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Production of High Quality Planting Materials Through Breeding for Four Important Herbal Species

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ABSTRACT

With the growing interest in these species as a source of new pharmaceutical products and the increasing demand for herbal products in Malaysia, the demand for its raw materials is also increasing. Therefore, initiatives have been taken by Forest Research Institute Malaysia (FRIM) to select and to produce high quality planting materials for future uses. To our knowledge, most of the raw materials used in producing herbal products and its development were being sourced from the wild, with little knowledge on the quality of the raw materials. In order to ensure the quality and sustainability of raw materials in the market, it is important to come out with suitable breeding strategy for the selected species. This paper highlights research by FRIM on collecting, screening, selecting and producing high quality planting materials for the four species based on their growth performance and bioactive compounds. Several research on the production of high quality planting materials for four important medicinal plants has been initiated by FRIM starting from the 11th Malaysian Plan until now. This paper discusses about four selected species namely *Chromolaena odorata* (kapal terbang), *Andrographis paniculata* (creat or green chiretta), *Beackea frutescens* (false ru) and *Senna alata* (candle bush). These species are found to have a significant benefit to the human's health. *C. odorata* (kapal terbang) leaf extract are found to have relatively strong inhibition on platelet-activating factor (PAF) receptor binding *in vitro*, indicating an anti-inflammatory activity, which is favourable for wound healing. A major bioactive compound in *A. paniculata*, andrographolide has shown anticancer potential in various research. Whereas, *B. frutescens* has a potential to be used as anti-gout remedies. Research conducted by FRIM showed that active compound in the leaves and stems of *B. frutescens* extract are effective in inhibiting uric acid formation and promoting uric acid secretion. Whereas for *S. alata*, the highest content of major flavonoid glycoside compound, kaempferol-3-O-gentiobioside (K3G), has an anti-inflammatory effect detected in leaf extracts. Germplasm plots for the species were also established in FRIM for future breeding activities. The outputs derived from this study will support the herbal industries in getting quality raw materials in the future. By using high quality plants will also increase the value of pharmaceutical products in the market. It is anticipated that herbal industries and interested party will seek

FRIM for high quality seeds and seedlings materials for the development of their products as well as for the establishment of commercial herbal plantation.

Keywords: breeding strategy, selection, growth performance, bioactive compound, product development, commercial plantation

INTRODUCTION

Plant breeding is the science of changing the traits of plants in order to produce desired characteristics (Phoelman & Sleper, 1995). It has been used to improve the quality of nutrition in products for humans and animals (Hartung & Schiemann, 2014). The goals of plant breeding are to produce crop varieties that has unique and superior traits for a variety of agricultural applications. The most frequently addressed those traits are related to biotic and abiotic stress tolerance, grain or biomass yield, end-use quality characteristics such as taste or the concentrations and others (Willy, 2010). Plant breeding basically starts with selection of high yielding individuals. It is a process of preservation of certain individual plants of desirable characters or in simplest form selection means choosing plants of one's choice. It is the basis of all crop improvement (Carol, 2000). In Malaysia, many plant improvement was made for vegetables, rice and fruits through selecting superior plants from local and foreign genetic resources (Nordin et al., 2007). Very limited effort was done to improve herbal plants. In National Agrofood Policy (NAP 2011-2020), herbal plants have been classified as one of the high value agriculture activity. Several strategies were outlined by the government to strengthen the herbal industry in Malaysia. One of the highlighted strategy was to ensure sustainable and consistent supply of raw materials to the downstream sector.

Currently, the industries are facing the issue of insufficient supply of high quality herbal raw herbal materials for the development of herbal products. This is because the industries have to depend on the supply from the wild where the quality of the herbal raw materials is uncertain and the supply is not sustainable. Study by Rohana et al. (2017) found that 83% of *L. pumila* raw materials were harvested from natural forests and only 17% were cultivated. Looking at this issue, Forest Research Institute Malaysia (FRIM) has initiated to select and to produce high quality planting materials for future uses. To our knowledge, most of the raw materials used in producing herbal products and its development were being sourced from the wild, with little knowledge on the quality of the raw materials. In order to ensure the quality and sustainability of raw materials in the market, it is important to come out with suitable breeding strategy for the selected species. Under the 11th Malaysian Plan project (2017-2020), four important herbal species were selected for breeding work. The four species are *Chromolaena odorata* (kapal terbang), *Andrographis paniculata* (creat or green chiretta), *Beackea frutescens* (false ru) and *Senna alata* (candle bush). These species are found to have a significant benefit to the human's health. *C. odorata* (kapal terbang) leaf extract are found to have relatively strong inhibition on platelet-activating factor (PAF) receptor binding *in vitro*, indicating an anti-inflammatory activity, which is favourable for wound healing (Ling et al. 2007). A major bioactive compound in *A. paniculata*, andrographolide has shown anticancer potential in various research (Zoya et al. 2021). Whereas, *B. frutescens* has a potential to be used as an anti-gout remedies. Research conducted by FRIM showed that active compound in the leaves and stems of *B. frutescens* extract are effective in inhibiting uric acid formation and promoting uric acid secretion (Fadzureena et al., 2013). Whereas for *S. alata*, the highest content of major flavonoid glycoside compound, kaempferol-3-O-gentiobioside (K3G), has an anti-inflammatory effect detected in leaf extracts (Moriyama et al. 2003). This paper highlights the achievement made by FRIM in producing high quality planting materials started from the collection process, chemical screening and selecting high quality planting materials for the four species based on their growth performance and bioactive compounds.

MATERIAL AND METHOD

Collection of accessions from natural populations in Peninsular Malaysia

Chromolaena odorata

A total of 34 accessions of *C. odorata* collected from three populations of Kota Tinggi, Johor (accessions 1, 2, 4, 5, 12, 13, 15, 16, 18, 19, 20, 21, 22, 25, 28), Jasin, Melaka (accessions 4, 9, 12, 23, 26, 28, 29, 30) and Maran, Pahang (accessions 1, 2, 3, 4, 5, 7, 10, 11, 18, 19, 21) were selected for screening their chemical constituents. The accessions were coded as JKT for accessions from Johor, MKT for accessions from Melaka and CKT for accessions from Pahang. The topographic information such as coordinates, altitudes and dates of assessment were also recorded. Leaf sample were collected from each of the populations for chemical screening process. The morphology characteristics of all mother plants such as number of clumps, height, diameter, leaf length, leaf width, crown diameter and number of nodes were measured (Table 1). These stumps were then planted and grown at FRIM's nursery under 50% shades.

Table 1: Morphological characteristics of *Chromolaena odorata* accessions from three populations

Population	No of samples	No. of Clumps	Height (m)	Diameter (mm)	Leaf Length (cm)	Leaf Width (cm)	Silara X (cm)	Silara Y cm)	No of Nodes
JKT	15	2-5	1.38-2.35	5.2-12.3	9.4-12.5	4.5-7.2	15-48	20-57	9-23
MKT	8	2-5	1.32-2.50	5.6-12.6	8.0-19.8	3.5-9.0	10-55	9-45	8-24
CKT	11	1-7	1.35-2.74	3.2-12.3	8.7-13.4	4.7-7.9	21-55	24-72	9-27

Fig. 1. Stumps of *Chromolaena odorata* were collected from three populations and replanted at FRIM's nursery for screening process

Andrographis paniculata

A total of 30 accessions of *A. paniculata* were collected from each of the five different populations in Peninsular Malaysia. The five populations were coded as JHB (Kota Tinggi, Johor), CHB (Kuantan, Pahang), AHB (Tapah, Perak), RHB (Mata Ayer, Perlis) and THB (Kuala Terengganu, Terengganu). Each location was tagged using Global Positioning System (GPS) and topographic information such as location coordinates and altitudes were also recorded. Morphological characteristics of all accessions such as height (cm), diameter (mm), leaf length (cm), and leaf width (cm) were measured (Table 2). Leaf samples were collected from each accessions for chemical screening process. The collected accessions were then acclimatized at FRIM's nursery in plastic trays (9.5 cm height x 40 cm width x 53 cm length) and watered daily.

Table 2: Morphological characteristics of *Andrographis paniculata* accessions from five populations

Population	**No of Samples**	**No. of clumps**	**Height (cm)**	**Diameter (mm)**	**Leaf length (cm)**	**Leaf width (cm)**
JHB	30	3-14	22-102	1.8-11.9	4.5-11.5	1.0-3.7
CHB	30	2-14	29-120	2.3-10.7	5.0-13.0	1.0-3.6
AHB	30	3-15	36-89	2.3-11.2	3.6-14.5	1.0-3.8
RHB	30	2-15	24-87	2.3-8.7	5.0-13.0	1.1-3.6
THB	30	1-10	23-90	2.4-7.0	3.0-10.3	1.0-4.9

Fig. 2. *Andrographis paniculata* accessions were collected from five populations and replanted at FRIM's nursery using plastic tray

Baeckea frutescens

A total of 84 accessions of *B. frutescens* were identified from three populations such as i) Gunung Pulut, Perak, ii) Setiu, Terengganu, and iii) Sungai Baging, Pahang. A few phenotypically superior trees showing good growth, full of branches, superior height, and bole diameter were selected for the study. Only leaves and secondary branches parts were cut from the mother tree and packed into the plastics bags (Figure 3) for chemical screening. All samples were labelled with different codes such as ACA1 to ACA30 (Gunung Pulut, Perak), TCA1 to TCA30 (Setiu, Terengganu), and CA1 to CA30 (Sungai Baging, Pahang) for easier identification (Table 3). The topographic information such as coordinates, altitudes, and dates of assessment were also recorded.

Table 3: Morphological characteristics of *Beackea frutescens* accessions from three populations

Population	No of Samples	Plant Height (cm)	Diameter (mm)
ACA	28	160-350	1.50-5.30
TCA	28	269-581	1.50-7.00
CCA	28	160-374	1.40-5.50

Fig. 3. Collection and preparation of *Baeckea frutescens* samples at field

Senna alata

A total of 120 accessions of *Senna alata* were identified from four populations such as i) Kuala Selangor, Selangor ii) Raub, Pahang and iii) Ketereh, Kelantan and iv) Kuala Pilah, Negeri Sembilan. A few phenotypically superior genotypes showing good growth, full of branches, superior height, and diameter were selected for the study. The leaf samples were collected for chemical screening and stumps of selected plants were dug out and brought back to FRIM (Figure 4). These materials were transplanted into polybags and placed at the nursery. All collected stumps were labelled differently such as BG1-BG30 (Kuala Selangor, Selangor), CG1-CG30 (Raub, Pahang), DG1-DG30 (Ketereh, Kelantan) and NSG1-NSG30 (Kuala Pilah, Negeri Sembilan). Morphological data were recorded as shown in Table 4.

Table 4: Morphological data of *Senna alata* accessions from four populations

Populations	No of Samples	No. of clumps	Height (m)	Diameter (cm)	Leaf length (cm)	Leaf width (cm)
BG	30	1-3	1.0-4.2	1.2-6.8	10.0-16.3	4.0-7.8
CG	30	1-5	0.5-5.3	0.7-7.5	10.5-17.2	3.5-6.5
DG	30	1-8	1.02-2.56	1.0-5.4	10.2-18.5	4.4-7.8
NSG	30	2-9	1.03-2.97	1.0-4.0	9.3-17.4	3.8-7.5

Fig. 4. Collection and preparation of *Senna alata* stumps from four populations

Screening of chemical constituents and/or bioactivity

The collected leaf samples from all accessions for the four species were carefully covered with tissue paper and packed in plastic bags before they were transported back to FRIM in Kepong, Selangor. Upon arrival, the samples were deposited to FRIM's chemistry laboratory for chemical analysis using High Performance Liquid Chromatography (HPLC) except for *B. frutescens,* where screening of xanthine oxidase inhibitory assay was carried out for this species. Chemical screening was not carried out due to the reason that chemical compounds of the species has been previously patented (PI 2014000187).

Screening of naringenin 4'-methyl ether and aromadendrin 4'-methyl of *Chromolaena odorata* extract

A total of 0.5 g *C. odorata* dry samples was dissolved in 5 mL of MeOH and sonicated for 15 minutes. Each of resulting solution were filtered through membrane filter (pore size 0.45 µm) prior to analysis. All samples were diluted 5 times for quantification of naringenin 4'-methyl ether and aromadendrin 4'-methyl ether prior to analysis. A series of working solution with concentration range from 5-1000 µg/mL of naringenin 4'-methyl ether and aromadendrin 4'-methyl ether were prepared in MeOH. The samples were analyzed by means of High Performance Liquid Chromatography (HPLC) system (Waters Delta 600 with 600 controllers) with Photodiode array detector (Water 2996). A WATER x- bridge C-18 was used and for elution of constituents, two solvent denoted as A and B was employed. A was 0.1% aqueous formic acid and B was acetonitrile. Initial conditions

were 80% A and 20% B. Analysis was performed using gradient solvent system in 45 minutes of running time. The flow rate used was 1.0 mL/min and the injection volume was 10 µL.

Screening of andrographolide of *Andrographis paniculata* extract

A total of 0.5 g of sieved powder material (500 µm) was prepared. About 5 mL of methanol was added and the mixture was ultra-sonicated for 15 minutes. The resulting solution was filtered using 0.45 µm syringe filter prior to analysis. The samples were analysed by means of a HPLC system (Waters 2535 quarternary gradient module, Waters 2707 Autosampler and Waters 2998 photodiode array detector). A Phenomenex Luna C18 column was used (4.6 mm i.d. x 250 mm) and for elution of the constituents, three solvents denoted as A, B and C were employed. A was 0.1% aqueous formic acid, B was acetonitrile and C was methanol. The flow rate used was 1.0 mL/min and the injection volume were 10 µL. The retention times and UV spectra of the targeted compounds were analysed at wavelength of 220 nm.

Screening of *in-vitro* xanthine oxidase inhibitory assay in *Beackea frutescens* extract

Extracts from *B. frutescens* leaves was obtained using the solvent extraction method, whereby fresh *B. frutescens* leave samples were cut into small pieces and dried in a ventilated oven for three days before being grounded into fine powder form. About 30 g of ground powder were soaked in a mixture of solvents at 1:10 ratio for a few hours at a set temperature. The extract was filtered using suitable filtering method to remove unwanted fragments in the extracts. Excess solvent was removed using rotary evaporator. The dark green crude extract was stored in a freezer at -20°C for further analysis. The average yield of *B. frutescensns* crude extract was about 4%.

Method of *in vitro* XO inhibitory analysis was adapted from Noro et al. (1983) with a few modifications. The *B. frutescens* extract was dissolved with dimethyl sulfoxide (DMSO) at 20 mg/mL as extract solution. The potassium phosphate monobasic (KH_2PO_4) buffer solution, pH 7.5 was used as the main buffer in the system. The XO enzyme solution was freshly prepared. The 96-well microplate was pipetted in with main buffer solution, extract solution and XO enzyme before incubated for 15 min at 25°C. After incubation period, the reaction of enzyme was induced with addition of substrate solution. The microplate was then incubated again for 10 min before analysis. The production of uric acid was measured using spectrophotometer at 295 nm. The allopurinol is a common drug used to treat gout patient. Therefore, allopurinol was selected as positive control in this assay system.

Screening of kaempferol 3-*O*-gentiobioside of *Senna alata* extract

A total of 12 accessions with the height of 1.0 m and above were selected from germplasm for chemical screening. HPLC analysis was conducted on a waters HPLC system equipped with a photodiode array (PDA). Samples were prepared in methanol by ultrasonication for 15 minutes. Sample solutions were then filtered using 0.45 µm PTFE membrane filter before eluted through a Phenomenex Luna C18 column (5 µm, 2 mm x 250 mm). A gradient elution was carried out with a mobile phase consists of 0.1% formic acid in water (A) and acetonitrile (B). The gradient profile used was 10% - 40% B in 20 min, 40% - 50% B in 20 min, 50% - 90% B in 10 min and equilibrate for 5 min. The flow rate was 1 mL/min and injection volume was 10 µL. Kaempferol 3-*O*-gentiobioside was detected by measuring the absorbance at 280 nm. For quantification analysis, a series of concentrations of kaempferol 3-*O*-gentiobioside in the range of 5 µg/mL to 1000 µg/mL were prepared and a calibration curves was plotted.

RESULTS AND DISCUSSION

Screening and selection of high yielding accessions for four herbal species

Chromolaena odorata

Ranges of concentration for both chemical constituents in 34 accessions of *C. odorata* are presented in Table 5. It showed that concentrations of aromadendrin 4'-methyl ether are ranging from 0.50-5.77% while naringenin 4'-methyl ether are ranging from 0.22-1.11% in the leaf samples of *C. odorata* (Table 5). The results also recorded that chemical constituent of aromadendrin 4'-methyl ether is higher in concentrations as compared to naringenin 4'-methyl ether. Findings of the study are in line with Hung et al. (2011), Johari et al. (2012) and Omukhua et al. (2016) where they reported the same chemical constituents were found in the species of *C. odorata* (L.) R.M. King & H. Rob.

Table 5. Range of concentration of two chemical constituents in 35 accessions of *Chromolaena odorata* leaf extract

Compound	Average Concentration in % Sampel ± RSD (W/W)
Naringenin 4'-methyl ether	0.22-1.11
Aromadendrin 4'-methyl ether	0.50-5.77

Based on the screening results, 14 accessions were identified for having more than average of 0.6% concentration of naringenin 4'-methyl ether chemical constituents, whereas for aromadendrin 4'-methyl ether, a total of 15 accessions which having more than average of 2.23% concentrations were selected as high yielding accessions (Table 6 & 7). Accession JKT22 which is from Kota Tinggi, Johor recorded the highest concentrations for both chemical constituents, naringenin 4'-methyl ether and aromadendrin 4'-methyl ether with 1.11 % and 5.77%, respectively.

Table 6. Selection of 14 high yielding accessions of *Chromolaena odorata* with high concentrations of naringenin 4'-methyl ether chemical constituent

No.	Accessions Code No.	Average Concentration in % Sampel ± RSD (W/W)
1.	JKT22	1.11 ± 0.01
2.	JKT25	0.91 ± 0.59
3.	CKT5	0.84 ± 0.44
4.	CKT11	0.80 ± 0.80
5.	JKT5	0.77 ± 5.43
6.	JKT20	0.76 ± 4.44
7.	MKT12	0.75 ± 1.72
8.	MKT28	0.75 ± 0.34
9.	MKT29	0.73 ± 1.76
10.	CKT19	0.69 ± 0.61
11.	CKT4	0.67 ± 2.98
12.	MKT9	0.66 ± 5.08
13.	CKT21	0.65 ± 4.78
14.	JKT4	0.62 ± 0.30

Table 7. Selection of 15 high yielding accessions of *Chromolaena odorata* with high concentrations of aromadendrin 4'-methyl ether chemical constituent

No.	Accessions Code No.	Average Concentration in % Sampel ± RSD (W/W)
1.	JKT22	5.77 ± 0.15
2.	CKT19	5.45 ± 0.35

3.	CKT1	4.15 ± 0.04
4.	CKT18	3.37 ± 1.35
5.	MKT26	3.18 ± 0.93
6.	JKT5	3.18 ± 0.72
7.	CKT3	3.12 ± 1.19
8.	CKT10	3.07 ± 1.12
9.	CKT2	2.92 ± 1.34
10.	CKT4	2.66 ± 0.84
11.	JKT2	2.55 ± 0.32
12.	CKT5	2.52 ± 0.36
13.	JKT18	2.48 ± 0.69
14.	CKT21	2.34 ± 0.07
15.	CKT11	2.24 ± 0.73

Andrographis paniculata

Table 8 indicated that all samples from four populations exhibited andrographolide constituent. Accessions from Terengganu (THB) and Perak (AHB) contained of andrographolide with more than the average (2.89 %). Accessions from Pahang (CHB) has recorded the same andrographolide value as the average which is about 2.83% ± 1.24. The lowest value of andrographolide was recorded by accessions from Johor (JHB) which is 2.59% ± 2.61. The concentration of andrographolide for four populations were presented during the injection time of nine to ten minutes between 220 and 230 nm. The concentration of andrographolide obtained in this study gave higher values as compared to results obtained by Ibrahim & Chong (2008) which reported that the concentration of andrographolide is more than 7.0 µg/mL under different dried sample storage temperature.

Table 8. Screening of high andrographolide chemical constituent in four populations of *Andrographis paniculata*

No.	Accessions Code No.	Average Concentration in % sampel ± RSD (W/W)
1.	THB	3.21 ± 0.75
2.	AHB	2.94 ± 1.18
3.	CHB	2.83 ± 1.24
4.	JHB	2.59 ± 2.61

Beackea frutescens

Based on the screening, it was recorded that 11 out of 84 genotypes of *B. frutescens* were selected as superior due to the value of more than 70% inhibitory activity on Xanthine Oxidase (XO) (Table 9). Accession TCA8 from Setiu, Terengganu showed the highest value (78.12 % ± 1.68) whereas TCA11 gave the lowest value (70.12% ± 2.02). It was observed that seven accessions were originated from population Setiu, Terengganu and three accessions from Gunung Pulut, Perak. Only one accession from Sungai Baging, Kuantan was categorized as superior. Beside *B. frutescens,* there were several herbs are proven to show as anti-gout potential mainly through XO inhibition assay. This assay is regarded as an essential standard for discovering anti-gout potential among medicinal plants (Bakar et al., 2018). It was reported that bitter melon (*Momordica charantia*) showed the highest percentage of XO inhibitory activity (96.5%) at 100 µg/mL using 70% methanol extract (Alsultanee et al., 2014). The highest XO inhibitory activity at 100 µg/mL (90.6%) was also discovered using the extract of aromatic ginger (*Kaempferia galangal*) (Nguyen et al., 2004). Superior genotypes usually referred to good growth characteristics and/or contained high quality active ingredients (Zobel and Talbert, 1984). High quality materials will be the added value to the end products. Therefore, it is important to screen the plants which has better characteristics than the common one. A few investigations on selection of superior plants was previously

conducted by FRIM on selected species such on *Citrus hystrix* (Farah Fazwa et al., 2005); *Citrus microcarpa* (Farah Fazwa et al., 2007); *Labisia pumila* (Farah Fazwa et al., 2012) and *Chromolaena odorata* (Farah Fazwa et al., 2019).

Table 9. Selection of eleven superior accessions of *Baeckea frutescens* with high inhibitory activity on xanthine oxidase

No.	Accessions code No.	Xanthine Oxidase (%) ± SEM[a]
1	TCA 8	78.12 ± 1.68
2	ACA 10	75.79 ± 4.79
3	TCA 15	74.14 ± 5.11
4	TCA 12	73.39 ± 2.98
5	TCA 6	72.24 ± 3.24
6	TCA 14	72.00 ± 1.95
7	CCA 9	71.77 ± 2.22
8	ACA 6	71.28 ± 1.28
9	TCA 13	71.10 ± 0.93
10	ACA 4	70.39 ± 3.57
11	TCA 11	70.12 ± 2.02

[a]Values are expressed as mean Inhibition (%) ± SEM of triplicate measurements from three independent experiments.

Senna alata

All samples from 11 accessions of *S. alata* exhibited the kaempferol-3-O-gentiobioside constituent. Results showed that the concentration of kaempferol-3-O-gentiobioside from 11 accessions have the minimum value of 0.43 ± 4.68% (w/w) and maximum value of 0.84 ± 7.47% (w/w) which were presented during the injection time of 17 minutes (Table 10). However, for further selection as superior accession, only six accessions were selected due to higher concentration of kaempferol-3-O-gentiobioside which are more than the average of 0.56% (w/w). The six accessions that categorized as superior were BG29 (0.84 ± 7.47% w/w), DG23 (0.80 ± 4.51% w/w), DG21 (0.76 ± 4.86% w/w), BG30 (0.71 ± 1.94% w/w), BG20 (0.66 ± 4.48% w/w) and BG25 (0.59 ± 5.11% w/w). Previous study conducted by Moriyama et al. (2003) reported that matured leaf of *S. alata* was found to contain the highest content of kaempferol-3-O-gentiobioside. The contents are reported to be at the ranged of 2.0 to 5.0% where the samples were taken from leaf of matured plants. However, it is not comparable with the results of this study, where the percentage of kaempferol-3-O-gentiobioside is lower because samples from this study were taken from relatively young plants.

Table 10. Percentage of kaempferol-3-O-gentiobioside ± RSD w/w for 11 selected *Senna alata* accessions

No	Accessions code No.	Percentage of KG3 ± RSD w/w
1	BG 29	0.84 ± 7.47
2	DG 23	0.80 ± 4.51
3	DG 21	0.76 ± 4.86
4	BG 30	0.71 ± 1.94
5	BG 20	0.66 ± 4.48
6	BG 25	0.59 ± 5.11
7	BG 6	0.52 ± 1.30
8	BG 24	0.50 ± 3.44
9	CG 30	0.50 ± 4.41
10	BG17	0.46 ± 2.91
11	NSG 12	0.43 ± 4.68

CONCLUSION

The superior accessions of the four herbal plants were identified based on its value of chemical and/or bioactive compounds. Currently, these accessions were planted in germplasm plots located in FRIM, Kepong, Selangor. The selected superior accessions will provide the opportunity for plant breeder to use these basic planting materials to initiate a breeding programme for the species. Through selection, plants from different origins can be improved to develop new clone or varieties. The established germplasm is also a method of genetic conservation and to sustain the production of quality planting materials in the future. As conclusion, all superior plants of *C. odorata*, *A. paniculata*, *B. frutescens* and *S. alata* produced from the studies are ready to be mass produced and promote to the interested industries.

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