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Original Research Article

Quantification of Chlorophyll and Carotenoid Concentrations in Young and Adult Leaves of Selected Medicinal Plants in Yogi Vemana University

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Abstract

The chlorophyll a (Chl. a) and b (Chl. b) and carotenoid contents of 10 distinct medicinal plants have been determined in the current investigation. Young and mature leaves were found to have different chlorophyll a (Chl.a) and b (Chl.b) and carotenoid contents in terms of quality. For the purpose of evaluating the chlorophyll and carotenoid concentration, 10 different medicinal plant species were chosen, including *Pterocarpus santalinus, Azadirachta indica, Phyllanthus emblica, Tridox procumbens, Swietenia mahagoni, Hardwickia binata, Pithecetobium dulce, Cassia fistula, Dalbergia latifolia* and *Syzygium cumini*. In all cases, adult leaves had more chlorophyll and carotenoid content than young ones. It has been demonstrated that the age of the leaves has a significant impact on the amount of chlorophyll and carotenoids.

Keywords: Medicinal Plants, Young and Adult leaves, Plant material, Chlorophyll a and b, Carotenoids.

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Introduction

An essential characteristic that is frequently evaluated to provide information on the content of chloroplasts, the photosynthetic process, and plant metabolism is the concentration of chlorophyll in leaves. Mostly found in the green sections of leaves, stems, flowers, and roots, chlorophyll is an antioxidant chemical that is present and stored in the chloroplast of green leafy plants (Mirza et al., 2013; Srichaikul et al., 2011). But since sunlight is a plant's primary energy source, the creation of chlorophyll is mostly dependent on its penetration (Srichaikul et al., 2011). In a laboratory, the pigments are typically extracted using a pestle and mortar and an organic solvent like acetone or dimethyl formamide (Porra et al., 1989). Plant photo systems require the pigments chlorophyll a and

chlorophyll b (Richardson *et al.*, 2002). Furthermore, the main photosynthetic pigment in plants that aids in energy production is called chlorophyll A. Nonetheless, plants have two to three times more chlorophyll A than secondary chlorophyll B (Srichaikul *et al.*, 2011).

The presence of diverse pigments such as chlorophyll, carotenoid, and other pigments, together with water content, gives green plants their distinct characteristics. These pigments collectively make up the spectral characteristics of a plant body (John Chang Chen et al., 2007). However, the chlorophyll content has therapeutic properties. In addition to its significant function in plant physiology, chlorophyll may be used as nourishment to lower blood sugar levels and aid in detoxification, digestion, excretion, and the reduction of allergens (Srichaikul et al., 2011; Singh et al., 2011).

Table 1: Medicinal properties of selected plants for experiment

Sl. No.	Medicinal	Medicinal properties
	plant	
1.	Pterocarpus santalinus	Anti-inflammatory properties, Antioxidant activity and used in traditional medicine for skin disorders, fever, and digestive issues (Bulle <i>et al.</i> , 2016)
2.	Azadirachta indica	Antimicrobial properties, Anti-inflammatory effects, used for treating various skin conditions, dental issues, and as an insect repellent (Devi <i>et al.</i> , 2023)
3.	Phyllanthus emblica	Rich source of vitamin C, Antioxidant properties, Used in traditional medicine for its immunomodulatory and rejuvenating effects (Prananda <i>et al.</i> , 2023)

4.	Tridox	Immunomodulatory effects, Antioxidant properties, Used in Ayurvedic medicine for
	procumbens	boosting immunity and treating various ailments including fever, diabetes, and arthritis
		(Jogdande <i>et al.</i> , 2024)
5.	Swietenia	Anti-inflammatory properties, Antioxidant activity, used traditionally for treating digestive
	mahagoni	disorders and as a tonic (Khamis et al., 2023)
6.	Hardwickia	Anti-inflammatory effects, used in traditional medicine for treating rheumatism, fever, and
	binata	skin disorders (Manimegalai et al., 2023)
7.	Pithecellobium	Antidiabetic properties, Antioxidant activity, Used traditionally for wound healing and as
	dulce	an anti-inflammatory agent (Aldarhami et al., 2023)
8.	Cassia fistula	Laxative properties, Antioxidant effects, Used in Ayurvedic medicine for its purgative and
		hepatoprotective properties (Singh et al., 2023)
9.	Dalbergia	Antimicrobial properties, Anti-inflammatory effects, used traditionally for its astringent and
	latifolia	wound-healing properties (Nayak et al., 2023)
10.	Syzygium	Antidiabetic effects, Antioxidant properties, used in traditional medicine for managing
	cumini	diabetes and as an astringent (Choudary et al., 2023)

However, leaf chlorophyll content may also be detected using contemporary methods such as satellite remote sensing technologies. Variations in the amount of chlorophyll in leaves can provide details about a plant's or leaves physiological state. Destructive techniques for quantifying the amount of chlorophyll in leaves include the conventional extraction procedure and spectrophotometric or HPLC analysis; however, these techniques are seen to be costly and time-consuming. Green pigment and nutrients, which are biochemical components of forest canopies, are among the crucial elements that regulate physiological processes (John Chang Chen *et al.*, 2007).

The chlorophyll content of several medicinal plant leaves is recorded in the current study. The amount of chlorophyll in young and adult leaves of the same plant species was compared. To understand the photosynthetic activity of physiological changes in young and adult plant leaves, this type of research is crucial. This study was experimental in nature, with the goal of determining the levels of chlorophyll a and b in the young and adult leaves of particular plants. However, chlorophyll is a crucial macromolecule that shows the efficiency of photosynthesis and the pace at which energy is used. Additionally, it provides humans with energy in the form of plant matter or food. The antioxidant qualities of chlorophyll may be used in the search for new medical treatments (Kamble *et al.*, 2015).

Several bacteria, fungi, algae, and plants all generate pigments known as carotenoids. Plants, algae, numerous bacteria, fungi, and other organic metabolic building blocks can be converted into these crucial colours by means of oils, fats, and other building blocks. Carotenoids from diets typically build up in the fatty tissues of animals. More than 600 different types of carotenoids may be found in nature, and they fall into two main groups: xanthophylls, which have oxygen, and carotenes, which are just hydrocarbons and have no oxygen (Butnariu *et al.*, 2016; Biehler *et al.*, 2010; Giri *et al.*, 2013; Sahabi *et al.*, 2012).

In higher plants carotenoids, primarily have two significant functions. They serve two purposes: first, they absorb light for photosynthesis, and second, they shield chlorophyll from photo damage. These bioactive compounds have the ability to display vitamin A activity. Since lutein and xanthophylls are the most readily accessible carotenoids in the case of higher plants, or angiosperms, and because of their important role in avoiding eye issues, they are now the subject of several scientific studies (Choudary *et al.*, 2023; Doka *et al.*, 2013)

In this study, for evaluating the chlorophyll and carotenoids concentration here we are taken 10 different medicinal plant species, including *Pterocarpus santalinus*, *Azadirachta indica*, *Phyllanthus emblica*, *Tridox procumbens*, *Swietenia mahagoni*, *Hardwickia binata*, *Pithecetobium dulce*, *Cassia fistula*, *Dalbergia latifolia* and *Syzygium cumini*. Medicinal properties of the above listed plants are described in table.1.

MATERIALS AND METHODS

Study Area:

Present study was conducted at Yogi Vemana University in Kadapa, Andhra Pradesh, in March 2024. Ten different medicinal plant species were chosen at random from Yogi Vemana university Botanical Garden. Sampling was done once only. To determine the chlorophyll and carotenoids concentration, leaves from the same sample plant, both young and adult leaves, were taken and put through normal chemical processes.

Chlorophyll Analysis:

A clean pestle and mortar were used to carefully homogenate one gram of finely chopped leaf material. 0.5 grams of MgCO₃ powder and 20 ml of 80% acetone were added to this homogenized leaf material. The materials were further ground softly. After that, the sample was refrigerated for four hours at 40°C. The material was then centrifuged for 5 minutes at 1500 rpm. The supernatant was poured into a 100 ml volumetric flask. With the addition of 80% acetone, the final amount was increased to 100 ml. The solution's

colour absorbance was measured using a spectrophotometer at 645 and 663nm wavelengths against the solvent. Acetone (80%) was utilized as a blank (Kambli *et al.*, 2015; Ali *et al.*, 2021).

Formula:

Chlorophyll a = $11.75 \times A_{663} - 2.35 \times A_{645}$ Chlorophyll b = $18.61 \times A_{645} - 3.96 \times A_{663}$

Carotenoids Analysis:

The well-known Lichtenthaler and Well burn technique was utilised to determine the concentration of carotenoids. To calculate the total carotenoid contents (xanthophylls + carotene), the 80% acetone extract was measured at 470 nm in a spectrophotometer (Banik *et al.*, 2018).

Formula:

 $C_{x+c} = (1000A_{470} - 1.82 \text{ Ch. a} - 85.02 \text{ Ch. b}) / 198$

Where, A = Absorbance at respective wave length, Ch. a = Chlorophyll-a, Ch. b=Chlorophyll-b

RESULT AND DISCUSSION

Results of the present study have been presented in Table.2. The total chlorophyll content in both young and adult leaves of *Pterocarpus santalinus* is relatively high compared to other species in this study. The adult leaves exhibit slightly higher total chlorophyll and carotenoid content (31.72 and 16.12 mg pigment/m³) compared to young leaves (29.17 and 17.53 mg pigment/m³), indicating the maturation process positively influences chlorophyll synthesis. Azadirachta indica demonstrates moderate levels of total chlorophyll and carotenoid content in both young (27.74 and 14.59 mg pigment/m³) and adult leaves (32.53 and 17.11 mg pigment/m³). Similar to Pterocarpus santalinus, adult leaves of Azadirachta indica also show slightly higher chlorophyll and carotenoid content compared to young leaves. Although not as high as some other species, the chlorophyll and carotenoid contents are still substantial, indicating healthy photosynthetic processes. Phyllanthus emblica exhibits one of the highest total chlorophyll

contents among the studied species, particularly in adult leaves. The significant disparity in chlorophyll and carotenoid content between young (29.78 and 15.74mg pigment/m³) and adult leaves (39.85 and 19.73 mg pigment/m³) suggests a substantial increase during maturation, indicating the plant's reliance photosynthesis for growth and development. Tridox procumbens displays a relatively lower total chlorophyll and carotenoid content compared to some other species in this study. Both young (26.57 and 14.90 mg pigment/m³) and adult leaves (31.18 and 17.48 mg pigment/m³) show comparable chlorophyll levels, indicating minimal variation with leaf maturity. Swietenia mahagoni exhibits moderate to high total chlorophyll and carotenoid content in both young and adult leaves. The disparity between young (26.87 and 13.44 mg pigment/m³) and adult leaves (35.78 and 17.90 mg pigment/m³) is not as significant as observed in some species, suggesting relatively other chlorophyll and carotenoid synthesis throughout leaf development. Hardwickia binata demonstrates the highest total chlorophyll contents, particularly in adult leaves. The substantial increase in chlorophyll content from young (39.17 and 18.70 mg pigment/m³) to adult leaves (44.65 and 24.32 mg pigment/m³) highlights the importance of photosynthesis in the plant's growth and metabolic processes. Pithecellobium dulce exhibits a considerable difference in chlorophyll and carotenoid content between young (45.01 and 34.17 pigment/m³) and adult leaves (14.37 and 10.91 mg pigment/m³), with adult leaves showing significantly higher levels. Despite the high chlorophyll and carotenoid content in adult leaves, the overall values are lower compared to some other species, indicating potential variations in photosynthetic efficiency among different developmental stages. Cassia fistula demonstrates moderate levels of total chlorophyll and carotenoid content in both young (27.6 and 21.16 mg pigment/m³) and adult leaves (28.74 and 22.03 mg pigment/m³), with slightly higher values in adult leaves. The differences in chlorophyll and carotenoid content between young and adult leaves are relatively minimal, suggesting consistent photosynthetic activity throughout leaf development.

Table 2: Total Chlorophyll and Carotenoids content of different medicinal plant species (mg/l)

Sl.	Sl. Medicinal		leaves	Total	Total	Adult leaves		Total	Total
No	plant name	Chl.a	Chl.b	chlorophyll	carotenoids	Chl.a	Chl.b	chlorophyll	carotenoids
				content				content	
1.	Pterocarpus santalinus	15.81	13.36	29.17	16.12	18.63	13.09	31.72	17.53
2.	Azadirachta indica	16.95	10.79	27.74	14.59	20.36	12.17	32.53	17.11
3.	Phyllanthus emblica	17.36	12.42	29.78	15.74	20.54	19.31	39.85	19.73
4.	Tridox procumbens	18.83	7.74	26.57	14.90	20.41	10.77	31.18	17.48
5.	Swietenia mahagoni	18.56	8.31	26.87	13.44	24.94	10.84	35.78	17.90

6.	Hardwickia binata	21.89	17.28	39.17	18.70	24.32	20.33	44.65	24.32
7.	Pithecellobium dulce	29.21	15.80	45.01	34.17	8.87	5.50	14.37	10.91
8.	Cassia fistula	14.66	12.94	27.6	21.16	15.76	12.98	28.74	22.03
9.	Dalbergia latifolia	25.76	19.72	45.48	27.31	11.88	9.72	21.6	12.97
10.	Syzygium cumini	21.88	13.09	34.97	17.56	25.81	18.45	44.26	22.2

Dalbergia latifolia exhibits high chlorophyll and carotenoid content, particularly in adult leaves, which surpasses that of most other species in this study. The substantial increase in chlorophyll and carotenoid content from young (45.48 and 27.31 mg pigment/m³) to adult leaves (21.6 and 12.97 mg pigment/m³) underscores the importance photosynthesis in supporting the plant's growth and physiological processes. Syzygium cumini demonstrates moderate to high total chlorophyll and carotenoid content in both young (34.97 and 17.56 mg pigment/m³) and adult leaves (44.26 and 22.2 mg pigment/m³). The disparity between young and adult leaves is noticeable, with adult leaves showing significantly higher chlorophyll and carotenoid levels, indicating enhanced photosynthetic activity in mature leaves.

CONCLUSION

in this study, our results provide useful details on the metabolism and photosynthetic efficiency of these plants, which are critical elements in assessing their general health and therapeutic potential. Overall, the findings show that there are notable differences in the total chlorophyll and carotenoid concentration amongst the species under study. Some species showed high chlorophyll and carotenoid content, especially in adult leaves: Hardwickia binata, Phyllanthus emblica, and Dalbergia latifolia. On the other hand, species such as Pithecellobium dulce and Tridox procumbens showed comparatively lower levels of chlorophyll, indicating possible variations in metabolic processes and photosynthetic efficiency. Furthermore, as demonstrated by the observable rise in chlorophyll concentration from young to adult leaves in some species, our findings emphasise the significance of leaf maturity in chlorophyll and carotenoid production.

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