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RESEARCH ARTICLE

Variation of bioactive secondary metabolites in Ceylon cinnamon (*Cinnamomum zeylanicum* Blume) in different climatic conditions, maturity status and propagation types

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ABSTRACT

This paper presents the variation of phytochemical quantities of leaves and bark of Cinnamomum zeylanicum Blume (Ceylon cinnamon) under different conditions such as ecological variation, maturity and physical state of the tree, and the propagation type. Among 49 climatic zones in Sri Lanka, six cinnamon growing zones including three wet zones (WL 1a, WL 2a, and WU 2b), two intermediate zones (IL 1a, IL 1b) and one dry zone (DL5) were selected for collecting cinnamon leaves and bark samples. The quantities of phytochemicals in leaves and bark extracts of Ceylon cinnamon according to the climatic difference, maturity stages (1-3, 3-5 and >5 y) and physical stage (flowering, fruiting, flushing), and seed and vegetative propagates (Sri Gamunu and Sri Wijaya) were evaluated. The results showed that climatic zones of WL 1a and IL 1a were the best climatic zones for obtaining high quantities of phytochemical classes in seeding and vegetative propagation types, respectively. Besides, the best phytochemical composition was observed in the leaves with >5 y and the bark of 3-5 y maturity of seed propagation varieties. Comparatively, bark from Sri Wijaya and leaves from Sri Gamunu which belonged to >5 y maturity contained better phytochemical composition in vegetative propagated varieties. According to the physical variations, the best phytochemical composition was observed in the bark samples which were collected in the flowering season, and the leaves samples collected in the fruiting season. Therefore, leaves in >5 y matured stage and bark in 3-5 y matured stage of seed propagation varieties of Ceylon cinnamon which are grown in WL 1a climatic zone are the best Cinnamon cultivar for the purpose of enhancing value chain and development cinnamon into different value adduct products.

***Keywords**: Cinnamomum zeylanicum, phytochemicals, agro-ecological zones, maturity stages, physical stages*

INTRODUCTION

Ceylon cinnamon or true cinnamon (*Cinnamomum zeylanicum* Blume) (Plate 1) is a tropical evergreen herb which belongs to the family Lauraceae and is indigenous to Sri Lanka as well as to southern parts of India (Meena *et al.*, 2018; Sooriyagoda *et al.*, 2021).

Cinnamon species have been recognised as medicinal herbs with a wide array of pharmacological properties (Keith, 2019; Blaszczyk *et al.*, 2021), many health care applications and uses in the traditional medicine (Ranasinghe *et al.*, 2013), and applications as food additives (Sooriyagoda *et al.*, 2021). Presently, the forms

of functional foods and nutraceuticals from cinnamon have drawn interest in the society as a preventive measure of health risks, mainly in "on-communicable diseases" as well as in the business world for the development of demanded consumers' products (Muhammad and Dewettinck, 2017).

Plate 1: *Cinnamomum zeylanicum* Blume.

About 250 species of the *Cinnamomum* genus have been recognized around the world; out of them, eight species are available in Sri Lanka. However, only *C. zeylanicum* Blume is commercially cultivated and the other seven are considered as wild (Liyange *et al*., 2017). *C. zeylanicum* Blume (Ceylon cinnamon) has been popular across the globe due to its characteristic properties such as fragrance, and pungent taste that are associated with its unique chemical compositions, especially in essential oils (Behbahani *et al*., 2020). Chemical composition of Ceylon cinnamon has been broadly explored and different part of the plant contain structurally diverse compounds that belong to the main class of phytochemicals such as terpenoids, alkaloids, flavonoids, polyphenols, saponins etc. (Husain *et al.*, 2018; Adarsh *et al*., 2020), and those are responsible for its immense medicinal value (Pathak and Sharma, 2021; Perera *et al*., 2021). Adequate literature is available on varying the composition and the quantity based on different parts of the plant such as roots, bark, leaves *etc*. (Rao and Gan, 2014). It can be anticipated that the composition and the quantity of phytochemicals produced by medicinal herbs may vary on the climatic conditions that are grown, the maturity stage of the trees as well as the genotype of plant, which is dependent on the propagation method. The related data are not available with respect to Ceylon cinnamon grown in Sri Lanka.

Cinnamon has been an economic crop in Sri Lanka for ages and it has been cultivated across the country for export purpose and local use. Sri Lanka has been credited as the largest producer and exporter of the high-grade cinnamon. About 90% of the world demand is supplied by Sri Lanka (Fonseka *et al*., 2018; Mohamed *et al*., 2019). Ceylon cinnamon is cultivated in different climatic zones in Sri Lanka using two different propagation methods, *i.e*. seeding and vegetative propagation (Senaratne and Pathirana, 2020). As Ceylon cinnamon is considered as unique due to its specific phytochemical profile, data on the quantities of

bioactive phyto-constituents produced in the tree based on its maturity stages, different ecological conditions, different physical stage of the tree and on propagative methods used for growing is found to be prime important to be used in enhancing its value chain and to make value addition. Therefore, the present study aimed on a quantitative investigation of important secondary metabolites that are present in the leaves and bark (Plate 2) extracts of Ceylon cinnamon under different conditions such as grown at different climatic zones of Sri Lanka, at different maturity stages and different physical stage of the tree, and based on the different propagation types used for growing.

Plate 2: Leaves (a) and bark (b) of the Ceylon cinnamon.

MATERIALS AND METHODS

Plant collections

Samples (inner bark and leaves of Ceylon cinnamon) were collected as below. All were done in triplicate (three sampling sites per each) from different ecological zones as given in Table 1, based on two propagative methods; seed propagative and vegetative propagative (Sri Gemunu and Sri Wijaya) methods. Five (5) ecological zones were considered for collecting seed propagative samples and two (2) ecological zones were considered for collecting vegetative propagated samples. Collected based on three maturity stages of the plant such as 1-3, 3-5, and >5 y per each type (seed propagation and vegetative propagation). All the samples were properly collected, cleaned, and labeled correctly at the "Cinnamon Research and Training Center", Thihagoda, Matara.

Sample preparation

After air drying for 7 d, all samples were finely grounded using a grinder and separately stored in amber-colored glass bottles until used.

Preparation of extracts for the analysis

The coarse powder (100 g) of each sample was macerated separately for 3 d in 200 mL of methanol using OHAUS magnetic stir (e-G51HS07C) and the fraction was filtered through Whatman® No.1 filter papers and concentrated up using HAHNVAPOR rotary evaporator (HS-2005S) under the vacuum pressure of 250 mmHg at 40 °C (Bulugapitiya, 2013).

Table 1: Harvested date, harvested location and agro-ecological zones of the samples used in the present study.

District	Agro-ecological zone	Sample collected site	Divisional secretariats division	Harvested date
Galle	WL1a	Galle-1	Pitigala	9th of Nov. 2018
	WL2a	Galle-2	Akmeemana	9th of Nov. 2018
Matara	IL1a	Hakmana-2	Hakmana	25th of Jan. 2018
	IL1a	Aththuduwa	Thihagoda	20th of Nov. 2017
	IL1a	Palolpitiya	Kamburupitiya	7th of Oct. 2017
	IL1b	Hakmana-1	Hakmana	25th of Jan. 2018
	WL2a	Makandura	Mulathiyana	14th of Dec. 2017
	WL2a	Deyyandara	Mulathiyana	14th of Dec. 2017
	WL1a	Akuressa	Akuressa	28th of Jan. 2019
Hambantota	IL1b	Katuwana	Katuwana	16th of May 2018
	IL1b	Middeniya	Katuwana	27th of Mar. 2018
	IL1b	Athpitiya	Walasmulla	18th of June 2018
	WL2a	Handugala	Walasmulla	18th of June 2018
	DL5	Suriyawewa	Suriyawewa	7th of April, 2018

Quantitative investigation of phytochemicals

Quantitative determination of phytochemicals in the methanolic extracts of leaves and bark of cinnamon under varying conditions were carried out by following standard gravimetric and colorimetric methods reported in the literature (Ezeonu and Ejikeme, 2016; Kokilananthan *et al.*, 2020).

Determination of alkaloids

To a 250 mL conical flask, 1 g of the powdered cinnamon sample was mixed with 100 mL of 10% acetic acid in ethanol. After standing for 4 h, the supernatant was separated and it was concentrated by heating in a water bath. About 25.0 mL of 25% ammonium hydroxide (excess volume) was added to the hot extract, covered and allowed to complete the precipitation. After standing for 24 h, the precipitate was filtered using 0.45 µm Sartorius cellulose nitrate filter paper and it was washed with 10 mL of 0.1 M ammonium hydroxide. The residue was dried in an oven at 40 ºC until it reached a constant weight. The percentage of alkaloids was calculated after weighing (Harbone, 1973; Ezeonu and Ejikeme, 2016).

Alkaloid % = (Weight of alkaloids/Dry weight of the sample) 100%

Determination of flavonoids

The powdered plant material (1 g) was treated with 50 mL of 80% methanol, and the solution was allowed to stand for 24 h. The supernatant was discarded and the residue was extracted with 10 mL of ethanol (thrice). After evaporation of solvents, the remains were cooled in a desiccator and the weight percentage was calculated (Harbone, 1973; Ezeonu and Ejikeme, 2016).

Flavonoids % = (Weight of flavonoids/Dry weight of the sample) 100%

Determination of saponins

About 1 g of powdered cinnamon sample was mixed with 20% ethanol solution and it was continuously stirred at 55 °C for 4 h. The resultant extract was separated through filtration (the procedure was repeated another two times). The combined extract was evaporated to one-half in a water bath at 90 ºC. After transferring to the separatory funnel, it was defatted with 10 mL of diethyl ether. To the resultant solution, 25.0 mL of n-butanol was added and it was washed twice with 5% sodium chloride. After standing, the residue with n-butanol was transferred to a crucible and evaporated. The solid matter formed was dried at 40 °C oven and chilled in a desiccator until it reached a constant weight.

Saponins % = (Weight of saponin/Dry weight of the sample) 100%

Determination of polyphenols (Ferrous tartrate method)

Standard calibration curve was prepared by following the procedure given (Figure 1). Tannic acid was used as the standard solution with concentrations ranging from 0.05-0.40 mg mL^{-1}. The stock solution was prepared by dissolving 1 g of tannic acid in a 1,000 mL volumetric flask and topped up with distilled water. Using serial dilution, the concentration series was prepared and each solution measured the absorbance at 540 nm. Blank solution: distilled water was used as the blank solution in replacement for cinnamon extract.

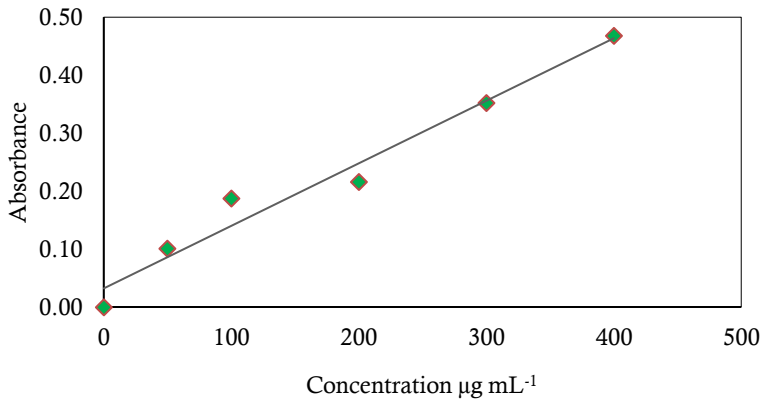

Figure 1: Calibration curve for tannic acid.

The aqueous solution containing 1 g of each cinnamon sample in 10 mL of distilled water was prepared by stirring for 3 h. The extraction was filtered and 1.0 mL of the extract was transferred to a 25 mL volumetric flask, then 5.0 mL dying solution and 15.0 mL buffer solution were added before volume up with distilled water. The solution was allowed to develop the coloration (Purple/violate) at room temperature for 20 min and the absorbance was measured using the spectrophotometer at 540 nm. The results were reported in

mg TAE/g FW (mg of tannic acid equivalence per g of fresh weight) (Liang *et al.*, 2003; Abugri and McElhenney, 2013).

Determination of oxalate

The powdered cinnamon (1 g) was suspended in 10 mL of 0.3 M HCL and warmed at 50 °C for 1 h while stirring continuously. The extraction was filtered and 5 mL of the filtrate was mixed with 1.0 mL of 25% ammonium hydroxide (for adjusting the pH of the solution), two drops of phenolphthalein indicator, 1.0 mL of acetic acid and 1.0 mL of 5% calcium chloride solutions, and then the mixture was allowed to stand for 3 h. It was then centrifuged at 3,000 rpm for 15 min. and the supernatant was discarded and the precipitate was washed properly with distilled water 3 times with centrifugation. The precipitate was dissolved by adding 2.0 mL of 5 M H_2SO_4 and was warmed in a water bath at 70 ºC. It was transferred into a 25 mL volumetric flask and topped up with distilled water and used for the titration given below.

Permanganate titration: About 5 mL of the above solution was titrated with freshly prepared 0.01 M potassium permanganate until the first pink color appeared. The solution was allowed to stand until the pink coloration disappeared and then the mixture was warmed in a water bath at 70 ºC for three minutes. Then, the mixture was re-titrated until the pink color appeared again and persisted for at least 30 s. Both the extraction and titration steps were triplicated and the average calcium oxalate percentage was calculated (Ezeonu and Ejikeme, 2016; Iwuozor, 2019).

Determination of cyanogenic glycosides

Powdered plant material (5 g) was added to 250 mL of distilled water and homogenized by adding tannic acid. The homogenate was transferred into a distillation flask and left for 3 h. Distillation was performed until 150 mL of the distillate was collected. About 20 mL of 0.02 M sodium hydroxide was added to the distillate and mixed well. The mixture was transferred to a 250 mL volumetric flask and was volume-up using distilled water. The resulting mixture was divided into three portions of 50 mL and each aliquot was added to a 4 mL of 6 M ammonium solution and 1 mL of 5% potassium iodide. The aliquots were titrated using 0.02 M silver nitrate. The amount of silver nitrate consumed to the endpoint of the titration was recorded. The equation below was used to determine HCN in the various parts in mg (Ezeonu and Ejikeme, 2016).

1 mL of 0.02 M silver nitrate =1.08 mg HCN

Determination of lipid

Lipids were extracted using the modified method of Folch *et al*. (1957). Briefly, 5 g of powdered cinnamon sample was added to the mixture of chloroform: methanol (2:1, v/v); followed by extraction with 20, 10 and 10 mL of above mixture, respectively. Each extraction was carried out for 10 min with the aid of

ultra-sonication. The solvent mixture containing extracted lipids was separated from the residual biomass by centrifugation and all the fractions were combined. The combined mixture was mixed with 10 mL of 1% KCl solution in a separating funnel while stirring vigorously; lower phase (chloroform) containing extracted lipid was separated, filtered using a Whatman® No.1 filter paper containing 1 g of anhydrous sodium sulfate. The residue was collected in glass vials and the solvent was removed in the fume hood, and subsequently the weight percentage was calculated (Floch *et al*., 1957).

Statistical analysis (mean, standard deviation and analysis of variance)

The percentage for MIC of each absorbance was calculated using Excel-2013 Microsoft Office package. The mean and standard deviation were calculated including only positive values using IBS SPSS 20.0 trial version of the statistical analysis software package for Windows. Tukey's post-hoc tests (Student's unpaired, t-test) were performed to evaluate the *p*-values, means and standard deviation. One-way analysis of variance (ANOVA) was carried out to compare the mean values of different extracts at the same concentration. This was done by performing Dunnet's Multiple Comparisons Test. Alkaloids, flavonoids, saponins, poly-phenols, oxalates, cyanogenic glycosides and lipid content were selected as the dependent variables. Type of the sample according to the climatic variations, maturity stages and physical stages of the plant were considered as the independent variables. Principal component analysis was performed to differentiate and classify the cinnamon samples.

RESULTS AND DISCUSSION

Variation of phytochemical profile according to the different climatic conditions in seed propagation types of *C. zeylanicum* Blume

The results of the quantitative analysis of phytochemicals of Ceylon cinnamon based on the climatic zone variations are shown in Table 2. Accordingly, phytochemical quantities have shown significant (P<0.05) differences based on climatic conditions and propagation type. The alkaloid quantity in the bark in seeding propagation type was in the range of 3.23 to 19.94 mg g^{-1} of dry weight (DW). When considering the agro-ecological zones or climatic variations, *C. zeylanicum* grown in WL 1a, WL 2a and IL 1b showed significantly (P<0.05) higher amounts of alkaloids than that of in other zones. Besides, flavonoids, poly-phenols and saponins were in the range of 0.93 to 3.27, 6.22 to 18.92 and 5.24 to 30.45 mg g^{-1}, respectively, and it showed the highest flavonoids, poly-phenols and saponin quantity in the bark extracts that grow in WL 1a and DL5 climatic zones. However, cyanogenic glycosides quantities were in the range of 40.05 to 122.49 mg 100 g^{-1} DW of the bark powder. Interestingly, the amount of oxalate in *C. zeylanicum* varied in a narrow range: 2.23 to 3.53 mg g^{-1} DW, and the climatic variations did not significantly (P<0.05) effect on the oxalate quantity.

Table 2: Quantitative analysis of phyto-constituents in leaves and bark of *C. zeylanicum* according to the climatic zones (CZ).

District	CZ	Alkaloids (mg g^{-1} DW)	Flavonoids (mg g^{-1} DW)	Polyphenol (mg g^{-1} DW)	Saponin (mg g^{-1} DW)	Oxalate (mg g^{-1} DW)	CG (mg 100 g^{-1} DW)	Lipid (mg g^{-1} DW)
Seed Propagation – Bark								
Galle	WL 1a	19.94 ± 4.75^{a}	3.27 ± 2.27^{a}	12.45 ± 0.35^{h}	10.25 ± 0.15^{c}	2.79 ± 0.13^{acdef}	70.04 ± 14.00^{d}	0.86 ± 0.09^{f}
	WL 2a	3.69 ± 0.28^{cdefg}	1.56 ± 0.11^{adefg}	25.59 ± 0.23^{d}	5.66 ± 0.31^{f}	2.86 ± 0.11^{acd}	40.05 ± 15.53^{f}	0.89 ± 0.07^{f}
Matara	IL 1a	3.23 ± 0.09^{cdefg}	0.93 ± 0.04^{dh}	14.68 ± 0.33^{f}	5.24 ± 0.18^{g}	2.32 ± 0.06^{fg}	136.44 ± 11.68^{ab}	1.19 ± 0.09^{bcde}
	IL 1b	4.07 ± 0.11^{cde}	0.96 ± 0.03^{dh}	18.92 ± 0.36^{e}	5.46 ± 0.41^{fg}	2.23 ± 0.15^{fg}	136.57 ± 10.50^{a}	1.27 ± 0.16^{bcde}
	WL 1a	4.13 ± 0.30^{cd}	1.67 ± 0.17^{adef}	13.64 ± 0.19^{g}	8.41 ± 0.04^{de}	2.77 ± 0.15^{acdef}	115.70 ± 11.34^{abc}	1.40 ± 0.05^{ab}
	WL 2a	3.33 ± 1.28^{cdefgh}	1.68 ± 0.06^{ade}	25.48 ± 0.30^{d}	8.83 ± 0.10^{d}	2.84 ± 0.13^{acde}	118.83 ± 9.53^{abc}	1.36 ± 0.08^{abced}
Hambantota	WL 2a	6.51 ± 0.24^{b}	2.35 ± 0.29^{adefg}	53.79 ± 0.10^{a}	8.32 ± 0.04^{e}	3.11 ± 0.26^{abc}	22.86 ± 0.16^{fg}	1.37 ± 0.05^{abc}
	IL 1b	5.66 ± 0.25^{bc}	2.32 ± 0.11^{abc}	27.73 ± 0.06^{c}	11.32 ± 0.57^{b}	3.30 ± 0.14^{ab}	35.52 ± 8.89^{fg}	0.65 ± 0.03^{g}
	DL5	3.78 ± 0.05^{cdef}	2.20 ± 0.03^{abcd}	52.67 ± 0.07^{b}	30.45 ± 0.19^{a}	3.56 ± 1.08^{a}	68.24 ± 0.80^{f}	1.52 ± 0.01^{a}
Seed propagation – Leaves								
Galle	WL 1a	3.79 ± 0.15^{bc}	4.39 ± 0.17^{ab}	4.71 ± 0.03^{ab}	11.03 ± 0.27^{a}	2.63 ± 0.11^{abcd}	53.32 ± 4.08^{bcdef}	5.76 ± 0.05^{b}
	WL 2a	3.62 ± 0.18^{bcd}	4.52 ± 0.30^{ab}	4.70 ± 0.28^{abc}	10.67 ± 0.17^{ab}	2.79 ± 0.07^{abc}	45.13 ± 0.32^{defg}	5.54 ± 0.05^{b}
Matara	IL 1a	1.95 ± 1.09^{efg}	1.66 ± 0.69^{ef}	1.50 ± 0.87^{gh}	6.07 ± 0.51^{f}	2.15 ± 0.95^{bcdefg}	94.68 ± 43.47^{abc}	1.99 ± 0.45^{ef}
	IL 1b	2.18 ± 0.15^{def}	3.65 ± 0.26^{bc}	3.36 ± 0.34^{bcd}	6.23 ± 0.16^{f}	3.10 ± 0.13^{a}	45.04 ± 0.38^{defg}	2.53 ± 0.25^{def}
	WL 1a	5.75 ± 0.02^{a}	5.46 ± 0.35^{a}	5.56 ± 0.26^{a}	9.48 ± 0.22^{abc}	1.75 ± 0.04^{bcdefg}	46.92 ± 11.31^{cdef}	7.50 ± 0.24^{a}
	WL 2a	1.32 ± 0.26^{g}	1.18 ± 0.24^{fg}	0.96 ± 0.32^{gh}	5.75 ± 0.53^{g}	1.33 ± 0.18^{dfg}	135.21 ± 14.39^{a}	1.92 ± 0.54^{f}
Hambantota	WL 2a	1.03 ± 0.05^{g}	1.28 ± 0.05^{efg}	2.90 ± 0.32^{gh}	6.38 ± 0.45^{f}	2.62 ± 0.10^{abcde}	83.30 ± 11.07^{abcd}	2.71 ± 0.22^{de}
	IL 1b	2.67 ± 0.85^{cde}	2.29 ± 1.02^{de}	2.63 ± 1.32^{def}	6.90 ± 1.27^{d}	2.85 ± 1.05^{ab}	64.54 ± 23.45^{bcde}	3.78 ± 0.50^{c}
	DL5	4.08 ± 0.06^{b}	3.22 ± 0.09^{bcd}	1.94 ± 0.21^{defg}	8.76 ± 0.07^{c}	2.28 ± 0.29^{abcdef}	109.10 ± 37.86^{ab}	3.33 ± 0.14^{cd}

Table 2 cont.

Vegetative propagation – Bark								
Palolpitiya (G)	IL 1a	7.94±0.08^{a}	1.63±0.27^{a}	5.41±0.13^{d}	9.55±0.60^{bc}	4.32±0.55^{ab}	317.55±70.15^{a}	1.31±0.39^{bc}
Palolpitiya (V)	IL 1a	3.75±0.28^{bcd}	1.45±0.47^{abc}	3.70±0.25^{e}	2.32±0.33^{e}	2.49±0.17^{de}	210.73±19.85^{b}	0.74±0.17^{cd}
Nilamba (G)	WU 2b	5.39±4.10^{ab}	0.96±0.41^{abcde}	4.10±0.13^{e}	7.68±0.75^{d}	4.07±0.41^{bc}	81.74±4.63^{c}	1.57±0.20^{b}
Nilamba (V)	WU 2b	2.88±0.67^{bcde}	1.28±0.38^{abcd}	11.56±0.57^{c}	44.34±1.37^{a}	4.96±0.29^{a}	56.38±9.25^{cde}	0.59±0.40^{d}
Narammala (G)	IL 1a	3.56±0.28^{bcde}	1.57±0.09^{ab}	15.52±0.39^{a}	8.22±0.25^{bcd}	3.53±0.16^{cd}	76.73±0.78^{cd}	2.24±0.13^{a}
Narammala (V)	IL 1a	4.84±0.13^{abc}	0.77±0.03^{de}	13.19±0.56^{b}	9.57±0.32^{b}	2.86±0.13^{de}	66.72±8.42^{cde}	1.15±0.06^{bcd}
Vegetative propagation – Leaves								
Palolpitiya (G)	IL 1a	3.68±1.11^{bc}	3.67±0.89^{bcd}	3.91±0.16^{a}	24.75±0.92^{b}	3.42±0.77^{bcd}	195.12±112.50^{abc}	2.41±0.51^{d}
Palolpitiya (V)	IL 1a	2.18±1.04^{ce}	1.94±0.27^{e}	2.61±0.45^{b}	6.58±0.88^{cde}	2.40±0.05^{e}	86.87±19.88^{cd}	2.11±0.08^{d}
Nilamba (G)	WU 2b	4.73±0.71^{b}	3.14±0.29^{cd}	3.94±0.36^{a}	6.71±0.61^{cd}	3.85±0.05^{bc}	228.11±20.44^{ab}	5.88±0.25^{b}
District	CZ	Alkaloids (mg g^{-1} DW)	Flavonoids (mg g^{-1} DW)	Polyphenol (mg g^{-1} DW)	Saponin (mg g^{-1} DW)	Oxalate (mg g^{-1} DW)	CG (mg 100 g^{-1} DW)	Lipid (mg g^{-1} DW)
Nilamba (V)	WU 2b	5.66±0.51^{a}	3.87±0.49^{bc}	2.58±0.01^{b}	7.53±0.10^{c}	5.47±0.13^{a}	246.43±9.94^{a}	3.87±0.45^{c}
Narammala (G)	IL 1a	3.65±0.10^{bcd}	5.29±0.16^{a}	1.67±0.13^{c}	7.35±0.11^{cd}	4.15±0.23^{b}	22.65±0.32^{e}	7.69±0.03^{a}
Narammala (V)	IL 1a	2.45±0.18^{cde}	4.39±0.20^{ab}	1.89±0.08^{c}	49.06±2.14^{a}	3.04±0.38^{cde}	45.76±0.18^{de}	3.92±0.08^{c}

CZ - climatic zone, CG - cyanogenic glycoside, DW - dry weight, G - Sri Gamunu, V - Sri Wijaya, Results are expressed as mean±standard deviation (n = 3).

Different letters (***a-h***) for each column in separated section donates significant (P>0.05) different between the tested phytochemical.

The leaves of Ceylon cinnamon were shown to produce a high yield of phytochemicals. The quantities of alkaloids, flavonoids, poly-phenols and saponins were in the range of 1.03 4.08 mg g^{-1} DW, 1.18-4.52 mg g^{-1} DW, 0.55-4.71 mg g^{-1} DW and 5.75-11.03 mg g^{-1} DW, respectively. Comparatively, WL 1a and DL5 climatic zones presented higher yields of alkaloids, flavonoids, saponins and poly-phenolics, than the other climatic zones. A lower range of oxalate was present in leaves than in bark, which was ranging from 1.27 to 3.10 mg g^{-1} DW; the lowest amount in IL 1b zone. In contrast, yield of cyanogenic glycosides and lipids in leaves were significantly (P<0.05) higher than that was in the bark being in the range from 45.04-135.21 mg 100 g^{-1} DW and 1.46-5.76 mg g^{-1} DW, respectively.

Variation of phytochemical profile according to the different climatic conditions in vegetative propagation types of *C. zeylanicum* Blume

Results for the quantitative phytochemical analysis of leaves and bark from two vegetative propagated varieties (Sri Wijaya and Sri Gamunu) according to the climatic zone variations is illustrated in the Table 2. The results showed a higher yield percentage of phytochemicals in both bark and leaves of Sri Gamunu than Sri Wijaya. The highest yield of alkaloids, flavonoids and cyanogenic glycosides were found in bark of Palolpitiya (IL 1a) Sri Gamunu variety, which had 7.94, 1.63 and 338.24 mg 100 g^{-1} DW, respectively. Out of studied samples, the highest yield of polyphenols and saponins were noted in Nilamba (WU 2b) Sri Wijaya. Comparatively, the leaves and bark of Sri Gamunu yielded higher lipid percentages than Sri Wijaya. Out of six leaves samples of vegetative propagated varieties, the highest alkaloids, flavonoids, polyphenols, saponins and cyanogenic glycosides were exhibited in Nilamba (V) WU 2b, Narammala (G) IL 1a and Palolpitiya (G) IL 1a climatic zones. However, the lowest oxalate yield was presented in Sri Wijaya; leaves as well as bark.

Variation of phytochemical profile according to the different Maturity stages in seed and vegetative propagation types of *C. zeylanicum* Blume

Variation of phytochemical content of leaves and bark based on the maturity stage (1-3, 3-5, and > 5 y) is shown in Table 3. Among the seed propagation type of normal Ceylon cinnamon varieties, the highest alkaloid content was observed in > 5 y matured bark and 1-3 y matured leaves whereas vegetative propagated varieties showed significantly high alkaloid content in bark which range from 2.38 to 8.42 mg g^{-1} DW in three maturity stages considered. Among the vegetative propagated varieties, GM3 and VM3 exhibited significantly higher alkaloid content at all maturity stages. In the leaves, VM3 demonstrated the highest alkaloid. A similar situation was observed for flavonoids and the highest content was observed in NM1 bark and NM2 leaves ranging from 1.0 to 2.87 mg g^{-1} DW.

Table 3: Quantitative analysis of phyto-constituents in leaves and bark of *C. zeylanicum* based on the maturity stage of the plant.

Sample	Maturity stage	Alkaloids (g 100 g^{-1} DW)	Flavonoids (g 100 g^{-1} DW)	Polyphenol (g 100 g^{-1} DW)	Saponin (g 100 g^{-1} DW)	Oxalate (g 100 g^{-1} DW)	CG (mg 100 g^{-1} DW)	Lipid (g 100 g^{-1} DW)
Seed propagation – Bark								
N	1-3 y	3.35 ± 0.05^{b}	1.24 ± 0.06^{b}	2.94 ± 0.18^{c}	6.43 ± 0.12^{a}	3.11 ± 0.10^{c}	143.44 ± 0.48^{b}	1.17 ± 0.53^{ab}
	3-5 y	2.47 ± 0.05^{c}	1.28 ± 0.15^{b}	3.92 ± 0.13^{b}	4.59 ± 0.13^{b}	3.02 ± 0.13^{a}	215.60 ± 5.40^{a}	0.99 ± 0.32^{ab}
	>5 y	3.92 ± 0.09^{a}	1.44 ± 0.08^{a}	6.97 ± 0.10^{a}	2.65 ± 0.11^{c}	2.49 ± 0.12^{b}	218.08 ± 7.92^{6}	1.15 ± 0.28^{a}
Seed propagation – Leaves								
N	1-3 y	1.54 ± 0.02^{c}	1.03 ± 0.29^{c}	6.87 ± 0.11^{b}	5.36 ± 0.27^{c}	1.44 ± 0.06^{b}	129.79 ± 0.61^{a}	2.09 ± 0.66^{ab}
	3-5 y	4.18 ± 0.03^{a}	2.83 ± 0.12^{a}	9.68 ± 0.06^{a}	8.19 ± 0.19^{b}	2.60 ± 0.13^{a}	98.56 ± 0.86^{b}	2.12 ± 0.26^{ab}
	>5 y	2.08 ± 0.06^{b}	1.94 ± 0.10^{b}	6.16 ± 0.08^{c}	9.57 ± 0.05^{a}	2.42 ± 0.08^{a}	83.07 ± 1.60^{c}	2.78 ± 0.39^{a}
Vegetative propagation – Bark								
GM3	1-3 y	8.42 ± 0.27^{a}	9.02 ± 0.43^{a}	5.51 ± 0.45^{a}	9.02 ± 0.42^{bc}	1.91 ± 0.18^{bcde}	279.26 ± 69.50^{ab}	1.20 ± 0.29^{ab}
GM2	3-5 y	7.33 ± 0.30^{c}	8.76 ± 0.39^{bc}	4.72 ± 0.19^{ab}	8.76 ± 0.39^{bc}	2.89 ± 0.95^{abc}	359.96 ± 89.24^{abc}	1.05 ± 0.28^{abc}
GM1	>5 y	7.94 ± 0.08^{ab}	9.55 ± 0.60^{cde}	5.41 ± 0.12^{ab}	9.55 ± 0.60^{b}	4.32 ± 0.55^{a}	338.24 ± 70.15^{a}	1.31 ± 0.39^{a}
VM3	1-3 y	8.26 ± 0.24^{ab}	1.78 ± 0.18^{bcd}	1.63 ± 0.32^{d}	13.84 ± 0.81^{a}	2.64 ± 0.44^{bcd}	156.77 ± 25.95^{bde}	0.71 ± 0.14^{abcde}
VM2	3-5 y	2.38 ± 0.13^{e}	2.30 ± 0.32^{b}	3.75 ± 0.53^{c}	1.39 ± 0.33^{d}	3.00 ± 0.29^{ab}	242.90 ± 84.40^{abcd}	0.60 ± 0.04^{cde}
VM1	>5 y	3.75 ± 0.28^{d}	1.44 ± 0.47^{cde}	3.70 ± 0.25^{c}	2.32 ± 0.33^{d}	2.49 ± 0.17^{bcde}	210.73 ± 19.85^{abcde}	0.74 ± 0.17^{abcd}
Vegetative propagation – Leaves								
GM3	1-3 y	1.99 ± 0.32^{abcde}	3.73 ± 1.21^{abc}	2.48 ± 0.09^{b}	4.42 ± 0.39^{e}	2.48 ± 0.18^{bc}	141.57 ± 3.78^{abc}	3.05 ± 0.59^{a}
GM2	3-5 y	1.27 ± 0.33^{abcde}	3.59 ± 0.24^{ab}	1.44 ± 0.30^{c}	8.05 ± 0.95^{cd}	2.47 ± 0.17^{bcd}	187.64 ± 42.93^{ab}	2.10 ± 0.13^{abcde}
GM1	>5 y	3.86 ± 1.28^{abc}	3.67 ± 1.07^{a}	3.90 ± 0.16^{a}	24.75 ± 0.92^{a}	3.42 ± 0.94^{a}	254.69 ± 44.69^{a}	2.33 ± 0.59^{abc}
VM3	1-3 y	6.22 ± 1.10^{ab}	1.40 ± 0.21^{d}	0.41 ± 0.26^{d}	10.08 ± 0.54^{b}	1.49 ± 0.07^{d}	102.55 ± 4.10^{abcd}	2.02 ± 1.07^{bcde}
VM2	3-5 y	4.43 ± 0.26^{a}	3.55 ± 0.22^{abc}	0.97 ± 0.05^{c}	9.29 ± 0.20^{bc}	2.64 ± 0.04^{ab}	98.85 ± 2.11^{abcde}	2.87 ± 0.19^{ab}
VM1	>5 y	2.28 ± 1.25^{bcd}	1.94 ± 0.27^{d}	2.61 ± 0.44^{b}	6.58 ± 0.88^{d}	2.40 ± 0.05^{bcd}	82.80 ± 2.22^{abcde}	2.11 ± 0.08^{abcd}

Comparatively, poly-phenolic content varied as NM2 leaves > NM1 bark > GM3 bark > GM1 leaves, which was in the range from 0.41 to 9.67 mg g^{-1} DW in all maturity stages (Table 3). When comparing the saponin content in different maturity stages, the highest value was given by GM1 leaves, while the lowest content was in NM1 bark. Out of tested samples, the lowest oxalate content was observed in NM3, and VM3 leaves whereas significantly high oxalate content was in all the bark samples in selected maturity stages. Cyanogenic glycoside content was significantly varied in the tested samples, ranging from 82.80 to 359.96 mg 100g $^{-1}$ DW in all maturity stages.

Variation of phytochemical profile according to the different physical stages in seed and vegetative propagation types of *C. zeylanicum* Blume

The values for phytochemical content based on the physical stage of the plant (FLW, FRT and FLSH) in leaves and bark belong to the seeding and vegetative propagated varieties are illustrated in Table 4. Among the bark samples, the highest alkaloid content was found in seeding propagated FLSH season and the moderate alkaloid content was found in vegetative propagated FLSH season. When considering leaves, significantly (P<0.05) higher alkaloid content was found in the FRT season of both seeding and vegetative propagated types. In the seeding propagated type, blue fruits containing varieties exhibited high alkaloid content while green fruits containing vegetative propagated types showed a significantly (P<0.05) high alkaloid content.

Among all the bark samples, flavonoid content was significantly (P<0.05) higher during the FLSH season. In contrast, leaves in the FRT season exhibited significantly (P<0.05) higher flavonoid content in both propagated types. When considering the poly-phenolic content in bark, the yield was in order "FLSH > FRT-g > FRT- b > FLW", "FRT-g > FRT- b > FLSH > FLW" and "FRT-g > FLSH > FRT-b >FLW" for Normal, Sri Gamunu and Sri Wijaya varieties, respectively. Among all varieties, the highest poly-phenolic content was exhibited in normal FLSH season; and there was no significant (P>0.05) difference in polyphenolic content of both seed and vegetative propagated plants in FRT season. When compared to bark, leaves samples exhibited moderate poly-phenolic content and among tested propagated types, high poly-phenolic content was observed in FRT season plants. When considering the saponin content in bark, all the varieties exhibited high saponin content (Normal > Sri Gamunu > Sri Wijaya) but the leaves showed the least amount. The lowest oxalate content was exhibited in the bark of normal varieties belonging to the FLW season. Interestingly, there was no significant (P>0.05) different in oxalate content of two vegetative propagated varieties. As regard to cyanogenic glycoside content in bark, FLW season of vegetative propagated varieties contained a significantly (P<0.05) high percentage of cyanogenic glycoside.

Table 4: Quantitative analysis of phyto-constituents in leaves and bark of *C. zeylanicum* based on the physical stage of the plant.

Sample	Physical stage	Alkaloids (g 100 g^{-1} DW)	Flavonoids (g 100 g^{-1} DW)	Polyphenol (g 100 g^{-1} DW)	Saponin (g 100 g^{-1} DW)	Oxalate (g 100 g^{-1} DW)	CG (mg 100 g^{-1} DW)	Lipid (g 100 g^{-1} DW)
Seed propagation – Bark								
N	FLW	8.40± 0.06b	4.48± 0.14a	6.43± 0.25c	8.50± 0.57c	2.27± 0.15a	164.01± 4.41c	2.58± 0.16ab
	FRT-b	8.88± 0.05a	3.78± 0.16b	8.84± 0.18b	8.93± 0.19bc	3.11± 0.18ab	200.01±15.37b	2.50± 0.05ab
	FRT-g	8.46± 0.08b	3.39± 0.15a	9.07± 0.17b	9.57± 0.20b	3.28± 0.18ab	205.57± 15.61a	2.64± 0.04a
	FLSH	8.89± 0.06c	4.86± 0.03c	11.46± 0.35a	10.51± 0.28a	3.36± 0.26c	23.02± 0.17a	2.82± 0.03ab
Vegetative propagation – Bark								
G	FLW	7.29 ± 0.06^{f}	2.51 ± 0.06^{f}	5.57 ± 0.38^{f}	8.56 ± 0.09^{ef}	2.63 ± 0.12^{a}	250.68 ± 35.03^{a}	1.30 ± 0.05^{g}
	FRT- b	6.87 ± 0.07^{g}	2.88 ± 0.08^{e}	7.14 ± 0.18^{e}	8.34 ± 0.10^{efg}	2.56 ± 0.06^{ac}	135.40 ± 6.61^{e}	1.76 ± 0.03^{cdef}
	FRT-g	7.66 ± 0.03^{e}	3.39 ± 0.16^{bcd}	7.67 ± 0.16^{cd}	9.60 ± 0.06^{b}	2.85 ± 0.12^{ab}	150.55 ± 15.64^{cde}	1.88 ± 0.01^{bc}
	FLSH	7.81 ± 0.02^{g}	3.55 ± 0.05^{ab}	$8.08\pm 0.35a^{bc}$	10.29 ± 0.34^{a}	2.92 ± 0.07^{ab}	161.49 ± 2.18^{bcd}	2.16 ± 0.22^{b}
V	FLW	8.55 ± 0.10^{a}	3.47 ± 0.21^{abc}	5.84 ± 0.11^{f}	8.38 ± 0.05^{efg}	2.52 ± 0.11^{acd}	174.15 ± 3.15^{b}	1.70 ± 0.08^{cdef}
	FRT-b	7.81 ± 0.16^{c}	3.22 ± 0.04^{cd}	7.94 ± 0.05^{bcd}	8.68 ± 0.05^{de}	2.32 ± 0.06^{adef}	150.77 ± 6.56^{cd}	1.78 ± 0.07^{cde}
	FRT-g	8.40 ± 0.15^{ab}	3.71 ± 0.15^{a}	8.50 ± 0.05^{a}	8.91 ± 0.06^{d}	2.30 ± 0.05^{adef}	165.50 ± 3.49^{bc}	1.87 ± 0.06^{cd}
	FLSH	8.39 ± 0.01^{ab}	3.69 ± 0.08^{ab}	8.31 ± 0.08^{ab}	9.27 ± 0.07^{c}	2.52 ± 0.07^{acde}	150.61 ± 1.90^{cd}	2.41 ± 0.22^{a}
N	FLW	2.11 ± 0.14^{c}	3.31 ± 0.16^{ab}	9.40 ± 0.13^{a}	9.44 ± 0.2^{c}	1.94 ± 0.04^{b}	67.31 ± 1.97^{b}	4.57 ± 0.05^{b}
	FRT-b	3.58 ± 0.30^{a}	3.43 ± 0.21^{a}	4.51 ± 0.30^{c}	11.17 ± 0.22^{b}	2.52 ± 0.18^{a}	90.98 ± 1.01^{a}	5.86 ± 0.03^{a}
	FRT-g	2.60 ± 0.08^{b}	2.56 ± 0.31^{b}	6.57 ± 0.25^{b}	13.57 ± 0.34^{a}	2.68 ± 0.11^{a}	45.15 ± 0.38^{d}	3.37 ± 0.09^{c}
	FLSH	2.23 ± 0.14^{b}	1.62 ± 0.01^{c}	0.83 ± 0.02^{d}	9.85 ± 0.14^{c}	1.62 ± 0.08^{c}	60.91 ± 0.22^{c}	4.47 ± 0.07^{b}
Vegetative propagation – Leaves								
G	FLW	0.83 ± 0.17^{g}	1.31 ± 0.09^{defg}	2.53 ± 0.15^{e}	13.24 ± 0.09^{a}	2.65 ± 0.12^{ab}	48.00 ± 8.60^{f}	1.82 ± 0.13^{g}
	FRT-b	1.38 ± 0.02^{c}	3.45 ± 0.19^{a}	4.55 ± 0.26^{d}	11.16 ± 0.22^{b}	2.62 ± 0.19^{abc}	91.35 ± 0.50^{a}	5.83 ± 0.04^{a}
	FRT-g	4.37 ± 0.01^{b}	1.51 ± 0.20^{de}	2.70 ± 0.26^{e}	6.32 ± 0.22^{g}	2.71 ± 0.22^{a}	22.69 ± 0.21^{g}	1.84 ± 0.02^{f}
	FLSH	1.21 ± 0.07^{e}	2.74 ± 0.30^{b}	0.42 ± 0.03^{f}	10.37 ± 0.13^{d}	2.32 ± 0.17^{c}	68.37 ± 0.50^{b}	3.80 ± 0.09^{b}
V	FLW	0.99 ± 0.04^{f}	1.66 ± 0.26^{d}	5.31 ± 0.07^{c}	7.84 ± 0.10^{f}	1.32 ± 0.06^{de}	66.71 ± 4.12^{bc}	3.61 ± 0.03^{de}
	FRT-b	4.31 ± 0.08^{b}	1.42 ± 0.06^{def}	6.33 ± 0.06^{b}	10.92 ± 0.03^{bc}	1.56 ± 0.03^{d}	65.74 ± 2.96^{bcd}	3.77 ± 0.05^{bc}
	FRT-g	5.04 ± 0.28^{a}	2.50 ± 0.16^{c}	7.31 ± 0.21^{a}	10.75 ± 0.09^{c}	1.28 ± 0.04^{def}	58.59 ± 5.43^{de}	3.74± 0.15bcd
	FLSH	2.44 ± 0.12^{f}	1.31 ± 0.03^{defg}	0.04 ± 0.00^{f}	8.42 ± 0.23^{e}	1.20 ± 0.09^{ef}	61.66 ± 0.29^{bcde}	3.60± 0.25de

N - *C. zeylanicum* seed propagated varieties, G - Sri Gamunu, V - Sri Wijaya, FLW - flowering season, FRT - fruiting season, FLSH - flushing season, b - blue color fruits, g - green color fruits. Results are expressed as Mean±Standard deviation (n = 3). Different letters (***a-h***) for each column in separated section donates significant different (P>0.05) between the tested phytochemicals

In lipid content, among the tested bark samples, FLSH season of vegetative and seeding propagated varieties had the highest lipid content ordering as normal > Sri Wijaya > Sri Gamunu. Besides, the highest lipid content was exhibited in leaves of blue color fruits containing cinnamon plants, which were in FRT season.

As a summary of the phytochemical analysis based on different conditions, the following could be summarized after statistics. The climatic zones, WL 1a (Galle district) and IL 1a (Matara district) were the best climatic zones for obtaining high yield phytochemical content in both seeding and vegetative propagation types of Ceylon cinnamon, respectively. Out of these, the best phytochemical composition could be found in the leaves of more than five years matured stage and bark of 3-5 y matured stage of seed propagation varieties. When considering vegetative propagation types, bark from Sri Wijaya and leaves from Sri Gamunu with more than five years matured stage contained better phytochemical composition. According to the seasonal variations, the best phytochemical composition was found in the bark samples which were collected in the flowering season, and the leaves samples which were collected in the fruiting season. Therefore, leaves in >5 y matured stage and bark in 3-5 y matured stage of seed propagation varieties of Ceylon cinnamon which are grown in WL 1a (Galle district, Pitigala) climatic zone are the best collection of Ceylon cinnamon having appreciable contents of important bioactive compounds, and hence to be utilized for further pharmacological studies and value addition.

Comparison of phytochemical classes in the leaves and bark of *C. zeylanicum*

Alkaloids

Comparison of the alkaloid content of the leaves and bark of *C. zeylanicum* is shown in Figure 2.

Accordingly, the highest alkaloid content was exhibited in bark and leaves which were collected from WL 1a climatic zone. When considering the maturity of the plant, bark of Sri Gamunu showed higher alkaloid quantities for all the tested maturity stages over other varieties. However, except NM2 and VM2, all the other maturity stages of normal, Sri Gamunu and Sri Wijaya varieties contained higher alkaloid percentages in the bark than leaves. Among the physical stages of the plant, the bark of flowering, fruiting, and flushing seasons exhibited, higher alkaloid content than the leaves.

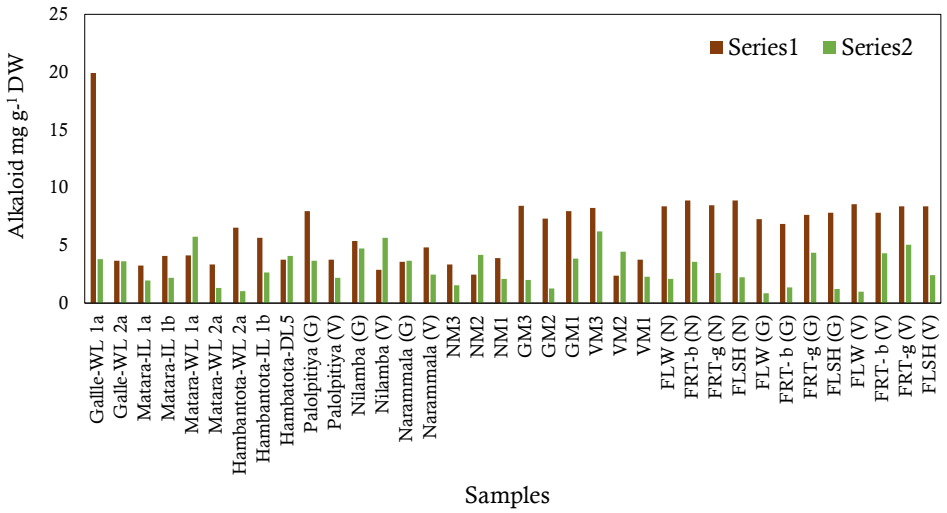

Figure 2: Comparison of alkaloid contents of leaves and bark of *C. zeylanicum* (Series 1: bark; Series 2: leaves).

Flavonoids

Comparison of flavonoid contents of the leaves and bark of *C. zeylanicum* is shown in Figure 3.

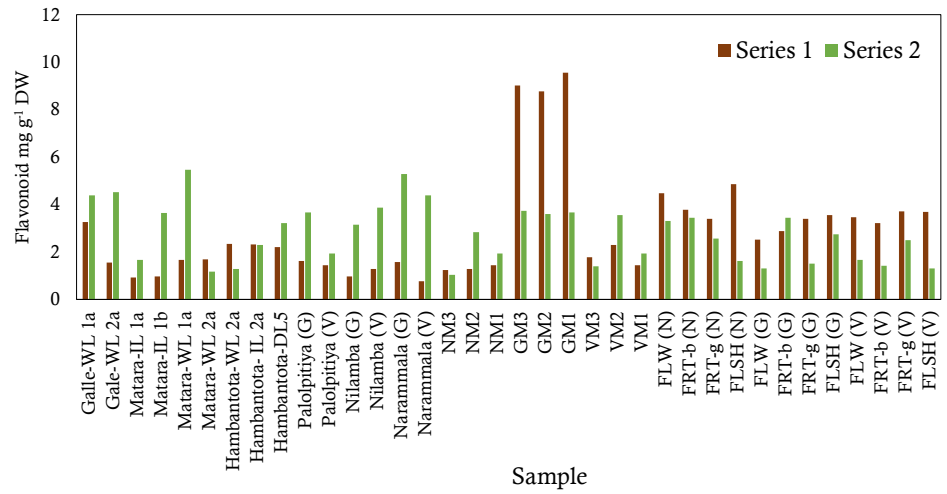

Figure 3: Comparison of flavonoid contents of leaves and bark of *C. zeylanicum* (Series 1: bark; Series 2: leaves).

Flavonoid content in leaves was greater than that of the bark which was collected from WL 1a climatic zone. However, the order differed in the case of WL 2a that had higher flavonoid content in bark than leaves. When considering the

vegetative propagated varieties, the flavonoid content of leaves was greater than bark, and it did not depend on the climatic zone. Bark of all the maturity stages in Sri Gamunu showed higher flavonoid content than leaves. Interestingly, flavonoid content of 1-3 y matured plants of seed propagation varieties and Sri Wijaya vegetative propagation variety exhibited greater flavonoid content than leaves. However, flavonoid content of leaves which were in 3-5 and >5 y showed higher values than that in the bark. In that sense, variation of flavonoid contents based on maturity stages of the plant in normal varieties in seed propagation and Sri Wijaya variety showed equality. When considering the physical stages of the plants, except FRT-b in Sri Gamunu, all other bark samples showed greater flavonoid content than leaves.

Polyphenols

Comparison of poly-phenolic contents of the leaves and bark of *C. zeylanicum* is shown in Figure 4.

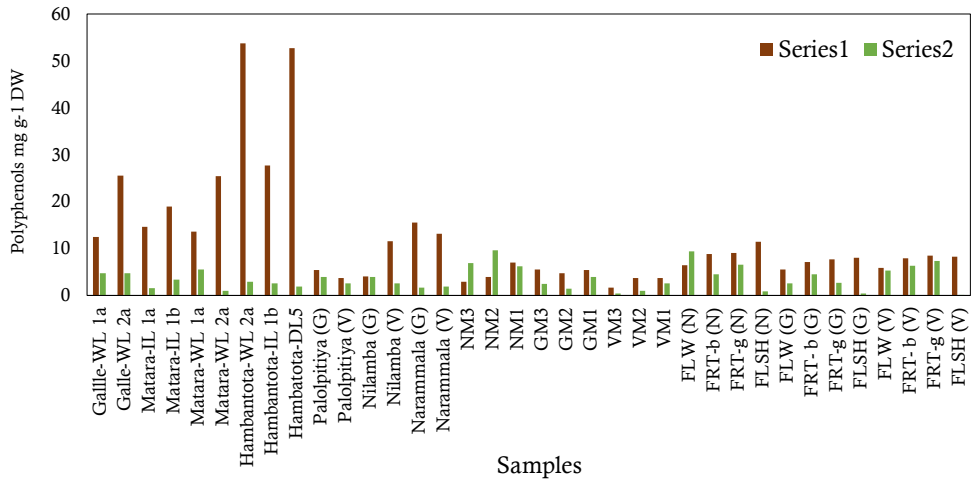

Figure 4: Comparison of polyphenolic contents of leaves and bark of *C. zeylanicum* (Series 1: bark; Series 2: leaves).

When considering climatic variations, the pol-phenolic content in bark was greater than that in the leaves. Polyphenolic content in 1-3 and 3-5 y seed propagation varieties exhibited higher polyphenolic content in leaves than the bark. When considering the physical stages of the plants, except flowering season of seed propagation variety, all the other samples exhibited greater polyphenolic content in bark than in leaves.

Saponins

Comparison of the saponin contents of the leaves and bark of *C. zeylanicum* is shown in Figure 5.

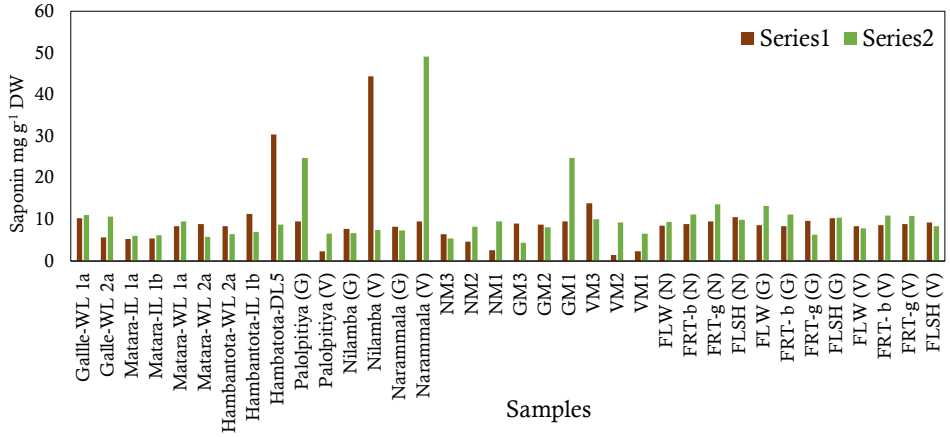

Figure 5: Comparison of saponin contents of leaves and bark of *C. zeylanicum* (Series 1: bark; Series 2: leaves).

Variation of saponin content in leaves of WL 1a climatic zone was greater than that of the bark. When considering the WL 2a climatic zone, saponin contents in bark were higher than in leaves, except Galle – WL 2a zone. In seed and vegetative propagation varieties, bark of 1-3 year matured plants exhibited higher saponin content than leaves. However, when increasing the age of the plant, saponin content in leaves was increased and in bark it was decreased, gradually. Except FRT-b (G), FLSH (N), FLSH (G), and FLSH (V) leaves of all the physical stages in seed and vegetative propagation varieties exhibited higher saponin content than the bark.

Oxalate

Comparison of the oxalate content of the leaves and bark of *C. zeylanicum* is shown in Figure 6.

Accordingly, almost all bark samples in seed propagation varieties showed greater oxalate content than leaves. When considering the vegetative propagation varieties, oxalate content in leaves of Narammala (G) and Narammala (V), which were in IL 1a climatic zone, exhibited greater oxalate content than the bark. However, it was contrast in the case of Palolpitiya (G) and Palolpitiya (V) which were in IL 1a, climatic zone. The reason could be the variation in soil conditions and the manner of fertilizer application. When considering about the maturity stages of the plants, except GM3, bark of the other samples exhibited greater oxalate content than the leaves. Interestingly, in leaves of all the physical stages showed higher oxalate content than the bark.

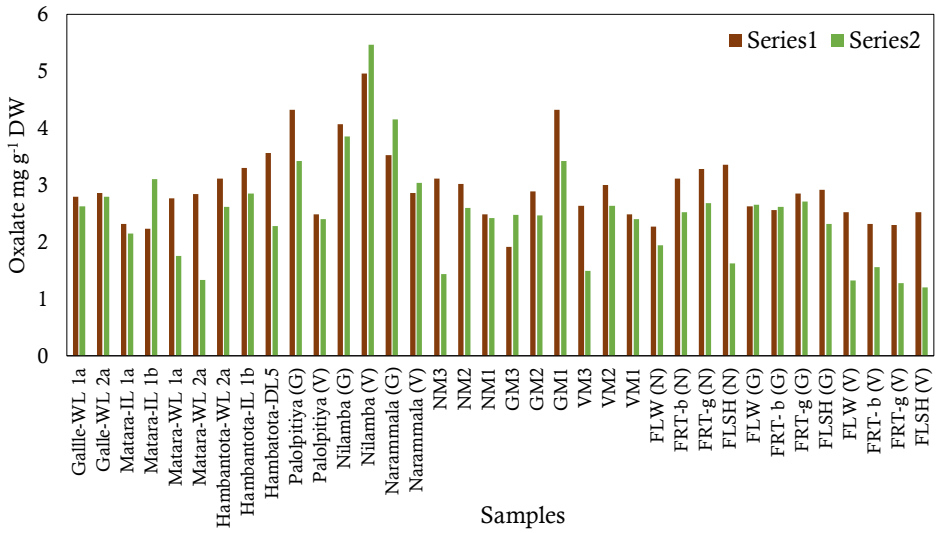

Figure 6: Comparison of oxalate contents of leaves and bark of *C. zeylanicum* (Series 1: bark; Series 2: leaves).

Cyanogenic glycoside

Comparison of the cyanogenic glycoside (CG) contents of the leaves and bark of *C. zeylanicum* is shown in Figure 7.

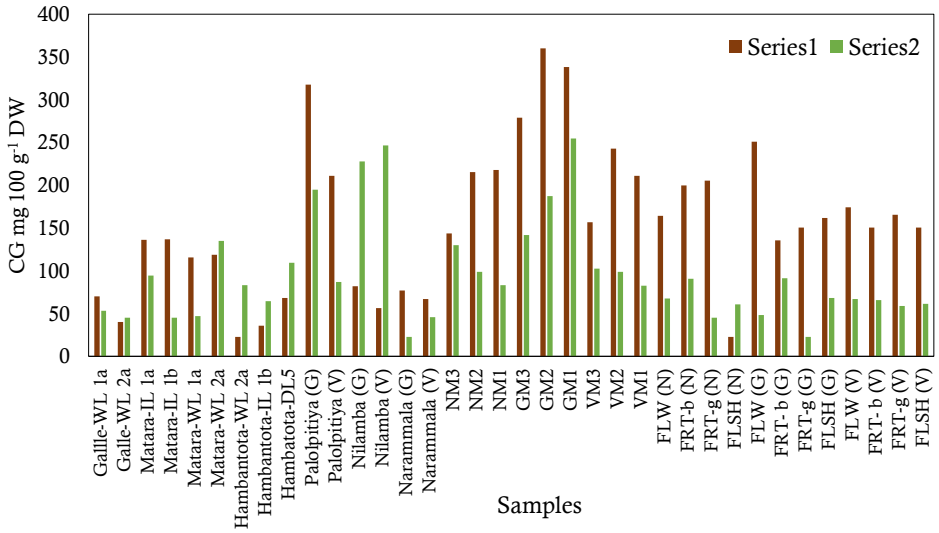

Figure 7: Comparison of cyanogenic glycoside contents of leaves and bark of *C. zeylanicum* (Series 1: bark; Series 2: leaves).

When considering the variation of CG content depending on the climatic variations, bark of WL 1a climatic zone showed greater CG content than the

leaves. However, CG content in leaves of the WL2a climatic zone was greater than that of the bark. Also, CG content in leaves in vegetative propagated varieties was higher than that in the bark. When considering the variation of CG based on the maturity stages, bark exhibited higher CG content than the leaves. Same as physical stages of the plants, leaves showed greater CG content than the bark.

Lipid

Comparison of the lipid contents of the leaves and bark of *C. zeylanicum* is shown in Figure 8.

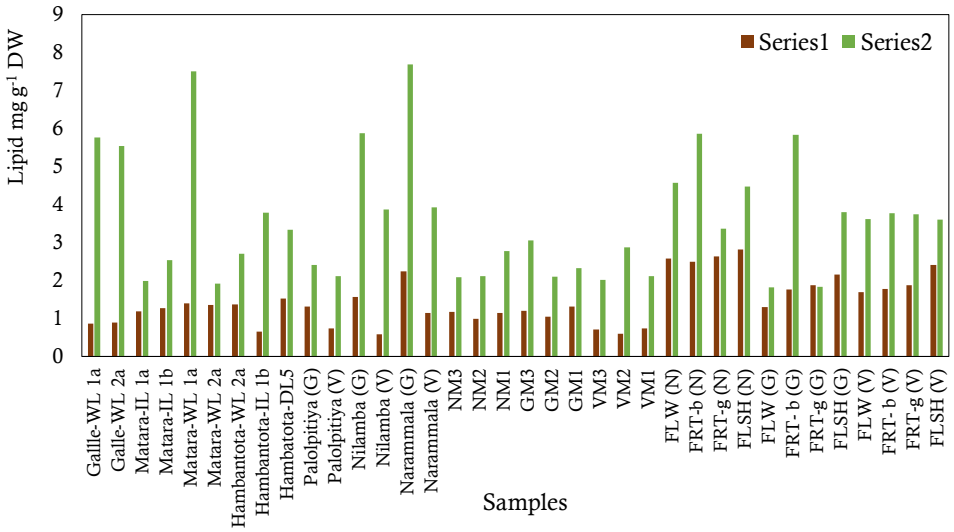

Figure 8: Comparison of lipid contents of leaves and bark of *C. zeylanicum* (Series 1: bark; Series 2: leaves).

Accordingly, lipid content in leaves was greater than that of the bark for all the samples and no variations between climatic zone, maturity stage and physical stage of the plant were noticed.

Principal Component Analysis

Principal component analysis (PCA) is considered as a "dimensionality reduction technique" used in statistical analysis of reduce the dimensions of dependent variables in large data sets to smaller orthogonal data sets of principal components. Two main components with eigenvalues >1:0 were extracted from the original data set using Kaiser's rule, and Kaiser-Mayer-Olkin measures the sampling adequacy equals to 0.505 for bark and 0.528 for leaves. Eigenvalues, percentage cumulative variance and loading values for the first two principal components for leaves and bark is shown in the Table 5.

Accordingly, the percentage cumulative variance for PC2 for leaves and bark, were almost 50 and 56%, respectively. When considering the principal

component analysis of leaves, PC1 strongly correlated with depending variables such as lipid, oxalate, and poly-phenols. Therefore, according to the Steven's suggestions, these 3 original dependent variables positively loaded significantly on the PC1. The PC1 of bark strongly correlated with depending variables in descending order such as saponin, poly-phenols, cyanogenic glycoside and oxalate that had high positive loaded values on the PC1.

Table 5: Eigenvalues, percentage cumulative variance and loading values for the two principal components for leaves and bark.

Dependent variable (Phyto-constituent)	Leaves		Bark	
	PC1	PC2	PC1	PC2
Eigenvalue	2.209	1.233	2.250	1.685
% Cumulative	31.551	49.163	32.137	56.202
Alkaloids	0.655	0.143	0.615	0.382
Flavonoids	0.862	0.214	0.764	0.526
Poly-phenol	0.369	-0.254	-0.469	0.584
Saponin	0.326	0.515	-0.468	0.593
Oxalate	0.421	-0.343	-0.479	0.204
Cyanogenic glycoside	0.174	0.824	0.477	-0.552
Lipid	0.783	-0.200	0.610	0.472

The score pots for leaves and bark resulting from principal component analysis are illustrated in Figure 9 and Figure 10, respectively.

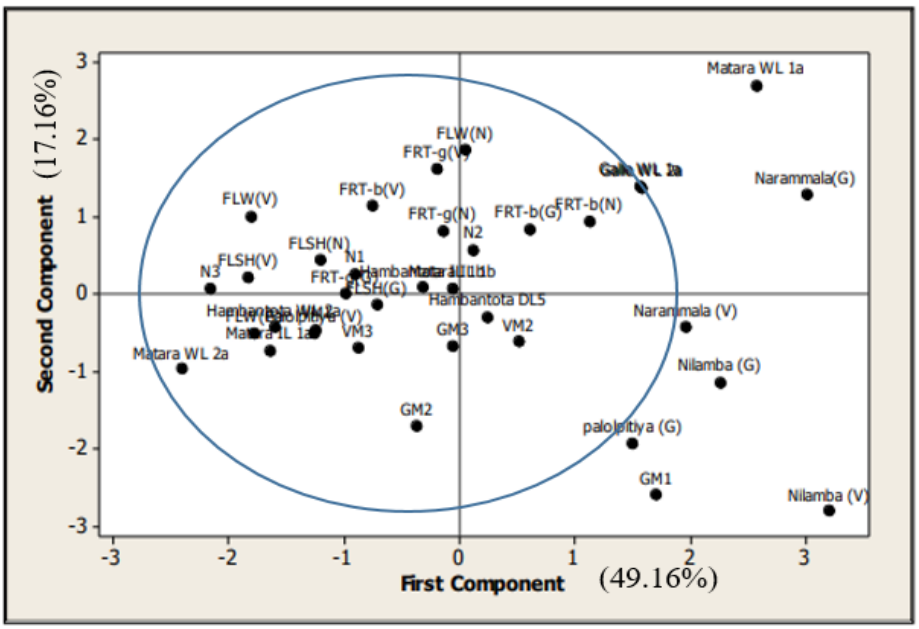

Figure 9: Score plot for leaf samples.

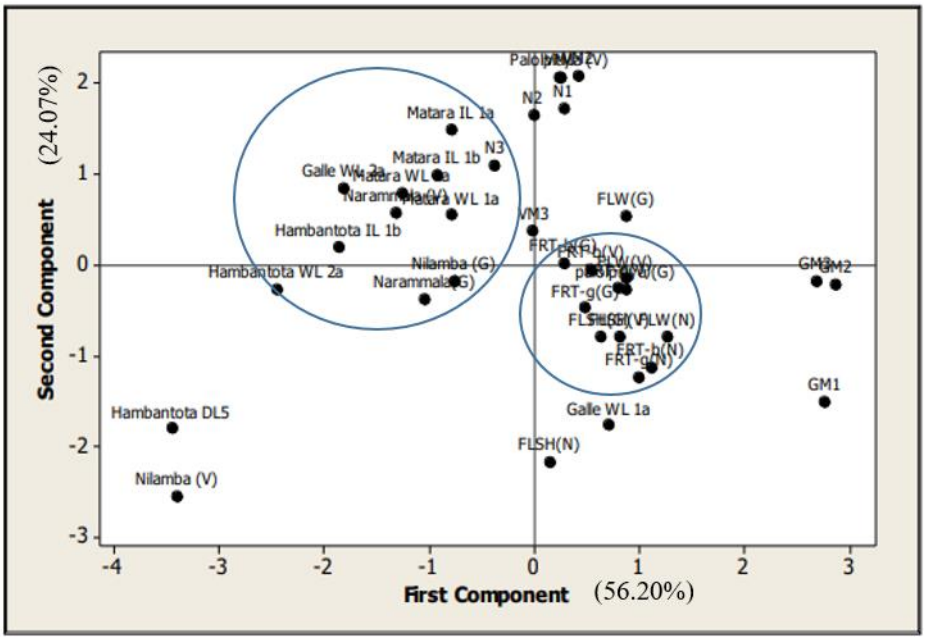

Figure 10: Score plot for bark samples.

According to the score plot of the leaves, the entire samples spread throughout the graph and most of the spots which belong to the physical stage of the plant have been placed on the positive side of the PC2 and negative side of the PC1. Samples belonging to the maturity stages of seed propagation (N1, N2, and N3) have been placed on the positive side of PC1, and vegetative propagation (G1, G2, G3, V1, V2, V3) have been placed on the negative side of PC1. Based on these factors, WL 1a, N2 and FRT-b (N) samples exhibited the greatest PC1 values; hence, that was the best leaves sample of cinnamon that can be obtained from 3-5 yrs. matured, seed propagation varieties which grown in WL 1a climatic zone in the fruiting season.

According to the score plot of the bark samples, almost all the samples have been divided into the main two clusters. Most of the samples except Galle WL 1a collected from different agro-ecological zones have been placed on the positive side of the PC1. All the samples belonging to the physical stages (flowering, fruiting and flushing seasons) have been placed on the negative side of the PC1. However, most of the samples were placed on the zero of PC1 and PC2 Figure 10 score plot for bark samples. GM1, GM2 and GM3, which are the different maturity stages of Sri Gemunu accession, exhibited the highest PC1 and can be considered as the best cinnamon bark sample. N1 and Galle WL 1a of the seed propagation varieties have been placed on the positive side of the PC1 as the best samples of climatic and maturity variations of seed propagation varieties. FLW-(N) has the highest PC1 among the cinnamon samples belonging to the various physical stages while all the samples of the same category are placed in the same

cluster. When considering all the factors (maturity, physical stage, and climatic variation), cinnamon bark of >5 y. matured seed propagation varieties which grown in WL 1a climatic zone district in fruiting season is the best variety.

CONCLUSIONS

This study provides very comprehensive data on how the phytochemical contents of leaves and bark of the Ceylon cinnamon vary with different climatic conditions in the country, different maturity stages of the plant, different propagation types and the physical stage of the plant. This study is credited as the first detailed and systematic study conducted on Ceylon cinnamon for its phytochemical composition variation based on the climatic conditions, genotype, maturity stage and physical stage of *C. zeylanicum* plants. The qualitative phytochemical screening showed the presence of diverse phytochemicals and the effect of climatic or maturity stage variation on the availability of phyto-constituent. The phytochemical content has greater impact on maturity, genotype and physical stage of the plant and climatic variation. In seeding propagation type, the climatic zone, WL 1a is the best agro-ecological zone where it showed higher alkaloids, flavonoids and poly-phenol. In vegetative propagation, Sri Gamunu in IL 1a and Sri Wijaya in WU 2b are the best zones for high yield of phytochemical content. When considering the variance of the maturity, >5 (bark) and 3 to 5 y (leaves) in seeding propagation type are the best maturity stages. Sri Gamunu is better than Sri Wijaya for both leaves and bark samples. According to the seasonal variance, FLSH and FRT-b are the best seasons for bark and leaves, respectively, in seed propagation type. The results of this study would be supportive to the scientific community for extending their studies towards identifying novel applications as well as development value added products in order to cater larger community and to increase the value chain.

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