Heliyon 9 (2023) e12488

Contents lists available at ScienceDirect

Heliyon

journal homepage: www.cell.com/heliyon

Research article

Effects of two contrasting potting media on the leaf development index, photosynthetic rate, and metabolite profile of camphor (*Dryobalanops aromatica*) seedlings

Tsamarah Nur Rahmah [a], Fifi Gus Dwiyanti [b,c], Mohamad Rafi [c,d], Ulfah Juniarti Siregar [b], Iskandar Z. Siregar [b,c,*]

[a] *Graduate Study Program of Tropical Silviculture, Faculty of Forestry and Environment, IPB University (Bogor Agricultural University), IPB Darmaga Campus, Bogor 16680, West Java, Indonesia*
[b] *Department of Silviculture, Faculty of Forestry and Environment, IPB University (Bogor Agricultural University), IPB Darmaga Campus, Bogor 16680, West Java, Indonesia*
[c] *Advanced Research Laboratory, IPB University (Bogor Agricultural University), IPB Darmaga Campus, Bogor 16680, West Java, Indonesia*
[d] *Department of Chemistry, Faculty of Mathematics and Natural Sciences, IPB University (Bogor Agricultural University), IPB Darmaga Campus, Bogor 16680, West Java, Indonesia*

A R T I C L E I N F O

Keywords:
Dryobalanops aromatica
LC–MS/MS
Metabolite
Photosynthetic rate
Plastochron index (PI)

A B S T R A C T

Camphor (*Dryobalanops aromatica* C. F. Gaertn.) is a vulnerable tropical tree species that has been exploited for its timber as well as its resin, which is used for medicinal uses. The use of camphor in Indonesia is limited owing to the decreasing size of the species' population in its native habitat. Therefore, replanting programs have been encouraged for this species owing to its adaptability to mineral soils and shallow peatlands. However, experimental evidence of the effect of different growing media on morphology, physiology, and biochemistry is very limited, which is needed to evaluate the replanting program's success. Therefore, this study aimed to determine the responses of camphor (*D. aromatica*) seedlings grown in two different types of potting media i.e. mineral and peat, for 8 weeks of planting. In particular, the types of bioactive compounds produced in camphor leaves and their levels were assessed by analyzing their metabolite profiles. Leaf growth was evaluated morphologically using the plastochron index, while photosynthetic rates were measured with LI-6800 Portable Photosynthesis System. Metabolites were identified by using liquid chromatography-tandem mass spectrometry. The percentage of LPI of 5 or more was lower in the peat medium at 8% than in the mineral medium at 12%. The photosynthetic rate of camphor seedlings was 1–9 μmol CO_2 m^{-2} s^{-1}, with a higher rate in the peat medium than in the mineral medium, suggesting that the peat medium was better for growth. Lastly, the metabolomic analysis in the leaf extract revealed the presence of 21 metabolites, which were dominated by flavonoid compounds.

1. Introduction

Dryobalanops aromatica C. F. Gaertn. commonly known as camphor, is a tropical tree species from the Dipterocarpaceae family,

* Corresponding author.
E-mail address: siregar@apps.ipb.ac.id (I.Z. Siregar).

https://doi.org/10.1016/j.heliyon.2022.e12488
Received 11 November 2020; Received in revised form 16 October 2021; Accepted 12 December 2022
Available online 20 December 2022

which is native to Borneo and Sumatra islands [1, 2]. Camphor produces wood for carpentry with strength class II-I [3], and, it is also a source of non-timber forest products (NTFPs) such as crystals and essential oils, namely, borneol [4, 5]. Borneol is a premium compound that effectively melts blood clots in the heart and brain; reduces menstrual, muscle, and joint pain; and prevents germ cell proliferation [4, 6]. The high market demand for camphor wood products and their derivatives must be balanced against raw materials' supply and availability. However, the population of camphor trees has been decreasing annually owing to exploitation of the species and land conversion, resulting in the species being placed in the vulnerable category based on the International Union for Conservation of Nature Red List of Threatened Species assessment [7]. Therefore, the conservation of this species through a replanting program is urgently needed.

In natural forests, camphor trees grow in groups on dry sandy or gravel soils [8]. However, recent evidence suggests that this species also grows naturally in the shallow peat swamp forest in the Singkil Wildlife Reserve (Suaka Margasatwa Singkil), Aceh Province, Sumatera Islands, Indonesia [9]. These findings provide important baseline information for future replanting plans for this species. Therefore, studies regarding the adaptability of this species in two different habitats and how these differences affect the morphology, physiology, and biochemistry of plants are needed, given that the morphological, physiological, and biochemical characteristics of plants are correlated with their adaptive responses to environmental conditions, and these responses are based on the plant's adaptive strategies, stress response systems, and ecological strategies [10]. For instance, soil properties' differences may affect plants' growth rates and physiological responses because plants experience different types and levels of stress [11, 12]. This stress directly impacts plants' photosynthesis rates, which is accompanied by a decrease in transpiration rate and stomatal conductance. Each type of plant in different habitat has a different rate of photosynthesis depending on its habitat. Furthermore, plant photosynthates are translocated toward vegetative and generative growth, including leaf formation [13].

The question regarding how camphor plants adapt to different habits was initially addressed through RNA sequencing (RNA-Seq) transcriptome analysis of seedlings planted in mineral and peat media [9]. However, to the best of our knowledge, phenotypic and chemical compound analyses have not yet been carried out. Therefore, this study aimed to investigate the responses of camphor seedlings to different planting media by examining their morphological, physiological, and chemical traits, such as growth rate, photosynthetic rate, leaf development index, and metabolic profile. The findings of this study may be fundamental for formulating strategies to conserve camphor populations and to sustainably maximize the utilization of this species in the future. In addition, the findings may support various restoration programs in Indonesia in terms of selecting economically valuable species that are adaptive to peatlands.

2. Materials and methods

2.1. Plant materials and potting media

Because natural regeneration could not be found in the Singkil Wildlife Reserve, we collected camphor seedlings from Lae Kombih Forest Park, Aceh Province, Sumatera Island, Indonesia. A total of 78 one-year-old seedlings were collected and transported to the greenhouse of the Department of Silviculture, Faculty of Forestry and Environment, IPB University, Bogor, Indonesia. Subsequently, the seedlings were treated with two different potting media, mineral soil ($n = 39$) or peat soil ($n = 39$), for 8 weeks at the greenhouse.

The mineral soil was obtained from an area in the district of Dramaga, Bogor, West Java Province, Indonesia, whereas the peat soil was collected from the peatland hydrology units of Sungai Mendahara-Sungai Batanghari (Kesatuan Hidrologis Gambut Sungai Mendahara–Sungai Batanghari), East Tanjung Jabung Regency, Jambi Province, Indonesia. Then, the two types of soil were transported to the greenhouse of the Silviculture Department, Faculty of Forestry and Environment, IPB University, Bogor, Indonesia, and subsequently used for planting media.

The physical characteristics of the two soils, including soil pH, texture, and water content (%) for the mineral soil and pH, peat maturity level, and water content (%) for the peat soil, were assessed in the field. Soil pH was measured using pH test papers, and soil texture was assessed using the pipette method [14]. Soil water content was measured using the gravimetric method [15], and peat maturity was assessed using the handheld method [16]. Based on the results, the mineral medium was classified as clay loam with a pH of 5.0 and 32.09% water content, whereas the peat medium was classified as fibric peat with a pH of 4.0 and 135.32% water content [9, 13].

Soil quality indicators, such as C-organic, cation exchange capacity (CEC), and base saturation, significantly affect plant growth and photosynthetic rates [17, 18]. In the current study, however, the chemical indicators of the two soils, including C-organic (%), CEC (cmol/kg), and base saturation (%), were evaluated using secondary data from the previous experiment. The mineral soil had a C-organic value of 2.43% (medium), base saturation of 10% (very low), and CEC of 10.21 cmol/kg (low), while the peat soil had a C-organic value of 15.22% (very high), base saturation of 6.15% (very low), and CEC of 16.40 cmol/kg (very high) [19, 20].

The mineral soil and dry peat were sieved using a 1×1-m screen with a galvanized 8×8 square wire mesh and a 1.00-mm sieve opening to remove the rocks and other soil impurities to obtain fine soils for use as planting media. Next, the seedlings were planted in 25 cm round pots with fine mineral or peat medium without fertilizer. Acclimatization of the seedlings was conducted in a closed, dark, and humid location inside the greenhouse for 1 month. Afterward, the seedlings were moved to a place with sufficient sunlight exposure. The seedlings were watered every evening.

2.1.1. Growth rate measurement

The growth development of the camphor seedlings was evaluated based only on the seedling's height recorded using a measuring tape. In addition, the number of leaves on each seedling was counted. The measurements were performed in the first and eighth weeks after planting.

2.2. Photosynthetic rate, transpiration rate, and stomatal conductance measurement

The photosynthetic rate, transpiration rate, and stomatal conductance of five randomly selected camphor seedlings from both mineral and peat media were measured between 09:00 and 10:00 a.m. The measurements were performed using healthy leaves with the LI-6800 Portable Photosynthesis System instrument (LI-COR Biosciences, Lincoln, NE, USA).

2.2.1. Leaf development index measurement

Leaf development was measured using the plastochron index (PI) and leaf PI (LPI). PI was used to estimate the time in terms of the total plastochron that has passed the reference length (λ) in growing plant populations. LPI was used to estimate leaf age based on morphological development, and it was expressed relative to the reference length [21]. The measurements were carried out on 5 leaves from 5 individual seedlings grown in each medium, using a digital caliper for every 2 days for a total of 20 days. A total of 50 leaves were used in this study. PI and LPI were calculated using the following formulas [21]:

$$\mathrm{PI} = \mathrm{n} + \frac{logLn - log\ \lambda}{logLn - logLn + 1}$$

$$\mathrm{LPI} = \mathrm{PI} - \tilde{\mathrm{n}}$$

where *n* represents the sequential index number calculated from the base of the shoot and has a length greater than or equal to the reference length (λ) of 20 mm; logLn is the logarithm of leaf length n, and logLn +1 is the logarithm of leaf length *n* that is less than λ [21]. Finally, leaf dimensions were measured according to the method described by Kremer *et al.* [22].

2.2.2. Metabolite separation and identification

Metabolites were analyzed at Advanced Research Laboratory, IPB University, Bogor, Indonesia. Two upper leaves collected from five randomly selected camphor seedlings were used for metabolite identification. The leaves were cleaned, sprayed with liquid nitrogen in a vacuum sealer to minimize contact with air and stored in a freezer at −80 °C. Next, the dried leaves were extracted using ultrasonication. A sample was splashed with liquid nitrogen to facilitate the grinding process, and 100 mg of the ground sample was then immersed in 1 mL of 70% ethanol. Ultrasonication extraction was carried out at 28 °C for 30 min using 5 repetitions. Afterward, the extract obtained underwent liquid chromatography-tandem mass spectrometry (LC–MS/MS) with the use of a Vanquish Flex Ultra High–Performance Liquid Chromatography (UHPLC) tandem Q Exactive Plus Orbitrap–High Resolution Mass Spectrometer (Thermo Scientific, Germany) for metabolite identification.

The metabolites were separated using a UHPLC converter for the mobile phase of LC–MS/MS, according to the method described by Jalal *et al.* [23]. The optimal mobile phase used was a ratio of water (A) and acetonitrile (ACN) (B). The ACN composition was as follows: 5%–25% B (0–3.500 min), 25%–40% B (3.500–4.733 min), 40%–75% B (4.733–6.933 min), 75%–100% B (6.933–7.800 min), 100%–5% B (7.800–8.233 min), and 5% B (8.233–14.000 min). The columns (Accucore ™; 100 × 2.1 mm × 2.6 μm) were run with a phenyl stationary phase injection volume of 0.2 μL and a flow rate of 0.080 mL/min at 35 °C. The data obtained were analyzed using Compound Discoverer 2.1 SP 1. The metabolites were identified using an untargeted approach and an online database. Lastly, the m/z tolerance of 5 ppm and intensity tolerance of 30% was applied to detect unknown compounds.

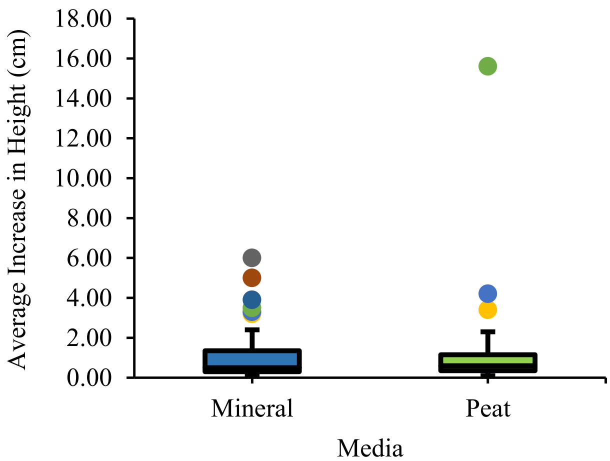


Figure 1. The average increase in height of the camphor seedlings grown in the mineral and peat media.

2.3. Data analysis

The distribution of growth data on the seedlings' height, diameter, and number of leaves was examined using descriptive analysis in the form of a graphs and boxplot diagrams using Microsoft Excel 2016. In addition, the effect of the media on the growth of camphor seedlings was analyzed by correlation analysis and a *t*-test, using SPSS 16.0 [24].

3. Results and discussion

3.1. Seedling growth performance

Photosynthate distribution in growing plants may be influenced by the ability of sink organs to attract photosynthate (sink strength) and by environmental factors [25, 26]. Seedling growth is indicated by the increase in the plant's height and diameter and the number of its leaves. The average increase in height of camphor seedlings planted in the peat medium was 1.28 cm, which was higher than that in the mineral medium at 1.18 cm (Figure 1). The average increase in seedlings' diameter in the peat medium was 0.49 mm, which was also higher than that in the mineral medium at 0.45 mm (Figure 2). Meanwhile, the average increase in the number of leaves of seedlings in the mineral media was 6 leaves, which was more than that of seedlings in the peat medium with 5 leaves (Figure 3). However, the increase in height, diameter, and leaf number of camphor seedlings grown in the peat and mineral media were not morphologically significant. Nevertheless, some seedlings grown in the peat medium tended to be taller and have larger diameters than those grown in the mineral medium, likely due to a higher rate of photosynthesis in the peat medium. As previously noted, a higher photosynthetic rate produces more photosynthates for growth [27]. In addition, the leaves of the camphor seedlings grown in the peat medium (Figure 4a) appeared darker and duller but larger than those of seedlings grown in the mineral medium (Figure 4b).

The availability of adequate nutrients, as well as differences in the acidity and soil absorption of certain nutrients, may affect the nutrient absorption by plants [28]. According to the *t*-test analysis, the two media in the current study had no significantly different effects on the growth performance of camphor seedlings in terms of height, diameter, and leaf number (Table 1), presumably because both media provided the same or sufficient nutrients to the seedlings. The camphor seedlings grown in the peat and mineral media demonstrated the same potential for increases in the height, diameter, and leaf number.

3.1.1. Leaf development index

Among the 5 camphor seedlings grown in the peat medium, the greatest average leaf length was found in individual G1 (75.3 mm) and the lowest was found in individual G3 (24.4 mm) (Figure 5). Meanwhile, the average leaf length of 5 camphor seedlings in the mineral medium was highest in individual M3 (82.3 mm) and the lowest was found in individual M1 (51.2 mm) (Figure 6). However, the leaf length was not significantly different between seedlings grown in peat and mineral media.

Results from PI analysis showed that the plastochron intervals had a linear pattern, while the LPI was calculated to determine leaf sizes and leaf development age based on the index length. Previous research showed that young leaves have an LPI value of 3–5, while fully developed (mature) leaves have LPI values between 10 and 14 [29]. The PI of the seedlings grown in the peat and mineral media with the reference length (λ) of 20 mm ranged from 4 to 6. The percentage of camphor leaves with an LPI of more than 5 was 8% in the peat media (Figure 7a), which was less than that in the mineral media at 12% (Figure 7b). Therefore, the leaves of camphor seedlings in both media were relatively young and still developing. Leaf age can affect plants' photosynthetic stage, particularly during gas exchange. Consequently, the limitations of photosynthetic induction are significantly higher in mature leaves than in young leaves.

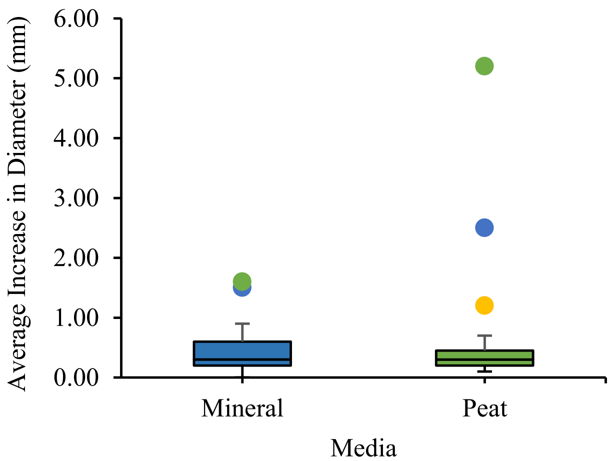


Figure 2. The average increase in diameter of camphor seedlings grown in the mineral and peat media.

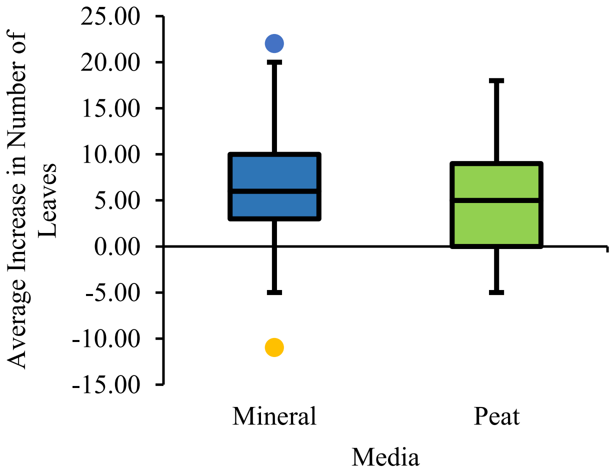


Figure 3. The average increase in the number of leaves of the camphor seedlings grown in the mineral and peat media.

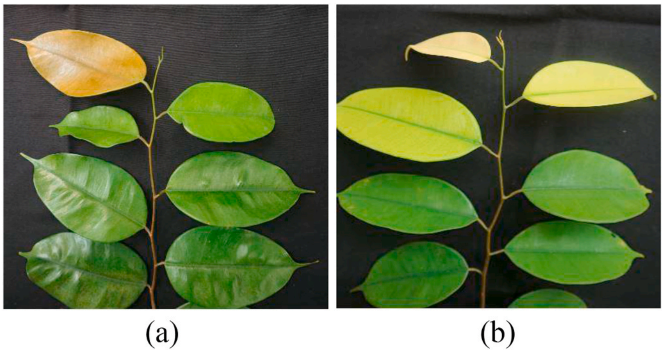


Figure 4. Comparison of the color of the leaves of the camphor seedlings grown in the peat (a) and mineral (b) media.

Table 1
The *t*-test analysis of the effect of the peat and mineral media on camphor seedling growth performance.

Growth Parameter	P-value
Height	0.461^{ns}
Diameter	0.371^{ns}
Leaf number	0.370^{ns}

ns, P-value >0.05, indicating a lack of significant difference at the 95% confidence interval.

3.1.2. Photosynthetic rate

The photosynthetic rates of the camphor seedlings grown in the peat and mineral media varied. Individual camphor seedlings planted in the peat medium had a higher photosynthetic rate than those in the mineral medium (Figure 8). The photosynthesis rate of camphor measured as the light energy (Q. leaves) absorbed was within a range of 190–440 μmol CO_2 m^{-2} s^{-1}. Meanwhile, the photosynthesis rate of camphor ranged from 1 to 9 μmol CO_2 m^{-2} s^{-1} (Figure 8), indicating that it is a slow-growing species. According to previous studies, the photosynthetic rate of trees in the tropics is 3–30 μmol CO_2 m^{-2} s^{-1}, and the photosynthetic rate of fast-growing species is higher than that of slow-growing species [30, 31]. Growth media can affect a plant's photosynthetic rate, leaf age, transpiration rate, and stomatal conductance, thus influencing its photosynthesis and development [32, 33, 34].

Stomatal conductance, which tends to be high, is in line with a high transpiration rate and impacts the photosynthetic rate. The stomatal conductance and transpiration rate were higher in camphor seedlings grown in the peat medium than those grown in the mineral medium. In this study, stomatal conductance was 0.15933 mol m^{-2} s^{-1} in seedlings in the peat medium, higher than that of seedling in the mineral medium at 0.03571 mol m^{-2} s^{-1} (Figure 9). High stomatal conductance can increase carbon dioxide (CO_2) diffusion, thus increasing the performance of the RuBisCo during carboxylation reaction and likely increasing the rate of

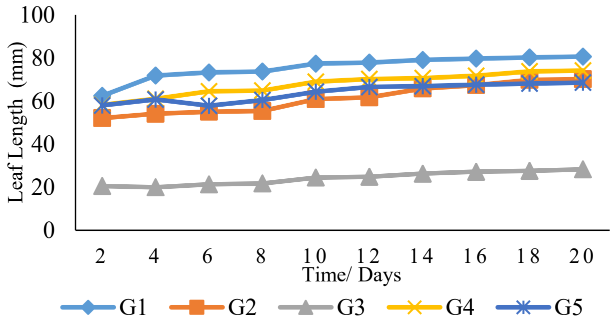


Figure 5. Average leaf length of five camphor seedlings grown in the peat medium.

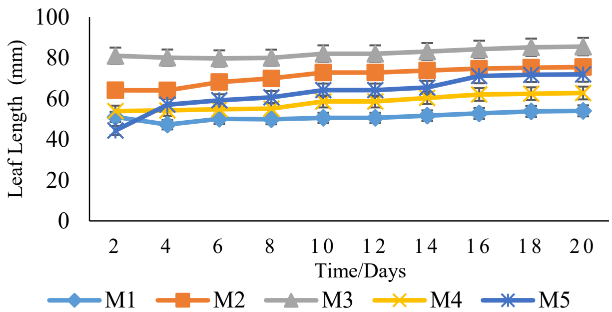


Figure 6. Average leaf length of five camphor seedlings grown in the mineral medium.

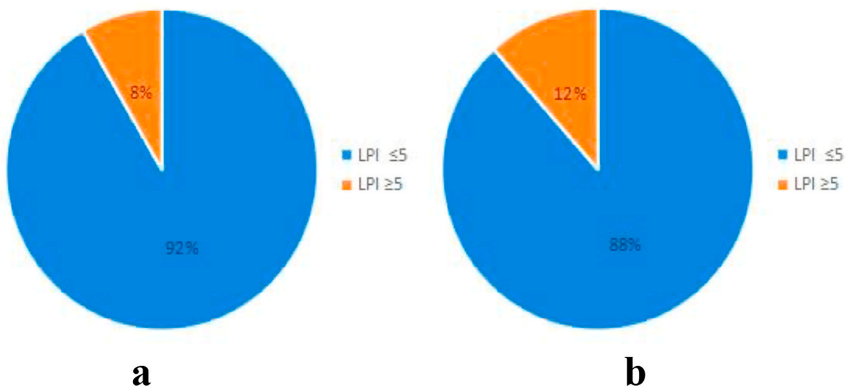


Figure 7. Percentage of camphor seedlings grown in the peat (a) and mineral (b) media with different leaf plastochron indexes (LPIs).

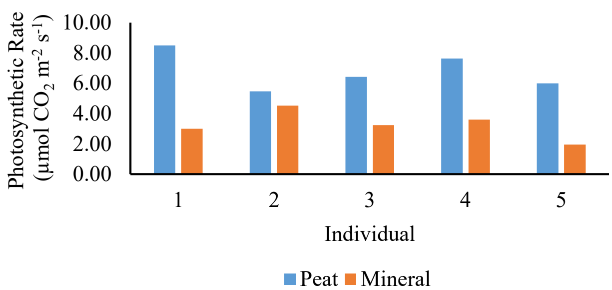


Figure 8. Photosynthetic rates of camphor seedlings grown in the peat and mineral media.

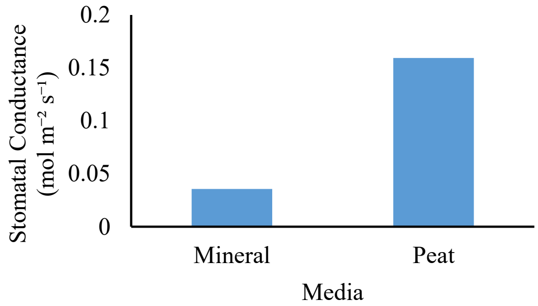


Figure 9. Stomatal conductance of camphor seedlings grown in mineral and peat media.

photosynthesis [35, 36].

The highest transpiration rate in the camphor seedlings was found in the peat medium at 0.0037 mol m^{-2} s^{-1} and the lowest transpiration rate was found in the mineral medium at 0.0006 mol m^{-2} s^{-1} (Figure 10). The transpiration rate is closely associated with water loss in the stomata. Water loss under an open-stomatal condition is higher than CO_2 absorption [37]. High CO_2 absorption can lead to more carbohydrate production. A plant's transpiration rate has a significant linear relationship with its photosynthetic rate and the 2 factors increase concurrently. In this study, the high transpiration rate of the seedlings grown in the peat medium affected the plants' photosynthetic rate.

Photosynthetic rates, transpiration rates, and stomatal conductance values were significantly different between camphor seedlings grown in the peat and mineral media (Table 2), suggesting that these leaf characteristics were affected by media. In addition, the peat medium was better for planting camphor seedling than the mineral medium in terms of the characteristics as mentioned above. The most striking differences between the peat and mineral media were associated with their pH, water content (%), peat maturity level, C-organic (%), and CEC (cmol/kg). These parameters significantly affect the availability of nutrients and water, important factors for photosynthesis.

The peat medium (pH = 4.0) was more acidic pH than the mineral medium (pH = 5.0). The acidity of fibric peat is generally known to be limited by basic cations from decomposing organic matter. Moreover, the water content of the peat medium was 135.32%, which was higher than that of the mineral medium (32.09%). This difference was likely due to high organic matter content of peat, which enables it to absorb more water. As an example, polar OH-phenol and hydrophilic cellulose in fibric peat absorb substantial amount of water from the soil. A lack of water can cause stomatal closure and damage to the chlorophyll and thylakoid membranes, thus inhibiting carbon dioxide (CO_2) absorption and decreasing transpiration and stomatal conductance [38, 39].

Peat maturity level is defined based on the level of decomposition of organic matter, the main component in affecting the fertility of peat soils and the availability of nutrients, and thus the determining factor in the productivity level of peatlands. The level of peat maturity is classified as fibric, which is the least mature; hemic, which is more mature; or sapric, which is the most mature [40]. The soil fertility and nutrient availability in fibric peatlands are lower than those in mature, hemic or sapric peat [41]. Finally, the C-organic and CEC in the peat medium were higher than those in the mineral medium, suggesting a higher level of soil fertility in the peat medium.

3.1.3. Metabolite identification

The identification of the metabolites in camphor leaf extract was conducted using an untargeted metabolomics approach and identifying all the metabolites detected by LC–MS/MS. The LC–MS/MS analysis yielded a chromatogram with bands resulting from the separation of camphor leaf metabolites. The chromatograms of camphor leaf extract with positive (Figure 11a) and negative (Figure 11b) ionizations had similar patterns, although intensities differed. The chromatograms were processed using the Compound Discoverer software, detecting 21 metabolites (Table 3). Then, the putative identification of metabolites was obtained from MS2 fragmentation, followed by comparison and confirmation based on compound-centric databases such as ChemCalc [42], METLIN [43], MassBank [44], mzCloud [45], and LIPID MAPS [46].

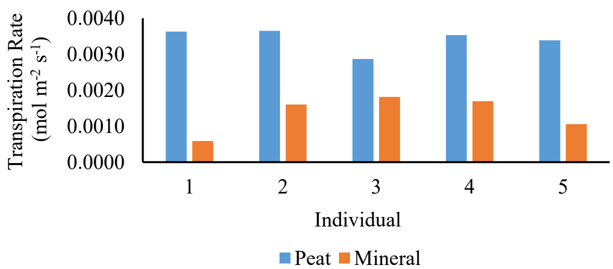


Figure 10. Transpiration rate of camphor seedlings grown in the peat and mineral media.

Table 2
The *t*-test analysis of the effect of the peat and mineral media on the photosynthetic parameters in camphor seedlings.

Parameter	P-value
Photosynthetic rate	0.001*
Transpiration rate	0.001*
Stomatal conductance	0.000*

* Significantly different at P-value >0.05 at the 95 % confidence interval.

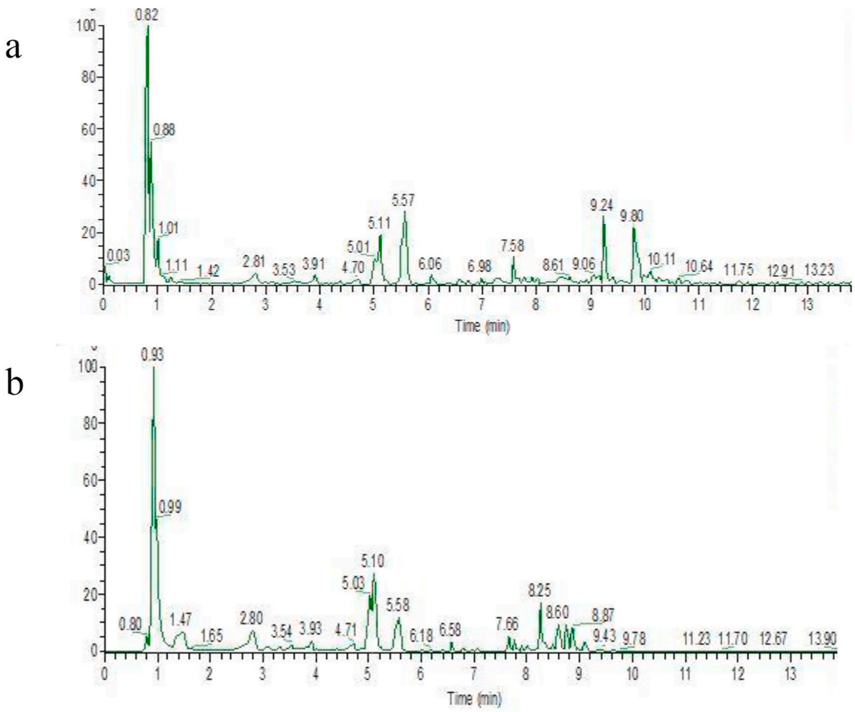


Figure 11. LC–MS/MS chromatogram with positive ionization (a) and negative ionization (b) in the camphor leaf extract.

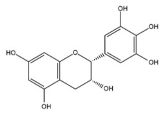
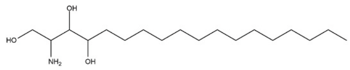
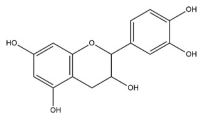
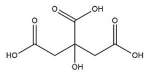
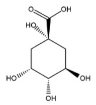
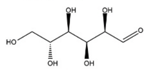
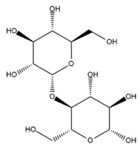
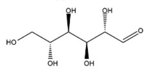
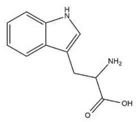
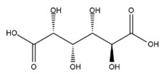
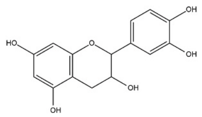
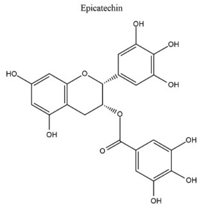
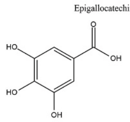
The identified compounds were classified into several groups, including amino acids, phenols, flavonoids, lipids, and sugars. These compounds were primary and secondary metabolites produced by plants in a certain amount under the condition of gripping. The identified amino acid compounds included Methionine sulfoxide, DL-tryptophan, and L-glutamic acid. The identified phenols were D-(−)-Quinic acid, Gallic acid, and Syringic acid. Meanwhile, the flavonoids identified were (−)-Epigallocatechin, Catechin, Citric acid, Epicatechin, Epigallocatechin gallate, Mearnsitrin, Quercetin-3β-D-glucoside, and *trans*-aconitic acid. The only lipid compound identified was 2-Amino-1,3,4-octadecanetriol. Several sugars were also identified: D-(+)-Glucose, D-(+)-Maltose, D-(+)-Mannose, D-Saccharic acid, L-Iditol, and α-Lactose. The identification procedure using LC−MS/MS did not successfully detect borneol compounds in the camphor leaf extract. Previously, borneol, which is volatile, was successfully identified in camphor leaf extracts using GC−MS [47]. Borneol is not extracted into ethanol solvents because it tends to be nonpolar.

The research on camphor's responses to two different growth media in terms of growth performance, leaf development index, and metabolite identification was conducted on seedlings grown at a greenhouse scale to estimate the growth response when the seedlings are planted in the field. Therefore, field-scale studies are needed to ascertain how these camphor seedlings respond to two media. In addition, it is also necessary to further optimize the extraction method of camphor metabolites, particularly borneol compounds, using ethyl acetate and n-hexane, considering that borneol compounds were not detected in this study.

4. Conclusions

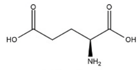
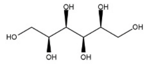
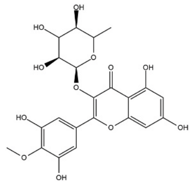
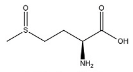
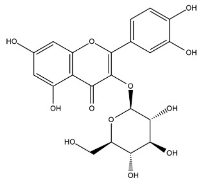
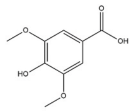
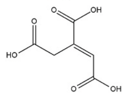
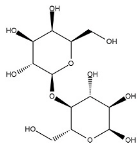
The camphor seedling leaves in the peat and mineral media were relatively young and still developing; the percentage of the leaves with an LPI of more than 5 in peat medium was 8%, which was lower than that of the leaves of seedlings grown in the mineral medium (12 %). The photosynthetic rate of the camphor seedlings in the peat and mineral media varied. The photosynthetic rate, ranging from

Table 3
Identification of metabolites (positive and negative ionization modes) in camphor leaf extract.

No	Structure	Compound	Formula	Category
1	 (-)-Epigallocatechin	(−)-Epigallocatechin	C15 H14 O7	Flavonoid
2	 2-Amino-1,3,4-octadecanetriol	2-Amino-1,3,4-octadecanetriol	C18 H39 NO3	Lipid
3	 Catechin	Catechin	C15 H14 O6	Flavonoid
4	 Citric acid	Citric acid	C6 H8 O7	Flavonoid
5	 D-(-)-Quinic acid	D-(−)-Quinic acid	C7 H12 O6	Phenol
6	 D-(+)-Glucose	D-(+)-Glucose	C6 H12 O6	Monosaccharide
7	 D-(+)-Maltose	D-(+)-Maltose	C12 H22 O11	Disaccharide
8	 D-(+)-Mannose	D-(+)-Mannose	C6 H12 O6	Monosaccharide
9	 DL-Tryptophan	DL-Tryptophan	C11 H12 N2 O2	Amino acid
10	 D-Saccharic acid	D-Saccharic acid	C6 H10 O8	Polysaccharide
11	 Epicatechin	Epicatechin	C15 H14 O6	Flavonoid
12	 Epigallocatechin gallate	Epigallocatechin gallate	C22 H18 O11	Flavonoid
13	 Gallic acid	Gallic acid	C7 H6 O5	Phenol

(*continued on next page*)

Table 3 (*continued*)

No	Structure	Compound	Formula	Category
14	 L-Glutamic acid	L-Glutamic acid	C5 H9 N O4	Amino acid
15	 L-Iditol	L-Iditol	C6 H14 O6	Monosaccharide
16	 Mearnsitrin	Mearnsitrin	C22 H22 O12	Flavonoid
17	 Methionine sulfoxide	Methionine Sulfoxide	C5 H11 N O3 S	Amino acid
18	 Quercetin-3-beta-D-glucoside	Quercetin-3β-D-glucoside	C21 H20 O12	Flavonoid
19	 Syringic acid	Syringic acid	C9 H10 O5	Phenol
20	 trans-Aconitic acid	*trans*-Aconitic acid	C12 H22 O11	Disaccharide
21	 alpha-lactose	α-Lactose	C12 H22 O11	Disaccharide

1 to 9 μmol CO_2 m^{-2} s^{-1}, was higher in the peat medium than in the mineral medium, indicating that planting camphor in the peat medium enables better growth than in the mineral medium. Therefore, camphor represents a species that can be recommended for restoration program on peatlands. Although the analysis of camphor leaf extract using LC–MS/MS was not successful in detecting borneol, a typical compound in camphor; however, the same technique succeeded in identifying 21 other metabolites.

Declarations

Author contribution statement

Tsamarah Nur Rahmah: Performed the experiments; Analyzed and interpreted the data.
Fifi Gus Dwiyanti: Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.
Mohamad Rafi: Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.
Ulfah J Siregar: Conceived and designed the experiments; Wrote the paper.
Iskandar Z Siregar: Conceived and designed the experiments; Contributed reagents, materials, analysis tools or data; Wrote the paper.

Funding statement

This research was supported by the Basic Research Scheme 2019–2020 [No: 3/E1/KP.PTNBH/2019 and No: 1/AMD/E1.KP.PTNBH/2020] awarded by the Ministry of Research and Technology/National Agency for Research and Innovation (RISTEK-BRIN) of the Republic of Indonesia and the National Competitive Basic Research Scheme 2022 [No. 001/E5/PG.02.00PT/2022] awarded by Ministry of Education, Culture, Research and Technology of Republic of Indonesia.

Data availability statement

Data included in article/supplementary material/referenced in article.

Declaration of interests statement

The authors declare no competing interest.

Additional information

No additional information is available for this paper.

Acknowledgements

The authors are grateful to Mr. Abdul Rahman Ali from Subulussalam, Aceh, for donating the *Dryobalanops aromatica* seedlings and Mr. Riya Kamba from Subulussalam, Aceh, for assisting in transporting the seedling from Subulussalam to Bogor. We further appreciate Ms. Ridha Wati and Ms. Nur Fadila Kustiyarini for maintaining the seedlings in the greenhouse of the Department of Silviculture, Faculty of Forestry and Environment, IPB University. The authors also acknowledge Advanced Research Laboratory (ArLab), IPB University, for facilitating access to the analytical equipment.

References

[1] C.F. Symington, Foresters' manual of dipterocarps, in: Revised by Ashton PS; Appanah P). Malayan Forest Records No. 16, second ed., Forest Research Institute Malaysia, Kuala Lumpur, 2004, p. 519.
[2] F.G. Dwiyanti, K. Kamiya, K. Harada, Phylogeographic structure of the commercially important tropical tree species, *Dryobalanops aromatica* gaertn. f. (Dipterocarpaceae) revealed by microsatellite markers, REINWARDTIA 14 (2014) 43–51.
[3] [P3HH] Pusat Penelitian dan Pengembangan Hasil Hutan, Practical Guidelines for Basic Characteristics of Indonesian Wood Types, P3HH and ISWA ITTO Project, Jakarta, ID, 2015.
[4] Gusmailina. Borneol—future potential of essential oils. Prosiding Seminar Nasional Masyarakat Biodiversitas Indonesia 1, 259-264.
[5] A. Aswandi, C.R. Kholibrina, New insights into Sumatran camphor (*Dryobalanops aromatica* Gaertn) management and conservation in western coast Sumatra, Indonesia, IOP Conf. Ser. Earth Environ. Sci. 739 (2021), 012061.
[6] Q.L. Zhang, B.M.M. Fu, Z.J. Zhang, Borneol a novel agent that improves central nervous system drug delivery by enhancing blood-brain barrier permeability, Drug Deliv. 24 (2017) 1037–1044.
[7] M. Barstow, A. Randi, Dryobalanops aromatica (errata version published in 2020). The IUCN Red List of Threatened Species 2018: e.T61998024A173026192, 2018. Downloaded on 10 October 2021.
[8] M.F. Newman, P.F. Burgess, T.C. Whitemore, Pedoman Identifikasi Pohon-Pohon Dipterocarpaceae: Pulau Kalimantan, Prosea Indonesia, Bogor, ID, 1999, p. 151.
[9] I.Z. Siregar, F.G. Dwiyanti, U.J. Siregar, D.D. Matra, De novo assembly of transcriptome dataset from leaves of *Dryobalanops aromatica* (Syn. *Dryobalanops sumatrensis*) seedlings grown in two contrasting potting media, BMC Res. Notes 13 (2020) 405.
[10] A.R. Dyer, D.E. Goldberg, R. Turkington, C. Sayre, Effects of growing conditions and source habitat on plant traits and functional group definition, Funct. Ecol. 15 (2001) 85–95.
[11] S.A. Anjum, X. Xie, L. Wang, M.F. Saleem, C. Man, W. Lei, Morphological physiology and biochemical responses of plants to drought stress, J. Afr. Agri. Res. 6 (2011) 2026–2032.
[12] R.O. Erickson, F.J. Michelini, The plastochron index, Am. J. Bot. 44 (1957) 297–305.
[13] I.Z. Siregar, N.F. Kustiyarini, R. Wati, R.R. Rachmat, U.J. Siregar, F.G. Dwiyanti, Vegetative propagation of *Dryobalanops sumatrensis* and *Dryobalanops oblongifolia* subsp. oblongifolia by shoot cuttings, IOP Conf. Ser. Earth Environ. Sci. 394 (2019), 012029.
[14] F. Agus, S. Yusrial Sutono, Determination of soil texture in Indonesia, in: U. Kurnia, F. Agus, A. Adimihardja Dariah (Eds.), Soil Physical Properties and Analysis Method in Indonesia, Balai Besar Litbang Sumberdaya Lahan Pertanian, Bogor, Indonesia, 2006.
[15] A. Abdurrahman, U. Haryati, I. Juarsah, Determination of groundwater content by gravimetry method in Indonesia, in: U. Kurnia, F. Agus, A. Adimihardja Dariah (Eds.), Soil Physical Properties and Analysis Method in Indonesia, Balai Besar Litbang Sumberdaya Lahan Pertanian, Bogor, Indonesia, 2006.
[16] Wahyunto, R. Sofyan, H. Suparto Subagyo, Distribution and Carbon Content of Peatlands in Sumatra and Kalimantan, Wetland International Indonesian Program, Bogor, ID, 2004.
[17] R. Prabowo, R. Subantoro, Soil analysis as an indicator of the fertility level of agricultural cultivation land in the city of Semarang, Cendekia Eksakta 2 (2017) 59–64.
[18] H. Yulina, W. Ambarsari, Relationship between N-total and C-organic content of soil on harvesting weight of pakcoy plants after combining with municipal waste compost and cow manure in alluvial, Indramayu, Agro Wiralodra J. 4 (2021) 25–30.
[19] Y.H. Milasari, C. Kusmana, Muhadiono, Earthworm as Bioindicator for Soil Fertility within Land Cover Types in Bogor Dramaga, IPB University, Bogor, Indonesia, 2013 [Thesis].
[20] S.W. Budi, A. Sukendro, S.H. Pasaribu, A.D. Nurhayati, F.G. Dwiyanti, A.D. Dwisutono, T. Purwanegara, H. Muhtahdin, Restoration of Peatlands with an Applied Nucleation Concept Approach in the KHG Mandahara River-Batanghari River, East Tanjung Jabung District, Jambi Province, Peat Restoration Agency and IPB University, Bogor, Indonesia, 2017.
[21] O.E.A. Ademilua, C.E.J. Botha, A re-evaluation of plastochron index determination in peas-a case for using leaveslet length, South Afr. J. Bot. 71 (2005) 76–80.
[22] A. Kremer, J.L. Dupouey, J.D. Deans, J. Cottrell, U. Csaikl, R. Finkeldey, S. Espinel, J. Jensen, J. Kleinschmit, B.V. Dam, et al., Leaves morphological differentiation bertwen *Quercus robur* and *Quercus petraea* is stable across western European mixed oak stands, Ann. For. Sci. 59 (2002) 777–787.

[23] R.S. Jalal, J.F.F. Weber, N. Manshoor, Dereplication of oligostilbenes in dipterocarpaceous plants using LCMS-ESI-Ion trap-database, J. Liq. Chromatogr. Relat. Technol. 41 (2018) 161–169.
[24] SPSS Inc., SPSS for Windows, Version 16.0, SPSS Inc., Chicago, 2007.
[25] P. Bridgemohan, Production and partitioning of dry matter in leren [*Calathea allouia* (Aubl.) Lindl], J. Agric. Univ. Puert. Rico 95 (2011) 35–44.
[26] R. Lemoine, S. La Camera, R. Atanassova, F. Dédaldéchamp, T. Allario, N. Pourtau, J.L. Bonnemain, M. Laloi, P. Coutos-Thévenot, L. Maurousset, Source-to-sink transport of sugar and regulation by environmental factors, Front. Plant Sci. 4 (2013) 1–21.
[27] M.T. Lin, A. Occhialini, P.J. Andralojc, M.A.J. Parry, M.R. Hanson, A faster Rubisco with potential to increase photosynthesis in crops, Nature 513 (2014) 547–550.
[28] Bachtiar, G. Munif, M. Maya, G. Dwi, S. Atang, Phosphorus sufficiency for growth and production of soybean under saturated soil culture in mineral and peaty soils, J. Soil Sci. Environ. 18 (2016) 21–27.
[29] O. Urban, M. Šprtová, M. Košvancová, I. Tomášková, H.K. Lichtenthaler, M.V. Marek, Comparison of photosynthetic induction and transient limitations during the induction phase in young and mature leaves from three poplar clones, Tree Physiol. 28 (2008) 1189–1197.
[30] Y. Matsumoto, Y. Maruyama, A. Uemura, H. Shigenaga, S. Okuda, H. Kawarasaki, L.H. Ang, S.K. Yap, Gas exchange and turgor maintenance of tropical tree species in Pasoh Forest Reserve, in: T. Okuda, N. Manokaran, Y. Matsumoto, K. Niiyama, S.C. Thomas, P.S. Ashton (Eds.), Ecological of Lowland Rain Forest in Southeast Asia, Springer-Verlag, Tokyo, JP, 2003.
[31] N. Hidayati, M. Reza, T. Juhaeti, M. Mansur, Carbon dioxide (CO_2) absorption by trees in "MekarSari" Fruit Garden, Bogor, in relation to green house gases mitigation, Jurnal Biologi Indonesia 7 (2011) 133–145.
[32] S. Xie, X. Luo, Effect of leaves position and age on anatomical structure, photosynthesis, stomatal conductance, and transpiration of Asian pear, Bot. Bull. Acad. Sin. (Taipei) 44 (2003) 297–303.
[33] F.J. Douglass, T. Landis, T. Luna, Nursery Manual for Native Plant, Department of Agriculture, Washington, 2009.
[34] Z. Shi, M. Haworth, Q. Feng, R. Cheng, M. Centritto, Growth habit and leaves economics determine gas exchange responses to high elevation in an evergreen tree, a deciduous shrub, and a herbaceous annual, AoB Plants 7 (2015) 1–14.
[35] S. Fujimura, P. Shi, K. Iwama, X. Zhang, J. Gopal, Y. Jitsuyama, Effect of altitude on the response of net photosynthetic rate to carbon dioxide increase by spring wheat, Plant Prod. Sci. 13 (2010) 141–149.
[36] P. Kostopoulou, M. Karatassiou, Photosynthetic response of *Bromus inermis* in grasslands of different altitudes, Turk. J. Agric. For. 40 (2016) 642–653.
[37] L. Taiz, E. Zeiger, Plant Physiology, Sinauer, Massachusetts, US, 2010.
[38] Pertamawati, The responses of potatoes (*Solanum tuberosum* L.) explant in vitro growth in photoautorof condition, Jurnal Sains dan Teknologi Indonesia 12 (2010) 31–37.
[39] M. Ashraf, P.J.C. Harris, Photoshynthetic under stressful environments: an overview, Photosynthetica 51 (2013) 163–190.
[40] Y. Astuti, D. Astiani, R. Herawatiningsih, The effect of repeat combustion on peatland on some characteristics of land in Rasau Jaya Umum village district, Kubu Raya Kalimantan Barat, Jurnal Hutan Lestari 3 (2020) 668–681.
[41] A. Dariah, E. Maftuah, Maswar, Peatland characteristics, in: N.L. Nurida, A. Wihardjaka (Eds.), Guidelines for Sustainable Management of Degraded Peatlands, Balai Besar Penelitian dan Pengembangan Sumberdaya Lahan Pertanian, Bogor, ID, 2014.
[42] L. Patiny, A. Borel, ChemCalc: a building block for tomorrow's chemical infrastructure, J. Chem. Inf. Model. 53 (2013) 1223–1228.
[43] Z.J. Zhu, A.W. Schultz, J. Wang, C.H. Johnson, S.M. Yannone, G.J. Patti, G. Siuzdak, Liquid chromatography quadrupole time-of-flight mass spectrometry characterization of metabolites guided by the METLIN database, Nat. Protoc. 8 (2013) 451–460.
[44] H. Horai, M. Arita, S. Kanaya, Y. Nihei, T. Ikeda, K. Suwa, et al., MassBank: a public repository for sharing mass spectral data for life sciences, J. Mass Spectrom. 45 (2010) 703–714.
[45] mzCloud - advanced mass spectral database, (n.d.). Available at https://www.mzcloud.org/ [Accessed on March 4, 2020].
[46] E. Fahy, M. Sud, D. Cotter, S. Subramaniam, LIPID MAPS online tools for lipid research, Nucleic Acids Res. 35 (2007) W606–W612.
[47] A.S. Kamariah, T. Ozek, B. Demirci, K.H.C. Baser, Chemical composition of leaves and seed oils of *Dryobalanops aromatica* Gaertn. (Dipterocarpaceae), ASEAN J. Sci. Technol. Dev. 29 (2012) 105–114.