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Morphological, anatomical, and preliminary phytochemical characterization of Olax psittacorum (Willd.) Vahl. A Wild leafy Vegetable

Jayendra G, Nakade[1]*, Praveenkumar N. Nasare[2]

[1]Research Scholar IHLR & SS Nilkanthrao Shinde Science & Arts College Bhadrawati Dist-Chandrapur Maharashtra, India

[2] Department of Botany, Nilkanthrao Shinde science & arts College Bhadrawati Dist-Chandrapur Maharashtra, India

[1]Department of Botany Bhawabhuti Mahavidyalaya, Amgaon. Dist. Gondia Maharashtra, India 441902

Corresponding Author: Email: jayendranakade26@gmail.com

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ABSTRACT

Olax psittacorum (willd.) Vahl.is a perennial shrubby climber, distributed all over the tropical and subtropical region. This study focused on the morphological, anatomical, and phytochemical characteristics of *Olax psittacorum*. The findings revealed that the plant typically grows to a height of 120 to 200 mostly erect and climbing in nature with white flowers having axillary inflorescence. Leaves showed a well-developed epidermis with unicellular trichomes. Mesophyll tissue is filled with oils & globules, crystals of calcium oxalate& chlorenchymatous tissue. Stem and root consist of open and bicollateral endarch vascular bundle. Phytochemical analysis of leaves crude powder extract shows the presence of carbohydrates, proteins. fats, alkaloids, flavonoids, tannins & resin were present. The result indicates that the leaves are rich in primary and secondary metabolites, Therefore, it is recommended as a plant for its wide use as a wild leafy vegetable and phytopharmaceutical important

INTRODUCTION

Plants have been a source of food for human beings from the start of the evolution of mankind. Besides food, it is a source of medicine used to treat various diseases. Plants have been investigated for their chemical moieties and nutraceutical values. Leafy vegetables are a natural source of antioxidants and are rich in phytochemicals (Mibei et al., 2012). Plants naturally possess primary and secondary metabolites, along with essential minerals such as calcium (Ca), iron (Fe), zinc (Zn), magnesium (Mg), copper (Cu), and manganese (Mn). They also contain nutrients like carbohydrates, proteins, fats, and crude fiber (Raghavendra et al., 2013; Sree & Author, 2018). According to the 2011 census, the tribal population in India constitutes 8.6% of the total population. Tribal communities in India live close to nature and possess hereditary traditional knowledge of consuming wild plants (Niveditha, 2017). Wild edible plants play a significant role in fulfilling the nutritional requirements of tribal populations. Various studies have shown that wild edible plants are a potential source of nutrition and, in many cases, are more nutritious than conventionally cultivated crops (Grivetti & Ogle, 2000). Pharmacological and phytochemical research efforts are underway to extract and characterize plant bioactive compounds, such as alkaloids, tannins, flavonoids, resins, phenols, fatty acids, and steroids. These compounds play a crucial role in human health and are nutritionally significant, contributing to developing new pharmaceutical drugs with high activity profiles.

(Alam & Us Saqib, 2015) Plant ingredients are used as raw material for drug formation therefore it is essential to do anatomical and phytochemical analysis to detect the adulteration (Pandiyan & Ilango, 2022)*Olax psittacorum (family Olacaceae) is a shrub commonly found across tropical regions of India. Traditionally, it has been used in folk medicine to treat ailments such as fever, constipation, cough, headaches, and mouth ulcers*. Diarrhoea, anaemia, filaria, joint pains, intestine, and liver diseases(Zareen et al., 2023, Naidu et al., 2016.) In this context, the plant *Olax psittacorum* was collected from the Gadchiroli District of Maharashtra state, India. It belongs to the Vidarbha region. It is widely and popularly consumed by all people and sold in the local market also This wild vegetable is an economic source for Adivasi people in Gadchiroli District and adjoining areas. Thus, this study investigates the macroscopic, microscopic, and phytochemical characteristics of *Olax psittacorum*. The findings may aid in the taxonomic identification of the plant and help confirm its suitability as an edible vegetable.

Material and Methods

Collection of plant

The fresh and healthy *Olax Psittacorum* (Willd.) Vahl. were collected from the village of Shirpur, Tah. Kurkheda. Dist. Gadchiroli (M.S.), India, and the plant was identified with the help of available literature such as flora of Maharashtra (Shing and Karthikeyan 2000), Flora of Nagpur (Ugemuge 1986), Flora of Kolhapur (Yadav and Sardesai 2002), etc. The leaves of the plant were shade-dried at room temperature for 10 to 15 days and

then ground into coarse powder using a grinder. The dried plant powder was stored in paper bags for phytochemical analysis.

Morphological Study

The morphological characteristics of plant specimens were studied in natural habitats as well as in laboratory conditions, which include plant height, leaf length, leaf breadth, leaf arrangement, inflorescence, flower and fruit size, etc.

Anatomical study

The fresh plant material was preserved in a solution of alcoholic acetic acid (50% alcohol, 40% formalin, and 2% acetic acid). The preserved material was then subjected to anatomical studies. The transverse sections were dehydrated using 30%, 50%, 70%, and 90% ethanol solutions. For double staining, safranin and fast green were used. The double-stained sections were then mounted on slides using DPX as the mounting medium

Phytochemical Analysis

One hundred grams of *Olax psittacorum* leaf powder were soaked in different solvents—water, ethanol, methanol, acetone, and chloroform—for 48 hours at room temperature. After soaking, the mixtures were refluxed for 2 hours at room temperature, then cooled and filtered. The extracted solution from each solvent was transferred to a 100 mL beaker, and the solvents were evaporated using a water bath until the extracts were reduced to dryness. The resulting dry extracts were stored in airtight containers and kept in a refrigerator until they were ready for phytochemical analysis. The following standard tests were conducted to detect various phytochemicals: alkaloids (Mayer's and Wagner's tests), flavonoids (ferric chloride and alkaline reagent tests), phenols (ferric chloride test), coumarins, glycosides (Keller-Killani test), phytosterols, saponins (froth test), resins, tannins, terpenoids (Salkowski test), and steroids (Liebermann-Burchard's test). The presence or absence of each phytoconstituent was determined based on color changes, with a (+) indicating presence and a (-) indicating absence (Harborne, 1984).

Result and Discussion

Olax psittacorum, commonly known as Hartfari, belongs to the Olacaceae family. The plant was examined through morphological, anatomical, and preliminary phytochemical characterization in the present study. The findings of the study are as follows:

Morphological Characterization

Olax psittacorum (Wild.) Vahl, Enum.34.1804; Moorthy in Singh et al., Fl. Maharashtra St.Dicot. 1:515. 2000.*Fisslia psittacorum* Wild., Sp. Pl.1:194. 1797.Olax *scandens* Roxb., Corom.2:2, t.102.1798; Mast. In Hook. f., Brit.1:575.1875; Cooke, Fl.Pres.Bombay 1:234.1958(Repr.). **Olacaceae**

Common Name: - Parrot Olax, Hindi:किटहरी, Marathi: haratfari, harduli

Systematic Position

Kingdom-Plantae
Phylum- Tracheophyta
Class-Magnoliopsida
Order-Santalales
Family- Olacaceae
Genus- *Olax psittacorum* (Willd.) Vahl.
Fls. & Frts.: - December -June
Locality: -Shirpur (Kurkheda)

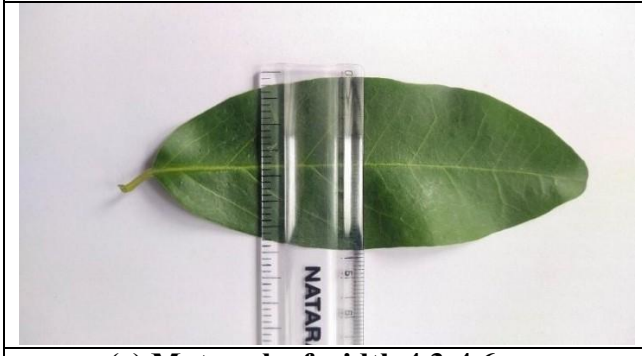
Sr.	Part	Characters
1	Habit	Shrubby, Climbing, Much Branched, Perennial,
2	Habitat	Wild, dry deciduous to moist deciduous forests
3	Plant hight	4- 5 meter
4.	Stem	Woody, climbing, old wood has stout prickles, soft, yellowish white and porous
5	Leaf	Simple, petiolate, alternate, glabrous, shiny, pale yellowish (young), oblong-elliptic, rounded at base, entire,
6	Leaf blade length	11-12 cm
7	Leaf blade width	4.3-4.6 cm
8	Inflorescence	Axillary racemes
9	Flower:	Flowers white, fragrant about 6 mm, in axillary racemes, shorter than leaves. Calyx cup-Shaped. Petals white. Stamen 8-10,3-perfect, remaining staminodes. Ovary ovoid, style shorts; Stigma 3-lobed.
10	Fruit	Fruits is drupe, sub globus, fleshy yellow or orange when ripe, more than half enclosed by accrescent calyx.
11	Fruit length	0.7 cm
12	Fruit diameter	0.9 cm
13	Seeds	One seeded.
15	Occurrence	Along edges of the forest, roadsides, near streams, and damp shady forest
16	Conservation status	IUCN Red List category
17	Traditional and Medicinal uses	Tender leaves and shoots are plucked and boiled and cooked as a vegetable(Deshpande, 2013) In Ayurvedic medicine, the bark is used in anaemia and as a supporting drug in diabetes; also, in the treatment of fever.

Figure 1. (a) Whole plant

(b) Phyllotaxy

(c) Mature leaf length 11-12 cm

(d) Young Leaf length 5.9 cm



(e) Mature leaf width 4.3-4.6 cm

(f)Young leaf width 2.5 cm

(g) Fruit dimeter 0.9 cm

(h) Fruit length 0.7 cm.

	
(i)Flower (6mm)	**(j) Gynoecium (stigma 3 lobed)**

Anatomical characterization

The anatomical study was useful for the authentication of plants based on tissue arrangement. Anatomical studies provide reliable and additional pieces of evidence for the taxonomic delimitations (Bhattacharya et al.,2015)

T.S. of leaves

T. S. of leave of *Olax psittacorum* consists of upper and lower epidermis covered with a thin layer of cuticle both epidermises are covered with unicellular trichomes. The epidermis was a single-layered compactly arranged barrel-shaped epidermal cell. A greater number of trichomes were present in the lower epidermis than in the upper epidermis. Stomata were confined to only at lower epidermis

Epidermis followed by mesophyll tissue differentiates with upper palisade tissue and lower spongy parenchyma. Mesophyll tissue differentiates about 1-2 layers of compactly arranged parenchymatous tissue which consists of oils globule, chloroplast pigment, and in some tissues rosette crystals of calcium oxalate were present. The lower epidermis was made up of 2–3-layer spongy parenchyma cells, loosely arranged with air space. The centrally positioned vascular bundle in the midrib area was open and bi-collateral in shape. Xylem tissue encircled the phloem. The ring-like structure of the phloem is made up of phloem fibers and sieve components. The protoxylem faces the top epidermis, while the metaxylem faces the lower epidermis. Thin uniseriate medullary rays, xylem parenchyma, and fibers separated the xylem bundle **Fig.2 (a,b)**

T.S. of petiole

T.S. of the petiole of *Olax psittacorum* is half-moon shaped and made up of upper and lower epidermis both the epidermis were single layers of compactly arranged barrel shape tissue, filled with yellow substance surrounded by a thick layer of cuticle and unicellular horn shaped trichome. Epidermis followed by 4-6 layers of cortex. which is made up of loosely arranged parenchymatous cells filled with crystals of calcium oxalate and some cells filled with oil globules, tannins, and resins. The endodermis, which is composed of long, thin-walled cells, is located inside the cortex. 4-6 layers of pericyclic fibers encircle the vascular bundle, creating a ring-like shape. The vascular bundle is bicollateral and open. With certain phloem fibers and a sieve element creating a ring-like structure, the phloem is encircled by a xylem. The protoxylem faces the top epidermis, while the metaxylem confronts the lower epidermis. Uniserrate medullary rays divided the xylem bundle.**Fig.3. (c, d)**

T.S. of Stem

The transverse section of the stem was somewhat circular in outline. which consists outermost layer of the epidermis made up of compactly arranged parenchymatous cells. Epidermis densely covered with hair-like horn-shaped trichomes. The cortex is differentiated into upper cortex and lower cortex. The upper cortex is made up of three to four layers of chlorenchymatous cells consisting of chloroplast pigment. The lower cortex is made up of four to six layers densely arranged with parenchymatous cells filled with prismatic crystals of calcium oxalate, rosette crystals, and some tannin. Endodermis is inner to the cortex with two to three layers of somewhat elongated cell thin-walled cells forming endodermis which covers the vascular bundle.

Vascular bundles had an endarch state, were open, bicollateral, and organized in a ring. The metaxylem faces the cortex, while the protoxylem faces the ground tissue. The number of medullary rays varied between 20 and 30. In the cortical region, phloem was found in a cluster. Parenchymatous pith occupied the central area. **Fig.4. (e, f)**

T.S. of root

The bark on the outside of the transverse section of the root was covered in cork (phellem) and cork cambium (phellogen). There was a secondary cortex (phelloderm) within the cork cambium, which was roughly 6–8 layers thick. Secondary vascular tissue covers the cortical area. Bicollateral, open vascular bundles with an endarch state were observed. The metaxylem faces the cortex, while the protoxylem faces the ground tissue. The number of medullary rays varied between 20 and 30. The phloem surrounds the xylem, forming a ring-like structure containing a few phloem fibers and a sieve element. There was a phloem in the shape of a cluster. Parenchymatous pith occupied the central area.**Fig.5. (g, h)**

Photo plate - 1

Anatomical characterization

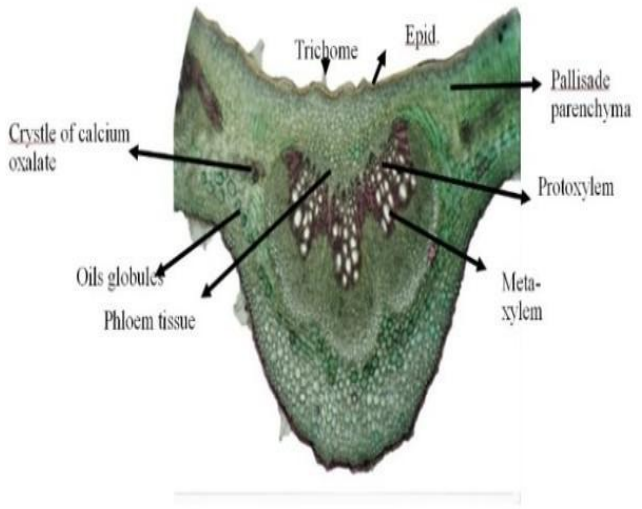
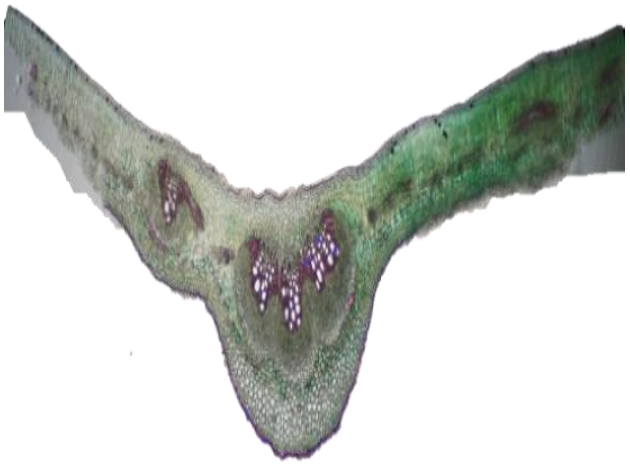


Fig. 2 (a) T. S. of leave through midrib



(b). T. S. of Leaves With lamina showing Prismatic Crystal

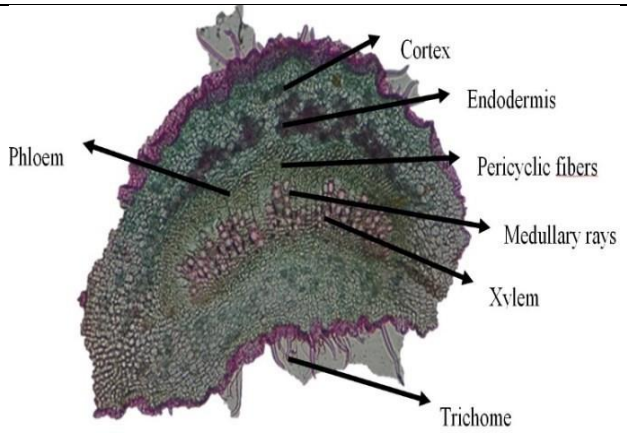
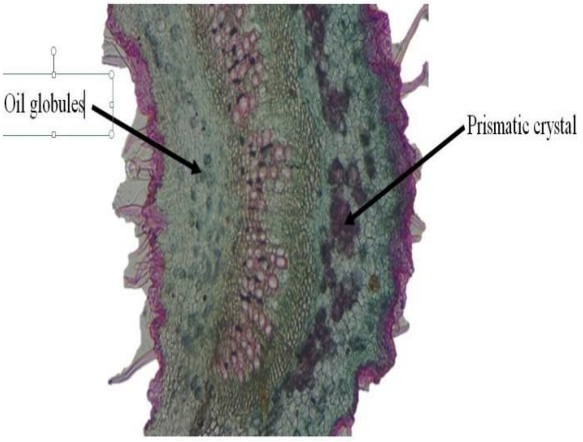


Fig.3. (c) T. S. of Petiole



(d) T. S. of Petiole showing Oil globules

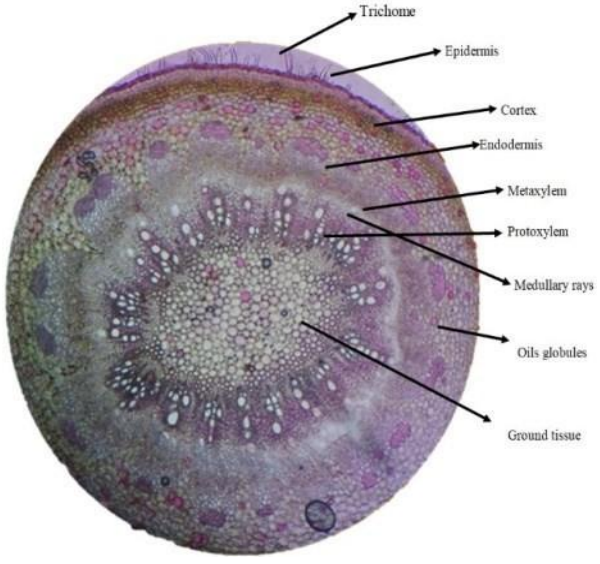
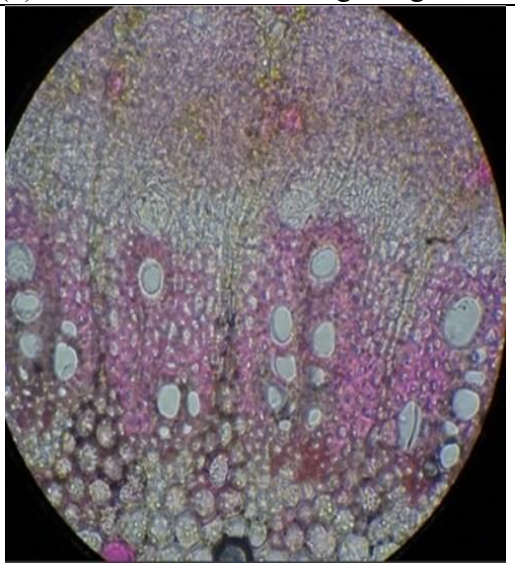


Fig.4. (e) T. S. of stem



(f) Enlarge view showing Medullary rays & Vascular bundle

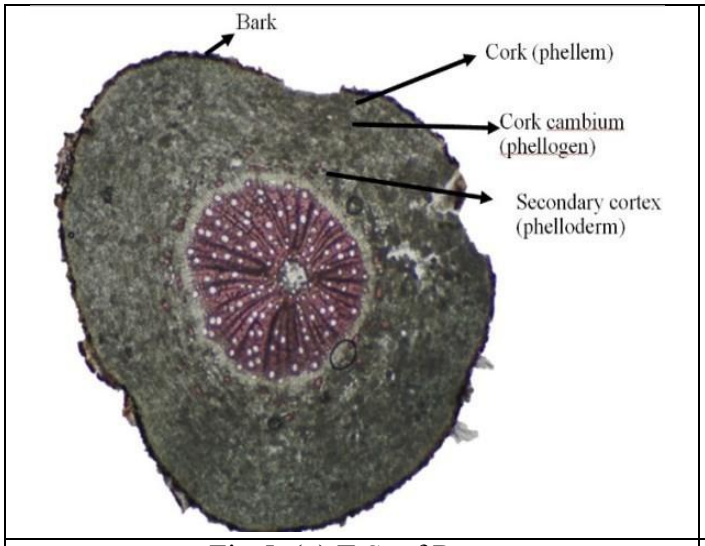
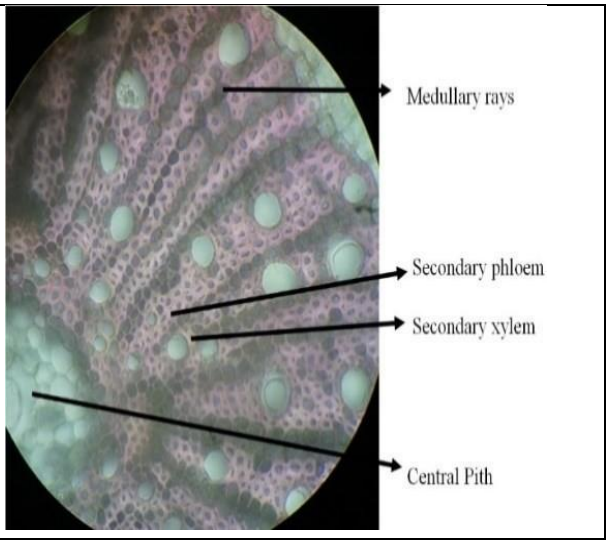


Fig.5. (g) T.S. of Root



(h). T.S. of root showing vascular bundle.

Phytochemical Analysis

The powder analysis and colour of different extracts of the crude drug of *Olax psittacorum* were studied with different chemical reagents and showed the presence of tannin, starch, oil, xanthoprotein & alkaloids **(Table 1 & 2)**

The preliminary phytochemical screening of leaf power of *Olax psittacorum* was studied in different extracts such as water, Ethanol, Methanol, Acetone & chloroform, and showed the presence of various phytoconstituents like carbohydrates, proteins, amino acids, resins, oils & fats, anthraquinone, alkaloids, flavonoids, tannins, anthocyanin, etc. **(Table 3. & 4.)** Some phytoconstituents such as Gams & Mucilage, Carboxylic acid, terpenoids Phlobatannins were absent in all extracts of leaves of *Olax psittacorum*. The Ethanolic extract showed positive for carbohydrates, protein, rensin, oils & fats, cardiac glycosides, tannin, flavonoids, phytosterol, steroids, anthocyanin & coumarin. The methanolic extract showed positive for protein, resins, oils & fats, alkaloids, cardiac glycoside, tannins flavonoids, phytosterol, and anthocyanin. The acetone extract yielded positive for carbohydrates, starch, protein, amino acid, resin, Oils & fats, anthocyanin, tannin flavonoids, phytosterols, cardiac glycosides, tannin, flavonoids, phytosterol & chalcone. In chloroform extract, very few phytoconstituents were reported. The results showed that the ethanol, methanol & acetone are suitable for the extraction of phytochemical in this extract most of the primary & secondary metabolites shows positive results. Chloroform is a not suitable solvent for extraction of phytoconstituents in *Olax psittacorum*. The results show alkaloids, tannins, flavonoids, anthocyanin, coumarin & phytosterol were reported to be dominant chemical constituents in ethanol, methanol & acetone extract.

***Olax psittacorum* Leaves Powder analysis with different chemical reagents**

Table No. 1

Sr.	Reagent	Colour/behavior	inference
1	Powder as such	Olive green	
2	Powder+5%fecl3	Sepia brown	Tannins present
3	Powder +5 % iodine	Raw umber brown	Starch present
4	Powder + Sudan	Lion brown	Oil is present
5	Powder + conc. HNO3+ammonia	Fulvous brown	Xanthoprotein present
6	Powder+40%NaOH+lead acetate	Buff brown	Cysteine absent
7	Powder + picric acid	Gold Yellow	Alkaloids present

Colour of crude in various extract

Table 2

Extract	Colour
Water	Light green
Ethanol	Olive green
Methanol	Dark green
Acetone	Green
Chloroform	Olive green

Qualitative Analysis of Primary Metabolites

Table 3

sr.no	Phytoconstituents	Water Extract	Ethanol Extract	Methanol Extract	Acetone Extract	Chloroform Extract
1	Carbohydrate	+	+	-	+	-
2	Starch	+	-	-	+	-
3	Protein	+	++	++	++	-
4	Amino acid	+	-	-	+	+
5	Fatty acid	-	-	-	-	-
6	Resins	-	+	+	+	-
7	Oils & Fats	+	+	+	+	-
8	Gams & Mucilage	-	-	-	-	-
9	Carboxylic acid	-	-	-	-	-

Qualitative analysis of Secondary Metabolites from *Olax psittacorum*

sr.no	Phytoconstituents	Water Extract	Ethanol Extract	Methanol Extract	Acetone Extract	Chloroform Extract
1	Anthraquinones	++	-	-	+	-
2	Quinone	+	-	-	+	+
3	Alkaloids	+	-	+	-	+
4	Glycoside	-	-	-	+	-
5	Cardiac glycoside	-	+	+	+	-
6	Tannins	+	+	+	+	-
7	Flavonoids	+	++	++	++	-
8	phytosterol	-	+	+	+	-
9	Phlobatannins	-	-	-	-	-
10	steroids	-	+	+	-	-
11	Chalcones	-	-	-	+	-
12	terpenoids	-	-	-	-	-
13	Triterpenoids	-	-	-	-	-
14	Anthocyanins	+	+	+	+	-
15	Coumarins	+	+	-	+	-
16	Emodins	-	-	-	+	-

CONCLUSION

Olax psittacorum is a wild leafy vegetable that has been studied for its Morphological anatomical and preliminary phytochemical characteristics. Here is a conclusion based on this aspect.
Morphological characterization It is a thorny climbing shrub with all its parts being actively used for medicinal purposes. Leaves extensively used as wild leafy vegetables are small elliptical, and oblong. flower small axillary inflorescence with white colour, present along roadside leaves particularly young evergreen leaves used as vegetables & consumed throughout the year. Anatomical Characterization of *Olax psittacorum* has provided insights into to internal structure of plants, showing a well-developed epidermis with unicellular trichomes. Leaves well differentiated into palisade & spongy parenchyma, crystals of calcium oxalate. Stem & Root with bicollateral type of vascular bundle. In preliminary phytochemical analysis of the crude drug of *Olax psittacorum* shows the presence of various phytoconstituents. Consequently, these studies will be helpful in the future to highlight the significance of plants,

Conflict of interest statement.

We declare that there are no financial, personal, or professional conflicts of interest related to this work,

Ethical Approvals

In this study, no experimental trial was performed on Animal & human subjects

Funding: - No Funding received

Originality: We declare that this work is original, has not been published elsewhere.

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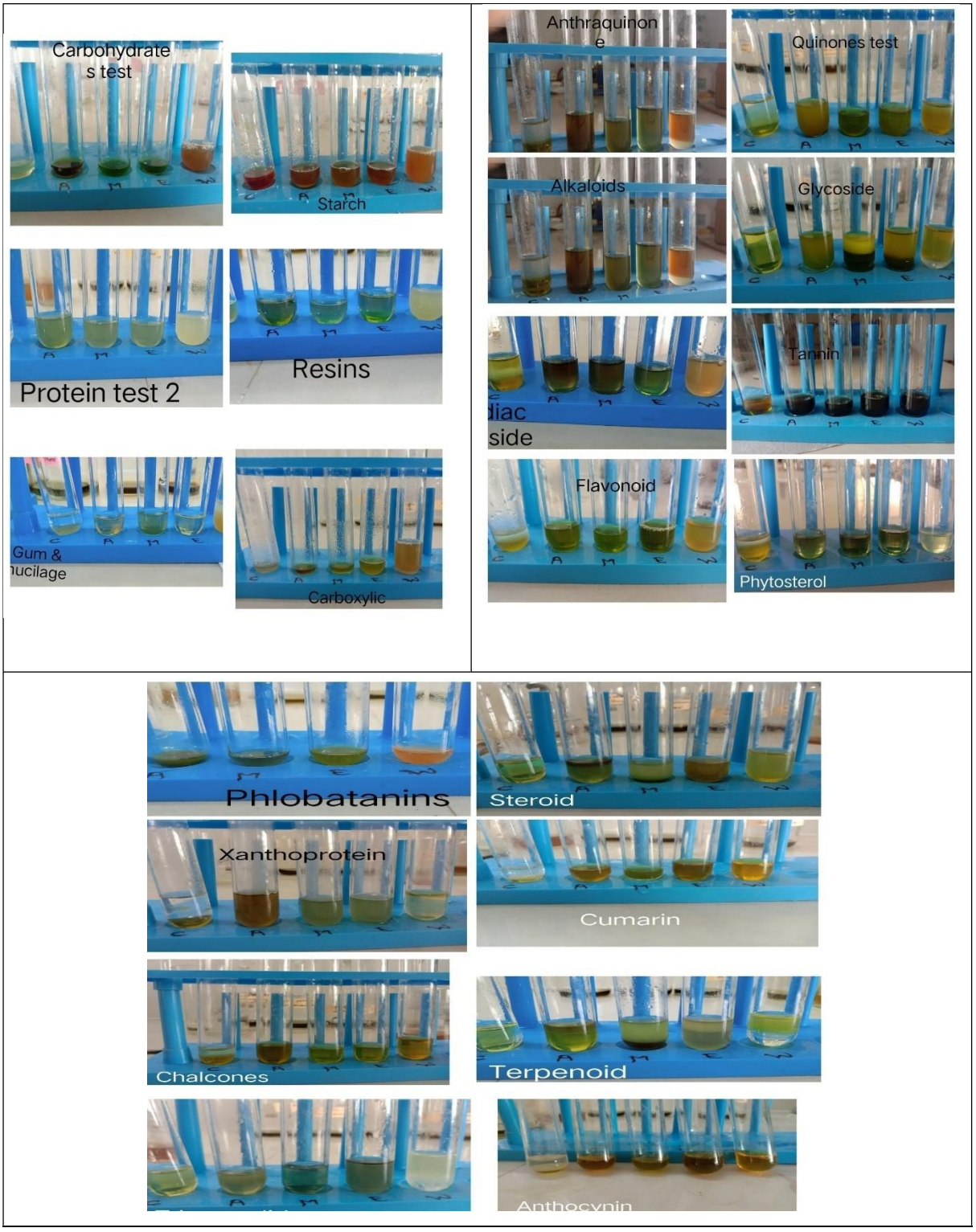

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