangladesh it is mostly found in Chittagong, Chittagong Hill Tracts, Madhupur and Sylhet (Hossain *et al*., 2015; Rahman *et al*., 2017) [42, 37]. Though *F. colorata* is treated as "Not Evaluate' by Ahmed *et al*. (2008) [2] but in the natural ecosystems the population of this species is reduced drastically due to habitat destruction (Hossain *et al*., 2017) [42]. Udal is a medium sized tree and the fresh young seeds of *F. colorata* are edible as like as almond. The bark yields cordage used to tie cattle, goat in Chittagong areas. Also the tribal people of Chittagong Hill Tracts use the bark to make strap of their long bamboo basket (called "turung") which they carry on their fore-head (Das and Alam, 2001) [12]. The plant is used in the treatment of hysteria, jaundice, seminal emission and spermaturia (Uddin and Rahman, 2006) [36]. Seeds are also eaten by hanuman langurs (Punekar, 2002) [33]. Nevertheless, it is a low priority species in plantation programs where the nursery raising methods are not determined yet.

In tropical reforestation projects, native species have been underutilized, despite the ability to grow successfully in degraded pastures (Butter field, 1995; Butter field and Espinoza, 1995) [9, 10]. It is well proven that application of inorganic fertilizers stimulated the seedling growth and significant increases in biomass of many tree species (Walker *et al*., 1993; Sanginanga *et*

al., 1989; Bungard *et al*., 2000; Ong *et al*., 2004; Hoque *et al*., 2004; Uddin *et al*., 2007;) [48, 8, 16]. Fertilizer recommendation for soils and crops is a dynamic process in view of the generation of the new knowledge, changes in soil nutrient status, changes in plants and planting patterns and associated management practices (Hoque *et al*., 2004) [16]. Nitrogen-fertilized trees are taller and increased at least twice the leaf area than unfertilized trees (Clearwater and Meinzer, 2001) [11]. Therefore, N has an important role in physiological processes vital to the life cycle of plants such as photosynthesis, ion absorption, respiration, cell multiplication and differentiation and inheritance (Epstein and Bloom, 2006) [14].

However, there are very scarce information is available for native lesser known species in Bangladesh as most of the studies were concentrated about the effect of inorganic fertilizer on fast growing species only (Aryal *et al*., 2000; Bhuiyan *et al*., 2000; Hossain *et al*., 2001; Huda *et al*., 2007; Uddin *et al*., 2007; Uddin *et al*., 2009) [5, 7, 42, 22]. Though *S. multiflora* and *F. colorata* has multipurpose value but preference has not been given yet by the Government and private organizations as a priority species for large scale of plantation program. Moreover, the nursery raising technique of these species is still unknown to the scientific community. Therefore, the aim of the present study was to determine the suitable combination of fertilizers that affects the growth performance of *S. multiflora* and *F. colorata* seedlings in the nursery.

Materials and methods

Study site

The study was conducted during the month of May 2017 to April 2018 in the nursery of Institute of Forestry and Environmental Sciences, University of Chittagong, Chittagong, Bangladesh (lies between 91°50´E longitude and 22°30´N latitude) (Hossain *et al*., 2005) [42] (Figure 1). The climate is tropical monsoon with a mean monthly maximum temperature of 29.75°C and a monthly minimum of 21.24 °C. The highest temperature usually occurs on May as 32.60 °C and minimum in January as 14.10 °C (Peel *et al*., 2007). The average annual rainfall of this area is about 2500-3000 mm which mostly takes place between June and September (Mozumder *et al*., 2018) [28].

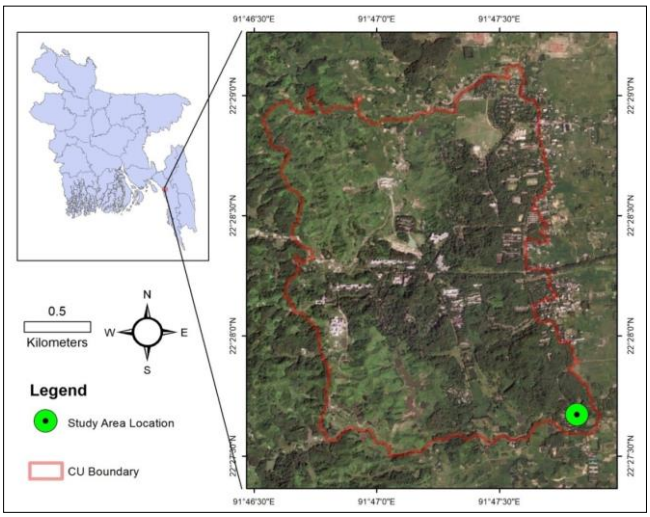

Fig 1: Map showing the location of nursery of IFESCU (Institute of Forestry and Environmental Sciences, Chittagong University)

Seed collection and experimental design

Seeds of *F. colorata* and *S. multiflora* were collected from Chittagong University Campus and Hazarikhil, Chittagong, respectively. Seeds were dried for three days in open sun and only healthy seeds were used in this experiment. The soil used for filling polybags were collected from forest floor of the University campus, dried and sieved well (<3 mm) and mixed with decomposed cow dung in a ratio of 3:1. Polybags of 15×10 cm in size were used for the experiment. Seeds were dibbed to 0.5 cm under the soil by pressing them with thumb. After that they were covered with a thin layer of soil. The seedlings were kept under nursery shade to protect strong sunlight and heavy rainfall. Proper maintenance and precaution were followed during the whole study period. Seedlings were watered regularly and weeding was done in every 4-5 days.

However, after seeds sowing in polybag, seedlings of both species has got the opportunity to grow freely in the nursery till to nine months age. Later on, fertilizers (Urea) of different combinations were applied to initiate growth performance. Urea were measured by 4-digit electronic balance and mixed with the soils of polybags. After mixing the fertilizers watering was done so carefully that water would not go out of polybag and just keep the media moist.

A Completely Randomized Block Design (CRBD) was adopted for both the experiments. During the direct application of fertilizers, the average heights of 9 months *S. multiflora* seedlings were 33.0 cm. There were 4 treatments and 4 replications used whereas in each replication there were 4 seedlings (Table 1). However, total 64 numbers of seedlings were used in this experiment.

Table 1: Level of fertilizer applied in each treatments for Ban-naringa (*S. multiflora*)

Treatments	Kg/ha of Urea	Urea per seedling (gm)
T_0	0	0
T_1	100	0.3367
T_2	200	0.6734
T_3	300	1.0101

Average height of the seedlings of *F. colorata* was 53.5 cm when the fertilizers were applied. However, there were 6 treatments and 5 replications were used and in each replication there were 5 seedlings (Table 2). The total number of seedlings used in this experiment was 150.

Table 2: Level of fertilizer applied in each treatment for Udal (*F. colorata*)

Treatments	Kg/ha of Urea	Urea per seedling (g)
T_0	0	0
T_1	100	0.3367
T_2	200	0.6734
T_3	300	1.0101
T_4	400	1.347
T_5	500	1.6835

Assessment of physiological growth performance

After the application of fertilizer treatments on nine month old seedlings of both species, height was recorded in every 10 days (for convenient measurement) interval where a total of 5 measurements were taken respectively. Finally, after 40 days of

fertilization, the overall height increments of seedlings were recorded. For both the experiments (*S. multiflora* and *F. colorata*), there were 32 and 40 seedlings were uprooted (3 seedlings were selected from each replication) respectively during harvesting period. After taking records of shoot length, root length, collar diameter, no. of branch, no. of nodes, no. of leaf, fresh weight of shoots and roots separately of selected seedlings, then oven-dried at 70 ºC for 48 hours until the constant weight is obtained.

Root-shoot ratio

Root-shoot ratio is the value obtained by dividing shoot (leaf and stem) with the root.

Seedlings volume, quality indices, and sturdiness

Volume index is the value obtained by multiplying shoot height or length (cm) with the square of collar diameter (mm^2) of the seedlings (Manavalan, 1990). The quality index (QI) as developed by Dickson *et al*. (1960) to quantify seedlings morphology was calculated as follows:

$$QI = \frac{\text{Total seedlings dry weight (g)}}{\frac{\text{Shoot height (cm)}}{\text{Collar diameter(mm)}} + \frac{\text{Shoot dry weight (g)}}{\text{Root dry weight (g)}}}$$

Sturdiness of seedling was calculated using the following formula:

$$\text{Sturdiness} = \frac{\text{Shoot height or length(cm)}}{\text{Collar diameter(cm) of the seedling}}$$

Statistical analysis

All the recorded data related to seedling growth attributes were analyzed statistically by using computer software SPSS ver. 23. Duncan's Multiple Range Test (DMRT) was employed to determine the statistical significance ($P<0.05$) of the differences among the mean values. Significant differences were indicated by different letters in the Table.

Results

Growth performance of Ban-naringa (*S. multiflora*)

Mean total length of seedlings was highest (55.11 cm) in T_1 followed by T_2 (53.64 cm) (Table 3). Collar diameter of seedlings was highest (4.9 mm) in T_3 and significantly ($p<0.05$) higher from T_0, T_1 and T_2. However, there were no significant difference was found in leaf number, root-shoot ratio and sturdiness of seedlings in different treatments (Table 3). Besides, average number of leaf per seedling and sturdiness was found highest (20.5 and 84.02) in T_1 respectively.

Table 3: Effect of fertilizer treatments on shoot length, root length, total length, collar diameter, leaf no, root-shoot ratio and sturdiness of Ban-naringa (*S. multiflora*) seedlings in the nursery.

Treatments	Length (cm)			Collar diameter (mm)	Leaf no.	Root/ Shoot	Sturdiness
	Shoot	Root	Total				
T_0	34.21 ^{a*}	14.38 ^{a}	48.59 ^{a}	4.31 ^{b}	17.75 ^{a}	0.53 ^{a}	80.23 ^{a}
T_1	39.24 ^{a}	15.88 ^{a}	55.11 ^{a}	4.76 ^{ab}	20.5 ^{a}	0.44 ^{a}	84.02 ^{a}
T_2	37.9 ^{a}	15.73 ^{a}	53.64 ^{a}	4.88 ^{ab}	16.13 ^{a}	0.48 ^{a}	79.18 ^{a}
T_3	37.11 ^{a}	12.86 ^{a}	49.98 ^{a}	4.9 ^{a}	16.62 ^{a}	0.51 ^{a}	75.86 ^{a}

***** Means followed by the same letter (s) in the same column do not vary significantly at $P<0.05$, according to Duncan's Multiple Range Test (DMRT).

Biomass production of Ban-naringa (*S. multiflora*)

The highest total fresh weight was recorded in T_1 (18.38 g) and significantly ($p<0.05$) different from other treatments. No significant difference were found in root dry weight, shoot dry weight and total dry weight of T_0, T_1, T_2 and T_3 treatments (Table 4). Besides, total oven dry weight of the seedlings was analyzed, where the highest value was found in T_1 (7.87). However, maximum volume index was recorded in T_3 (900.61) and minimum in T_0 (645.78). Similar trends of result were found for quality index, where the highest value was recorded in T_3 (0.757) and lowest in T_0 (0.726) (Table 4).

Table 4: Effects of fertilizer treatments on Fresh weight, Dry weight, quality index and volume index of Ban-naringa (*S. multiflora*) seedlings in the nursery.

Treatments	Fresh weight (g)			Dry weight (g)			Quality index	Volume index
	Shoot	Root	Total	Shoot	Root	Total		
T_0	10.61 ^{b*}	4.95 ^{a}	15.56 ^{b}	4.87 ^{a}	2.54 ^{a}	7.42 ^{a}	0.726 ^{a}	645.78 ^{a}
T_1	13.1 ^{a}	5.27 ^{a}	18.37 ^{a}	5.44 ^{a}	2.44 ^{a}	7.87 ^{a}	0.736 ^{a}	889.79 ^{a}
T_2	10.86 ^{b}	4.12 ^{a}	15.00 ^{b}	4.49 ^{a}	2.03 ^{a}	6.52 ^{a}	0.649 ^{a}	888.65 ^{a}
T_3	10.49 ^{b}	3.8 ^{a}	14.29 ^{b}	4.87 ^{a}	2.4 ^{a}	7.24 ^{a}	0.757 ^{a}	900.61 ^{a}

***** Means followed by the same letter (s) in the same column do not vary significantly at $P<0.05$, according to Duncan's Multiple Range Test (DMRT).

Height (cm) increment of Ban-naringa (*S. multiflora*)

Though initially few seedlings were died due to the effect of treatments but after 40 days, T_1 showed the best height increment (2.96 cm) among all the treatments where T_2 and at T_3 showed negative effect (over doze) of fertilizer application (Figure 2).

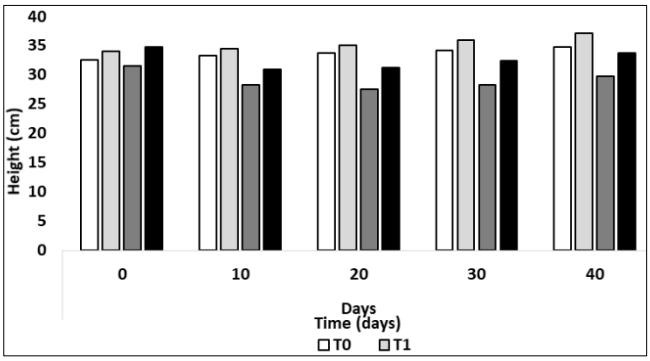

Fig 2: Height growth of Ban-naringa (*S. multiflora*) seedlings in response to different treatments.

Growth performance of *F. colorata*

Considering the total length of *F. colorata* seedlings, the value was highest (68.37 cm) in T_3 followed by T_4 (67.19 cm) (Table

5). Collar diameter of seedlings was maximum (6.86 mm) in T_1 and significantly ($p<0.05$) higher from T_0, T_2, T_3, T_4, T_5. Besides, highest volume index (2679.52) was recorded in treatment T_4. However, root shoot ratio and sturdiness was found maximum (0.931 and 100.53) in T_3 respectively, whereas these values were significantly different ($p<0.05$) from other treatments (Table 5).

Table 5: Effect of fertilizer treatment on shoot length, root length, total length, collar diameter, volume index, root-shoot ratio and sturdiness of Udal (*F. colorata)* seedlings in the nursery.

Treatment	Length (cm)			Collar diameter (mm)	Volume index	Root/Shoot	Sturdiness
	Shoot	Root	Total				
T_0	53.57 ^{a*}	12.08 ^{a}	65.65 ^{a}	5.76 ^{b}	1845.39 ^{a}	1.0185 ^{ab}	95.91 ^{ab}
T_1	54.03 ^{a}	11.66 ^{a}	65.69 ^{a}	6.86 ^{a}	2668.30 ^{a}	0.915 ^{ab}	81.39 ^{b}
T_2	53.68 ^{a}	10.90 ^{a}	64.58 ^{a}	6.69 ^{ab}	2405.02 ^{a}	1.208 ^{a}	81.83 ^{b}
T_3	58.07 ^{a}	10.30 ^{a}	68.37 ^{a}	5.80 ^{b}	1997.26 ^{a}	0.931 ^{ab}	100.53 ^{a}
T_4	56.24 ^{a}	10.95 ^{a}	67.19 ^{a}	6.79 ^{a}	2679.52 ^{a}	0.918 ^{ab}	82.97 ^{b}
T_5	52.63 ^{a}	11.51 ^{a}	64.14 ^{a}	6.53 ^{ab}	2290.03 ^{a}	0.868 ^{b}	81.30 ^{b}

*****Means followed by the same letter (s) in the same column do not vary significantly at P<0.05, according to Duncan's Multiple Range Test (DMRT).

Biomass production of *F. colorata*

Highest total fresh weight was recorded in T_4 (30.3 g) and significantly ($p<0.05$) different from other treatments. Similar trends were found in case of oven dry weight of the seedlings where the maximum value was recorded in treatment T_4 (8.907 g). The highest quality index was recorded in T_4 (0.946) followed by T_2 (0.896) and was significantly ($p<0.05$) different from other treatments (Table 6).

Table 6: Fertilizer treatments effects on Fresh weight, Dry weight and quality index of Udal (*F. colorata)* seedlings in the nursery.

Treatments	Fresh weight (g)			Dry weight (g)			Quality index
	Shoot	Root	Total	Shoot	Root	Total	
T_0	9.60 ^{c*}	10.20 ^{bc}	19.80 ^{b}	3.01 ^{b}	2.89 ^{b}	5.905 ^{c}	0.591 ^{bc}
T_1	11.82 ^{bc}	10.01 ^{bc}	21.83 ^{b}	3.88 ^{ab}	3.23 ^{b}	7.127 ^{bc}	0.825 ^{ab}
T_2	13.20 ^{ab}	14.30 ^{a}	27.50 ^{a}	3.59 ^{b}	4.24 ^{a}	7.842 ^{ab}	0.896 ^{a}
T_3	11.56 ^{bc}	8.45 ^{c}	20.01 ^{b}	3.01 ^{b}	2.79 ^{b}	5.809 ^{c}	0.521 ^{c}
T_4	15.65 ^{a}	14.65 ^{a}	30.30 ^{a}	4.69 ^{a}	4.21 ^{a}	8.907 ^{a}	0.946 ^{a}
T_5	13.10 ^{ab}	11.90 ^{ab}	25.00 ^{ab}	3.89 ^{ab}	3.25 ^{b}	7.146 ^{bc}	0.785 ^{ab}

***** Means followed by the same letter (s) in the same column do not vary significantly at P<0.05, according to Duncan's Multiple Range Test (DMRT).

Height (cm) increment of *F. colorata*

Height increment for *F. colorata* shows an increasing trend after the application of Urea to the end of 40 days. Height data at different intervals showed the best height increment among all treatments was found in T_4 (2.75 cm) where the minimum increment was recorded in T_5 (Figure 3).

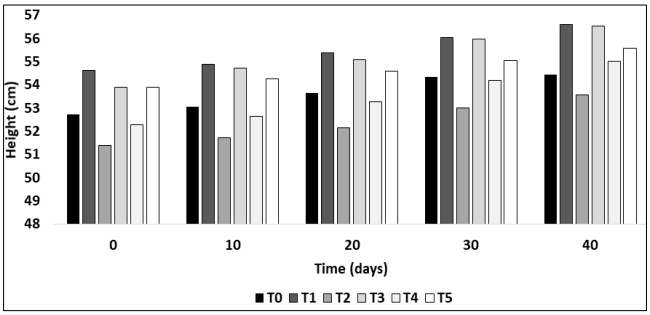

Fig 3: Height growth of Muliudal (*F. colorata)* seedlings in response to different treatments.

Discussion

Physical traits are the determinable attributes of tree seedlings in nursery (Afa *et al*., 2011) [1]. Different morphological criteria used to grade seedling quality in the nursery include shoot height, collar diameter, leaf area, leaf number, and seedling biomass (Hoque *et al*., 2004) [16]. However, the results of the present study demonstrate that *S. multiflora* seedlings, when treated with T_1 (100 kg N ha^{-1}), and *F. colorata* with T_4 (400 kg N ha^{-1}) showed better performances in growth, and biomass production.

The results of the study is consistent with the findings of Hoque *et al*. (2004) [16] who found significant height increase in seedlings of *Michelia champaca* after N and P fertilization. Hossain and Hossain (2006) [42] investigated the effect of NPK fertilizer on growth of Telsur seedlings where they found N has significantly increased the seedling height, leaf and shoot dry matter production which supported present findings. Andrew *et al*. (2018) [3] studied the response of four species of tropical timber seedlings to Urea and Folivert fertilizers in nursery where they revealed plants treated with 3 g of urea produced the highest number of leaves in *Albizia zygia* seedlings. Afa *et al*. (2011) [1] revealed the effects of organic and inorganic fertilizers on early growth characteristics of *Khaya ivorensis* in nursery where they found all the growth parameter increased with the use of 75 mg urea added to seedlings and supported by present study. Earlier, The response of few major timber species to N fertilization have been assessed, including lodge pole pine (Van den Driessche, 1977) [45], white spruce (Armson and Sadreika, 1979) [4] where present study is agreed with their findings. However, several studies have also found some sort of positive responses of fertilization reported Awang and Katim (1986) [6], Zwierink (1984) [50], Onuwaje and Uzu (1982) [31], and Van den Driessche (1982) [45] earlier.

In addition, there was no toxic effect of fertilizer was observed for *F. colorata (*Udal*)* seedlings but for *S. multiflora* with the application of 200 kg N ha^{-1} (T_2) and 300 kg N ha^{-1} (T_3). It may initially hinders the seedling (*S. multiflora*) growth and supported the findings of Van den Driessche (1980) [45], who revealed both positive and negative impact of nursery fertilizers on seedlings. Present study also supported the finding of Kadeba (1978) [24] and Hartley (1977) which shown that addition of excess fertilizer on *Pinus caribaea* depressed growth and increased mortality on Nigerian Savannah sites. High dose of fertilizers application damaged young seedlings survival capacity (Evans, 1982). Urea

caused 50% mortality of seedlings if applied wrongly (Kadeba, 1978; Oju and Jackson, 1973) [24, 29]. Further research may be focused on nursery and plant production systems for native plant species in order to rich genetic diversity and reduce future environmental stresses (Hay and Probert, 2013). From over all aspects, though it has some negative effects itself, it is found that, inorganic fertilizers obviously have beneficial effect to increase the collar diameter, and total biomass production of the seedlings for both the species.

Conclusion

Once the species *S. multiflora* and *F. colorata* were available in natural forest but now become very rare. Nowadays, it is well understood that the loss of flora may be minimized in a sustainable manner and increasing the production forestry through domestication of indigenous timber species with proper nursery technique establishment. Therefore, the application of N for the seedlings at the rate of 400 kg ha^{-1} for *F. colorata* (Udal) and 100 kg ha^{-1} for *S. multiflora* may be recommended to boost up the height, diameter and seedling dry matter production in the nursery. However, application of Urea should be done carefully as it has toxic effects if applied wrongly or over dozes.

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Conflict of interest

There is no conflict of interest.

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