Efficient agroforestry system using fiber grow bag of medicinal plant, Asparagus cochinchinensis (Lour.) Merr

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Research Article

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Abstract Forest cultivation of medicinal plants is environmentally friendly, but selecting the optimal cultivation site is difficult and it is necessary to overcome competition with weeds. Fiber growth bags are pots made of non-woven fabric that are light, inexpensive, and have excellent drainage properties. In this study, we investigated the optimal conditions for forest cultivation of *Asparagus cochinchinensis* using fiber growth bags. The soil temperature of the fiber growth bag was not significantly different from the soil temperature of the experimental site. The survival rate of the above-ground part was highest in large fiber growth pockets, and the growth of the underground part also showed differences depending on the size of the fiber growth pocket. The above-ground growth of *A. cochinchinensis* did not show significant differences depending on planting density. Planting under conifers was suitable in terms of above-ground growth. Above-ground shoot growth was higher when planted on a northern slope than on a southern slope. The photosynthetic rate of *A. cochinchinensis* was slightly lower than that of other wild plants. The maximum quantum yield was over 0.8, confirming that there were no problems with photosynthetic growth during forest cultivation using fiber growth bags. The content of asparagine, an important bioactive substance, in *A. cochinchinensis* was approximately 9.9 times higher than that grown on farms. In other words, in the case of *A. cochinchinensis* forest cultivation, the production method using fiber growth bags is an excellent production method and is considered to be apply to other wild vegetable cultivation.

Keywords Fiber grow bag, Forest cultivation system, *Asparagus cochinchinensis*, culture condition, The photosynthetic reaction

Declarations

Conflict of interest: The authors declare no competing interests.

Introduction

Asparagi tuber is a tuber of *Asparagus cochinchinensis* (Lour.) Merr. a perennial herb belonging to the genus Asparagus in the family Liliaceae (Fig. 1A). The dried roots of *A. cochinchinensis* have been used in Traditional Oriental Medicine for more than two thousand years to treat fevers, renal failure, heart diseases, respiratory diseases, skin diseases and lung cancer (Kook, 2012; Park and Lee, 2000; Xiong et al., 2011). *A. cochinchinensis* contains active ingredients such as steroidal saponins and β-sitosterol. It has been reported that β-sitosterol is effective for lung cancer (Le Son and Anh, 2013). In addition, research results show that it inhibits the growth of bacteria related to respiratory diseases such as *Staphylococcus aureus* and *Staphylococcus bovis*. (Bradford and Awad, 2007; Saeidnia, et al 2014;

Wang et al., 2000). *A. cochinchinensis* is currently in danger of extinction as its native habitat has almost disappeared due to indiscriminate harvesting.

Traditional forestry focusing on timber production has a long payback period, so general private forest owners are reluctant to invest in forestry. However, Agro-foresty management using fruit trees, wild vegetables, and wild medicinal plants can overcome the economic vulnerability of long-term investment in forests and thus is in the spotlight as a representative short-term forest income source in rural areas (Jung et al., 2011). Agro-forestry management, including forest cultivation of wild medicinal plants, does not use pesticides, etc. and is carried out in a clean mountainous area, so it is possible to produce pollution-free organic food to meet modern consumer preferences. By reducing production costs, it is possible to increase farm household income (Jang and Lee, 2000).

In forest cultivation, the supply of natural medicinal plants must be increased to meet the growing demand. Forest cultivation is preferable because the production cost is lower than field culture (Song et al., 2016). However, most plant species are difficult to cultivate due to specific biological or ecological requirements, such as slow growth rates, special soil requirements, low germination rates, and susceptibility to pests (Schippmann et al., 2002). Therefore, it is necessary to search for suitable conditions for each medicinal plant's intensive cultivation (Park et al., 2014).

Since the productivity of the lower stand varies depending on the interaction between the abiotic environment, such as light, moisture, and temperature of the forest and the vegetation distributed there, the cultivation of medicinal plants should be approached scientifically (Song et al., 2015). Other study was reported on physiological responses such as chlorophyll fluorescence and chlorophyll content is one of the elements necessary for selecting plants suitable for forest cultivation according to the stand characteristics of trees and the development of cultivation technology (Kim et al., 2015).

Cultivation methods are also important in the forest cultivation of medicinal plants. A fiber grow bag can be defined as a flower pot made of non-woven fabric and is also called a non-woven flower pot (Fig. 1B). Fiber grow bag is lighter and less expensive than general plastic products and has excellent flame retardancy, durability, insulation, breathability, and drainage. It is possible to produce products of various sizes and capacities without a mold. The durability of the fiber grow bag is more than 7 to 10 years, and 2 to 4 handles are attached to make it easy and convenient to move. The advantages of fiber grow bag cultivation are that high-quality plants can be produced, eco-friendly cultivation is possible, it is very advantageous for weed management, forest damage can be minimized, few location restrictions, and year-round cultivation is possible. Some disadvantages are the initial cost fiber grow bag, soil, irrigation, etc.) and the disadvantage of mechanization in harvesting. However, forest cultivation of medicinal plants using fiber grow bags seems highly possible. However, there is no research report on forest cultivation of medicinal plants using fiber grow bags. This study was

conducted to investigate the optimal conditions for forest cultivation using the fiber grow bag of *A. cochinchinensis*, a medicinal plant.

Materials and Methods

Experimental site and plant material

The test site for this study was conducted at the Gajwa Experimental Forest of Gyeongsang National University (1056-1 Gajwa-dong, Jinju-si, Gyeongsangnam-do, GPS: 35o 8'52.55" N / 128o 5'55.21" E). A forest cultivation trial of fiber grow bag was conducted in a mixed forest with a southeast slope. *A. cochinchinensis* seedlings are healthy one-year-old seedlings and healthy seedlings with tubers and stems were produced by the Gyeongnam Forest Environment Research Institute.

Table 1

Fig. 1.

As for the environment of the test site, the daily average temperature (℃) ranges from 0.7 degrees in January to 24.5 degrees in July, the minimum daily temperature is January –6.3 to July 20.7℃, and the daily maximum temperature (℃) is from 8.9 ℃ in January to 29 ℃ in July. In the test site, the annual precipitation was 1,636 mm, and the average daily humidity was 52.5% in January and 85.6% in August.

The test site soil showed weak acidity at pH 5.2. In the case of EC, the level was similar to that of other plants, and the concentration of adequate phosphorus was higher than that of other forest cultivation sites (Table 1). In the case of substitution cations, calcium, potassium, and sodium were appropriate, but magnesium was slightly insufficient.

Fiber grow bag planting

Three sizes of fiber grow bags were used in the experiment. Fiber grow bags were used in large (diameter 55 cm × height 40 cm), medium (diameter 50 cm × height 40 cm), and small (diameter 40 × height 35 cm) . The material of the fiber grow bag was non-woven fabric, and one with a handle was used (Fig. 1B).

For the fiber grow bag planting of *A. cochinchinensis*, the fiber grow bag was moved to the forest test site, and seedlings were planted after filling the test soil halfway. Then, *A. cochinchinensis* seedling were placed in the center of the fiber grow bag, filled it with hill soil to the top 2-3 cm height, and pressed lightly (Fig. 1C).

In order to identify the optimal conditions for growing bag forest cultivation of *A. cochinchinensis*, the optimal fiber grow bag size and planting density were investigated. First, in order to identify the optimal fiber growth bag size conditions, 15 plants were planted in each large, medium, and small fiber grow bag. Second, planting densities per fiber grow bag was set to loose (10 plants/bag), medium (15 plants/bag) and dense (20 plants/bag). Planting density was measured using a medium-sized fiber grow bag most suitable for growth. Fiber grow bag size and planting density treatments were conducted at south slope in mixed forest. Third, forest floors were divided into coniferous forest, broad-leaved forest, and mixed forest. Coniferous forest was composed of *Quercus myrsinifolia*, broad-leaved forest was included with *Pinus densiflora*, and mixed forest was included with *P. densiflora*, *Castanea crenata*, *Quercus variabilis* Blume, *Prunus serrulata* Lindl. f. *spontanea* (Maxim.), and *Populus tomentiglandulosa* T. Lee. Fourth, slope positions were determined in north and south. Angle of north and south is 22 – 24° and 17 – 21°, respectively. 15 plants in medium sized fiber grow bag to forest floor and slope treatments.

Fertilization was not performed during the test period. The weeding was done once in August. The growth survey of *A. cochinchinensis* planted in a fiber grow bag was conducted every month from the end of April until the end of October. In order to measure the roots, the plants were collected from the fiber grow bag, the roots were separated, and washed with water to remove impurities. After filling a 20 × 25 × 3 cm transparent glass tray with water, the washed roots were placed to avoid overlapping as much as possible and scanned (Epson Perfection V700, Seiko Epson Crop., Japan). The morphology of root development was analyzed using the WinRHIZO program (Regent, CA/WinRHIZO Pro. LA2400 Scanner). The scanned image shows the total root length, projected root area, total root surface area, total root volume, and mean root diameter.

Investigation of temperature and moisture content of fiber grow bags

The characteristics of fiber grow bags exposed to forest plantation was investigated. A soil moisture meter (VG Merter-200, Vegetronix) and a data logger (HOBO UA-002–08) were installed in the test site, and the soil temperature and soil moisture inside the medium-sized fiber grow bag were measured.

Photosynthetic reaction

Photosynthetic rate, dark respiration rate, stomatal conductance, transpiration rate, etc., were measured using a portable photosynthesis meter (Li-6400, LI-COR Inc., Lincoln, NE, USA). The photosynthetic reaction was measured at 9 levels (0, 50, 100, 250, 500, 800, 1000, 1,500, and 2,000 $\mu mol \cdot m^{-2} \cdot s^{-1}$) were measured with a difference. Photosynthesis-related factors were calculated using the value at the luminous intensity of 600 $\mu mol \cdot m^{-2} \cdot s^{-1}$, the same level as in the lobe chamber, among the nine luminous

intensity levels. The flow rate of air flowing into the leaf chamber of the photosynthesis meter was set at 500 μmol·m^{-2}·s^{-1}, the temperature was set at 25℃, and the relative humidity was 60% so that there was no influence due to environmental changes in the outside air. In addition, to prevent sudden changes in CO_2 concentration, a CO_2 injector system was attached to the photosynthesis meter to maintain a stable CO_2 concentration within the range of 400±2 μmol·m^{-2}·s^{-1}. The photosynthesis reaction was measured 5 times for each species from 9:00 am to 2:00 pm when the plant growth was most active.

Chlorophyll fluorescence reaction

Chlorophyll fluorescence analysis was measured by the quenching kinetics analysis method under dark adaptation (20 min dark treatment) using Handy FluorCam (Photon Systems Instruments, spol. Sro., Drásov, Czech Republic) (Barbagallo et al., 2003). Actinic light and continuous light sources were used in the measurement to induce chlorophyll fluorescence. At this time, the Handy FluorCam analysis conditions were chemical light (actinic light, red LED): 200 μmol m^{-2}·s^{-1}, continuous light source (saturating light, moderate light): 1,250 μmol m^{-2}·s^{-1}).

Asparagine analysis

To compare asparagine contents of *A. cochinchinensis* according to cultivation environment, roots of *A. cochinchinensis* grown in fiber grow bag for 2 years were collected in forest and farm cultivated, respectively. For the content of free amino acids, 50 ml of 70 % ethanol was added to 1 g of sample powder, heated in an ultrasonic bath at 80 ° C for 3 hours, cooled at room temperature, and centrifuged at 3,000 rpm for 15 minutes. Supernatant 1 ml were mixed with lithium citrate buffer 1 ml (pH 2.2), and the solution was filtered using.0.2 μm membrane filter.

The asparagine analysis was used by amino acid analyzer Biochrom 30 (Pharmacia Biotech, Cambridge, UK). Sample analysis were conducted following manufacturer standard protocols. Experiments were performed with three times.

Statistical analysis

One-way analysis of variance was used using IBM SPSS Statistics (SPSS Statistics Program, version 14.0, SPSS Inc, USA), a statistical analysis program, and the significance between each treatment was determined by DMRT (duncan's multiple range test) was carried out at the 5% level.

Results

Soil Temperature and Moisture Content in Fiber Grow Bags

Changes in soil temperature and soil moisture in the fiber grow bag were investigated in the forest cultivation test strip (Fig. 2). The soil temperature in the fiber grow bag did not differ significantly from that in the experimental site (Fig. 2A). The soil temperature in the fiber grow bag did not show a significant temperature difference with forest soil in summer (June-July). However, after September, it tended to be higher, around 0.3 to 1 ℃, but there was no significant difference. The relative humidity of the test strip showed a significant difference in spring. The deviation decreased toward autumn (Fig. 2B).

Fig. 2

As a result of examining the moisture content of the soil in the fiber grow bag and the forest soil in the experimental site, the moisture content of the soil in the fiber grow bag was measured to be slightly higher than that of the forest soil. Relative humidity showed a significant difference between morning and evening in spring and autumn but little difference in summer, the rainy season. However, it was found that the soil temperature and moisture of the fiber grow bag were not significantly affected by the relative humidity.

Growth of *A. cochinchinensis* according to the size of the fiber grow bag

The growth of *A. cochinchinensis* differed greatly depending on the size of the fiber grow bag (Fig. 3). In the case of the aerial part of *A. cochinchinensis*, the survival rate was the highest in the large fiber grow bag. There was no significant difference between the medium and small fiber grow bags (Fig. 3A). Also, the survival rate of *A. cochinchinensis* decreased as the cultivation period passed.

The shoot growth of *A. cochinchinensis* was best in the small fiber grow bag (Fig. 3B), followed by the large fiber grow bag, and the lowest in the medium fiber grow bag. The growth of *A. cochinchinensis* in the fiber grow bag was slow until July but showed vigorous growth until August. After August, growth tended to decrease due to dryness and falling of the above-ground part.

The number of leaves did not show a significant difference according to the fiber grow bag size (Fig. 3C). The number of leaves showed a pattern similar to that of shoot growth and was the highest in August.

Chlorophyll also showed different patterns according to the size of the fiber grow bag (Fig. 3D). In the case of SPAD values, there was no significant difference until July, but there was a clear difference in August. Chlorophyll content was highest in *A. cochinchinensis* grown in large fiber grow bags and lowest in small ones.

Fig. 3

The growth of the underground part of *A. cochinchinensis* also showed a difference according to the size of the fiber grow bag (Table 2; Fig. 4). As for the total root length, the fiber grow bag of the medium size grew best at 180.56cm, and there was a significant difference at 139.98 and 85.45cm in the large and small sizes, respectively. The root area was the largest in the medium-sized fiber grow bag at 51.17 cm^2, followed by large and small fiber grow bags. The root volume was the thickest at 1.281 cm^3 in the large fiber grow bag, followed by the medium and small fiber grow bags in that order. The root diameter was 1.08 mm in the large fiber grow bag, followed by 0.902 mm in the medium fiber grow bag and 0.834 mm in the small fiber grow bag.

Effect of planting density

The above-ground growth of *A. cochinchinensis* did not show a significant difference according to the planting density (Fig. 4). However, there was no significant difference in stem growth according to planting density. The number of leaves did not significantly differ according to the planting density. Chlorophyll content also showed a similar trend.

Fig. 4

The growth of *A. cochinchinensis* under-ground showed significant differences according to the planting density (Table 2; Fig. 5). The root length was the longest at 220.13 cm at loose density, followed by 129.81 cm at normal dense density and 59.01 cm at normal density. The root area was the widest at 56.05 cm^2 at loose planting density, followed by 36.4 cm^2 at dense density and 24.45 cm^2 at normal density. The root volume was the thickest at a loose density of 1.14 cm^3 and similar at dense and normal densities. Root diameter was the thickest at 1.32 mm when the planting density was normal, followed by 0.89 mm at the dense density and the smallest at the loose density.

Fig. 5

Effect of forest floor

A. *cochinchinensis* planted in a fiber growth bag showed growth differences in the aboveground part depending on the forest floor (Fig. 6). A. *cochinchinensis* was suitable for planting under coniferous forests in terms of above-ground growth, number of leaves, and chlorophyll content. It was found that the shoot growth and number of leaves of A. *cochinchinensis* showed a very significant difference after 8 weeks of cultivation.

Fig. 6

The growth of the underground part of *A. cochinchinensis* planted in a growth bag showed differences depending on the forest floor (Table 2, Fig. 8). The root length was 87.29 cm in coniferous forests, 49.4 cm in broad-leaved forests, and 55.27 cm in mixed forests. Root area was larger in broadleaved forest than in coniferous forest and mixed forest. Root volume was largest in coniferous forest, followed by mixed forest and broadleaved forest. Root diameter was larger in coniferous forests than in broadleaved forest and mixed forest. There was significant difference in root characteristics between coniferous forest, broadleaf forest, and mixed forest.

Effect of slope

The growth of the aboveground part of *A. cochinchinensis* planted in the fiber growing bag also showed differences depending on the slope (Table 2). The stem growth and number of leaves were higher in plantings on the northern slope than on the southern slope. A. *cochinchinensis* showed significant growth differences depending on the slope after 4 months.

Fig. 7.

The growth of the underground part of *A. cochinchinensis* planted in the fiber growing bag also showed differences depending on the slope (Table 2, Fig. 8). The root length of *A. cochinchinensis* was about twice as different on the northern slope than on the southern slope. There was no significant difference in root area between the northern and southern slopes, and the root volume was larger on the northern slope than on the southern slope. The root diameter on the northern slope was approximately 1.14 mm larger than on the southern slope.

Table 2

Fig. 8.

Photosynthesis reaction of *A. cochinchinensis* during fiber grow bag cultivation

Photosynthesis-related factors were investigated in forest cultivation using a fiber grow bag (Fig. 9). The photosynthetic curve increased rapidly from 0 to 250 μmol $m^{-2}s^{-1}$ and gradually increased until 501000 and reached light saturation after 1000 μmol m^{-2} s^{-1} (Fig. 9A). The stomatal conductance decreased to the light compensation point. Then, it increased until it reached the light saturation point (Fig. 9B). The amount of CO_2 in mesophyll cells steadily decreased until the light saturation point (Fig 9C).

The light compensation point of *A. cochinchinensis* was 64 PPFD (Photosynthetic Photon Flux Density / μmol m^{-2} s^{-1}), and the light saturation point was measured as 1933 PPFD (Table 3). Dark Respiration Rate was 0.85, the photosynthetic ability was 4.41 μmol m^{-2} s^{-1}, and the apparent quantum yield was 0.013.

Fig. 9

Table 3

Chlorophyll fluorescence reaction

The initial fluorescence yield (Fo) value was 2950, the maximum fluorescence yield (Fm) was 15110, the variable fluorescence yield (Fv) was 12160, and the maximum quantum yield (Fv/Fm) was 0.802 (Table 4).

Table 4

Asparagine content in forest and farm cultivation

Forest cultivation of *A. cochinchinensis* using fiber grow bags showed differences from farm cultivation regarding asparagine and amino acid content (Table 5). The amino acid content of forest-cultivated *A. cochinchinensis* was about 8.7 times higher than farm-cultivated *A. cochinchinensis*. The asparagine content of forest-cultivated *A. cochinchinensis* was also about 9.9 times higher than farm-cultivated *A. cochinchinensis*. The content of L-arginine, another major amino acid, was about 9.7 times higher in forest-grown *A. cochinchinensis* than farm-cultivated, and taurine was also higher in forest-cultivated. However, γ-Amino-n-butyric Acid was lower in forest-cultivated *A. cochinchinensis* than in farm-cultivated *A. cochinchinensis*. Most interestingly, amino acids were detected in forest-cultivated *A. cochinchinensis*, but few were analyzed in farm-cultivated *A. cochinchinensis*.

Table 5

Discussion

There was a significant difference in the growth of *A. cochinchinensis* according to the size of the fiber grow bag. In the case of the above-ground part of *A. cochinchinensis*, the survival rate was the highest

in the large fiber grow bag. For the growth of *A. cochinchinensis,* the growth of the above-ground and underground parts also showed a difference depending on the size of the fiber grow bag, and the medium-sized fiber grow bag showed the best growth. This result is consistent with previous studies (Weston and Zandstra, 1986; Kemble et al., 1994). In the case of tomato seedlings, the larger the pot size, the more soil is filled, so the supply of nutrients is higher than that in a relatively small pot, and the large rhizosphere volume provides good root growth, which leads to better growth in the above-ground part. As the pot size is small, the growth of the underground part is inhibited, which affects plant growth. Therefore, it is judged that the optimal number of growing days may vary depending on the size of the container. Jeong et al. (2016) also stated that when both the number of days of growth and the size of the pot are considered, the quality is excellent during short-term growth when the pot is small, but the quality drops sharply when the growing period is long. Suitable pot size and several days of growth are required for excellent quality in forest cultivation of useful resource plants and forest cultivation of *A. cochinchinensis* using fiber grow bags.

The above-ground growth of *A. cochinchinensis* did not significantly differ according to the planting density. Generally, it is known that, when growing container seedlings of plants, the higher the planting density per unit area, the higher the live weight increases. However, when the density is exceeded, the growth decreases. These results have been reported in many previous studies. It was reported that in *Chamaecrista nomame* (Cho et al., 2000) and soybean (Kang et al., 1998), as the seeding amount per unit area increased, the fresh weight increased to a certain amount. However, when the appropriate seeding amount for each plant was exceeded, the fresh weight gradually decreased. Urbaniak et al. (2008) also said that the seeding amount per unit area affects the growth and density of individuals.

On the other hand, there are reports that there is no significant difference in growth according to planting density at the initial planting stage. Noh and Cho (2020) stated that there was no difference in survival rate according to planting density for 3 years after planting zelkova seedlings.

As a result of examining the moisture content of forest soil and fiber grow bag soil, the moisture content of the soil contained in the fiber grow bag was measured to be slightly higher than that of the forest soil. The gutter cultivation method, such as the fiber grow bag, is a bottom irrigation method in which a small amount of amniotic water is continuously supplied into the medium by the capillary action of the wick. This was developed over 30 years ago and is known as a system with low water stress for plants. The material of the fiber grow bag used in this study is polyester, and it is known that capillary action is easy (Jung et al., 2005). This study shows that the capillary phenomenon can be continuously induced because the underside of the fiber grows bag is in close contact with the forest soil. The fiber grow bag also does not have a significant temperature change compared to the forest soil, so it is judged that there will be no significant problem in the cultivation of useful forest resource plants.

The cultivation environment is critical in forest cultivation. In this study, it was good to cultivate under conifers in clinical practice. Hur et al. (2012) said that coniferous forests dominated by Hinoki cypress trees are more suitable for growing wild garlic. Forest cultivation using fiber grow bags has a different forest floor effect from general forest cultivation. This is judged to be due to the difference in the wavelength and intensity of light passing through coniferous and broad-leaved forests.

The photosynthetic capacity of *A. cochinchinensis* was 3-4 μmol m^{-2} s^{-1}. The photosynthetic curve tended to increase rapidly from 0 to 250 μmol $m^{-2}s^{-1}$, then gradually increased until 500-1000, and reached photo saturation after 1000 μmol m^{-2} s^{-1}. This is similar to or lower than medicinal and vegetable plants cultivated in forests in the mountains of Korea. The photosynthetic capacity of the test plants grown in the electric light treatment area in July was 6.1 μmol m^{-2} s^{-1} for wild garlic and 11.8 μmol $m^{-2}s^{-1}$ for Gomchi(*Ligularia fischeri*) and Gondalbi(*Ligularia stenocephala*), 12.2 μmol $m^{-2}s^{-1}$, respectively(Kim, 2008). As reported by Larcher (1995), in the case of Heliophytes, the light compensation point is 20-40 μmol $m^{-2}s^{-1}$, and the light saturation point is distributed in 1000-1500 μmol $m^{-2}s^{-1}$. In the case of Sciophytes, the light compensation point is 5-10 μmol $m^{-2}s^{-1}$, and the light saturation point is 100-200 μmol $m^{-2}s^{-1}$. The light compensation point was similar to that of the shady plant, but the light saturation point was found to belong to the sunny plant.

In addition, the optical compensation point of *A. cochinchinensis* was measured to be 64 PPFD (Photosynthetic Photon Flux Density / μmol m^{-2} s^{-1}), and the optical saturation point was measured to be 1933 PPFD. The stomatal conductance decreased until the photo compensation point and then increased until it reached the photo saturation point. The amount of CO_2 in mesophyll cells steadily decreased until the photo saturation point.

It is known that the lower vegetation's light conditions significantly influence the lower vegetation's development and physiological state, and the light requirement for the development and growth of the lower vegetation varies greatly depending on the species (Choi, 2001). Therefore, research on the physiological characteristics of plants, such as photosynthesis, chlorophyll fluorescence, and chlorophyll content in the lower vegetation, which are limited by the light environment, is one of the factors necessary for the selection of plants suitable for forest cultivation and the development of cultivation technology (Kwon et al., 2009). The luminosity and photo physiology of *A. cochinchinensis* were similar to those of *Ligularia fischeri*, a plant cultivated in forests in Korea (Kim, 2008). The photosynthetic rate of *Ligularia fischeri* tended to increase as the light intensity increased. However, the photosynthetic rate may vary depending on the environmental conditions, temperature, and soil moisture conditions of the plantation (Kim et al., 2008). Photo-physiology according to the light intensity is different for each plant species, but it is expected to contribute to creating an appropriate light environment.

The maximum quantum yield (Fv/Fm) of *A. cochinchinensis* was 0.802. Due to environmental changes in plants, photosynthetic efficiency is lowered, such as electron transfer inhibition during photosynthesis, chlorophyll content change, and charge separation decrease (Perboni et al., 2012), and plants (Rouse et al., 1974; Suárez et al., 2009).

Chlorophyll fluorescence and photochemical reaction in plants: Fluorescence emitted from chlorophyll is part of the light energy not used in the initial photochemical reaction. An increase in chlorophyll fluorescence indirectly means a decrease in photosynthetic activity (Baker and Oxborough, 2004). Recently, chlorophyll fluorescence analysis, which can be monitored non-destructively, has been used to evaluate the health status of plants caused by environmental stress under various environmental conditions such as salt damage, drought, high temperature, and low temperature (Prakash et al., 2003; Yoo et al., 2014). It has been reported that the chlorophyll content decreases as the shade increases with changes in the photographic environment (Cho et al., 2008).

The Fv/Fm value measured in scotopic leaves represents the photosynthetic efficiency of PSII, which means the maximum photosynthetic potential of plant leaves (Dini-Papanastasi et al., 2012). Fv/Fm is typically between 0.75 and 0.85 for healthy plants, below which the plant is considered stressed (Baker and Oxborough, 2004; Johnson et al., 1993). *A. cochinchinensis* is judged to be relatively unstressed as its Fv/Fm value is close to 0.8, which is the index standard for photoinhibition. It is judged that there is no problem in forest cultivation using fiber grow bags.

Asparagine content in forest-grown *A. cochinchinensis* was about 9.9 times higher than in farm-grown *A. cochinchinensis*. Studies on the biosynthesis of secondary metabolites of medicinal plants by forest cultivation are scarce. The most studied medicinal plant in forest cultivation is mountain-cultivated ginseng. Mountain-cultivated ginseng has similar growth conditions and periods to ginseng grown in fields but is advantageous to antibiotic and abiotic stress and contains more ginsenosides (Fan et al., 2019; Xu et al., 2020). The reason that forest cultivated has higher secondary metabolite content than cultivated ginseng is considered a large environmental factor. Because medicinal plant secondary metabolite content is influenced by environmental factors, including climatic and ecological issues, possible changes in environmental conditions may cause problems in the production of some medicinal species (Pant et al., 2021). In this study, the content of amino acids, including asparagine, in forest-cultivated *A. cochinchinensis* was more than 8 times higher than that of farm-grown *A. cochinchinensis*. This is presumed because amino acids such as asparagine are involved in stress (Lea and Azevedo, 2007). In other words, compared to farm cultivation, forest cultivation is subjected to abiotic stress, such as dryness, low temperature and high temperature, and biological stress, such as pests and diseases.

Conclusion

The fiber grow bag is a non-woven flower pot, which is light and inexpensive, and has excellent durability, heat insulation, breathability and drainage. Forest cultivation of useful forest resource plants is incredibly interesting because it is very eco-friendly, but cultivation methods still need to be established. In particular, fiber grow bag cultivation can solve problems that occur in forest cultivation through early survival and competition with weeds. In this study, we investigated the optimal culture conditions using fiber grow bags for forest cultivation of *A. cochinchinensis*, a medicinal plant expected to be used as a biomaterial but with low productivity. It was found that productivity can be increased by selecting a fiber-to-grow bag of an appropriate size, planting it at an appropriate density, and cultivating it in an appropriate light environment. The results of this study show that a light environment is essential for forest cultivation of useful forest plants. However, more research is needed on the growth status of various edible and medicinal plants according to the light environment in the future. In conclusion, fiber grow bag cultivation can be applied to cultivating *A. cochinchinensis* and other useful forest resources.

Acknowledgments

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450 **Table 1.** Experimental site soil environment

pH	Total nitrogen content (%)	EC (μm/ms)	Exchangeable cation amount (cmol+/kg)				Available phosphate (g/kg)
			Ca^{2+}	Mg^{2+}	K^{+}	Na^{+}	
5.02	0.12	16.83	3.297	0.947	0.256	0.187	1.75

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452

453 **Table 2**. *A. cochinchinensis* root characteristics according to forest cultivation factors.

		Total root length (cm)	Surface area of roots (cm^2)	Volume of roots (cm^3)	Average diameter (mm)
Size of fiber grow bag	Large	139.98 ± 8.88^b	47.47 ± 2.16^a	1.28 ± 0.08^a	1.08 ± 0.07^a
	Medium	180.56 ± 1.89^a	51.17 ± 2.33^a	1.15 ± 0.05^a	0.9 ± 0.04^a
	Small	85.45 ± 5.60^c	22.39 ± 3.78^b	0.47 ± 0.11^b	0.83 ± 0.21^a
Planting density	Loose	220.13 ± 2.65^a	56.05 ± 2.66^a	1.14 ± 0.055^a	0.81 ± 0.02^b
	Normal	59.01 ± 1.85^c	24.45 ± 4.33^c	0.81 ± 0.025^b	1.32 ± 0.123^a
	Dense	129.81 ± 3.01^b	36.4 ± 2.61^b	0.81 ± 0.015^b	0.89 ± 0.033^b
Forest floor	Coniferous forest	87.29 ± 4.72^a	9.62 ± 0.04^b	0.78 ± 0.01^a	2.83 ± 0.02^a
	Broad-leaved forest	49.4 ± 5.11^b	9.86 ± 0.03^a	0.26 ± 0.01^c	1.57 ± 0.01^b
	Mixed forest	55.27 ± 2.96^b	9.65 ± 0.06^b	0.49 ± 0.02^b	1.58 ± 0.01^b
Slope	North	92.35 ± 3.52^a	9.63 ± 0.08^a	0.72 ± 0.01^a	4.22 ± 0.02^a
	South	59.28 ± 2.06^b	10.33 ± 0.47^a	0.54 ± 0.01^b	3.08 ± 0.17^b

454 Fiber grow bags used in the experiment were medium size (height 50 x diameter 40 cm). The planting density per
455 fiber grow bag was set as Loose (10 plants/bag), Normal (15 plants/bag), and dense (20 plants/bag).
456 The data are presented as means ± SD of three times of independent test. Means with different letters are
457 significantly different at a=0.05 by Duncan's multiple range test.
458

459

460 **Table 3.** Photosynthesis-related factors in forest cultivation using fiber grow bag.

Light compensation point ($\mu mol\ m^{-2}\ s^{-1}$)	Light saturation point ($\mu mol\ m^{-2}\ s^{-1}$)	Dark Respiration Rate ($\mu mol\ m^{-2}\ s^{-1}$)	Photosynthetic capacity ($\mu mol\ CO_2\ m^{-2}\ s^{-1}$)	Apparent quantum yield ($\mu mol\ mol^{-1}$)
64.06 ± 48.8	1933.33 ± 208.17	0.85 ± 0.35	4.41 ± 0.77	0.013

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463

464 **Table 4.** Chlorophyll fluorescence reaction of *A. cochinchinensis* planted in the fiber grow bag.

Chlorophyll fluorescence reaction	Values
Fo	2950.4 ± 1532.9
Fm	15110.8 ± 8136.4
Fv	12160.4 ± 6612.27
Fv/Fm	0.8 ± 0.01

465 *Fo: initial fluorescence yield, Fm: maximum fluorescence yield, Fv: variable fluorescence yield,
466 Fv/Fm: maximum quantum yield

467

Table 5. Asparagine and amino acid content of forest-cultivated and farm-cultivated *A. cochinchinensis*

Amino acid	Cultivation	
	Forest (mg/g)	Farm (mg/g)
Taurine	159.04±18.66	110.03±33.49
L-Threonine	20.85±5.37	-
L-Serine	66.78±17.58	-
L-Asparagine	2184.93±213.99[a]	219.21±57.67[b]
Glycine	14.92±2.93	-
L-Alanine	102.86±20.73[a]	15.44±4.68[b]
L-Citrulline	53.36±11.91	-
L-Valine	21.23±7.51	-
L-Isoleucine	42.63±19.38	-
L-Leucine	18.75±4.86	-
ɣ-Amino-n-butyric Acid	60.19±17.1[b]	156.42±46.26[a]
Ammonium Chloride	243.87±101.32	-
L-Ornithine	9.91±4.96	-
L-Histidine	32.84±8.77	-
L-Anserine	137.45±35.45	-
L-Arginine	182.76±13.95[a]	18.8±2.22[b]

Samples used for analysis were analyzed using medium-sized grow bags cultivated in the forest for 2 years.

[*]Mean ± standard deviation (n = 3), [**]No detection

The data are presented as means ± SD of three times of independent test. Means with different letters are significantly different at a=0.05 by Duncan's multiple range test.

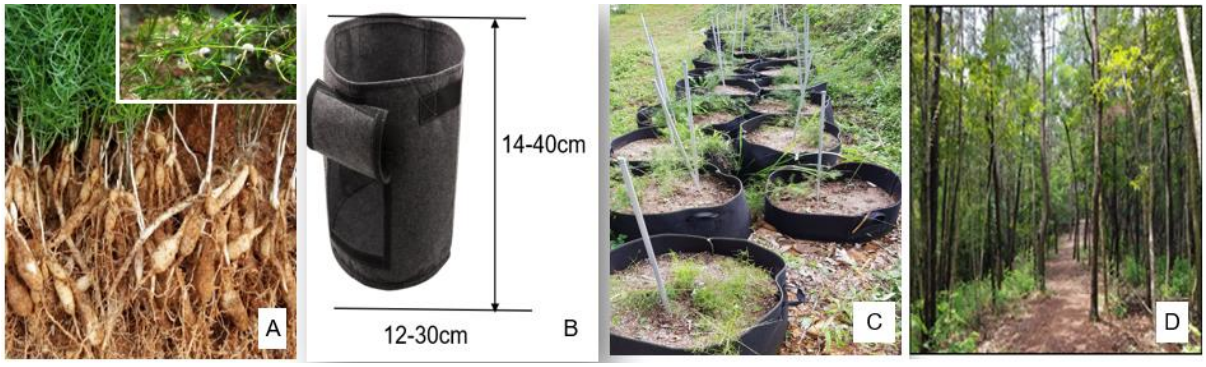

Fig. 1. The Grow-bag used for forest cultivation of *A. cochinchinensis* and the forest cultivation eexperiment site.

A: *A. cochinchinensis* plants, **B**: Fiber grow bag used in this study, **C**: Fiber grow bags planted with *A. cochinchinensis* plants, and **D**: A view of plantation in the forest

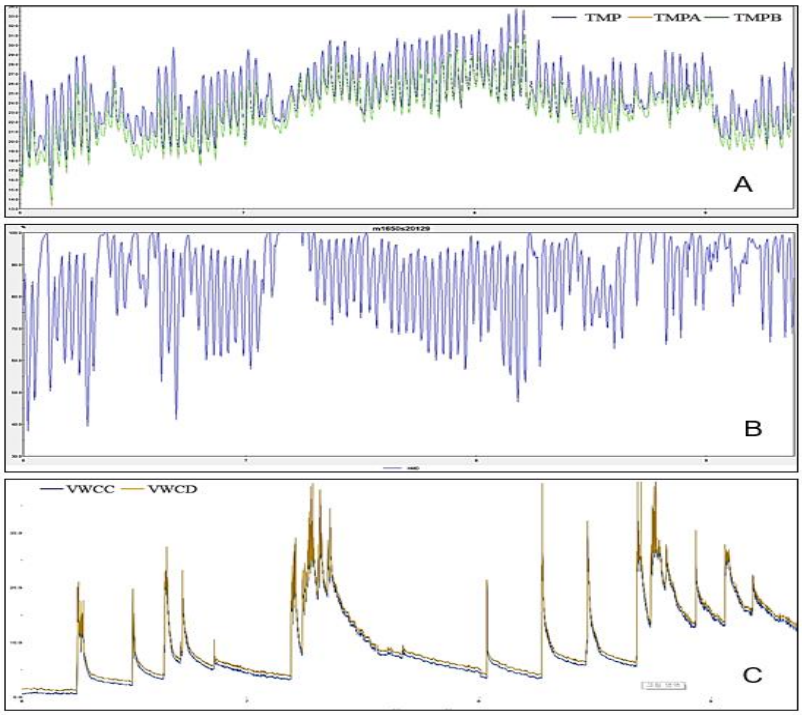

Fig. 2. Air and soil temperature (A), relative humidity changes (B), and aboveground and changes in moisture content of fiber grow bags and forest soils (C).

TMP: Air temperature, TMPA: Forest soil temperature, TMPB: Fiber grow bag soil temperature

VWCC: Forest Soil, VWCD: fiber grow bag Soil

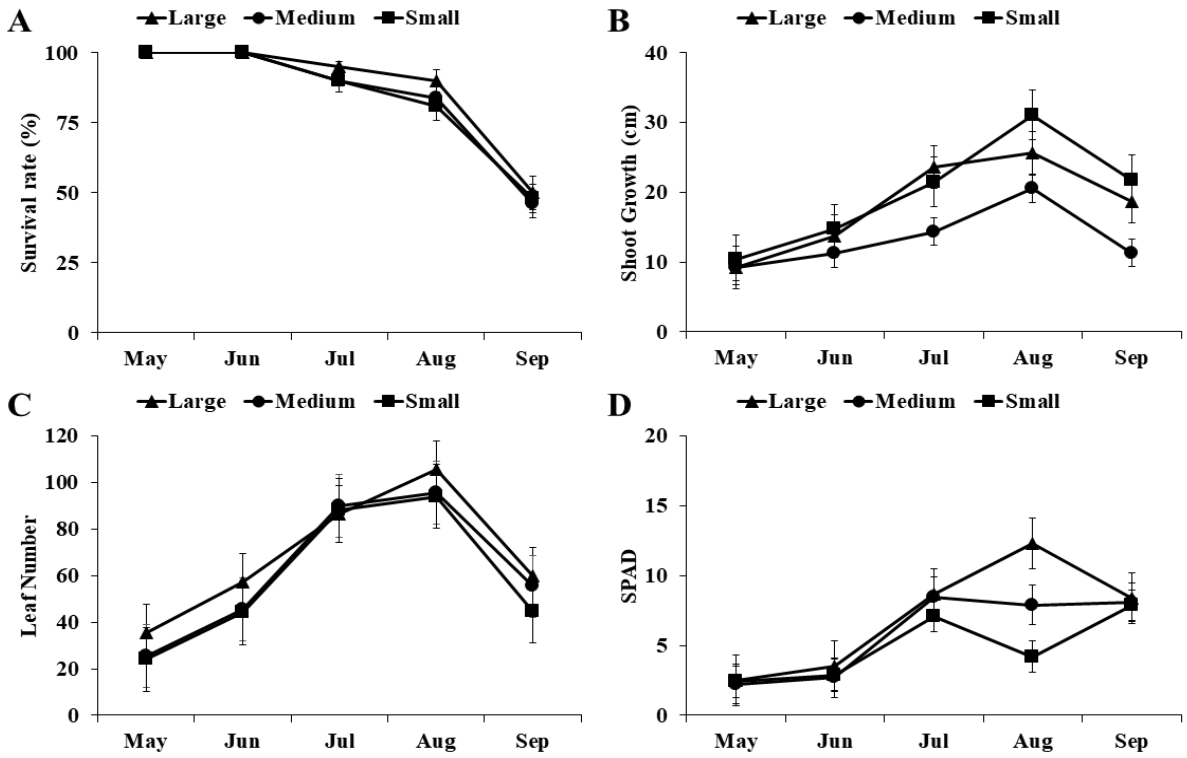

Fig. 3. Growth characteristics of the above-ground part of *A. cochinchinensis* according to the size of the fiber grow bag. **A**: survival rate, **B**: shoot growth, **C**: leaf number, and **D**: chlorophyll content Fiber grow bags used in the experiment were medium-size bag (height 30 × diameter 35 cm).

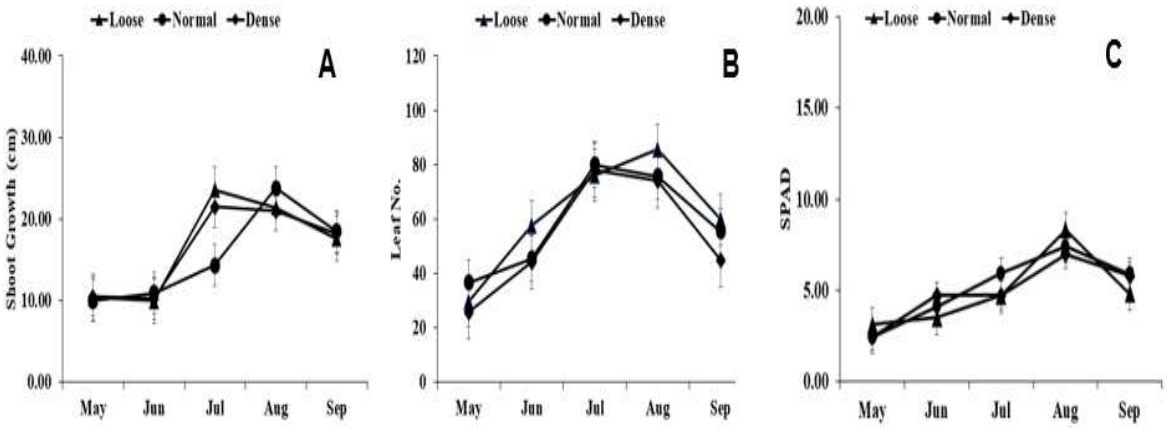

Fig. 4. Growth characteristics of the above-ground part of *A. cochinchinensis* according to planting density. **A**: shoot growth, **B**: leaf number, and **C**: SPAD (chlorophyll content)

Fiber grow bags used in the experiment were medium size (height 50 x diameter 40 cm) at south slope in mixed forest.

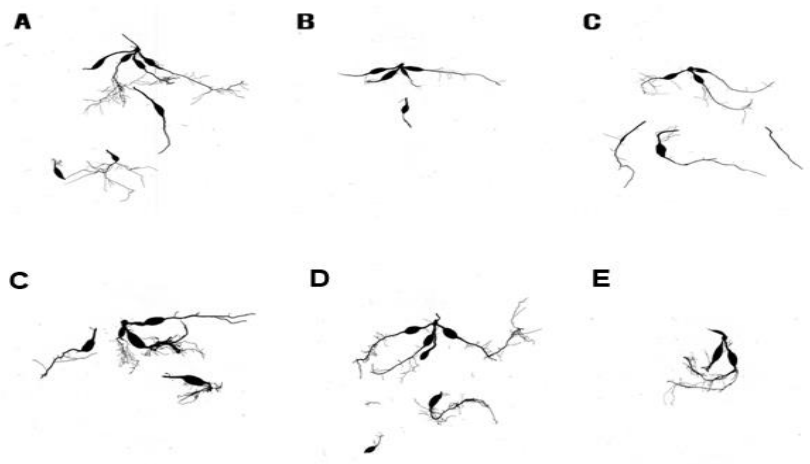

Fig. 5. *A. cochinchinensis* root growth according to fiber grow bag size and planting density. **A**: large, **B**: medium, **C**: small, **D**: dense, **E**: normal, and **F**: loose

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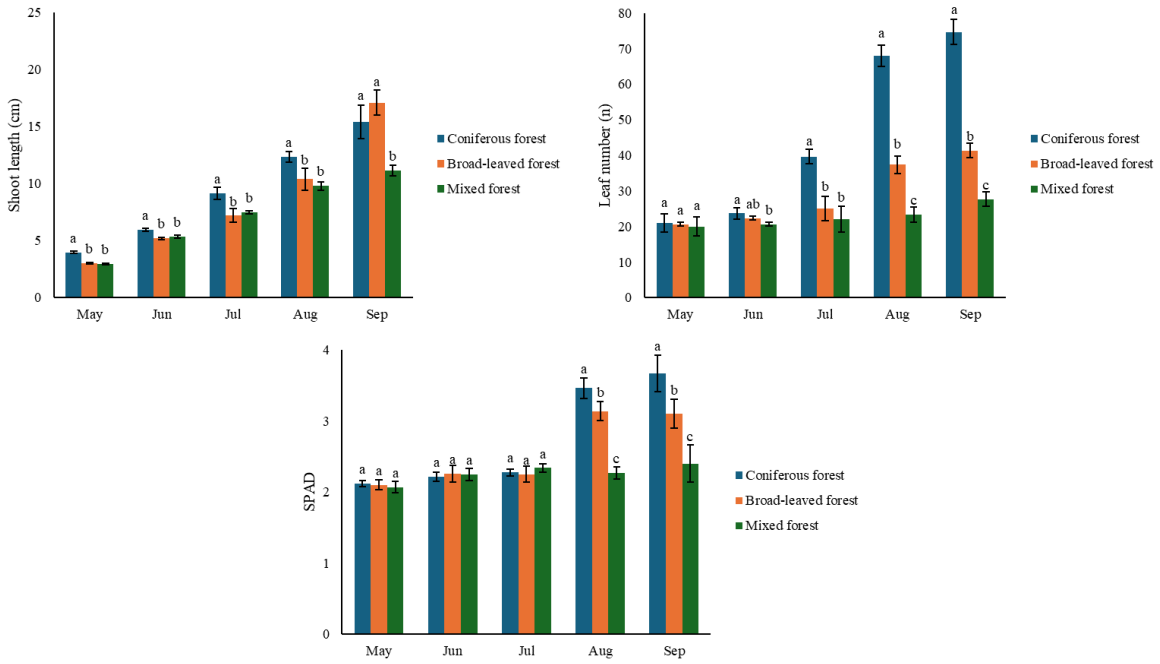

508

509 **Fig 6.** Growth characteristics of the above-ground part of *A. cochinchinensis* according to forest floor.

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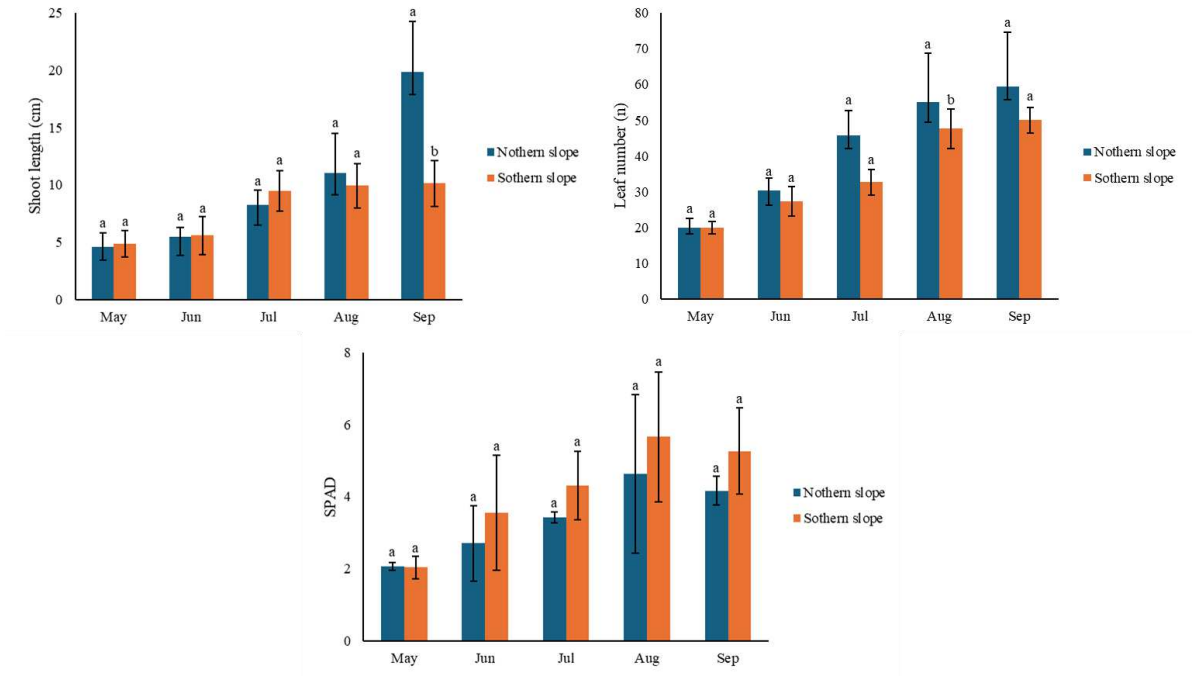

Fig. 7. Growth characteristics of the above-ground part of *A. cochinchinensis* according to planting slope.

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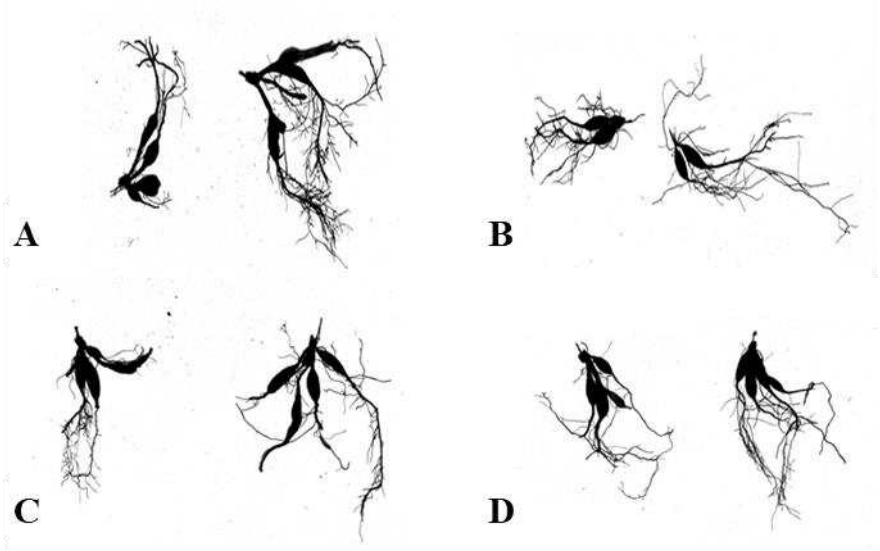

517

518 Fig. 8. *A. cochinchinensis* root growth according to floor and slope

519 (A: coniferous forest, B: broad-leaved forest, C: north slope, D: south slope)

520

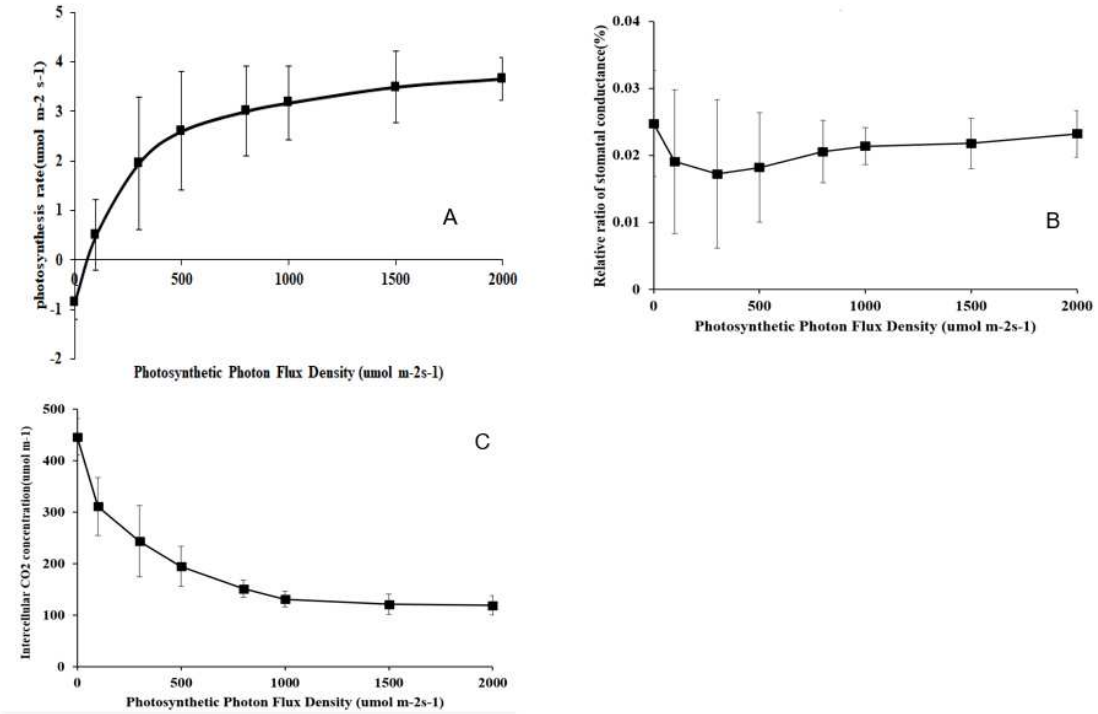

Fig. 9. Changes in photosynthesis in *A. cochinchinensis* according to light intensity. **A**: photosynthetic rate, **B**: stomatal conductivity and **C**: CO_2 change in mesophyll cells.

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