

Qualitative Analysis of Phytonutrient such as Xanthophyll, Carotenoids and Chlorophyll b in *Sesbania grandiflora* and *Erythrina variegata* a Comparative Study Under Alternate Temperature

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Abstract:- This study investigates the Phytonutrient such as Xanthophyll, Carotenoid and Chlorophyll b in *Erythrina variegata* and *Sesbania grandiflora* under varies conditions, which are commonly consumed in tropical and subtropical regions for their health benefits. Our primary aim was to assess the loss of chlorophyll pigments during boiling and shade drying processes. Thin Layer Chromatography (TLC) using silicon-coated sheets was employed as an analytical tool to detect and quantify the pigments in the samples. The R_f (Retention Factor) values for various pigments were calculated and recorded to derive the results.

Our findings indicate that temperature-sensitive pigments like xanthophylls degrade when the leaves are boiled, while shade-dried leaves showed a reduction in chlorophyll pigments (Chlorophyll a and Chlorophyll b) but retained xanthophylls. Additionally, ethanol was used as a solvent to extract pigments, revealing the presence of carotene in *Sesbania grandiflora* leaves. The study highlights the importance of optimizing cooking and preservation methods to retain phytonutrients, thereby maximizing the therapeutic and nutritional benefits of plant-based foods. Future research could focus on refining these methods to reduce nutrient loss, contributing to a more holistic approach to diet and health.

Keywords:- Phytonutrient, Xanthophyll, Carotenoid, Chlorophyll b, *Erythrina variegata* and *Sesbania grandiflora*, TLC, Temperature.

I. INTRODUCTION

The preservation of essential phytonutrients in plants is vital for ensuring that the nutritional value of food is retained from farm to table. Among these phytonutrients, chlorophyll stands out due to its significant role not only in plant biology but also in human health. As the green pigment responsible for capturing light energy during photosynthesis, chlorophyll is central to plant growth and oxygen production. However, its importance extends beyond plants, as research increasingly highlights chlorophyll's potential health benefits for humans.

Chlorophyll is recognized for its potent antioxidant properties, helping neutralize harmful free radicals in the body, which, if left unchecked, can lead to cellular damage and contribute to chronic diseases like cancer, cardiovascular disease, and aging. Its anti-inflammatory effects make it valuable in reducing inflammation, which is a root cause of numerous health issues, including arthritis and autoimmune conditions. Additionally, chlorophyll plays a significant role in detoxification, particularly in eliminating toxins from the body, thus supporting liver health and reducing the risk of toxin-related diseases.

The preservation of chlorophyll consumption is crucial, as its stability can be compromised by factors such as heat, light and pH changes during cooking and storage. Different methods of cooking and preservation can either protect or degrade chlorophyll pigments, affecting both the color and the nutritional quality of plant-based foods. Maintaining the stability of chlorophyll ensures that the health benefits, including its antioxidant, anti-inflammatory, and detoxifying effects, are preserved, enhancing the overall nutritional impact on the diet.

Erythrina variegata (Indian Coral Tree) and *Sesbania grandiflora* (Agathi or Hummingbird Tree) are two species of leguminous trees commonly found in tropical and subtropical regions. Both plants have been historically used in traditional medicine and are also incorporated into the local diet in many cultures. Their leaves are known to be rich in essential nutrients, including vitamins, minerals and phytochemicals like chlorophyll, making them valuable food sources. The consumption of these leaves is often linked to various health benefits, including anti-inflammatory, hepato protective and digestive health-promoting effects.

Despite their recognized nutritional value, the impact of routine culinary practices such as cooking, drying, and refrigeration on the chlorophyll content of *Erythrina variegata* and *Sesbania grandiflora* leaves has not been thoroughly examined. This study aims to bridge this knowledge gap by analyzing the chlorophyll retention in these leaves after subjecting them to various cooking and preservation methods. By employing advanced chromatographic techniques such as Thin Layer

Chromatography (TLC) we seek to quantitatively assess the degradation or retention of chlorophyll pigments.

This comparative analysis will provide insights into how traditional culinary and preservation practices influence the nutritional integrity of leafy vegetables. Furthermore, the findings of this study could guide recommendations for optimizing cooking and preservation methods to retain maximum chlorophyll content, thereby preserving the health-promoting properties of these plants. With increasing awareness of the health benefits associated with chlorophyll and other phytonutrients, understanding how food preparation techniques affect nutrient retention is critical. This study not only addresses the specific context of *Erythrina variegata* and *Sesbania grandiflora*, but it also contributes to broader discussions on food sustainability, nutrition, and public health by offering practical recommendations for improving dietary practices.

➤ Distribution

Erythrina variegata is the species which is native to the tropical and subtropical regions of East Africa, North Australia and the islands of Indian Ocean and the Western Pacific Ocean to Fiji. (*United States Department of Agriculture*) Retrieved 18 December 2017. *Erythrina variegata* is commonly known as Tiger's claw (EPPO). Retrieved 26 August 2021.) and Indian Coral tree.



Fig 1 Leaves of *Erythrina variegata*

Sesbania grandiflora is commonly known as vegetable humming bird, (*plants.usda.gov*) katurai or agathi and it is native to Maritime Southeast Asia and Northern Australia. It is a small leguminous tree. It has the edible part of flowers and leaves and is commonly eaten in south east Asia and South Asia. (*Bureau of Plant Industry, Department of Agriculture, Republic of the Philippines*)



Fig 2 Leaves of *Sesbania grandiflora*

➤ Physical Description

Erythrina variegata is a deciduous tree growing to the height of 27m (89ft) tall. The plant shows the pinnate leaves with a 20cm petiole and three leaflet upto 20cm long and broad. It has dense cluster of scarlet or crimson flower and black seeds (Huxley, A., ed. (1992)).

Systematic Position

Kingdom	Plantae
Clade	Tracheophyta
Clade	Angiosperm
Order	Fabales
Family	Fabaceae
Genus	<i>Erythrina</i>
Species	<i>Variegata</i>

Sesbania grandiflora is one of the fast growing trees. The leaves are regular and rounded. The flowers are pink, red or white in colour. The fruit is bean and is long thin and green in colour. The tree is frost sensitive and thrives under full exposure to sunshine. It is a small soft wooded tree grows upto 16 to 82 ft tall. Leaves are 15 to 30cm long and around 10 to 20 pairs of leaflets are found on the rachis odd leaf on at the tip. Flowers are found to be long and oblong with two to four flowers in racemose inflorescence. The calyx is companulate and shallowly two lipped. Pod are slender, falcate or straight and long with a thick suture. Seeds are 8mm in size.

Systematic Position

Kingdom	Plantae
Clade	Tracheophyta
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Order	Fabales
Family	Fabaceae
Genus	<i>Sesbania</i>
Species	<i>grandiflora</i>

➤ Phytonutrient

Dark green leafy vegetables, such as spinach, kale, bok choy, broccoli, Swiss chard, and romaine lettuce, are rich sources of phytonutrients. One of the most important phytonutrients found in these vegetables is chlorophyll, a pigment essential for photosynthesis. Chlorophyll allows plants to absorb energy from light, giving them their characteristic green color. Beyond its crucial role in photosynthesis, chlorophyll has been associated with numerous health benefits when consumed as part of a diet rich in green vegetables (Ferruzzi & Blakeslee, 2007).

Chlorophyll is the green pigment found in the chloroplast, several other related pigments are found in the cyanobacteria. Its name is derived from the Greek words. (Muneer S, Kim EJ, Park JS, Lee JH (March 2014)). Chlorophyll enable the absorption of light energy.

Chlorophyll absorbs light in the red as well as blue portion of the electromagnetic spectrum. It is the poor absorber of green and near green light in electromagnetic spectrum. Chlorophyll containing tissues appear green because green light diffusively reflected by structures like cell

walls, is less absorbed. Two types of chlorophyll appears in photosystem of green plants chlorophyll a and b.

A photosynthetic pigment that are accessory pigment, Chloroplast pigment, antenna pigment present in the chloroplast in the photosynthetic bacteria and captures the photons from light which is essential for photosynthesis. Photosynthetic pigment .Carotene – an orange pigment, Xanthophyll – a yellow pigment , Phaeophytin a –a gray brown pigment, Phaeophytin b- yellow brown pigment, Chlorophyll a Blue green pigment, Chlorophyll b yellow green.

Chlorophyll ‘a’ is the most commonly found pigment, present in every plant that performs photosynthesis. Chlorophyll pigments absorb light more efficiently in a different part of the electro magnetic spectrum. Chlorophyll a absorbs well in the ranges of 400 to 450 nm and at 650 to 700nm;Chlorophyll ‘b’ absorbs well in the range of 450 to 500 nm and at 600 to 650nm. Xanthophyll absorbs well at 430 to 530 nm. None of the pigments absorb well in green – yellow region .(Virtanen, Olli; Constantinidou, Emanuella; Tyystjarvi, Esa (2022)). (Elsevier: PMID 10393259.)

Chlorophyll pigments are abundantly present in the green leafy vegetables and fruits which forms the integral part of our diet. Chlorophyll play crucial role in photosynthesis. Although limited, existing studies suggest that these photosynthetic pigments and their derivatives possess therapeutic properties. These bioactive molecules exhibit a wide range of beneficial effects, including antioxidant, antimutagenic, antigenotoxic, anti-cancer, and antiobesogenic activities. However, it is unfortunate that leafy materials and fruit peels often go to waste in the food supply chain, contributing to the prevailing issue of food waste in modern societies. Nevertheless, these overlooked materials contain valuable bioactive compounds, including chlorophylls, which offer significant health benefits

II. REVIEW OF LITERATURE

Erythrina, is known to exhibit its culinary application. The young flowers and the young leaves of the plant are used for the cooking purpose in the regions like South Asia and Pacific islands. (Chopra, R. N., Nayar, S. L., & Chopra, I. C. (1956)). The tender leaves can be used to prepare curry and also soups. These leaves are also used to prepare herbal teas. chlorophyll pigments that are essential for photosynthesis. (Duke, J. A., & Ayensu, E. S. (1985)).

The flowers of *S. grandiflora* are used as a vegetable in Southern part of Asia and SouthEast Asia. The leaves are edible. Cucio, Ardy L.; Aragonese, Julie Ann(February 2021)). . The main types of chlorophyll pigments found in the plant *Erythrina variegata* and *Sesbania grandiflora* are chlorophyll a ,chlorophyll b. Chlorophyll a is a primary pigment involved in photosynthesis, it is able to absorb the blue to violet electromagnetic spectrum and it reflects the green colour. Whereas chlorophyll pigment b acts as the accessory pigment assist chlorophyll pigment a by expanding the range of light wavelength a plant can use. The balance

between chlorophyll a and b allows the plant to optimize light absorption efficiently and carryout photosynthesis and the perfect balance among the chlorophyll pigments helps the plant to adapt varying environmental conditions such as light.

Research on the Phytonutrient content in *Erythrina variegata* focus on it potential uses in medicinal applications and how it can supplement nutrient to provide various benefits of consumption of chlorophyll as phytonutrient. The leaves of *Sesbania grandiflora* are rich in nutrients, and their chlorophyll content contributes to their antioxidant properties, making the plant valuable in traditional medicine and nutrition.(Islam, M.T., et al. (2020), Manikandan, P., & Prabhakaran, R. (2015).

Chlorophyll, the green pigment present in plants, provides numerous health advantages when taken as a supplement. It is recognized for its possible detoxifying effects, assisting in the removal of harmful toxins from the body by binding to cancer-causing substances. Chlorophyll also supports digestion by enhancing gut health, potentially due to its role in maintaining a balanced gut microbiome. Some research indicates that chlorophyll may improve wound healing by promoting tissue regeneration and reducing inflammation. Moreover, it serves as a potent antioxidant, which helps shield cells from damage caused by free radicals, contributing to overall health and potentially lowering the risk of certain chronic illnesses. Additional studies suggest that chlorophyll might benefit skin health and help control body odor by reducing internal odor production and combating bacteria. (Das, B., & Mandal, S. (2020)).

Chlorophyll, a complex green pigment found in plants ,algae and in certain bacteria plays crucial role of converting the light energy into the chemical energy .(L.O., Pa'ageorgiou G.C., Blankenship R.E., Govindjee A) . Recent research shows potential benefits of chlorophyll as the chemopreventive agent have provided the molecular mechanism of chlorophyll activity . (Nagin et al.)

In vitro and in vivo studies shows that the anticancerous in man remains scarce. Dietary supplements containing chlorophyll and chlorophyllin are available and generally considered safe, with no reported adverse side effects over several decades of human use . However, skepticism about their effectiveness persists due to the lack of robust scientific evidence supporting their claimed health benefits. (MacKeen D.)

Despite the potential health benefits associated with chlorophyll, a significant number of chlorophyll-rich vegetables, leafy materials, and fruits are lost throughout the food supply chain (FAO). This loss occurs despite the underutilized potential of these agro-food residues .

Harnessing and utilizing this discarded material could contribute to the transition towards a more sustainable circular economy.

➤ *Aim*

The main aim of our study is to analyse of Phytonutrient such as Xanthophyll, Carotenoid and Chlorophyll b in *Erythrina variegata* and *Sesbania grandiflora* under various conditions.

➤ *Objectives :*

To test the presence of various phytonutrient chlorophyll in the leaves of *Erythrina variegata* and in *Sesbania grandiflora* by Chromatographic study specifically by TLC.

To compare the presence of such a substance which shows therapeutic property in both the samples of leaves belonging to the family Fabaceae

To compare the presence of chlorophyll pigments in both the samples of leaves (*Erythrina variegata* and *Sesbania grandiflora*) when soaked in boiling water for 5 minutes.

To compare the presence of phytonutrient in both the samples of leaves (*Erythrina variegata* and *Sesbania grandiflora*) when dried under shade for 8 hours and powdered using blender (preservative method).

III. MATERIALS AND METHODS

➤ *Sample Collection and Preparation*

- Fresh leaves of *Erythrina variegata* and *Sesbania grandiflora* were collected from Chennai.

- 100 grams of each type of leaf were cleaned and air-dried under shade for 8 hours.
- Once dried, the leaves were powdered using a clean blender to prepare samples for analysis.

➤ *Preparation of Extracts*

- *Fresh Leaf Extract:*

- ✓ 2 grams of freshly collected leaves were mixed with 2 mL ethanol in a conical flask.
- ✓ The mixture was kept at 25°C in the refrigerator for 1 hour.
- ✓ After 1 hour, the extract was filtered, collected, and labeled for TLC analysis.

- *Cooked Leaf Extract:*

- ✓ 5 grams of fresh leaves were soaked in boiling water at 100°C for 5 minutes.
- ✓ After boiling, the leaves were ground along with the water to create a homogenized extract.
- ✓ The extract was collected and labeled for further analysis.

- *Preserved Leaf Extract (Shade Dried):*

- ✓ 2 grams of powdered shade-dried leaves were treated with boiling water.
- ✓ The solution was filtered, and the extract was collected for TLC analysis.

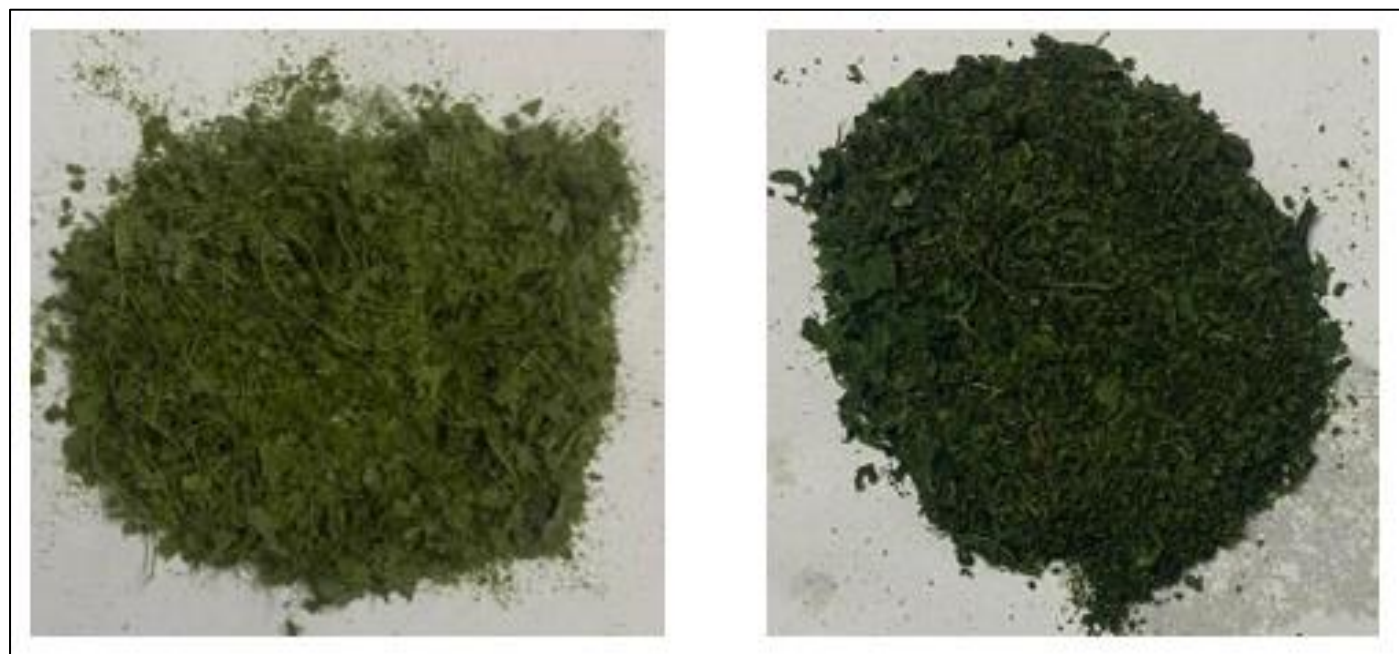


Fig 3 Preserved Leaf Powder of *Erythrina variegata* and *Sesbania grandiflora*



Fig 4 Solvent Extract of Leaf samples, preserved Leaf Extract (Aqueous), Cooked Leaf Extract (Aqueous)

➤ Chromatographic Analysis Using TLC

• Principle of TLC

Thin Layer Chromatography (TLC) separates components of a mixture based on their varying affinities toward the stationary and mobile phases. **Silica gel** was used as the stationary phase, and solvents were used as the mobile phase. Components with **higher affinity for the stationary phase** move slower, while those with **lower affinity** travel faster, resulting in distinct spots on the TLC plate.

➤ Procedure for TLC Analysis

• Stationary Phase:

- ✓ A ready-made TLC plate coated with silica gel was used.
- ✓ A baseline was drawn with a pencil, and a spot was marked for sample application.

• Sample Application:

- ✓ Extracts from *Erythrina variegata* and *Sesbania grandiflora* (fresh, cooked, and preserved) were applied using capillary tubes at marked spots on the TLC plate.

• Mobile Phase Preparation:

- ✓ A solvent mixture was prepared and poured into a TLC chamber. A moistened filter paper was placed inside the chamber to ensure uniform solvent rise.

➤ Development of TLC Plate:

- ✓ The TLC plate was placed upright in the chamber, ensuring that the solvent front did not cross the baseline.
- ✓ The chamber was sealed to prevent solvent evaporation, and the plate was allowed to develop.

➤ Visualization:

- ✓ Once the solvent front reached the desired level, the plate was removed and airdried.
- ✓ The developed spots were visualized under UV light to identify pigment separation.

➤ R_f Value Calculation:

- The **R_f value** (Retention Factor) for each pigment was calculated using the formula:

$R_f = \text{Distance travelled by the substance from reference line (cm)} / \text{Distance travelled by the solvent front from reference line (cm)}$

IV. RESULT

This study involved the comparative chromatographic analysis of chlorophyll pigments in the leaves of *Erythrina variegata* and *Sesbania grandiflora*. Extracts from fresh, shade-dried, and cooked leaves were analyzed using Thin Layer Chromatography (TLC). The R_f values for each pigment were calculated to assess the impact of cooking and preservation methods on chlorophyll stability.

➤ TLC Analysis of Fresh Leaf Extracts

• *Erythrina variegata*

The TLC of fresh *Erythrina variegata* extract revealed **five distinct pigment bands** with the following R_f values:

- ✓ **Xanthophyll** (Yellow) – R_f = 0.35
- ✓ **Phaeophytin a** (Gray-brown) – R_f = 0.56
- ✓ **Phaeophytin b** (Yellow-brown) – R_f = 0.79
- ✓ **Chlorophyll a** (Blue-green) – R_f = 0.69
- ✓ **Chlorophyll b** (Yellow-green) – R_f = 0.41

• *Sesbania grandiflora*

The TLC of fresh *Sesbania grandiflora* extract displayed **six pigment bands** with the following R_f values:

- ✓ **Carotene** (Orange) – R_f = 0.97
- ✓ **Xanthophyll** (Yellow) – R_f = 0.30
- ✓ **Phaeophytin a** (Gray-brown) – R_f = 0.59
- ✓ **Phaeophytin b** (Yellow-brown) – R_f = 0.81
- ✓ **Chlorophyll a** (Blue-green) – R_f = 0.69
- ✓ **Chlorophyll b** (Yellow-green) – R_f = 0.42

➤ TLC Analysis of Shade-Dried Leaf Extracts

• *Erythrina variegata*

The aqueous extract of shade-dried *Erythrina variegata* powder exhibited **two pigment bands**:

- ✓ **Xanthophyll** (Yellow) – R_f = 0.22
- ✓ **Phaeophytin b** (Yellow-brown) – R_f = 0.80

- *Sesbania grandiflora*

The aqueous extract of shade-dried *Sesbania grandiflora* powder revealed **three pigment bands**:

- ✓ **Carotene** (Orange) – Rf = 0.92
- ✓ **Xanthophyll** (Yellow) – Rf = 0.22
- ✓ **Phaeophytin b** (Yellow-brown) – Rf = 0.83

➤ *TLC Analysis of Cooked Leaf Extracts*

- *Erythrina variegata*

The aqueous extract from cooked *Erythrina variegata* leaves produced **three pigment bands**:

- ✓ **Phaeophytin a** (Gray-brown) – Rf = 0.57
- ✓ **Phaeophytin b** (Yellow-brown) – Rf = 0.78
- ✓ **Chlorophyll b** (Yellow-green) – Rf = 0.43

- *Sesbania grandiflora*

The aqueous extract from cooked *Sesbania grandiflora* leaves showed **three pigment bands**:

- ✓ **Phaeophytin a** (Gray-brown) – Rf = 0.58
- ✓ **Phaeophytin b** (Yellow-brown) – Rf = 0.81

- ✓ **Chlorophyll b** (Yellow-green) – Rf = 0.42

V. SUMMARY OF FINDINGS

- **Fresh extracts** of both plants contained a variety of chlorophyll pigments, with *Sesbania grandiflora* showing an additional **carotene band** compared to *Erythrina variegata*.
- **Shade-drying** reduced the number of detectable pigments, with only **two bands** observed for *Erythrina variegata* and **three bands** for *Sesbania grandiflora*.
- **Cooking** further degraded chlorophyll pigments, with **Phaeophytin a and b** becoming the dominant pigments in both samples, indicating **chlorophyll degradation** during thermal treatment.

These results demonstrate that both **shade-drying** and **cooking** affect the stability and composition of chlorophyll pigments in leaves, with cooking leading to the conversion of chlorophyll into **phaeophytins**, and shade-drying reducing pigment diversity. *Sesbania grandiflora* exhibited a higher retention of carotenoids across all treatments compared to *Erythrina variegata*.

Table 1 Solvent Extract of Leaves of sample 1 *Erythrina variegata* and Sample 2 *Sesbania grandiflora*

S NO.	COLOUR OF THE BAND	NAME OF THE PIGMENT	SAMPLE 1 <i>ERYTHRINA VARIEGATA</i>	SAMPLE 2 <i>SESBENIA GRANDIFLORA</i>
			RF VALUE (CM)	RF VALUE (CM)
11.	Yellow brown	Phaeophytin b	0.79	0.81
22.	Blue Green	Chlorophyll a	0.69	0.69
33.	Grey Brown	Phaeophytin a	0.56	0.59
44.	Yellow Green	Chlorophyll b	0.41	0.42
55.	Yellow	Xanthophyll	0.35	0.30
66.	Orange	Carotene	Nil	0.97

Table 2 Aqueous Extract from Shade Dried Powder of Sample 1 *Erythrina variegata* and Sample 2 *Sesbania grandiflora*

S.NO.	COLOUR OF THE BAND	NAME OF THE PIGMENT	SAMPLE 1 <i>ERYTHRINA VARIEGATA</i> RF VALUE (CM)	SAMPLE 2 <i>SESBENIA GRANDIFLORA</i> RF VALUE (CM)
11.	Orange	Carotene	Nil	0.92
22.	Yellow brown	Phaeophytin b	0.80	0.83
33.	Yellow	Xanthophyll	0.22	0.22

Table 3 Aqueous Extract of the Leaves of Sample 1 *Erythrina variegata* and Sample 2 *Sesbania grandiflora*

S.NO.	COLOUR OF THE BAND	NAME OF THE PIGMENT	SAMPLE 1 <i>ERYTHRINA VARIEGATA</i> RF VALUE (CM)	SAMPLE 2 <i>SESBENIA GRANDIFLORA</i> RF VALUE (CM)
11.	Grey brown	Phaeophytin a	0.57	0.58
22.	Yellow brown	Phaeophytin b	0.78	0.81
33.	Yellow green	Chlorophyll b	0.43	0.42

➤ Graphical Representation

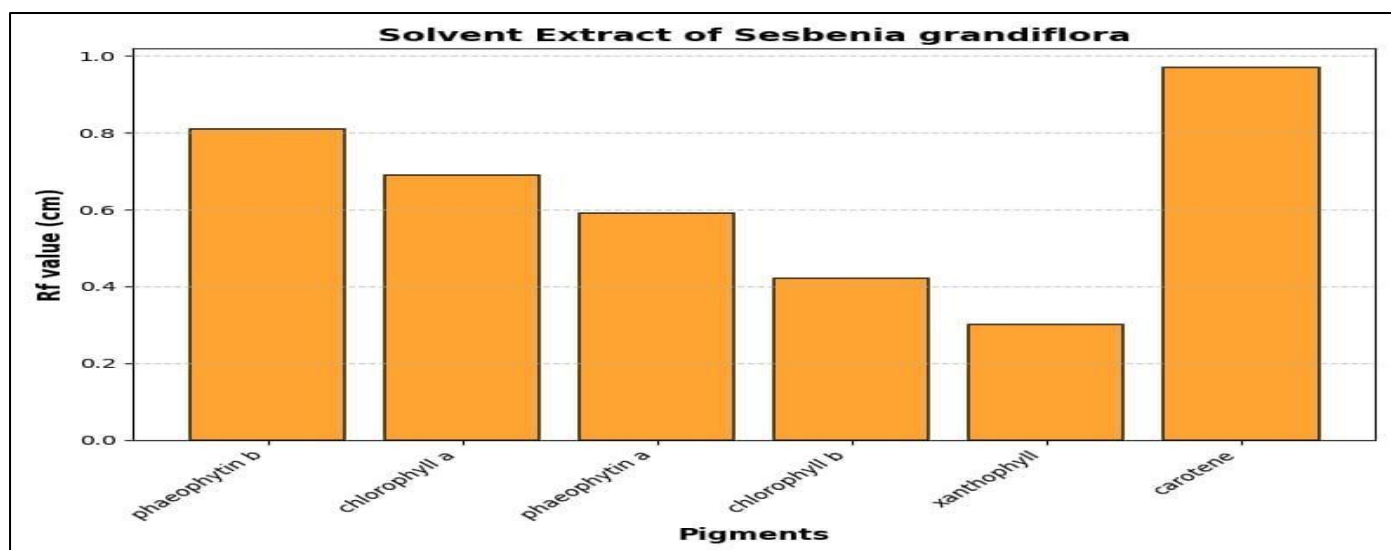


Fig 5 Solvent Extract of *Sesbenia grandiflora*

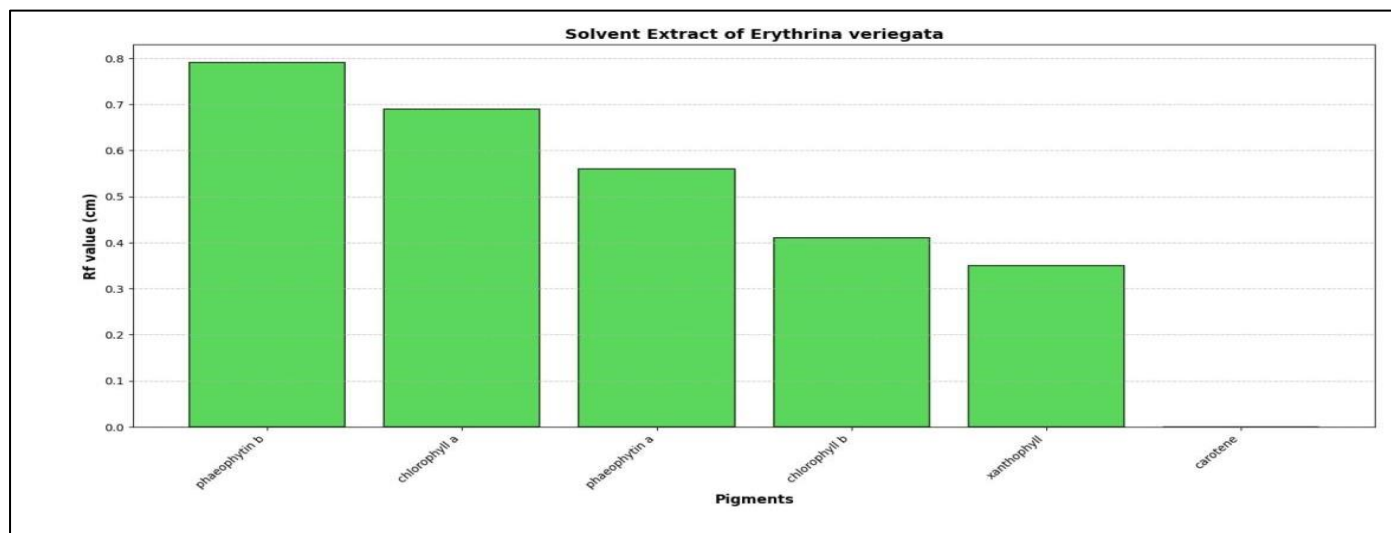


Fig 6 Solvent extract of *Erythrina variegata*

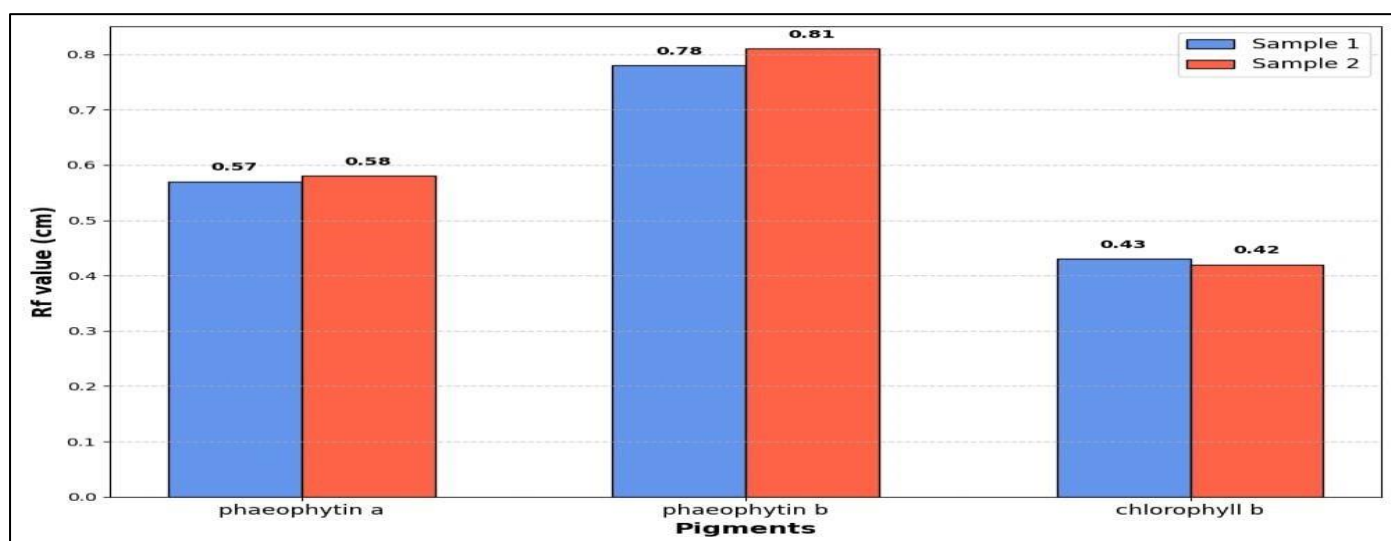


Fig 7 Comparative analysis of pigments in cooked leaves *Erythrina variegata* (sample1) and *Sesbenia grandiflora* (sample 2)

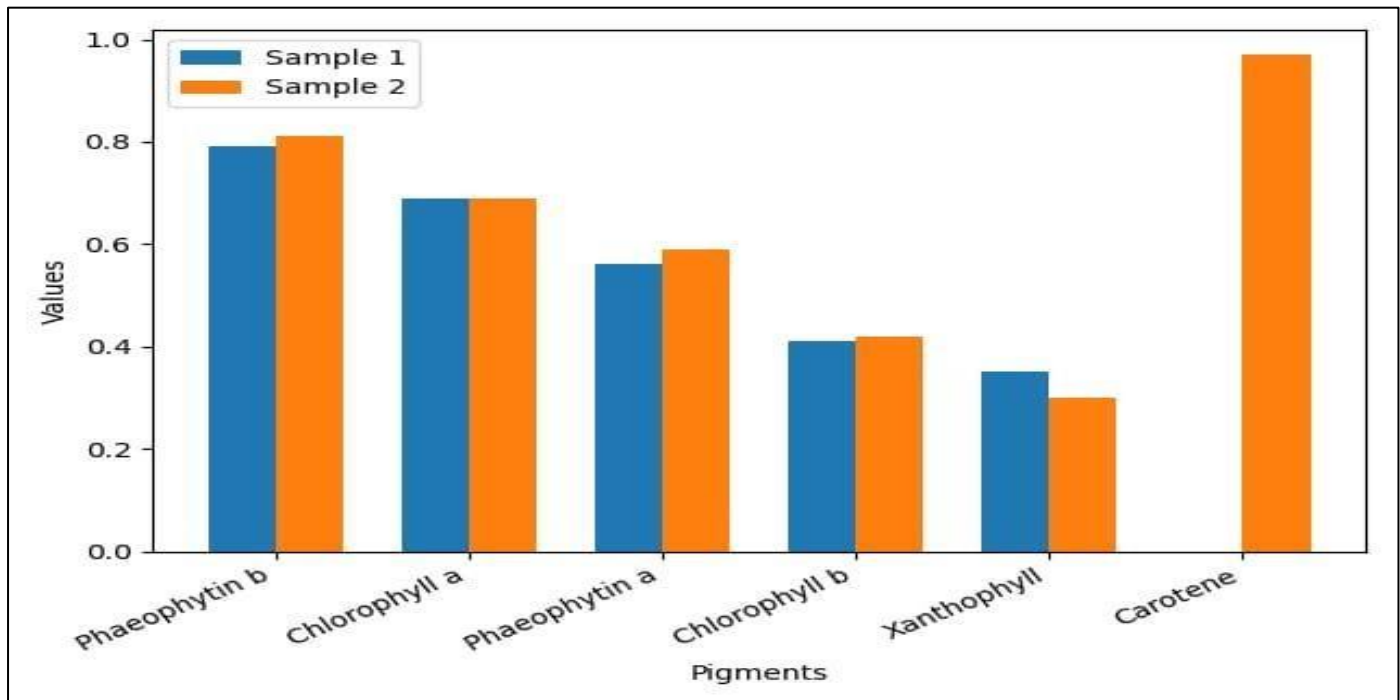


Fig 8 Comparative analysis of chlorophyll pigments in *Erythrina variegata* (sample1) and *Sesbenia grandiflora* (sample 2)

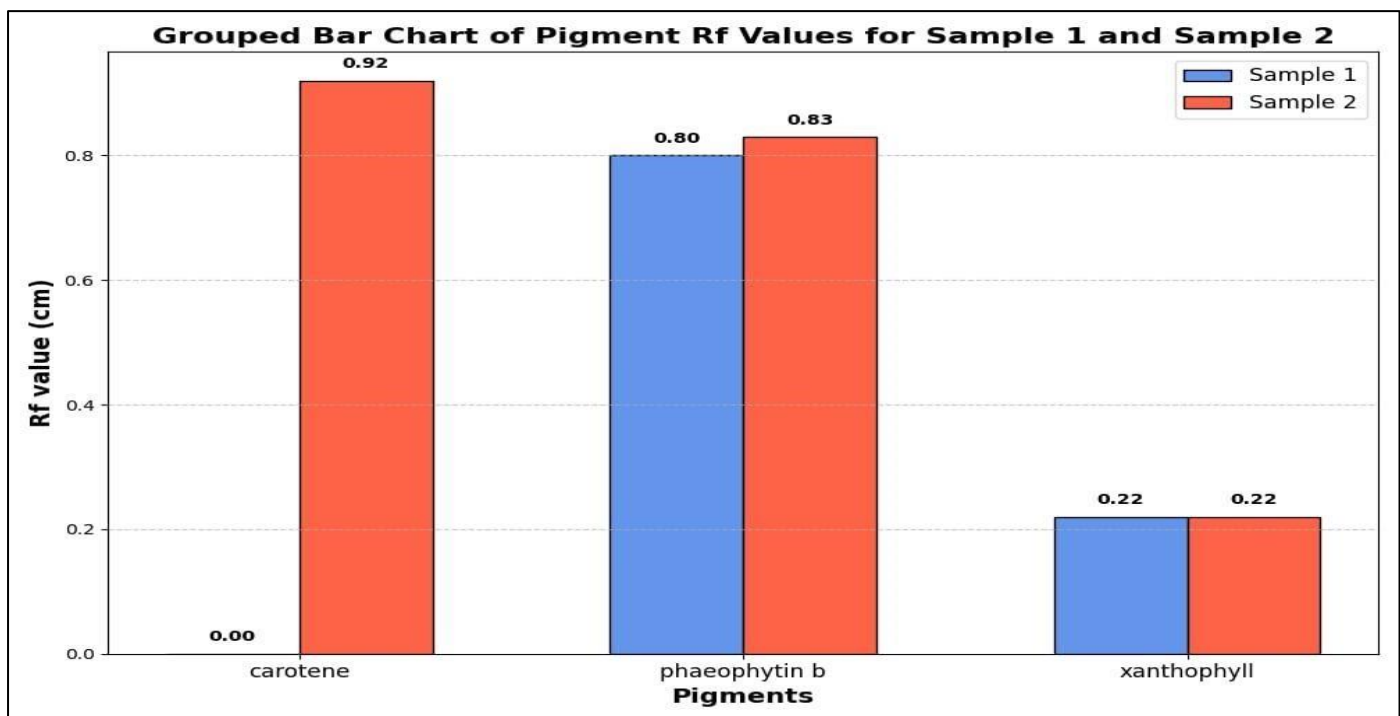


Fig 9 Comparative analysis of pigments in preserved leaves of *Erythrina variegata* (sample1) and *Sesbenia grandiflora* (sample 2)

VI. DISCUSSION

The chromatographic analysis of *Erythrina variegata* and *Sesbania grandiflora* leaf extracts revealed significant differences in pigment profiles across fresh, shade-dried, and cooked samples. The observed **Rf values** closely align with standard ranges, confirming the identity of chlorophyll-related pigments, including **chlorophyll a**, **chlorophyll b**, **phaeophytin a**, **phaeophytin b**, **carotene**, and **xanthophylls**.

Cooking and shade-drying had a notable impact on pigment stability. Chlorophyll pigments were susceptible to degradation under both conditions. In cooked samples, chlorophyll was converted into phaeophytins due to thermal stress, which is consistent with the known effect of magnesium loss and hydrogen replacement in the chlorophyll molecule. Similarly, shade-drying resulted in the loss of chlorophyll pigments, though xanthophylls and carotene were better retained, likely due to their higher resistance to oxidative stress.

Species-specific differences were observed, with *Sesbania grandiflora* showing better carotenoid retention than *Erythrina variegata* across different treatments. This suggests that *Sesbania grandiflora* may have a higher capacity for antioxidant protection, allowing it to preserve pigments better during processing.

The results align with previous studies highlighting the role of temperature, light, and oxidative conditions in influencing pigment retention (Taiz et al., 2015). **Carotenoids and xanthophylls** are more stable under heat and light exposure, while chlorophylls degrade more readily. These findings suggest that cooking and preservation methods significantly alter the nutritional and functional properties of these leaves.

VII. CONCLUSION

This study demonstrates that thermal cooking and shade-drying methods impact chlorophyll content in the leaves of *Erythrina variegata* and *Sesbania grandiflora*. Cooking promotes the formation of phaeophytins, while shade-drying reduces overall pigment diversity, leaving behind more stable pigments such as carotenoids and xanthophylls. These changes are relevant for evaluating the nutritional value of leafy vegetables subjected to common processing methods.

The study also highlights species-specific differences, with *Sesbania grandiflora* showing better retention of carotenoids, indicating its potential for nutritional and therapeutic applications.

Future research could use HPLC or mass spectrometry for more precise quantification of pigments and their degradation products. Additionally, antioxidant activity assays could provide insights into the bioactivity of phaeophytins and other degraded pigments, contributing to our understanding of how cooking and preservation affect the functional properties of plant-based foods.

RECOMMENDATIONS

The synergistic action of bioactive compounds such as chlorophyll and its derivatives plays a critical role in promoting human health. With the increasing prevalence of cardiovascular diseases, atherosclerosis, obesity, and cancer, there is a growing need to adopt dietary patterns rich in bioactive compounds. Chlorophyll pigments, along with their degradation products like phaeophytins, possess significant antioxidant, antimutagenic, antigenotoxic, anticarcinogenic, and anti-obesogenic properties, making them valuable for therapeutic applications.

However, as demonstrated in our study, nutrient loss occurs due to common cooking and preservation techniques, such as boiling and shade-drying. Thus, future research should explore alternative processing methods to minimize nutrient degradation and retain the functional benefits of chlorophyll and related compounds.

➤ *The following Recommendations are:*

- *Development of Optimized Cooking Methods:*

Further studies should investigate low-temperature steaming, blanching, or microwave cooking, which may help reduce chlorophyll degradation while retaining other bioactive compounds.

- *Exploration of Advanced Preservation Techniques:*

Employing techniques like freeze-drying or vacuum drying could help retain chlorophyll pigments and reduce nutrient loss during long-term storage.

- *Nutritional and Antioxidant Profiling of Processed Samples:*

Future research should assess the **antioxidant activity** of leaves subjected to different cooking and preservation methods to determine their biofunctional properties postprocessing.

- *Public Awareness and Culinary Education:*

Disseminating knowledge about the health benefits of minimally processed leafy **greens** could encourage better dietary habits and reduce the nutrient loss commonly associated with cooking practices.

- *Therapeutic Potential of Degraded Pigments:*

Investigating the **bioactivity** of phaeophytins and other degraded chlorophyll compounds could open avenues for novel nutraceuticals and functional foods.

By addressing the nutrient loss through optimized methods and promoting holistic cooking practices, we can harness the full medicinal and therapeutic potential of leafy vegetables such as *Erythrina variegata* and *Sesbania grandiflora*, contributing to improved nutrition and public health.

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