

## **TROPICAL AGRICULTURAL SCIENCE**

Journal homepage: http://www.pertanika.upm.edu.my/

# Physical, Chemical and Microbiological Properties of Different Combination of Soilless Media and Their Effect on the Vegetative Component and Nutrient Content of Hempedu Bumi (*Andrographis paniculata*)

Shara S. A.<sup>1</sup>, Zaharah S. S.<sup>1\*</sup>, Radziah O.<sup>2</sup> and Puteri E. M. W.<sup>1</sup>

<sup>1</sup>Department of Crop Science, Faculty of Agriculture, Universiti Putra Malaysia, 43400 UPM, Serdang, Selangor, Malaysia <sup>2</sup>Department of Land Management, Faculty of Agriculture, Universiti Putra Malaysia, 43400 UPM, Serdang, Selangor, Malaysia

#### ABSTRACT

Soilless media (SM) is a common worldwide growing method for industrial horticultural production. It is a good growing medium that relies on the properties that benefit plant growth i.e. physical property, chemical properties and microbial activity. There are several SM with good characteristics such as empty fruit bunch compost (EFBC), coconut coir dust (CCD) and peat. EFBC is one of the organic residues of oil palm that provide beneficial microorganisms a good source of bacteria-rich, high nutrient content. The composting process reduces the pH of EFB by 6.86-7.20 to 4.5-6.0. CCD is recommended as a substitute for other media because of it is excellent for holding water and drainage and has high air porosity due to its large surface area. Microbiologically, CCD is the absence of weeds and pathogens and has anti-fungal properties that prevent soil-borne diseases. It also maintains greater oxygen levels and is reusable after sanitisation. Despite the ideal characteristics of CCD, there are some chemical limitations of this medium, including low pH and low potassium content. Peat has good aeration characteristics that are good for root growth. An experiment was conducted to evaluate the best medium combinations for growth performance and nutrient content of hempedu bumi (*Andrographis paniculata*). Three types

#### ARTICLE INFO

Article history: Received: 05 February 2015 Accepted: 31 October 2016

*E-mail addresses*: shara8620@yahoo.com (Shara S. A), szaharah@upm.edu.my (Zaharah S. S.), radziah@upm.edu.my (Radziah O.), putri@upm.edu.my (Puteri E. M. W.) \* Corresponding author of SM with five different combinationa were used as growing media for the plant; they included C1=CCD (1: -) as control; C2=EFBC + CCD (7:3); C3=EFBC + CCD (3:7); C4=CCD + Peat (7:3) and C5=CCD + Peat (3:7). Prior to the experiment, the physical, chemical and microbiological

ISSN: 1511-3701 © Universiti Putra Malaysia Press

properties of the media were determined. The experiment was conducted in RCBD with five replications. In this experiment, the parameters of vegetative components (plant height, number of leaves and total leaf area), dry matter yield and partitioning [root, shoot dry weight and root to shoot ratio (R:S)] and the macronutrient nitrogen [(N), phosphorus (P), potassium (K), calcium (Ca) and magnesium (Mg)] of hempedu bumi were determined. C3 showed the highest vegetative component [plant height (39.5 cm), number of leaves (72.7 leaf plant-1), total leaf area (79.8 cm2) and shoot dry weight (3.03 g)] of hempedu bumi. The highest macronutrient content (1.17% N, 0.07% P, 2.45% K, 2.77% Ca and 0.58% Mg) was in the leaf tissues of hempedu bumi when grown in the C3 media. In conclusion, a combination of EFBC+CCD (3:7) is recommended as a suitable growing medium for hempedu bumi due to the greater vegetative components well as the higher macronutrient content it yielded in the leaf tissues of the plant.

*Keywords*: soilless media, *hempedu bumi*, empty fruit bunch, coconut coir dust, peat

#### INTRODUCTION

The characteristics of growing media is one of the important factors that affect the growth performance of the plants. Plants require sufficient nutrients and moisture from the medium in which they are grown. Soilless media have several physical, chemical and biological functions. The physical function of soilless media is to provide support to the plant with good root aeration, gas exchange to and from the roots and sufficient water for the roost. The chemical function of soilless media is to supply adequate oxygen and nutrients for proper root functions. Biologically, availability of soilless media is important for beneficial microorganisms as a host to release nutrients from the media. Combinations of SM may affect the production of plants in which the right combination of SM can indicate the suitability of the growing medium to fulfil the needs of the roots to grow. Several SM have been studied, such as empty fruit bunch compost (EFBC), coconut coir dust (CCD) and peat. Mixing different ratios of these SM affects the characteristics of the media and the growth of the plants that are grown in it (Suhaimi & Ong, 2001). For example, EFBC, one of the organic residues of oil palm, provides beneficial microorganism with a good source of bacteria and nutrients and has an acidity value of 4.5-6 as well as a moderately fine texture such as that of sand or dust particles of size 2 mm (Kavitha et al., 2013). The chemical properties of the media have huge influence on the ability of the media to increase the production of the plant. The EFBC medium contains 0.76% N, 0.21% P, 2.60% K, 0.58% Ca and 0.20% Mg as a single substrate.

CCD is recommended for use as a substitute for other media because it has suitable physical properties such as that it can hold and release water up to eightfold of its mass and has excellent drainage and high air porosity due to the large surface area of its particles. Coir dust is light brown

in colour with a particle size of (0.2-2.0)mm) and when compared to sedge peat and sphagnum peat, has higher CEC, superior structural stability, water absorption ability and drainage and is clear of sticks and other extraneous matter (Meerow, 1994; Noguera et al., 1998). Microbiologically, CCD is the absence of weeds and pathogens and has anti-fungal properties for a conducive environment for plant growth (Wira et al., 2011). CCD also prevents soil-borne diseases when mixed with soil (Mokhtari, 2010). Despite the ideal characteristics of CCD, there are some chemical limitation of this medium, including low pH and low potassium content.

Peat has good aeration characteristics and is suitable for root growth when the growing medium is mixed with other media (Molitor & Brückner, 1997). Peat is suitable for growing plants that require more acidic conditions and is generally not suitable for growing crops if not mixed with other media. The physical and chemical properties of peat have been determined by several researchers and in each study, various results were gathered because the properties of this substrate depend on the degree of intensity of decomposition (Ismail et al., 2001). As reported by Ismail (2011), CCD and EFB alone have the lowest bulk density, with 0.92 and 0.97 g cm<sup>-3</sup>, respectively. The original pH for EFB has been reported to be in the range of 6.86 (Ishak et al., 2014) to 7.20 (Kavitha et al., 2013). The addition of peat has been reported to improve bulk density of CCD and EFB. In terms of water availability, which reflexes the ability of a medium to hold water, CCD and a mixture of CCD and peat presented the highest percentage (15.18% and 13.99%). Ismail also reported that soilless media such as EFB, peat and a combination both have very poor water holding capacity and therefore, were not suitable for use as a growing medium for plants. This is because these media can easily dry up and cause water stress to the plant.

The decomposition degree of peat is positively related to the medium's carbon content and particle size fraction (<250 µm). Kala et al. (2009), who conducted a study on co-composting of different oil palm wastes with sewage sludge for use as potting media for ornamental plants, reported that EFB and peat had a particle size of 40 mm and <20 mm, respectively. Additionally, observations on the physical properties of peat by Sekhar and Sai Gopal (2013) showed that the bulk density of peat was significantly high when it was compared with other soilless substrates such as CCD. Yasmeen et al. (2009), in their study to prove the efficient conversion of empty fruit bunch of oil palm into fertiliser-enriched compost, reported that EFB exhibited higher bulk density and moisture content after the composting process than before it. Similar results were also shown by Zulkarami et al. (2010) when peat was used in the composting process. Good media must have at least a proportion of 45% of organic matter, 35% of water and 20% of aeration to allow healthy plant growth as suggested by Kumar and Kumar (2013).

With this in mind, the objectives of this study were (i) to compare the physical, chemical and microbiological properties of different compositions of soilless media of CCD as well as combinations of EFBC+CCD and CCD+peat, and (ii) to determine the vegetative components of *hempedu bumi* grown in different combinations of SM.

#### **MATERIALS AND METHODS**

The experiment was conducted in the rain shelter, Faculty of Agriculture, Universiti Putra Malaysia. Three types of SM with five different combinations were used as growing media C1=CCD (1: -) as control; C2=EFBC+CCD (7:3); C3=EFBC+CCD (3:7); C4=CCD+Peat (7:3) and C5=CCD+Peat (3:7). Physical (bulk density and available water), chemical (macronutrient content, electric conductivity and pH) and microbial (fungi, bacteria and actinomycetes population) properties were determined from each of the replicates. Seeds of Andrographis paniculata were obtained from the Agro Gene Bank, Faculty of Agriculture, Universiti Putra Malaysia, Serdang, Selangor, Malaysia. The fundamental principle of this method is the removal of the seed coat using the rough surface of sand paper number 120 with size  $30 \text{ cm} \times 30 \text{ cm}$ . After germination, the seedlings, with two expanded cotyledons, were transferred into a tray with equal proportiona of CCD, EFBC and peat (1:1:1 w/w/w) until the plant had grown to six to eight leaves. Then, healthy seedlings were transplanted into plastic pots measuring 12

 $cm \times 17$  cm with different ratios of SM. The experiment was arranged in Randomised Complete Block Design (RCBD) with four replications. The plants were well watered twice a day and other practice management was done when necessary. No mineral fertiliser was added throughout the experimental period. The physical (available water and bulk density) as well as chemical [electric conductivity (EC), pH and nutrient content] properties were recorded prior to the beginning of the experiment. The treatment effects of different media combination on vegetative components (plant height, number of leaves and total leaf area), dry matter yield and partitioning [root, shoot dry weight and root to shoot ratio (R:S)] and microbial activity (bacteria, fungi and actinomycetes) and macronutrient content [nitrogen (N), phosphorus (P), potassium (K), calcium (Ca) and magnesium (Mg)] in the leaf tissue of hempedu bumi were determined 60 days after planting (DAP).

#### **Soilless Media Analysis**

**Physical properties.** The moisture content and water availability of the SM was determined using the modified method of De Boodt and Verdonck (1971). Each mixture of SM was added to the rubber ring and saturated with water for 24 h. After overnight incubation, the media were transferred to the pressure plate with different pressure levels of pF 0 kPa, pF 1 kPa, pF 2 kPa, pF 2.54 kPa and pF 4.19 kPa. They were allowed to remain in the pressure plate for one week. The SM were then taken out from the pressure plate and put into aluminium dishes. The initial weight (W1) of the SM was recorded and then oven-dried for 24 h at 105°C. The dry weight (W2) was obtained; the weight of the empty dish (W3) had been measured earlier. At the end of the process, the water content percentage (%) of the SM was obtained at each of the different pressure levels. The moisture content (MC) and water availability (WA) for plant use was then calculated using the following formula:

$$MC (\%) = \frac{W1 - W2}{W1 - W3} \times 100$$

WA = MC at field capacity (pF 2.54 kPa)MC at permanent wilting point (pF 4.19 kPa).

The determination of bulk density was done using the modified method of De Boodt and Verdonck (1971) by fixing two copper rings together with a tape and a mesh filter at the bottom. Each composition of SM was added to the rings and saturated with water and left for 24 h. The separation of the two copper rings was conducted by removing the tape from the rings and the saturated SM was sliced to the exact level of the single ring. The water-saturated SM inside the ring was weighed (W1) and oven-dried at 105°C for 24 h. The oven-dried SM inside the ring was weighed after removing each sample from the ring (W2). The diameter and height of the single ring were also measured before the sampling for determination of ring volume (V).

Bulk Density (g cm<sup>-3</sup>) = 
$$\frac{W1-W2}{V}$$

where, W1 = Weight before oven drying (g), W2 = Weight after oven drying (g), V =  $\pi$  r<sup>2</sup> h (cm<sup>3</sup>)

Chemical properties. Each mixture proportion of soilless media was ovendried for 48 h at 60°C and ground to a size of <1 mm. Soilless media (0.5 g) was put into a digestion flask with 5.0 mL of concentrated sulfuric acid (H<sub>2</sub>SO<sub>4</sub>) and left for at least 2 h. Before starting the digestion, 2 mL of 50% hydrogen peroxide  $(H_2O_2)$  was added slowly down the sides of the tube while it was rotating and the heating process was allowed to proceed in the digestion block for 45 min. Another 2 mL of 50%  $H_2O_2$  was added to the tube and left for 45 min. The process was repeated until the digestion was clear or colourless. In this study, the process of adding 2 mL of 50% H<sub>2</sub>O<sub>2</sub> was repeated five times until the digestion changed to a colourless solution. Finally, the volume of the solution was made up to 100 mL with distilled water and filtered by Whatman filter paper of medium size 500 mm diameter. Nitrogen (N) and phosphorus (P) were determined using the Auto analyser (AutoAnalyzer, QuikChem FIA + 8000 Series, USA). Potassium (K), calcium (Ca) and magnesium (Mg) were determined using the Atomic Absorption Spectrophotometer (Perkin Elemer, 310, PC, California, USA). The results of the nutrient analysis were presented as percentage (%) per gram of dry weight.

The pH value was determined by adding the SM into 0.01 M CaCl<sub>2</sub> in a ratio of 1:5 v/v and left to stand for 1 hour, and then the acidity was determined using the Digital pH meter (GLP 21, Crison, Barcelona, Spain). Electrical conductivity (EC) was determined by adding 100 mL of distilled water to 10 g of SM (1:10 v/v). The mixture was then agitated using a mechanical shaker for 30 min and incubated for 24 h. After incubation, the sample was filtered and the EC was determined using an EC meter (GLP 31, Crison, Barcelona, Spain). The reading was expressed as ds m<sup>-3</sup>.

Microbiological properties. The microbial population of the soilless media was determined by serial dilution. A total of 10 g of each sample was suspended in 100 mL of distilled water and incubated in a mechanical shaker for 30 min. An agar plate containing a nutrient agar, Rose Bengal streptomycin agar, and actinomycetes was prepared for the determination of bacteria, fungi and actinomycetes, respectively, and a serial tenfold dilution of each sample was prepared. All of this and 0.1 mL aliquots and were spread on each plate. The inoculated petri plates were incubated in an incubator at 28°C for three days to count the number of bacteria and five days to the count number of fungi and actinomycetes. After the incubation period, the colony forming units were counted and expressed as log10 CFU g<sup>-1</sup> of soilless media on a moisture-free basis.

**Plant growth analysis.** Plant height (cm) was measured using a standard ruler from

the growth medium surface to the tip of the main stem. The number of leaves produced or maintained by each plant was also counted numerically. After harvesting, the leaves were individually separated from the stem, and the total leaf area (TLA) per plant was estimated using the Automatic Leaf Area Meter (LI-3100C Area Meter, Lincoln, Nebraska). Four plants were harvested from each treatment and separated into root and shoots. The stem and leaves were considered part of the shoots. After separation of the plant parts, the plants were dried individually under sunlight for one day and then oven-dried at 40°C for 48 h, and the dry shoot was recorded using a digital balance (QC 35EDE-S Sartorius, Germany). The roots were washed gently and the dry weight of the root was weighed using a digital balance (QC 35EDE-S Sartorius, Germany). The root and shoot dry weight was presented as gram (g). The ratio of root to shoot was calculated based on shoot and root dry weight using the following formula:

Total	Root	Drv '	Weight	(g)
				(D)

Total Shoot Dry Weight (g)

**Macronutrient content of leaf tissue.** The dried leaves (50 g) of *Andrographis paniculata* were ground to powder and stored in well-stoppered polyethylene vials and kept at room temperature (24°C). The ground leaf (0.25 g) tissue was put into a digestion flask with 5.0 mL of concentrated sulfuric acid (H<sub>2</sub>SO<sub>4</sub>) and left for 2 h. Before starting the digestion, 2 mL of 50%

Root: Shoot Ratio (R:S) =

hydrogen peroxide  $(H_2O_2)$  was slowly added down the sides of the tube while it was rotating. The tube was then left in the digestion block for 45 min for heating. Another 2 mL of 50% H<sub>2</sub>O<sub>2</sub> was added to the tube and left for another 45 min. The process was repeated until the solution was clear or colourless. Finally, the solution was brought up to 100 mL with distilled water. The nutrient element of N and P were determined using the Auto analyser (Lachat instruments, QuikChem<sup>®</sup> FIA + 8000 Series). The determination of K, Ca and Mg was done using the Atomic Absorption Spectrophotometer (Perkin Elmer Model 310, California, USA).

Statistical analysis. Statistical analysis was determined using a one-way ANOVA using Statistical Analysis System (SAS) (release 9.3, SAS Institute Inc., Cary, NC, USA). Fisher's Least Significant Differences (LSD) was used for comparison of treatments mean where the F values were significant at P $\leq$ 0.05. All the assumptions of ANOVA were checked by carefully analysing the results of the statistical analysis. Sigma Plot (version 12.5, Systat Software, San Jose, California USA) was used for the data that were presented in a graph.

#### **RESULTS AND DISCUSSION**

#### **Physical Property of Soilless Media**

The capacity of the media to supply sufficient amounts of water for plant growth can be indicated by the percentage of available water present in the field capacity and wilting point. There was a significant difference in the content of available water (Figure 1A) between each composition of the SM. The lowest water content was recorded in C5, containing 70% of peat, which has low water-holding capacity. A similar observation was confirmed by Iberahim (2001), who noted available water content of a single proportion of peat in his study. The available water for the plant and aeration in the growth medium were estimated by the physical criteria derived from the physical properties (Sharma & Kumawat, 2012).

The highest bulk density (Figure 1B) among different compositions of SM was found in C5 followed by C2, C3 and C4 as compared to the control media (C1). A study showed that C5, C2, C3 and C4 had significantly higher bulk density by 161%, 125%, 99.9% and 65%, respectively than their control. Higher amounts of peat increase bulk density of a soilless mixture; this was reported by Noorhanin et al. (2013) and Zulkarami et al. (2010). This is probably due to the characteristic of peat itself, which has higher bulk density than CCD (Abad et al., 2005). The bulk density of compost is very important where it comprises a larger proportion of the growing medium. Meanwhile, C1, which comprised a single composition of CCD, had the lowest bulk density; a similar result was confirmed in the findings of Al Rawahy et al. (2009), Iberahim (2001) and Riaz et al. (2008). Increasing and decreasing the bulk density of SM is attributed to the medium components, particularly particle size and presence of particles of different size, which lends to higher bulk density than a medium with particles of only one size.



*Figure 1*. The physical properties of soilless media: available water (A), bulk density (B) prior the beginning of the experiment. Vertical bars represent S.E. and means value with the same letter are not significantly different at P = 0.05

trend of the changes shows that the control (C1) had the highest content of moisture followed by C3, C5, C4 and C2 throughout the pF levels. The highest percentage of moisture content at field capacity (pF 2.54) was observed in 100% CCD (C1) followed by C3 medium comprising EFBC and CCD (7:3, v/v). According to the results, both media probably had a sufficient amount of water to support growth performance of *Andrographis paniculata* even under water-

The moisture content of each composition of SM was determined under different pressure levels (Figure 2). The



*Figure 2*. The moisture content of the soilless media prior the beginning of the experiment. Vertical bars represent S.E. of mean and are invisible when the values are smaller than the symbol

stress conditions. In other words, plants grown in these media will not be prone to severe wilting if the plant undergoes a sudden increase in the transpiration rate (Raviv & Blom, 2001). Similar findings have been reported by Phurailatpam et al. (2013), who found that CCD and EFB were successfully used as growing media for the production of cauliflower. It has been reported that the water-holding capacity of CCD is high because it can hold and release water eightfold of its mass (Mokhtari, 2010).

#### **Chemical Properties of Soilless Media**

Results of the macronutrient contents N, P, K, Ca and Mg of the soilless media are shown in Table 1. The highest N content was

SM	Ν	Р	K	Ca	Mg
C1	$0.11 \pm 0.004e$	$0.10\pm0.002c$	$0.22\pm0.008a$	$0.12\pm0.007c$	$0.09\pm0.004d$
C2	$0.57\pm0.008a$	$0.45\pm0.010a$	$0.24\pm0.003a$	$0.22\pm0.006ab$	$0.14\pm0.002a$
C3	$0.27\pm0.004\text{d}$	$0.10\pm0.002c$	$0.08\pm0.004b$	$0.18\pm0.011 bc$	$0.11\pm0.002 bc$
C4	$0.34\pm0.008\text{c}$	$0.30\pm0.011b$	$0.23\pm0.007a$	$0.21\pm0.009ab$	$0.13\pm0002ab$
C5	$0.42\pm0.007b$	$0.13\pm0.002\text{c}$	$0.03\pm0.002\text{c}$	$0.28\pm0.025a$	$0.1\pm0.008 dc$

Table 1Nutrient Contents of the Soilless Media (% on Dry Weight Basis) Prior to the Beginning of the Experiment

Mean values  $\pm$  S. E followed by the same lower case within columns are not significantly different at P=0.05

observed in C2 (0.57%), which contained EFBC and CCD (7:3, v/v). High proportions of EFBC increased the N content of the C2 mixture of SM. On the contrary, the lowest value of N was observed in C1 (0.11%), which contained a single composition of CCD. However, Wira et al. (2011) reported that the N content of CCD did not differ significantly from the mixture of CCD with EFBC. Smith et al. (1989) suggested that CCD requires mixing with a nutrient-rich material in order to become suitable for use as a SM. Furthermore, Tang et al. (2006) indicated that N usually increased during the composting process. C5 exhibited higher N content (0.42%) than C3 (0.27%) and C4 (0.34%). Higher N content in C5 was probably due to the fact that that medium contained 70% of peat compared to C3 and C4, which contained 70% of CCD.

The results also showed that C5 and C1 (control) exhibited the highest (0.28%) and the lowest (0.12%) content of Ca, respectively (Table 1). C2 contained significantly higher Mg (0.14%) than the control (C1) and the other mixtures. Reduction of Mg content was probably due to an increase in the pH value of the

substrate used. However, an earlier study by Wira et al. (2011) claimed that there were no specific high macronutrient (N, P, K, Ca and Mg) contents in EFBC and the CCD mixture and in the single composition of CCD. In this study, the K content was found to be higher in C2 (0.24%), followed by C4 (0.23%), C1 (0.22%) and C5 (0.03%), with no significant difference among them. Davies et al. (2000) observed that increases in the K content are expected when a combination of EFB and CCD is used as the growing medium compared to a medium with single CCD components.

Acidity (pH) of the media, which is a measure of the acidity or alkalinity of the media, is one of the most important chemical properties of growing media. The pH value for each SM composition is shown in Figure 3A. The results of this study showed that C2, C5, C3 and C4 have significantly higher pH by 36%, 19.4%, 10.9% and 8.4%, respectively, than the control. The high pH value of C2 enhanced the availability of essential nutrients in the growing medium (Griffiths et al., 2003). The increases of the pH value of the EFBC might be due to the active microbial activities that caused the

release of ammonia (NH<sub>3</sub>). CCD has been reported to have a high pH value (Mokhtari, 2010), and this could also be the reason the C4 medium, which consisted of CCD and peat (7:3, v/v), exhibited a higher pH value than the control (C1). However, in this study, the C2 SM gave a low pH value, even though it had higher proportions of CCD content. In general, the pH of the growing medium played an essential role in nutrient availability, which is essential for good plant growth (Ismail et al., 2013).

The salinity or electrical conductivity (EC) value was significantly different among the SM samples (Figure 3B). The



*Figure 3.* The pH value (A) and EC value (B) of soilless media prior the beginning of the experiment. Vertical bars represent S.E and means value with the same letter are not significantly different at P = 0.05

highest value of EC was shown in C2, followed by C3, while the lowest value was found in C5. Electrical conductivity (EC) is a measurement of dissolved salt concentration in a growing substrate and is an indicator of the available amount of fertiliser for plant growth (Mafakheri et al., 2010). The high EC of C2 and C3 was also probably due to the loss of EFB weight and release of other mineral salt and organic matter throughout the decomposition process of the EFB (Zarrouk et al., 2005).

## Microbiological Property of Soilless Media

The population of bacteria was significantly higher in C2  $[\log_{10}$ CFU g<sup>-1</sup>=6.9 or CPU=10<sup>6.9</sup> colonies g<sup>-1</sup>] than in the other treatments (Figure 4A). On the contrary, there was no significant difference among the bacterial population of C1 [CFU=10<sup>4.7</sup> colonies g<sup>-1</sup>], C3 [CFU=10<sup>5.4</sup> colonies g<sup>-1</sup>], C4 [CFU=10<sup>4.5</sup> colonies g<sup>-1</sup>] and C5 [CFU=10<sup>5.2</sup> colonies g<sup>-1</sup>]. According to the results shown in Figure 4B, medium C2 gave the highest population of fungi [CFU=10<sup>7.2</sup> colonies g<sup>-1</sup>] followed by C3 [CFU=10<sup>6.4</sup> colonies g<sup>-1</sup>]. However, the fungi population of medium C5 [CFU=10<sup>5.6</sup> colonies g<sup>-1</sup>] and medium C4 [CFU=10<sup>5.2</sup> colonies g<sup>-1</sup>] and medium C1 [CFU=10<sup>4.4</sup> colonies g<sup>-1</sup>] was not significant and relatively lower than in C2 and C3. The actinomycete population of the SM is represented in Figure 4C. The highest population of actinomycetes was detected in C2 [CFU=10<sup>7.1</sup> colonies g<sup>-1</sup>] followed by C3 [CFU=10<sup>5.8</sup> colonies



*Figure 4*. The bacteria (A), fungi (B) and actinomycetes (C) population of soilless media on 60 DAP. Vertical bars represent S.E. and means value with the same letter are not significantly different at P = 0.05

g<sup>-1</sup>], while the lowest was recorded in C5 [CFU=10<sup>5.6</sup> colonies g<sup>-1</sup>], C4 [CFU=10<sup>4.6</sup> colonies g<sup>-1</sup>] and C1 [CFU=10<sup>4.5</sup> colonies g<sup>-1</sup>]. Increased microbial population of C2 could have been due to an increase in the pH of these media as a result of the addition of EFBC. Similar results were obtained by Saravanan et al. (2009), in whose study the microbial population of the media increased at the highest level of EFBC.

In addition, availability of inherited microorganisms in the EFBC also contributed to increase the soil's microbial population (Abdalla, 2005). The lowest microbial population of C1, which contained 100% CCD might be related to low percentage of water availability as presented in Figure 1A. Other findings claimed that low availability of microbial populations of peat correlated with the moisture content of peat (Stankovic, 2011). Kyparissis et al. (1995) also reported that microbial population of peat increased with an increase in moisture content. However, the population of microbes in peat was also influenced by pH (Stankovic, 2011). The enhancement of the microbial population of C2, which contained a high ratio of EFBC might have been due to the availability of high organic nutrient matter in the media (Douds Jr et al., 1997). The high microbial population of the media was also influenced by the moisture content, pH and EC of the media where media with high moisture altered the population of microbes (Griffiths et al., 2003). Nutrient content was another factor that affected the microbial population of the media where the availability of beneficial microbes in the compost converted unavailable forms of nutrient to available forms. Despite the high moisture content of CCD, the fungal population of CCD was low and this was attributed to the anti-fungal properties of the CCD (Mokhtari, 2010). Addition of beneficial microbes was also previously recommended due to the cleanness of the CCD (Prabhu & Thomas, 2002).

### **Plant Vegetative Component**

The plant height of *Andrographis paniculata* was significantly affected by different

combinations of SM (Figure 5A). The highest mean of plant height was observed in C3 (39.50 cm) followed by C4 (29.83 cm) and C5 (26.33 cm), while the lowest was observed in C2 (15.00 cm). Plant height varied significantly due to different proportion of SM. This finding obviously varied from the previous study done by Noorhanin (2013). They found that the highest *Andrographis paniculata* plant was observed when the plant grown in 100% CCD was fed inorganic fertiliser. Plant height varied significantly due to different proportion of SM at different growth stages. The increased plant height in C3 compared to other SM treatments was probably due to the perfect combination and balance between nutrient availability, suitable pH and EC provided by the medium. A similar observation was recorded by Zulkarami et al. (2010); in their study the highest rock melon plant was observed in the EFB medium with an EC value between 1 and 2.5 ds m-1. Bloom et al. (1985) stated that plants responded to their environment in such a way as to optimise their resource use. Good results in plant height can also have been due to the response of the shoots



*Figure 5*. The plant height (A), number of leaves (B) and total leaf area (C), shoot dry weight (D), root dry weight (E) and root to shoot ratio (F) of hempedu bumi on 60 DAP grown under different combination of soilless media of soilless media. Vertical bars represent S.E. and means value with the same letter are not significantly different at P = 0.05

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and roots to optimal nutrient availability based on the carbon balance between both parts (Cannell & Dewar, 1994). In this study, healthy roots easily grew when the *hempedu bumi* plants were grown in C3 and the nutrients were absorbed well, thus aiding the growth in plant height. Results from another study have revealed that EFB enhanced the production of cabbage when used as its growing medium (Wahid et al., 2011).

Medium C3 and C4 produced 40.86% and 14.09% higher number of leaves per plant compared to the control (C1), respectively (Figure 5B). Similarly, as reported earlier, using EFB as a growing medium resulted in the highest number of rock melon leaves compared to using the CCD medium (Zulkarami et al., 2010). The result of this study illustrated that the increase in the number of leaves of plants grown in C3 and C5 media was probably due to increased nutrient release from C3 and C5 media; this correlates with the results of a study done by Abdul Mutalib et al. (2009). On the reverse side, the plant grown in medium C2 and medium C5 exhibited 7.04% and 2.81% lower number of leaves than in the control.

The total leaf area (TLA) of *Andrographis paniculata* was significantly affected by different combinations of SM (Figure 5C). The mean of the TLA was higher (64.95% and 50.57%) in the plants grown in medium C3 and medium C4 and lower (22.52% and 25.11%) in plants grown in medium C2 and medium C5 as compared to the control medium (C1). A similar result

was obtained by Wira et al. (2011), who recorded that the highest TLA of rock melon was observed in combinations of EFBC and CCD (3:7, v/v). In general, CCD was associated with high water-holding capacity but it was poor in aeration, causing decreases in oxygen diffusion to the roots (Abad et al., 2005). The combination of both substrates can increase the diffusion of oxygen to the roots, which leads to good plant growth. In addition, at the vegetative stage, the leaves and roots scramble for carbohydrate assimilation. To gain an increase in leaf area, the growth of the leaves and roots should be in good balance (Noorhanin et al., 2013). The increase in the TLA when the plant was grown in the C3 medium might be attributed to the availability of beneficial microorganisms in the EFBC media. These results are similar to those of a study done by Zydlik and Zydlik (2008) in which the beneficial microorganisms also significantly resulted in releasing nutrients from the soil to an available form for the growth of apple trees. Increase in the TLA of Vigna mungo was also observed with application of beneficial effective microorganisms (Karthick Raja, 2012).

The significant and highest shoot dry weight was found in the plant grown in the C3 medium (2.37 g plant-1) compared to the plant grown in the control (1.87 g plant-1) and other treatments. However, there were no significant differences between the C4, C5, C2 and C1 plants (Figure 5D). These results were in agreement with the findings of Wira et al. (2011). They recorded that application of EFBC with CCD (3:7, v/v)

enhanced the biomass production of rock melon. Results of this study were somewhat similar to results of Iberahim (2001), who suggested that the application of EFBC as SM increased the biomass production of cauliflower. Even though the nutrient content of C2 was higher, the nutrients available in C2 were not efficiently absorbed by the plants. This was probably due to the antagonistic influence of other nutrients that were available in the medium. In addition to this, toxicity to plants will also occur whenever a high amount of essential nutrients is supplied. An optimum ratio of EFBC enhanced the productivity of dry weight of shoot biomass of Andrographis paniculata, a result similar to that recorded by Zulkarami et al. (2010), who noted that application of EFBC significantly increased the production of rock melon.

Figure 5E showed there was no significant difference between treatments as influenced by different combinations of SM. The highest mean of root dry weight was obtained by C3  $(0.57 \text{ g plant}^{-1})$ followed by C2 (0.52 g plant<sup>-1</sup>), C1 (0.40 g plant<sup>-1</sup>) and C4 (0.39 g plant<sup>-1</sup>), while the lowest (0.34 g plant<sup>-1</sup>) root dry weight was obtained by the plant grown in medium C5. According to Awang et al. (2009), media that have a good balance of air and waterholding capacity would increase root dry weight of C. cristata. In addition, EFBC has moderately fine texture-like sand or dust particle of size 2 mm. A compact medium was probably attained, which led to low favourable conditions for root growth.

Unlike other parameters, the highest R:S (0.24) was obtained in plants grown in the C2 medium (Figure 5F). There were no significant differences in the R:S between different compositions of SM. These results are in conformity with the reports by Wira et al. (2011), who reported that applications of EFBC and CCD as a growing medium for rock melon decreased the R:S of the plant. What influenced the R:S ration was probably the physical conditions and chemical composition of the plant itself, which may have experienced various stresses during the growth process such as lack of nutrients or water for the root. Decrease in the R:S of the plant grown in the C3 medium indicated that the roots were able to supply the shoots of the plant with water, nutrients, stored carbohydrates and certain growth regulators (Harris, 1992). Increase in the R:S of C2 could be an indication of a healthier plant, provided the increase came from greater root size and not from a decrease in shoot weight. This was also due to the highest population of bacteria (A), fungi (B) and actinomycetes (C) in the SM, which may have helped the root achieve healthier growth while the shoot too was able to absorb nutrients and water. This result also has a relationship with the nutrient content accumulation in the SM that is presented in the next section.

#### **Macronutrient Content of Leaf Tissue**

The macronutrient content of leaf tissue of *Andrographis paniculata* is shown in Table 2. The highest N, P, K, Ca and Mg content (1.17%, 0.07%, 2.45%, 2.77%)

SM	Ν	Р	Κ	Са	Mg
C1	$0.87\pm0.022b$	$0.04\pm0.005ab$	1.85 ±0.106ab	$2.00\pm0.11~b$	$0.33\pm0.037c$
C2	$0.53\pm0.054\ c$	$0.06 \pm 0.010 \text{ a}$	$2.00\pm0.137ab$	$2.12 \pm 0.122 \text{ ab}$	$0.44 \pm 0.016 abc$
C3	$1.17\pm0.037a$	$0.07\pm0.009a$	$2.45\pm0.218a$	$2.77\pm0.118a$	$0.58 \pm 0.010 \; a$
C4	$1.11 \pm 0.036a$	$0.04\pm0.007ab$	$2.15\pm0.104ab$	$2.34\pm0.154ab$	$0.49\pm0.013ab$
C5	$0.63\pm0.038c$	$0.01\pm0.001\ b$	$1.62\pm0.095b$	$1.91\pm0.051b$	$0.42\pm0.042bc$

Table 2The Leaf Tissue Nutrient Content (% on Dry Weight Basis) on 60 DAP

Mean values  $\pm$  S. E followed by the same lower case within columns are not significantly different at P=0.05

and 0.58%) of leaf tissue was observed in the plants grown in C3 compared to C1 (control). The medium that yielded the next highest amounts of N, K, Ca and Mg in leaf tissue was C4 (1.11%, 2.15%, 2.34% and 0.49%), followed by C2 in P content (0.06%). The lowest N content (0.53%)was obtained from leaf tissue of plants grown in the C2 medium, while the lowest value of P, K and Ca (0.01%, 1.62%, and 1.91%, respectively) was found in plants grown in the C5 medium. The minimum content of Mg (0.33%) was observed in C1. Increases in the macronutrient content of leaf tissue of plants grown in the C3 medium associated with the availability of several microorganisms in the medium. These results were supported by Shen et al. (2004), whose study found that a growth medium with beneficial microorganisms significantly increased the nutrient content of plant tissue.

#### CONCLUSION

The results of this study showed that the biomass production and leaf tissue nutrient content of *Andrographis paniculata* could be significantly affected by growing the plant in

different combinations of SM. Growing the plant in EFBC and CCD (3:7, v/v) improved plant vegetative growth, dry matter yield and leaf tissue nutrient content. Application of EFBC in high or low volume released nutrients, increased pH and enhanced the microbial population of the potting medium. This suggests that EFBC can be used as an alternative to peat and CCD as a substrate in a soilless culture system.

#### ACKNOWLEDGEMENT

The author is grateful to Universiti Putra Malaysia, Serdang, Selangor for providing a conducive environment and good facilities and scientific equipment that proved indispensible in conducting this study.

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