The effects of growing media on growth and seedling quality of root cutting of *Cyclea* barbata Miers at nursery stage

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ABSTRACT

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Pham Thi Thuy Duong Email: pttduong@hcmuaf.edu.vn Cyclea barbata Miers can be used in pharmaceutical industries and food. They are of tropical origin, and thus suitable for cultivation in many places in Vietnam. This study was conducted to evaluate the effects of different types of growing media on growth, seedling quality of root cutting of Cyclea barbata and financial efficiency at the nursery stage. A one-factor experiment was arranged in a completely randomized design with three replicates and nine treatments which were different mixtures of sand, rice husk ash, coco peat and vermicompost with different ratios in volume. The results showed that root cutting of Cyclea barbata was planted on the mixed media of 25% rice husk ash: 50% coco peat: 25% vermicompost gave the highest number of roots (17.23 roots/cutting), shoot length (100.48 cm), number of leaves (14.70 leaves/cutting), fresh and dry root matter (3.71 and 0.48 g/cutting), fresh and dry shoot matter (5.45 and 1.50 g/cutting), seedling quality index (0.019) at 75 days after planting, the profit (3,665,800 VND/1000 cuttings) and a profit margin of (0.73 times).

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1. Introduction

Cyclea barbata Miers is a plant species native to tropical climates, distributed in some Southeast Asian countries and India (Manilal & Sabu, 1985), so it is suitable for climatic conditions in many areas in Vietnam. Pham (2006) indicated that jelly made from Cyclea barbata leaves can lower body heat and diuretic; Root of Cyclea barbata can cure liver diseases, haemorrhoids. In addition, leaf jelly is also used in the food industry as a food thickener and stabilizer (Arkarapantth et al., 2005). Many southern provinces such as Binh Phuoc, Ba Ria - Vung Tau, and Dong Nai in which Cyclea barbata is grown.

Cyclea barbata is commonly propagated by seeds and root cuttings. Root cutting is an asex-

ual propagation method that helps the plantlets to retain the desired characteristics of the mother plants. The root cutting technique of Cyclea barbata is simple, easy to implement and does not require any modern equipment. Many factors effect the quality of seedlings at the nursery stage. In particular, the growing media plays an important role in providing air and water, allowing optimal root development and responding to the physical properties of the plant (Olle et al., 2015). In addition, appropriate use of growing media also helps to make effective use of agricultural waste (Pham et al., 2018). However, each type of material used to mix the growing media has different characteristics, so it is necessary to determine the appropriate ratio of mixing media to help the cuttings grow well and achieve the best quality and

gain the highest economic benefit.

2. Materials and Methods

2.1. Experimental design

The experiment was carried out from July to September 2020 at the Experimental Station of the Faculty of Agronomy, Nong Lam University, Ho Chi Minh City.

The experiment was single-factorial and arranged in a Completely Randomized Design with 9 treatments corresponding to 9 types of media (denoted G) and 3 replicates. Nine types of media were described in Table 1.

The cuttings of *Cyclea barbata* root was 3 cm in length and 1 cm diameter. Each cutting was planted in a nylon grow bag contained 60 cm^3 of growing media. Each experiment plot included 30 cuttings planted in 30 grow bags.

2.2. Land preparation and field management

The ingredients were mixed by volumetric proportions corresponding to the formulated media. After 3 days of mixing and incubation, the compost would be supplemented with Trichoderma at a rate of 2 kg per m³ and Super Phosphate at a rate of 3 kg per m³. Then the mixture was mixed well every ten days for 30 days. The composts were covered using plastic film to prevent heat and nitrogen loss.

After 30 days, each type of media was sampled and analysed physical and chemical properties at the Department of Soil Science - Fertilizer, Faculty of Agriculture, Nong Lam University. The media properties are presented in Table 2.

Analytical methods include pH meter $(pH_{H_2O}(1:5))$, EC meter (EC (1:5)), Kjeldahl (total N), Tiurin (total organic), colorimeter (total P₂O₅), flame photometer (total K₂O), metal ring (bulk density), picnometer (density), drying method (humidity).

The results of Table 2 suggested that all media were slightly acidic but not salty (Slavich & Petterson, 1993). Total nitrogen content of the mixed media: G1 (50% sand + 25% rice husk ash + 25% coco peat (the control)); G2 (25% sand + 50% husk ash + 25% coco peat; G3 (25% sand + 25% husk ash + 50% coco peat); G4 (50% coco peat + 50% rice husk ash) were an average while the other media had high nitrogen content. Total phosphorus and potassium content of the media G1 (50% sand + 25% rice husk ash + 25% coco peat (the control)); G2 (25% sand + 50% husk ash + 25% coco peat); G3 (25% sand + 25% husk ash + 50% coco peat); G4 (50% rice husk ash + 50% coco peat) were relatively low, while the other media were high. All media had a high total organic content, ranging from 8.97 to 29.42%, and C/N ratio was from 16.44 to 60.58 (Rayment & Lyons, 2011). Besides, the media after incubation all had average porosity and moisture content. In general, the types of media were supplemented with vermicompost that had a higher content of macronutrients and porosity than the rest of the media.

Preparing the cuttings, planting and experiment managements: Root cuttings were harvested under the ground by carefully digging to avoid wounded inducing to the roots. Choose roots with a diameter of about 1.0 - 1.2 cm then cuttings without scratches, crushed on the outside into sections of about 3 cm long, then dipped in NAA solution the concentration of 1000 ppm in 2 sec for initiating rooting. After that, the 3 cm length cuttings were planted on sand and watered twice a day. After 20 days, the cuttings were transplanted into grow bag containing 20 cm^3 of media. Slow-release organic fertilizer (Bounce Back) was applied at a dose of 2 g/bag at 30 and 60 days after planting (DAPs). Two weeks after transplanted, all of the cuttings were watered twice a day in the early morning and late afternoon.

2.3. Data collection and statistics

During 75 days DAPs, data were measured from one-third of the cuttings (10 cuttings per plot).

Root number (roots/cutting): Count all of the main roots growing from the cutting; Root length (cm): Measure the length of main roots from the root neck to the longest point of the root; Shoot length (cm): Measure the length of shoot from the base of the trunk to the highest point of the shoot; Number of leaves/cutting (leaves/cutting): Count all of the leaves growing from the shoot; Chlorophyll Content Index (CCI): Use a chlorophyll meter (CCM-200 Plus) to measure at the center of the leaves.

Fresh roots matter (g/cutting): Weigh all the roots after removing the media.; Dry roots matter

Symbol	Growing media
G1 (control)	50% sand $+ 25%$ rice husk ash $+ 25%$ coco peat
G2	25% sand $+$ 50% rice husk ash $+$ 25% coco peat
G3	25% sand + $25%$ rice husk ash + $50%$ coco peat
G4	50% rice husk ash + $50%$ coco peat
G5	50% rice husk ash + $50%$ vermicompost
G6	50% coco peat+ $50%$ vermicompost
$\mathbf{G7}$	25% rice husk ash + $25%$ coco peat + $50%$ vermicompost
G8	25% rice husk ash + 50% coco peat + 25% vermicompost
G9	50%rice husk as h $+$ 25% coco peat $+$ 25% ver micompost

 Table 1. Nine types of media in the experiment

(g/cutting): Weigh all the roots after removed the media and dried at 70°C until the weight is constant; Fresh shoot matter (g/cutting): Weigh all the shoot; Dry shoot matter (g/cutting) (g/cut): Weigh all the shoot after dried at 70°C until the weight is constant.

Dickson Quality Index: DQI = Total dry matter/[(Shoot height/ stem base diameter) + (Dry shoot matter/Dry root matter)]; Percentage of live cuttings (%) = (Total number of live cuttings/30) × 100; Commercialarable ratio (%) = (Total qualified cuttings/30) × 100.

Economic efficiency: Total expenditure (VND/1000 cuttings): Cost of agricultural materials + electricity and water + labor; Total revenue (VND/1000 cuttings): Number of cuttings qualified x selling price; Profit (VND/1000 cuttings) = Total revenue minus total expenditure; Profit ratio (times) = Profit/Total expenditure.

The collected data were statistically calculated using Microsoft Excel, and analysed with ANOVA and Duncan's test at the significance level $\alpha = 0.05$ using the SAS 9.1 program.

3. Results and Discussions

3.1. Effect of growing media on root number and root length of *Cyclea barbata*

Different types of media have different physical and chemical properties, especially in terms of bulk density and porosity. Long (1993) stated that media plays an important role in the success of propagation by cuttings. The number of roots and root length of the cuttings at the time of 75 DAPs are presented in Table 3.

The results of Table 3 show that the number of roots on the cuttings is significantly different (P

= 0.01) when grown on different types of media. Cuttings were grown on the combined media of 25% rice husk ash + 25% coco peat + 50% vermicompost obtained the highest number of roots (18.27 roots/cutting) and higher than the control (14.67 roots/cutting). While, the mixture of 50% sand: 25% rice husk ash: 25% coco peat (14.67%) gave lest root number. This suggested that when sand was replaced with organic substances helped to enhance organic content, nutrients as well as improve the water holding capacity of the growing media. It turned out that a higher organic content media could support the cuttings to form more roots.

However, different types of media did not affect the root length of cuttings. At the time of 75 DAPs, the root length ranged from 18.03 to 23.07 cm, relatively equivalent to the deep of the media. The porosity of the different types of media ranged from 53,330 to 61,268% (Table 2). There was no significant difference in the ability of roots to penetrate inside the growing media.

3.2. Effect of growing media on shoot length, number of leave and chlorophyll content of *Cyclea barbata*

The leaves are the main harvested part of *Cy*clea barbata. Leaves are distributed on the shoot, so the shoot has a high length, which is the basis for forming a large number of leaves. Besides, the number and the chlorophyll content index of leaves indicate the photosynthetic capacity of the plant which relates to the harvest yield. When growing on a suitable media, the plant usually has a large number of leaves and a high chlorophyll content index.

The results of Table 4 show that the length of shoots and the number of leaves of the cuttings were significantly different under influence of dif-

Parameter	Unit	G1	G2	G3	G4	G5	G6	G7	G8	G9
$pH_{H_2O}(1:5)$		6.492	6.411	6.214	6.134	6.690	6.296	6.493	6.215	6.285
EC(1:5)	$\mathrm{mS/cm}$	1.703	2.224	2.336	2.557	3.372	3.496	3.484	2.776	3.052
Total N	%	0.113	0.115	0.223	0.225	0.699	0.913	0.806	0.669	0.605
Total organic	%	8.966	8.967	17.930	17.932	11.490	29.415	20.452	20.673	16.537
C/N ratio		58.713	57.667	60.575	59.746	16.440	28.218	25.377	30.901	27.334
Total P_2O_5	%	0.033	0.037	0.062	0.066	0.868	0.920	0.894	0.493	0.411
Total K_2O	%	0.090	0.091	0.178	0.178	0.769	0.943	0.856	0.560	0.610
Bulk density	g/cm^3	0.981	0.801	0.768	0.588	0.619	0.552	0.586	0.570	0.562
Density	g/cm^3	2.101	1.817	1.775	1.491	1.461	1.378	1.419	1.434	1.451
Porosity	%	53.330	55.923	56.770	60.586	57.634	59.920	58.743	60.266	61.268
$\operatorname{Humidity}$	%	30.869	33.888	33.967	36.986	33.700	33.858	33.779	35.422	32.103

ferent types of media at the time of 75 DAPs. The cuttings grew on a mixed media including 25% husk ash + 25% coco peat + 50% vermicompost reached the highest shoot length (116.41 cm) and higher than the control (83.05 cm). Cuttings planted cut a mixed media of 25% sand + 50% rice husk ash + 25% coco peat got the lowest shoot length (72.17 cm).

Similarly, cuttings were planted on a mixed media of 25% husk ash + 25% coco peat + 50% vermicompost gave the highest number of leaves (15.53 leaves/cutting), even this was not statistically different from other types of media. While the cuttings planted on mixed media of 50% rice husk ash + 50% coco peat got the lowest leaf number (8.33 leaves/cutting).

Thus, it was clear that addition of vermicompost to growing media helped plants grow better. It was also obvious that the shoot length and number of leaves of cuttings grew in media containing vermicompost thanks to an improvement in the content of essential macro as well as micronutrients to the plantlets. This result was in agreement with the conclusions by Bhadwaj (2014), Vo & Wang (2014) and Pham & Nguyen (2018) in the propagation of various plants. Leaves of the cutting planted on different types of growing media showed no statistically significant differences in leaf chlorophyll index (CCI). At the time of 75 DAPs, the leaf chlorophyll index of the cuttings ranged from 25.17 to 28.77 CCI.

3.3. Effect of growing media on fresh roots matter (g/cutting), dry roots matter, fresh shoot matter and dry shoot matter of *Cyclea barbata*

Biomass of shoots and leaves indicates the ability of the plant to absorb water and nutrients. Types of media have different physical and chemical properties effecting the biomass accumulation of cuttings.

The results of Table 5 show that the fresh and dry roots matter of cuttings was highest on mixed media of 25% rice husk ash + 25% coco peat + 50% vermicompost (4.08 and 0.54 g/cutting, respectively) and significantly different from the control. The cuttings planted on mixed media containing 25% sand + 25% rice husk ash + 50% coco peat gained the lowest fresh and dry root weight (1.60 and 0.21 g/cutting, respectively). The result indicated that different growing me-

	Parameters		
Growing media	Number of roots (root/cutting)	Length of root (cm)	
50% S : 25% HA : 25% CP (Control)	14.67^{bcd}	21.63	
$25\%~\mathrm{S}$: 50% HA : 25% CP	14.00^{cd}	19.70	
$25\%~\mathrm{S}$: $25\%~\mathrm{HA}$: $50\%~\mathrm{CP}$	12.80^{d}	18.03	
50% CP : 50% HA	16.83^{ab}	19.93	
50% HA : $50%$ VC	$16.43^{ m abc}$	22.37	
50% CP : 50% VC	$16.33^{ m abc}$	18.87	
25% HA : $25%$ CP : $50%$ VC	18.27^{a}	23.07	
25% HA : $50%$ CP : $25%$ VC	17.23^{ab}	22.57	
50% HA : $25%$ CP : $25%$ VC	16.00^{abc}	22.07	
CV (%)	9.03	11.23	
$\mathrm{F}_{\mathrm{value}}$	4.30^{**}	1.78^{ns}	

Table 3. Effect of growing media on number of roots and root length of *Cyclea barbata* 75 days after planting

S: Sand, HA: Husk ash, CP: Coco peat, PT: vermicompost; In the same column, numbers with the same character are statistically insignificant difference; ns: non-significant, **: the difference is statistically significant at P = 0.01.

Table 4. Effect of growing media on shoot length, number of leave and Content of Chlorophyll Index ofCyclea barbata 75 days after planting

Growing media –		Parameters	
	Shoot length (cm)	Number of leave (leave/cutting)	Content of Chlorophyll Index (CCI)
50% S : 25% HA : 25% CP (Control)	83.05^{bc}	11.43^{bc}	25.17
25% S : 50% HA : 25% CP	$79.93^{ m bc}$	$11.27^{\rm bc}$	28.77
25% S : 25% HA : 50% CP	72.17^{c}	$9.33^{ m cd}$	27.23
50% CP : 50% HA	80.13^{bc}	8.33^{d}	28.67
50% HA : $50%$ VC	$101.85^{\rm ab}$	$13.60^{\rm ab}$	28.53
50% CP : 50% VC	98.97^{ab}	$13.90^{\rm ab}$	25.37
25% HA : $25%$ CP : $50%$ VC	$116.41^{\rm a}$	15.53^{a}	25.73
25% HA : $50%$ CP : $25%$ VC	$100.48^{\rm ab}$	$14.70^{\rm a}$	27.17
50% HA : $25%$ CP : $25%$ VC	$89.41^{\rm bc}$	$13.73^{\rm ab}$	27.73
CV (%)	12.50	12.34	9.22
Fvalue	4.54**	7.77**	0.97 ^{ns}

S: Sand, HA: Husk ash, CP: Coco peat, PT: vermicompost; In the same column, numbers with the same character are statistically insignificant difference; ns: non-significant, **: the difference was statistically significant at P = 0.01.

dia did not impact statistically significant to both fresh and dry weights of the cuttings.

Similarly, fresh and dry leaf weights differed significantly when the cuttings were grown on different types of media. Fresh weight and dry were highest when cuttings were planted on a growing media containing 25% husk ash + 25% coco peat + 50% vermicompost (5.91 and 1.68 g/cutting, respectively) and higher than the control. However, this was not statistically different from weight of cuttings plated on mixed media of 25% rice husk ash + 50% coco peat + 25% vermicompost (5.45 and 1.50 g/cutting). It was clear that the media supplemented with vermi-

compost became lack of nutrients in the media is lower, thus affecting the growth of the cuttings. Cuttings planted on mixed media of 25%sand + 25% rice husk ash + 50% coco peat (3.26 and 0.79 g/tree, respectively) gained the lowest weight. These results were consistent with that ofreported by Vo & Wang (2014).

3.4. Effect of growing media on Dickson Quality Index, dry roots matter, fresh shoot matter and dry shoot matter of *Cyclea barbata*

The plantlet quality estimated based on Dickson Quality Index (DQI) is considered as a mea-

Crowing modia	Parameters			
Growing media	Fresh roots	Dry roots	Fresh shoot	Dry shoot
	matter	matter	matter	matter
	(g/cutting)	(g/cutting)	(g/cutting)	(g/cutting)
50% S : 25% HA : 25% CP (Control)	2.08^{d}	$0.25^{\rm dc}$	4.35^{bc}	1.10^{bcd}
$25\%~\mathrm{S}$: 50% HA : 25% CP	1.88^{de}	$0.23^{\rm dc}$	$3.72^{\rm cd}$	$0.90^{\rm cd}$
$25\%~\mathrm{S}$: $25\%~\mathrm{HA}$: $50\%~\mathrm{CP}$	1.60^{e}	$0.21^{\rm d}$	3.26^{d}	$0.79^{\rm d}$
50% CP : 50% HA	$2.87^{\rm c}$	$0.32^{\rm c}$	$3.48^{\rm cd}$	$0.86^{\rm d}$
50% HA : $50%$ VC	3.31^{b}	0.43^{b}	5.05^{ab}	1.39^{ab}
50% CP : 50% VC	$3.91^{\rm a}$	0.49^{ab}	5.09^{ab}	$1.42^{\rm ab}$
25% HA : $25%$ CP : $50%$ VC	$4.08^{\rm a}$	0.54^{a}	5.91^{a}	1.68^{a}
25% HA : $50%$ CP : $25%$ VC	$3.71^{\rm ab}$	0.48^{ab}	5.45^{a}	1.50^{ab}
$50\%~{\rm HA}$: $25\%~{\rm CP}$: $25\%~{\rm VC}$	3.34^{b}	$0.43^{\rm b}$	4.88^{ab}	1.29^{ab}
CV (%)	7.88	13.88	12.55	19.13
F_{value}	46.38^{**}	16.94^{**}	7.75**	5.58^{**}

Table 5. Effect of growing media on fresh roots matter (g/cutting), dry roots matter, fresh shoot matter and dry shoot matter of *Cyclea barbata* 75 days after planting

S: Sand, HA: Husk ash, CP: Coco peat, PT: vermicompost; Values followed by different lowercase letters in superscripts were significantly different at P < 0.01.

Table 6. Effect of growing media on Dickson Quality Index, dry roots matter, fresh shoot matter and
dry shoot matter of Cyclea barbata 75 days after planting

Growing media		Parameters	
Growing media	Dickson Quality Index	Percentage of live cuttings (%)	Percentage of Commer- cialarable cuttings (%)
50% S : 25% HA : 25% CP (Control)	0.016^{ab}	58.89	50.00
25% S : 50% HA : 25% CP	$0.014^{\rm b}$	60.00	51.11
25% S : 25% HA : 50% CP	0.013^{b}	56.67	50.00
50% CP : 50% HA	0.014^{b}	60.00	52.22
50% HA : $50%$ VC	0.017^{ab}	62.22	54.45
50% CP : 50% VC	0.019^{a}	61.11	56.67
25% HA : $25%$ CP : $50%$ VC	$0.019^{\rm a}$	64.44	57.78
25% HA : $50%$ CP : $25%$ VC	$0.019^{\rm a}$	63.33	57.78
50% HA : $25%$ CP : $25%$ VC	$0.019^{\rm a}$	61.11	54.44
CV (%)	13.662	6.23	9.46
Fvalue	3.680^{*}	1.14^{ns}	1.15^{ns}

S: Sand, HA: Husk ash, CP: Coco peat, PT: vermicompost; In the same column, numbers with the same character are statistically insignificant difference; ns: non-significant, *: the difference is statistically significant at P = 0.05.

sure for evaluating morphological characteristics (Johnson & Cline, 1991), which is a good indicator of seedling and plantlet quality based on the calculation of the healthiness and biomass distribution (Fonseca, 2002).

Table 6 shows that cuttings grown on different growing media differed statistically significantly (P < 0.05). In which, cuttings were grown on a mixed media containing vermicompost reached the best quality (0.019) and was not significantly different from the control (0.016). The mixed media of 25% sand + 25% rice husk ash + 50% coco peat showed the lowest plantlet quality (0.013).

Table 6 also shows that the survival percentage and ratio of Commercialarable plantlets achieved from different media were not statistically different. At the time 75 DAPs, the survival rate of cuttings ranged from 58.89 to 64.44% and the percentage of Commercialarable plantlets ranged from 50.11 to 57.78%.

Growing media	Total revenue (VND/1000 cuttings)	Total ex- penditure (VND/1000 cuttings)	Profit (VND/1000 cuttings)	Profit ratio (time)
50% S : 25% HA : 25% CP (Control)	$7,\!500,\!000$	$4,\!526,\!200$	$2,\!973,\!800$	$0,\!66$
25% S : 50% HA : 25% CP	$7,\!666,\!500$	$4,\!526,\!200$	$3,\!140,\!300$	$0,\!69$
25% S : 25% HA : 50% CP	7,500,000	4,601,200	$2,\!898,\!800$	$0,\!63$
50% CP : 50% HA	7,833,000	4,601,200	3,231,800	0,70
50% HA : $50%$ VC	8,166,000	5,251,200	2,914,800	0,56
50% CP : 50% VC	8,500,500	5,401,200	3,099,300	0,57
25% HA : $25%$ CP : $50%$ VC	8,667,000	5,326,200	3,340,800	$0,\!63$
25% HA : $50%$ CP : $25%$ VC	8,667,000	5,001,200	$3,\!665,\!800$	0,73
50% HA : 25% CP : 25% VC	8,166,000	4,926,200	3,239,800	0,66

Table 7. Financial efficiency for 1,000 cuttings

S: Sand, HA: Husk ash, CP: Coco peat, PT: vermicompost.

3.5. Financial efficiency for 1,000 cuttings of Cyclea barbata

Financial efficiency is an important factor that is interested in producers. Finding a suitable type of media can help reduce expenditure and increase revenue to achieve high profits.

The financial efficiency of Cyclea barbata cuttings on different types of media is presented in Table 7. The results showed that the plantlets planted on mixed media of 25% husk ash + 50% coco peat + 25% vermicompost achieved the highest profit (3,665,800 VND/1,000 plantlets) with a profit margin of 0.73 times. The plantlets planted on mixed media of 25% sand + 25% rice husk ash + 50% coco peat got the lowest profit (only 2,898,800 VND/1,000 plantlets).

The growing media 25% husk ash : 25% coco peat : 50% vermicompost gave the best growth and seedling quality, but not statistically different from the 25% husk ash growing media: 50% coco peat: 25% vermicompost. On the other hand, the addition of large amounts of vermicompost (50%) to the media increases investment costs, thereby reducing profits and profit margins.

4. Conclusions

Cuttings of *Cyclea barbata* planted the mixed media of 25% husk ash + 50% coco peat + 25% vermicompost was the best among tested media, the plantlet planted on media got highest number of roots (17.23 roots/cutting), longest shoot length (100.48 cm), highest number of leaves (14.70 leaves/cutting) as well as fresh and dry root matter (3.71 and 0.48 g/cutting), fresh and dry shoot matter (5.45 and 1.50 g/cutting). DQI coefficient reached the highest (0.019) at 75 DAPs, the profit was 3,665,800 VND/1000 cuttings with a profit margin of 0.73 times.

Conflict of interest

The authors declare no conflict of interest.

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