Morphological, Microscopical Characters and Elemental Analysis of *Limnocharis flava* (L.) Buchenau

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Abstract

The botanical study on *Limnocharis flava* (L.) Buchenau belongs to the family Alismataceae. had been undertaken. These plants were wild grown in places with water. They were collected from Hlegu Township, Yangon Region. The morphological characters were examined to ascertain their correct identification. The plant was hydrophyte. Leaves were simple and exstipulate. Inflorescences were cymose and flowers were ebractolate. Microscopical characters of the leaves were also conducted by free hand section method according to Esua. The epidermal cells of the lamina were polygonal in shaped. Stomata present on both surfaces were numerous and anomocytic type. In transverse section of midrib and petiole, the vascular bundles were collateral type. The elemental analysis was conducted by using the Energy Dispessive X-Ray (EDXRF) technique. Potassium (K) and calcium (Ca) were found as macronutrients elements and bromine (Br), copper (Cu) and zinc (Zn) were found as micronutrients elements in leaves. Medicinal plants contain numerous biologically active constituents which are helpful in improving the human life.

Key words: *Limnocharis flava*, nutrient, anomocytic

Introduction

The yellow velvetleaf plant is a type of plant that lives in water. This plant can be used as a vegetable for daily consumption in community. This plant is a pharmacological effect related to treatment and health maintained for the community because it has chemical compounds such as saponins, phenoland also mineral content (Jamila *etal*, 2021).

Aquatic angiosperms are derived from terrestrial ancestors, and have adopted the water habit at various times subsequent to their first appearance as flowering plants. These plants are the conspicuous plants that dominate wetlands, shallow lakes and streams. Lack of aquatic plants indicate water quality problems and changes in the ecological status of the water body. The vegetation of lakes and ponds are rich in aquatic flora and constitute very important resources of food and medicine for the rural population (Cook and kairiukstis, 1990).

Limnocharitaceae contain about 2 genera and 2 species in tropical and subtropical region. This family had about 3 genera with 12 species and then, genera Limnocharis had 2 species. *Limnochar flava* (L.) Buchenau is a perennial, robust, erect, lactiferous, marsh herb, tall, rooting in mud and strongly tillering. Leaves basal, glabrous, long petioled, entire, yellowish-green. Inflorescence axillary, long-peduncled, umbelliform, glabrous, 5-15 flowered, erect. Flowers rather large, in the axils of the membranous bracts, pedicle long, sepal 3, enlarge and yellow green, petals3, pale yellow. Stamens are more than 15, surrounded by a whorl of staminodes. Ovaries are numerous in one whorl, superior, laterally much compressed, stigmas narrowly linear (Dassanayake, 2000).

Limnocharis flava (L.) Buchenau is a perennial, robust, erect, lactiferous, marsh herb, 20-100 cm tall, rooting in mud and strongly tillering. Leaves basal, rosulate, glabrous, long-petioled, orbicular or ovate, entire, yellowish-green, folded lengthwise in bud. Inflorescence axillary, long-peduncled, umbelliform, glabrous, 5-

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15-flowered, erect, recurved after fruiting until it reaches the water or mud; peduncles 10-90 cm long. Flowers rather large, in the axils of membranous bracts; sepal 3, enlarged yellow-green, ovate-elliptic; petals 3, pale yellow, obovate, very thin, apex rounded, base darker coloured. Stamens more than 15, surrounded by a whorl of staminodes. Ovaries numerous in one whorl, superior, laterally much compressed, simulating a single, deeply incised ovary; stigmas narrowly linear, sessile. *Limnocharis flava* (L.) Buchenau is spread throughout South-East Asia (Malaysia, Indonesia, Thailand, South Myanmar, Sri Lanka, India and Vietnam), especially in rice-growing areas, and in South America and the USA (Waterhouse, 2003).

These flowering plants are several commercial products such as food, lumber, cosmetics, medicinal and ornamental plants. The first necessity for microscopically examination and analysis of powdered drugs is a practical knowledge of histology of plant acquaintance with the type of cell and cell contents which provide the diagnostic characters of drug. The size and form of the cell type and the nature of their contents then enable the worker to form an opinion as to the identity of the plant of which some particular member is present (Trease and Evans, 1978).

The leaf is a specialized organ in which the function of photosynthesis is centered**.** This is the most important of plant functions, since all other functions depend upon it or contribute to it either directly or indirectly. In dicotyledonous leaves of the broadly expended type, stomata normally occur in largest numbers on the dorsal surface. In the majority of leaves, the stomata are arranged apparently without regular pattern of orientation. They are spaced more or less equidistantly from each other and are only rarely found upon the veins. Xerophytes and some other plants often have stomata in groups or rows or sunken in furrows or otherwise protected (Eames, 1947).

Limnocharis flava (L.) Buchenau, native to tropical America, is naturalized as a noxious weed in Sri Lanka, India and Southeast Asian countries. It is widespread in flood plains, wetlands and agricultural wetlands resulting in poor drainage. Yellow velvetleaf is a fast-growing plant. It cannot be optimally used, particularly the old leaves. Old yellow velvetleaf waste has not been used appropriately, even though it has more benefits and value. The leaves of yellow velvetleaf contain natural color of carotenoids and flavonoids which can be used for natural dyes. Yellow Velvetleaf is categorized as swamp plants. It is a native American plant that grows in tropical and sub-tropical climates. The plant is found in India, Vietnam, Thailand, Laos, Cambodia, Malaysia and Indonesia. Young Yellow Velvetleaf is an edible vegetable as used by people. *Limnocharis flava* (L.) Buchenau showed morphological adaptations in different water conditions, including significant differences in the relative biomass allocation for root, petioles and leaves (Rusydi, 2014).

In chemical constituents, the active constituent is a mixture of alkaloids, fats and carbohydrate, have been isolated from the products (Kirtikar and Basu,1935).

In Myanmar, there were no researchers reported this species not only morphological characters and microscopical characters but also phytochemical investigation in this region. Therefore, it was selected to study and to do research.

For the facts in these research, morphological and microscopical characters of fresh specimens of leaves and elementary analysis of leaves of *Limnocharis flava* (L.) Buchenau were carried out. The aim of the plant of *Limnocharis flava* (L.) Buchenau to identify the morphological, microscopical characters of leaves, and elementary analysis of the leaves powdered.

Materials and Methods

Botanical Studies

Collection and identification of specimens

The specimens of *Limnocharis flava* (L.) Buchenau were collected from Hlegu Township, Yangon Region during the flowering and fruiting periods from May 2022 to September 2022.

After collection, the specimens were identified with the help of available literatures such as Kirtikar and Basu (1935), Cook and Kairiukstis (1990) and Rusydi, (2014). Both the vegetative and reproductive parts of the fresh specimens were used for the morphological and microscopical studies.

Microscopical studies

The samples were thoroughly washed with water, weighted and air dried in an open shaded area for about three weeks. When constant weight was obtained, the dry samples were pulverized by grinding machine into powder and kept in air tight bottle for further study the powdered characteristic. The powders were cleared in chloral hydrate solution on a glass slide and observed under the compound microscope.

For microscopical studies, free hand sections of fresh specimen from lamina, midrib and petiole were made and examined under microscope. The following chemical and reagents were used to examine for freehand sections.

(i) Chloral- hydrate solution B.P as clearing reagent.
(ii) Solution of phloroglucinol B.P followed by with concentrated hydrochloric acid for lignin.
(iii) Acetic acid and 80% sulphuric acid B.P for calcium oxalate crystals.

The lamina, midrib and petiole were examined by preparing free hand sections from the fresh specimens, according to the methods of Esau (1965), Trease and Evans (1978), Pandey (1978) and Tandon (2011).

Elemental analysis of *Limnocharis flava* (L.) Buchenau powdered leaves

The analysis of elemental concentration of the leaves on *Limnocharis flava* (L.) Buchenau were carried out by using Energy Dispersive X-ray Fluorescence (EDXRF) spectrometer technique at the Universities Research Center, Yangon.

Sample Preparation of Elemental analysis

EDXRF X-ray Fluorescence Spectrometer is and instrument the quantitatively determines the elements comprising a sample. The EDX - 700 spectrometer is used for determination of element from sodium (Na) to uranium (N). The parameters of each part of the spectrometer are given below.

Detector type	:	Si (Li) detector
LN_2 supply	:	Only during measurement
Dewar capacity	:	3 liters
LN_2 consumption	:	less than 1 liter per day
Detection area	:	10 mm^2
Resolution	:	less than 155 ev (Min K, 1500 H_Z)

The powdered samples were pressed into pellet (30 mm in diameter and 5 mm in thickness) and were used in the EDX- 700 spectrometer as specimens.

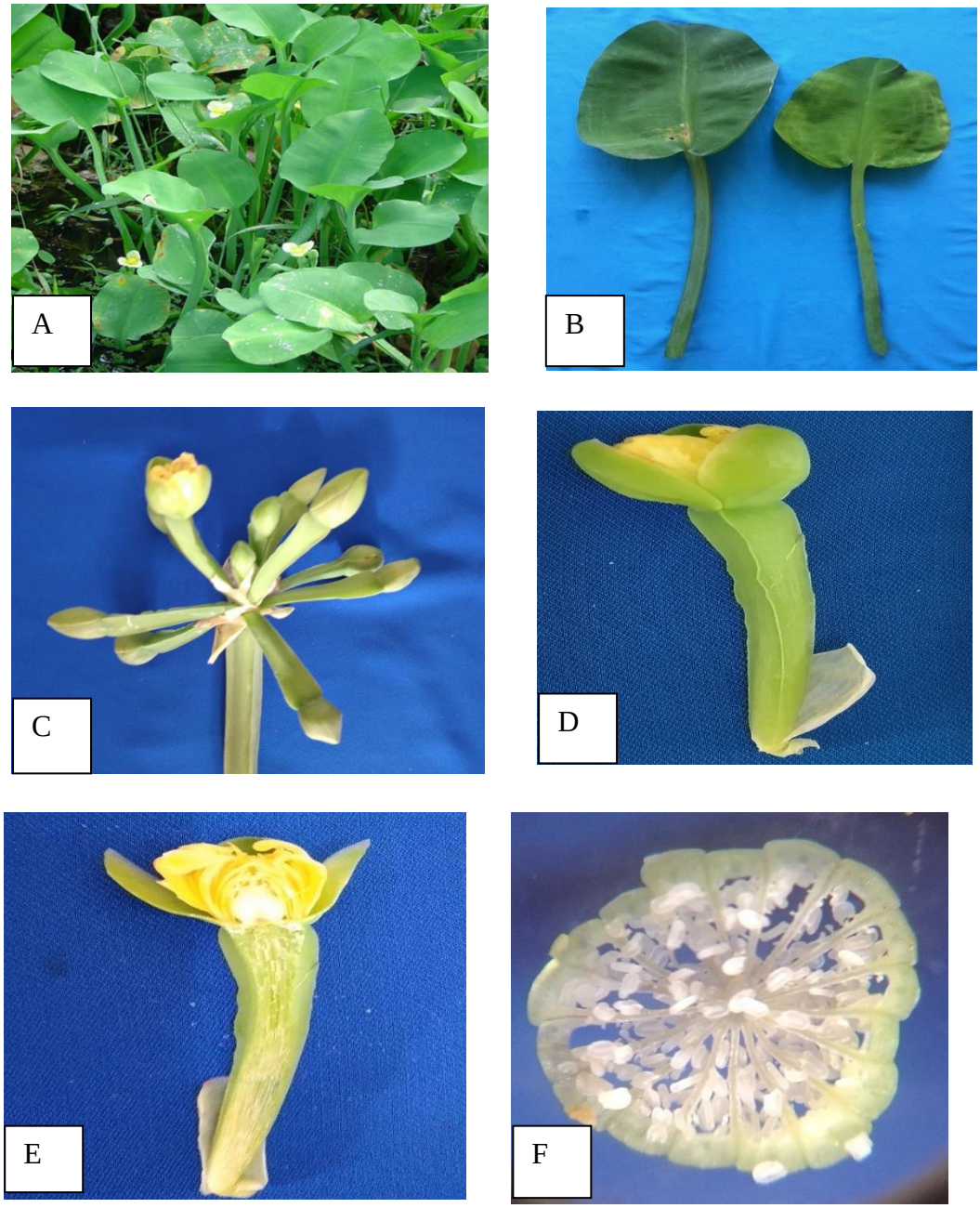
Results

Botanical Studies

Morphological Characters of *Limnocharis flava* (L.) Buchenau

Scientific name	-	*Limnocharis flava* (L.) Buchenau
Myanmar name	-	Tet-pya
English name	-	Yellow velvet Leaf
Family	-	Limnocharitaceae
Flowering and Fruiting Period	-	June to December
Part used	-	Leaves and Flowers

Habit; Perennial rhizomatous aquatic herbs; **Leaves**; simple, spiral, petiolate (very long), exstipulate; **Inflorescence**; axillary umbel-like cymose; **Flowers**; bracteate, ebracterolate, complete, bisexual, regular, actinomorphic, tri-merous, cyclic, hypogynous; Sepals 3, aposepalous, sepaloid, persistent, inferior; Petals 3, apopetalous, petaloid (yellow), inferior; Stamens α, free, surrounded by a whorl of flattened staminodes, the anther fertile, filament unequal, anther dithecous, introrse, basifixed, longitudinal dehiscence, inferior; Carpels (α), syncarpous, multilocular, superficial placentation, many ovules in each locule in transverse section, style short, stigma radiate, superior.



(A) Habit (B) Leaves (C) Inflorescence (D) Flowers (E) L.S of flower (F) T.S of ovary

Figure (1) Morphological Characters of *Limnocharis flava* (L.) Buchenau

Microscopical Characters of leaves of *Limnocharis flava* (L.) Buchenau

Lamina

In surface view, the upper and lower epidermal cells were parenchymatous, polygonal in shaped, thin-walled and completely arranged. Stomata were present on both surface. The type of stomata was paracytic. Stomata elliptic in shape, guard cells were reniform shape with chloroplast.

In transverse section, thin cuticle smooth was present on both surfaces. The arrangement of the lamina tissue was dorsiventrally arrangement. The epidermal cells were made up of one layer parenchymatous cells and barrel in shape. The mesophyll layer consisted of palisade and spongy parenchyma. Palisade parenchyma cells are found on upper side and two-layered, the cells vertically erect, compact, chloroplast

abundant. The spongy mesophyll consisted of many layers of arenchymatous cells, irregular to isodiametric shape and loosely arranged.

The vascular bundles of lateral vein were embedded in mesophyll cells and oval in shape. They were concentric and closed type. The phloem cells were very small. The xylem tissue composed of vessel, tracheids, xylem fibers and xylem parenchyma cells. The phloem tissues consisted of sieve tube, companion cells, phloem fiber and phloem parenchyma cells. The xylem always toward the inner side and phloem always toward the outer.

The midrib

In surface view, cuticle was smooth on both surfaces. The epidermal cells of both surfaces were made up of thin walled parenchymatous cells. They were polygonal to rectangular in shape, elongated along the length of the midrib.

In transverse section of midrib, both surfaces were covered with thin cuticle. The epidermal cells were only one layered, rounded to oval shaped and compactly arranged. The lower epidermal cells were similar to these the upper cells. Below the epidermis the cortex was differentiated into collenchyma and arenchymatous cells. The collenchymatous cells were 2-3 layers in thickness towards the upper surface and 1-2 layers in thickness towards the lower surface. The arenchyma cells were numerous.

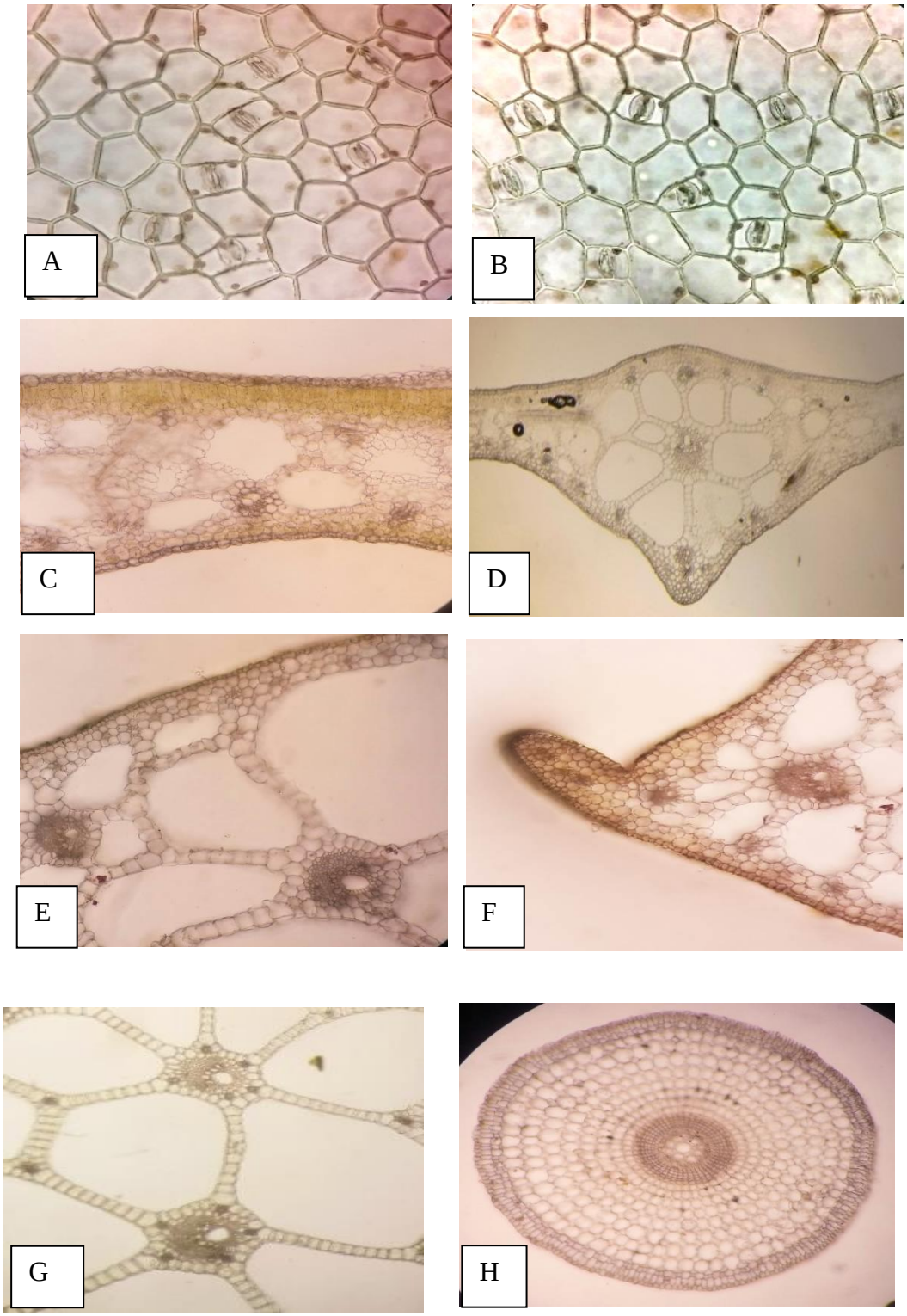
The vascular bundles were oval in outline and embedded in the cortex. The vascular bundles were collateral and closed type. The phloem cells were consisted sieved tube elements and companion cells. Xylem cells were composed of vessel, tracheids, xylem fibers and xylem parenchyma.

Petiole

In surface view, the epidermal cells were parenchymatous thin-walled and mostly barrel in shaped and elongated along the length of the petiole.

In transverse section, the petiole was triangular shape in outline. The cuticle layer was thin. The cortex was made up of two different types of tissues, outer collenchymatous and inner parenchymatous tissues. The outer collenchymatous cells below the epidermis consisted of 1-2 layers in thickness on both surfaces. The arenchymatous tissues were composed.

Vascular bundle was oval in outline and embedded in the arenchymatous tissue. Vascular bundle was collateral type. The phloem cells were thin walled and consisted of sieve tubes and companion cells. The xylem composed of vessels, tracheids, xylem fiber and xylem parenchymatous cells.



(A) Surface view of upper epidermis
(B) Surface view of lower epidermis
(C) T. S of lamina
(D) T.S of midrib
(E) T.S of petiole showing epidermal cell
(F) T.S of Petiole
(G) Close up view of vascular bundle
(H) T. S of primary root

Figure (2) Microscopical Characters of leaves of *Limnocharis flava* (L.) Buchenau

Elemental Analysis of *Limnocharis flava (L.) Buchenau* leaves

Elemental Analysis

The elements present in powdered leaves were quantitatively determined by EDXRF. It was found that Potassium (K), Calcium (Ca), Manganese (Mn) and Iron (Fe) were principal elements and Rubidium (Rb), Strontium (Sr), Bromine (Br), Copper (Cu) and Zinc (Zn) were trace elements. The results were shown in Figure (3) and Table (2).

Table (2) Concentration of elements contained in *Limnocharis flava (L.) Buchenau*

No.	Elements	Content%
1	Potassium (K)	1.720
2	Calcium (Ca)	0.158
3	Maganese (Mn)	0.036
4	Iron (Fe)	0.015
5	Rubidium (Rb)	0.004
6	***Strontium (Sr)***	0.002
7	***Bromine (Br)***	0.001
8	***copper (Cu)***	0.001
9	***Zinc (Zn)***	0.001

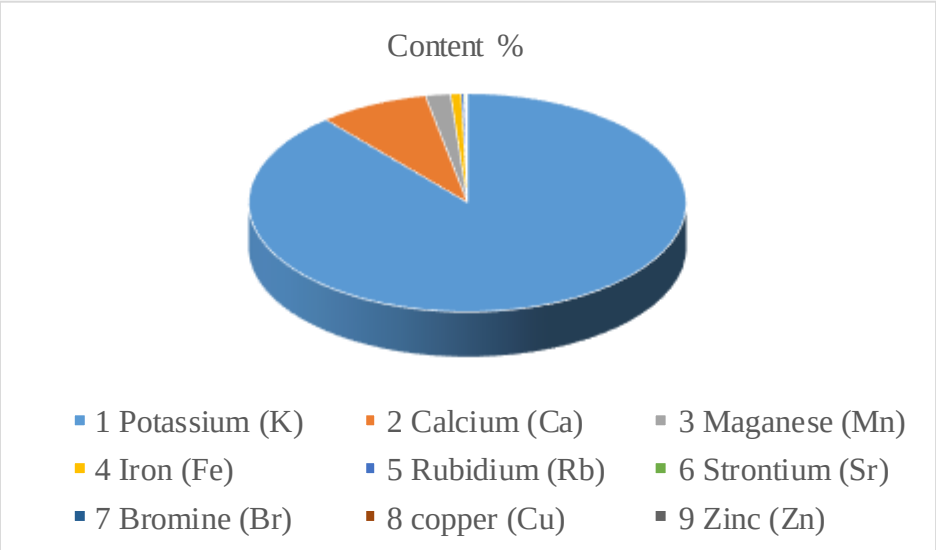


Figure 3. Elemental analysis of *Limnocharis flava* (L.) Buchenau powdered leaves

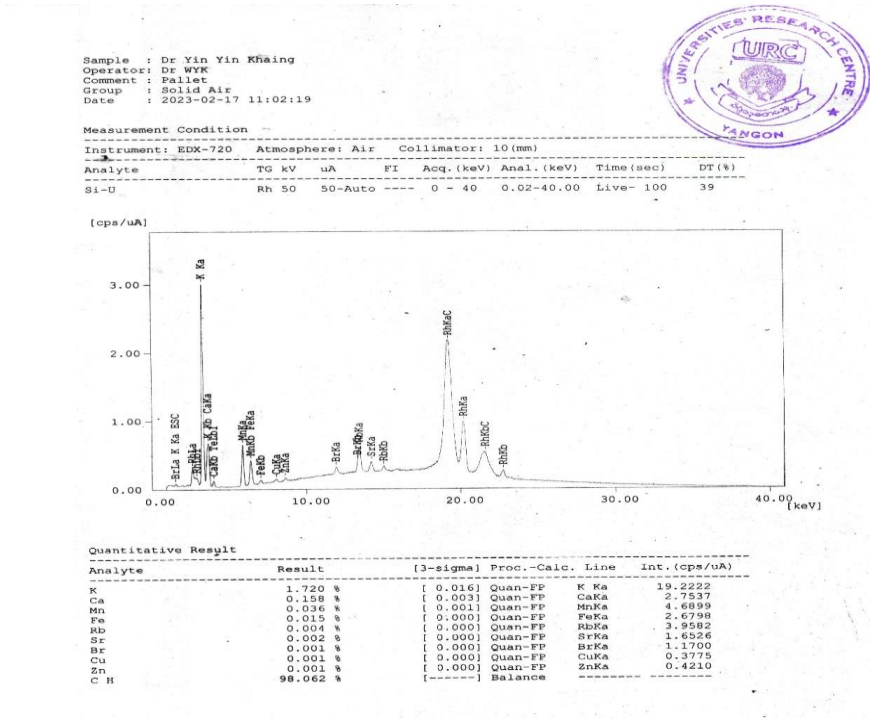


Figure 4. Elemental analysis of *Limnocharis flava* (L.) Buchenau powdered leaves

Discussion and Conclusion

The specimens of *Limnocharis flava* (L.) Buchenau belonging to the family Limnocharaticeae is aquatic herb and collected from Hlegu Township, Yangon Region. Flowering and fruiting period is May 2022 to September 2022.

The morphology studies on both vegetative and reproductive parts of the plants, the microscopical characters of leaves and elementary analysis of the leaves has been studied in this research.

In the morphological study, the plant of the *Limnocharis flava* (L.) Buchenau are perennial. The leaves are simple and exstipulate. The inflorescences are cymose. The flowers are yellow in color, ebracteolate and bisexual. The stamens are numerous, anther dithecous. Ovary are many carpels, unilocular and superficial placentation. These characters were in agreement with those mentioned by (Dassanayake, 2000 and Waterhouse, 2003).

The leaves are dorsiventral. The epidermal cells of upper and lower surfaces are polygolan in shaped. The stomata occur on both surfaces. Stomata are numerous and anomocytic types and arenchymatous cells were numerous. In transverse section of midrib and petiole, the vascular bundles were collateral type. These characters are agreements with those mentioned by (Metcalfe & chalk, 1950 and Eusa, 1965).

According to the EDXRF results were found that Potassium, Calcium, Manganese, Iron, Rubidium, Strontium, Bromine, Copper and Zinc elements. Potassium, Calcium, Manganese and Iron are generally known principal elements in human body. Bromine, Copper and Zinc elements are small amount of trace elements, many of which are essential to life (Maung Maung Htay, 2001). In this study, the elemental analysis of the leaves, potassium is found to be the highest in percentage and Bromine, Copper and Zinc are present in small quantities.

Potassium (K) requires maintaining health. For most people, a healthy diet rich in vegetables and fruits provides all of the potassium needed. People should be careful when taking medication that may further affect potassium leaves in the body.

A deficiency of potassium may result in fatigue, leg cramps, muscle weakness, slow reflexes, dry skin and irregular heart-beat.

Calcium (Ca) is the principal supporting material of the skeleton and teeth. It is one of the important components of a healthy diet. Calcium is needed for muscle construction, blood vessel construction and expansion, the secretion of hormones and enzymes and sending message through the nervous system. The maximum level of calcium is 2.5 g/day for people. Calcium deficiency can lead to osteoporosis.

Iron (Fe) present in small quantity has been considered an essential mineral for human body since a century ago. It is a mineral vital to good health. Symptom of iron deficiency may include fatigue, weakness, headaches, higher sensitivity to cold temperature and other health problems.

These minerals occur in some foods; it is essential that the diet should include a wide variety of foods. Potassium (K), Calcium (Ca) and Iron (Fe) are found in velvet leaves. So, this may be considered useful for medical purposes.

In conclusion, the morphological and microscopical studies, elemental analysis of leaves can give valuable information of *Limnocharis flava* (L.) Buchenau. Furthermore, medicinal plants contained numerous biologically active compounds which are helpful in improving the human life. by human health. It can be concluded that the extracts of *Limnocharis flava* (L.) Buchenau., especially the methanolic extracts, hold promise for development into natural preservatives with further research.

Acknoledgements

I would like to express appreciation to Dr. Thar Tun Maung , Rector, Dr. Myo Myin, Dr. San San Hmawe, Dr. San San Lwin, Pro-Rector, Dagon University Dagon University for permission to carry out of this research work. I wish to express my special thanks to Dr Tine Moe Aye Head of Professor, Dr Yee Yee Thu Professor Department of Botany, Dagon University, for her advice and encouragement. I would like to express my deepest gratitude to organizing committee of 4th Myanmar – Korea Conference hosted by Dagon University for participating this special event.

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