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SHORT COMMUNICATION

PROXIMATE AND PHYTOCHEMICAL ANALYSIS OF SELECTED WILD EDIBLE GREEN LEAFY VEGETABLES IN BATTICALOA

*K. Ravivarman**, *U. Mathiventhan*

Department of Botany, Faculty of Science, Eastern University, Vantharumoolai, Sri Lanka

ABSTRACT

Plant-derived natural products provide many novel bioactive molecules that are available in the market today as medicines or food. Wild Green Leafy Vegetables (WGLVs) have essential nutrients which are necessary for a healthy life. Three WGLVs such as *Allmania nodiflora*, *Borreria hispida,* and *Rivea ornata* were selected for the proximate and phytochemical analysis based on the market survey. Selected dry WGLVs were used to investigate proximate such as moisture, ash, fat, and fiber and phytochemicals such as alkaloids, flavonoids, tannins, steroids, terpenoids, phenolic compounds, and saponins analysis by using standard methods. The results were shown as follows: moisture 71.7±0.95- 80.3±2.84%, ash 8.84±0.69 - 11.05±0.21 %, fat 1.36±0.48 - 2.56±0.28 % and fiber 6.85±0.46 - 8.04±0.11 %. The qualitative screening of water, ethanol, and acetone extracts revealed that select WGLVs contained alkaloids, flavonoids, tannins, steroids, terpenoids, phenolic compounds, and saponin. *Rivea ornata* lacked quinones, while anthraquinones were only found in the water extract of *Borreria hispida*, and none of the three WGLVs were found to contain glycosides.

Keywords: *Wild green leafy vegetables, Phytoconstituents, Nutrients*

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1. INTRODUCTION

The food needs of a rapidly growing population are increasing over time, but the available land resources are decreasing day by day due to drought [1]. Both raw and cooked leafy greens are used as the cheapest source of energy to overcome malnutrition. Additionally, leafy greens are considered part of a balanced diet and are a source of ess-

Corresponding author: krishanthinyk@esn.ac.lk

ential nutrients [2]. Green leafy vegetables are rich sources of antioxidant vitamins [3]. The wild edible plants growing in their natural conditions have been a source of food to the rural people inhabiting remote areas of the world including Sri Lanka.

Many people over the world are suffering from nutrient deficiencies [4]. Leafy vegetables are the food with the highest nutritional value [5]. According to Artemis [6], wild greens are a considerable source of vitamins, minerals, antioxidants, and phytochemicals. They are a good source of protein, very low in fat and carbs, and high in fiber [7]. Phytochemicals are bioactive compounds found in plants that work with nutrients and dietary fiber to protect against diseases. They are non-nutritive compounds. These phytochemicals are the secondary metabolites present in smaller quantities in higher plants and they include alkaloids, Steroids, flavonoids, terpenoids, tannins, and many others [8]. Many phytochemicals have antioxidant activity and reduce the risk of many diseases. It is crucial to know the type of phytochemical constituent, thus knowing the type of biological activity which might be exhibited by the plant [9]. Wild plants contain many natural antioxidant compounds identified as free radicals or active oxygen scavengers [10].

Mathiventhan et al [11] reported that 30 species of edible green leaves were obtained from home gardens; river sides and forest lands for consumption in the Eastern Province of Sri Lanka. There are more wild edible leafy vegetables available in the Batticaloa district among them few of them are subjected to investigation of nutritional and phytochemicals compounds. Therefore, this study aimed to investigate the presence of nutritional such as ash, fat, moisture, and fiber and phytochemicals such as alkaloids, flavonoids, tannins, steroids, terpenoids, phenolic compounds, and saponins in selected WGLVs.

2. MATERIAL AND METHODS

2.1 Sample Selection

A Preliminary market survey was carried out from Kallar to Oddamavadi of the Batticaloa District, Sri Lanka in order to find out the availability of GLVs. Interviews were carried out with vegetable vendors, mainly focusing to find out wild greens. Based on the survey, three WGLVs were selected for further studies.

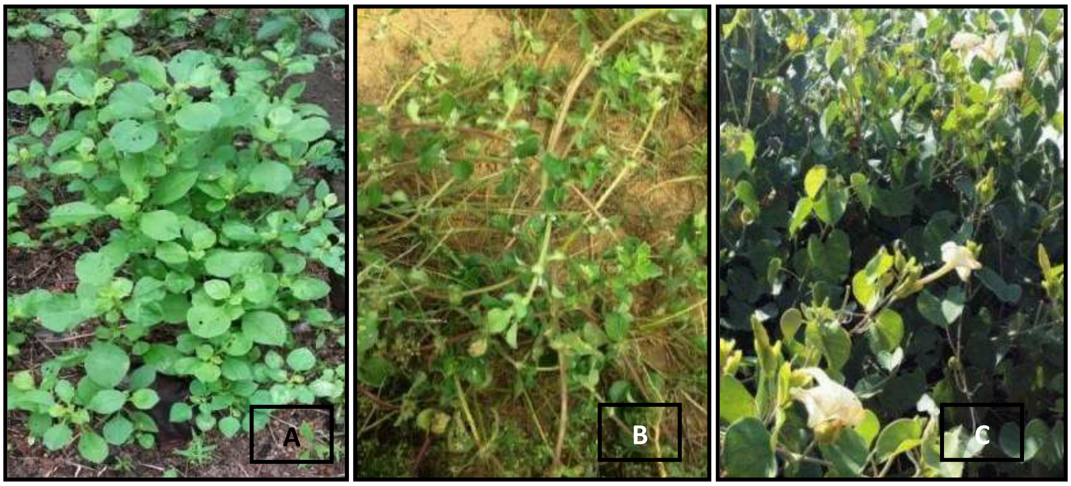

Figure 1: Selected wild edible green leafy vegetables for nutritional and phytochemical analysis: (A) *Allmania nodiflora* (Kumattikkirai-T), (B) *Borreria hispida* (Nathaichoori- T), and (C) *Rivea ornata* (Musuttai-T)

2.2 Identification of Wild Green Leafy Vegetables (WGLVs)

Preliminary identification of the WGLVs was done with the help of vendors in the market and with the help of available sources such as herbarium specimens from the Department of Botany, Eastern University, and other published documents.

2.3 Sample Collection and Treatment

Healthy, disease-free three different Wild Green Leafy vegetables (WGLVs) were collected from local markets in the Batticaloa district (Batticaloa, Aarayampathy, and Chenkalady). The samples (randomly selected samples from each WGLV) were identified and leaves were separated from the plant and washed thoroughly under running tap water and distilled water. These samples were shade dried for 1 week without any contamination. The dried samples were used for Nutritional and qualitative phytochemical analysis.

2.4 Nutritional Analysis

By using the standard methods of Association of the Analytical Chemists (AOAC) [12], nutritional analysis was carried out. All the analysis was done in triplicates.

Moisture: The moisture content of samples was determined using the oven (Model YCO-010- 200L USA) at 105^{o}C for 8 to 12 hours. After drying, the sample was transferred into the desiccators to cool and be weighed. The weighing was continued until a constant weight was obtained. The percentage moisture content was calculated as follows:

$$\% \text{ Moisture (w/w)} = \frac{\text{Weight of wet sample - Weight of dry sample}}{\text{Weight of wet sample}} \times 100$$

Ash: Finely powdered, dried sample of 2 g was taken in a weighed crucible and ignited using a muffle furnace at 500 ^{o}C for 6 hrs. The crucible was cooled in desiccators and the mass was ground with a pestle. The ash weight was recorded. The percentage ash content was calculated as follows:

$$\% \text{ Ash (w/w)} = \frac{\text{Weight of ash}}{\text{Weight of dried sample}} \times 100$$

Fat (Soxhlet extraction): The empty Soxhlet flask was weighed. A dried leaf sample of 5g was weighed and wrapped in filter paper. It was taken into extraction thimble and transferred into the Soxhlet distillation unit. Petroleum ether (bp 40 – 60 ^{o}C, 200 ml) was taken into the Soxhlet flask and fixed in the apparatus. It was heated in an electric heater for 6 hrs. The extraction was completed when a drop of the ether taken from the drippings of the extractor leaves no greasy stain on a filter paper. The boiling was stopped and the flask was disconnected. Then the excess solvent was evaporated in a water bath and dried in the oven at 70 ^{o}C. The dry weight of the Soxhlet flask with oil was recorded. The percentage fat content was calculated as follows:

$$\% \text{ Fat (w/w)} = \frac{\text{Weight of ether extract}}{\text{Weight of dried sample}} \times 100$$

Fiber: A moisture and a fat-free sample of 3 g were weighed and taken into a beaker (500 ml). H_2SO_4 (1.25 %, 200 ml) was added to it. The contents were boiled for 30 minutes. Then the contents were filtered through a muslin cloth and washed with distilled water until the washings are no longer acidic. The residue was transferred into the beaker containing NaOH (1.25 %, 200 ml) and boiled for 30 minutes. The residue was transferred into a crucible and oven dried at 105^{o}C. Then it was cooled and weighed. The weight represents the fiber plus ash content in comparison to the initial weight. Finally, the residue was ignited at 500 ^{o}C for 6 hrs using a muffle furnace. The percentage fiber content was calculated as follows:

$$\% \text{ Fiber (w/w)} = \frac{\text{(Oven dried weight – Ash weight)}}{\text{Weight of sample}} \times 100$$

2.5 Qualitative phytochemical analysis

Phytochemical analysis was carried out for the extract (water, ethanol, and acetone) as per standard methods described by [13], [14].

Table 1: Methodology for the phytochemical analysis

Phytochemicals	Test	Positive results
Alkaloids	Mayer's Test / Wagner's test	Prominent yellow ppt
Flavonoids	Lead acetate test	Yellow colour precipitate
Steroids	Salkowski s Test	Purple/Dark green
Terpenoids	Salkowski s Test	Reddish brown colouration at the interface
Anthraquinones	Borntrager s Test	Pink colour
Quinones	Salkowski test	Reddish brown colouration at the interface
Phenols	Ferric chloride test Lead acetate test	Bluish black colour yellow colour precipitate
Saponins	Foam test	Formation of frothing
Tannins	Ferric chloride test	Dark green colour
Glycosides	Liebermann's Test	Green colour

3. RESULTS AND DISCUSSION

3.1 Selection of Wild Green Leafy Vegetables (WGLVs)

According to a preliminary market survey, there are around 25 different types of green leafy vegetables are available in the market [11]. However, due to seasonality, some GLVs, such as *Allmania nodiflora*, *Borreria hispida*, *Rivea ornate*, *Pisonia grandis*, and

Aerva latana are infrequently offered on the market. Hence it was revealed that there was a demand for such Leafy Vegetables due to a lack of availability. By considering the consumable preference and the demand, *Allmania nodiflora*, *Borreria hispida*, and *Rivea ornata were* selected for the study (Table 2, Figures 1, 2, and 3).

Table 2: Wild Green Leafy Vegetables collected for the study

Species Name	Family	Local Name (Tamil and Sinhala)
Allmania nodiflora	Amaranthaceae	Kumattikkirai (T), Kumatiya(S)
Borreria hispida	Rubiaceae	Nathaichoori (T), Hithhadakola(S)
Rivea ornata	Convolvulaceae	Musuttai (T)

3.2 Proximate analysis of WGLVs

The Proximate analysis of WGLVs except for moisture content (WW) was determined on a dry basis (DW) and the results showed marked variations (Table 3).

Table 3: Nutritional (%) Analysis of selected WGLVs. Each value represents the mean ±SD

Nutritional Composition (%)	*Allmania nodiflora*	*Borreria hispida*	*Rivea ornata*
Moisture	71.7±0.95	79.8±1.71	80.3±2.84
Ash	11.05±0.21	8.84±0.69	9.45±0.47
Fat	2.56±0.28	1.36±0.48	1.53±0.15
Fiber	7.28±0.49	6.85±0.46	8.04±0.11

Vegetables are quick sources of energy and are highly beneficial for the normal growth and development of the human body [15]. The present study revealed that the moisture content varied from 71.7± 0.95 % for *Allmania nodiflora* to 80.3 ± 2.84 % for *Rivea ornata*. These values supported the results (60 – 90 %) of investigated vegetables as indicated by FAO [16].

A previous study done by Sharma & Patel [17] showed that the moisture content of *Rivea ornata* is 82.65% [17]. This is also nearly similar to the finding. The ash content is generally recognized as a measure of quality for the assessment of the functional properties of foods [18]. In the present study, ash content was ranging from 8.84 ± 0.69% for *Borreria hispida* to 11.05±0.21% for *Allmania nodiflora*. Previous studies showed that the total ash content of *Allmania nodiflora* leaf was 17.43% [19], *Rivea ornata* was 15.45% [19] and *Spermacoce hispida* was 9.6457% **but** the present studied WGLVs showed the ash content found to be lower (11.05%, 9.45%, and 8.84% respectively) than the previously studied values. This variability might be due to the differences in leaf habits, harvest time, and soil structure [20].

According to Aliyu [21], the fiber content of vegetables is known to support digestion and prevent constipation. The fiber content of selected WGLVs varied from 6.85±0.46 % for *Borreria hispida* to 8.04±0.11 % for *Rivea ornata.* A previous study showed the fiber content of *Allmania nodiflora* was ranging from 0.62-5.59%. The crude fiber content of *Allmania nodiflora* was 5.47% [19] but the present study showed, the fiber content of *Allmania nodiflora* is 7.28±0.49% so this value is higher than the previously studied leafy vegetable.

Vegetable fats and oils lower blood lipid levels and help to minimize the diseases caused by the malfunction of coronary veins [22 Adenipenkun]. The fat content obtained was 1.36±0.48% for *Borreria hispida*, 1.53±0.15 for *Rivea ornate, and* 2.56±0.28 % for *Allmania nodiflora*. The fat content of these WGLVs also confirmed many authors' findings indicating that leafy vegetables are the sources of poor lipids content [23].

3.3 Qualitative Phytochemical analysis

The phytochemical analysis consists of identifying plant chemical compounds showing pharmacological interest. In the present investigation, the qualitative screening of the water-ethanol and acetone extracts revealed the presence of a wide range of phytoconstituents given in (Table 4).

Phytochemicals are bioactive compounds found in plants that work with nutrients and dietary fiber to protect against diseases. They are non-nutritive compounds. These phytochemicals are the secondary metabolites present in smaller quantities in higher plants and they include alkaloids, Steroids, flavonoids, terpenoids, tannins, and many others [24].

Table 4: Qualitative phytochemical test for selected WGLVs in water, ethanol, and acetone solvent

Phytochemicals	Test	*Allmania nodiflora*			*Borreria hispida*			*Rivea ornata*		
		W	E	A	W	E	A	W	E	A
Alkaloids	Mayer's test	+	+	+	+	+	+	+	+	+
Flavonoids	Lead acetate	+	+	+	+	+	+	+	+	+
Steroids	Salkowski Test	+	-	-	+	+	+	+	+	+
Terpenoids	Extract + Chloroform + Conc. H_2SO_4	+	+	+	+	+	+	+	+	+
Anthraquinones	Bontrager's Reagent	-	-	-	+	-	-	-	-	-
Quinones	Extract + Conc. H_2SO_4	+	+	+	+	+	+	-	-	-
Phenols	Extract + $FeCl_3$ Extract + Lead acetate	+	+	+	+	+	+	+	+	+
Saponins	Foam Test	+	+	+	+	+	+	+	+	+
Tannins	Extract + $FeCl_3$	+	+	+	+	+	+	+	+	+
Glycosides	Liebermann's Test	-	-	-	-	-	-	-	-	-

(+) Presence or (-) Absence of phytochemical (W-Water, E- Ethanol, A- Acetone)

The results showed the presence of alkaloids, flavonoids, steroids, terpenoids, phenol, saponins, and tannins in all studied WGLVs the meantime presence of quinones in *A. nodiflora* and *R.ornata,* anthraquinones were only present in water extract of *B. hispida* and absence of glycosides in all three selected WGLVs. Here *Borreria hispida* shows qualitatively higher selected phytochemicals than the other two WGLVs.

CONCLUSION

The present study indicated that *Rivea ornata* has higher moisture, and fiber content than other selected WGLVs and *Allmania nodiflora* has high ash and fat content than the other two WGLVs. In a general observation that leafy vegetables are low lipid-containing foods. The qualitative phytochemical screening of the different extracts revealed the presence of a wide range of phytoconstituents. The results suggested that if consume these WGLVs in appropriate quantities would provide a wide range of health advantages for a secure and healthy life.

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