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Research Article

Pharmacognostical and Physiochemical Study on the Leaves of *Nyctanthes arbor-tristis* Linn

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Abstract

Aim: To rationalize the macroscopical, microscopical and physico-chemical studies on leaves of plant *Nyctanthes arbor-tristis* Linn. **Methods:** The pharmacognostic characters were determined in terms of macroscopy, microscopy and powder microscopy of plant *Nyctanthes arbor-tristis* leaves. The crude ethanolic extract of leaves of *Nyctanthes arbor-tristis* Linn. was using physico-chemical parameters, and preliminary phytochemical investigation (TLC). **Results:** The microscopic study shows the general characteristic of *Nyctanthes arbor-tristis* leaves. Phytochemical analysis revealed the presence of various phytochemical groups like alkaloids, glycosides, steroids, phenolic, tannins constituents. **Conclusion:** It can be concluded that the established pharmacognostic profile of *Nyctanthes arbor-tristis* leaves will be helpful in developing pharmacopoeial standards for correct identification and quality control. The present observation will also be helpful in macroscopical and microscopical on leaves of *Nyctanthes arbor-tristis* Linn.

Keywords: *Nyctanthes arbor-tristis*, Pharmacognostical Study, Macroscopical and Microscopical studies

1. INTRODUCTION:

Nyctanthes arbor-tristis L. (Oleaceae) is an essential medicinal plant that has been used for a variety of purposes since antiquity. Various parts of this plant have been used as traditional and local medicines. *Nyctanthes arbor-tristis* is used as a diuretic, digestive, laxative, anti-venom, mild bitter tonic, and expectorant in the Siddhya-Ayurveda, Ayurveda, and Yunani systems of medicine. ^{1,2} *Nyctanthes arbor-tristis* Linn. (Division: Magnoliophyta; Class: Magnoliopsida; Order: Lamiales; Family: Oleaceae), commonly known as Harshingar and Night jasmine ³, is a well-documented plant. It is a terrestrial woody perennial plant with a lifespan of 5–20 years that grows up to 10 meters tall. It is a well-known tree with a wide range of pharmacological activities that is widely cultivated throughout the world in tropical and subtropical regions. ^{4,5} It can be found in the outer Himalayas and parts of Jammu and Kashmir, Nepal to the east of Assam, Bengal, and Tripura, which stretch from the central region to the Godavari in the south ⁶. It prefers arid and semi-arid climatic conditions and grows in red and black soils with a pH of 5.6–7.5 ⁷.

Nyctanthes arbor-tristis, as previously mentioned, has a wide range of pharmacological activities and the ability to cure a variety of diseases. Since this plant is so essential to humans in their daily lives, it can be a good source of income. As a result, adulteration of this plant with various species of the genus *Nyctanthes* can occur in order to make more money. As a result, standard criteria for proper authentication of this plant or a part of it have to be established ⁸. The aim of this study is to investigate the morphological, microscopical, and physiochemical characteristics of *Nyctanthes arbor-tristis* leaves in order to ensure that they cannot be easily tampered with.

2. MATERIALS AND METHODS:

2.1 Materials:

2.1.1 Plant Collection: Fresh leaves of "*Nyctanthes arbor-tristis*" Linn was composed from local area of Lucknow (India) in the month July 2019. The plant material identified and authenticated by CSIR- National Botanical Research Institute (NBRI), Lucknow, Authentication No. (LWG) 104617. All the reagents, solvents, and chemicals used are of

A.R. grade (Merck, Loba, Qualigens) purchased from local supplier.

2.2 Methods:

2.2.1 Macroscopical study: The leaves' macroscopic characteristics were observed (appearance, fracture, colour, odour, scale, shape, texture, taste). The technique described by Trease and Evans was used to perform quantitative microscopy⁹.

2.2.2 Microscopical Study: A thin transverse segment of a fresh leaves was prepared and stained with concentrated hydrochloric acid: Phloroglucinol for microscopic examination (1:1). The photographs were taken with great care. The dried leaves were powdered and treated with a 5% KOH solution, then stained for 5 minutes with concentrated hydrochloric acid - Phloroglucinol (1:1) and placed in a 50% glycerine solution. "Histological studies such as category along with tissue preparation, the incidence of quantity classification such as calcium oxalate crystals using a magnification microscope were involved"^{10, 11, 12}.

2.2.3 Preparation of the Plant Extract: "The leaves of *Nyctanthes arbor-tristis* Linn were dried at room hotness for 25 days along with were ground keen on coarse fine particles among a chopper". "150 gm of coarse powder was macerated through ethanol (99.9% v/v) and kept for 72 hours on area heat (28-30°C)". The extract was filtered also evaporate towards dehydration at temperature 30-35 °C, the theoretical and practical yield of the extract was also calculated.¹³

2.2.4 Phytochemical Analysis: Phytochemical screening of the various extract of plants for the presence of carbohydrate, steroids, glycosides amino acids, saponin and flavonoid, alkaloid, proteins, tannin and phenolic compound which are responsible for the biological activity.

2.2.5 TLC Profile: "*Nyctanthes arbor-tristis* Linn TLC was performed with a mixture of ethyl acetate: methanol: water (77:15:8) and a solvent method (mobile phase)" using precoated silica gel G plate (stationary phase). For detection, ninhydrin reagents were used. The R_f value was calculated for a separate location. The "thin layer chromatography was conducted and extreme strain" was observed.

2.2.6 TLC Preparation of Extract: "Activation of pre-coated silica gel 60 TLC plates by injection at 110-120 ° C during furnace usage for 30 minutes proceeding to sample spotting". "The sample was applied on 60 TLC capillary silica gel precoated plates". "Plates with a solvent method of ethyl acetate: methanol: water (77:15:8)" were residential. The "R_f value of the fraction constituent variable" was detected¹⁴.

3. RESULTS:

3.1 Macroscopical Study: The macroscopic and "organoleptic characters of the leaves were experimental, i.e. shape, scale, colour, odour, margin, texture, taste, apex even petiole".



Figure 1: *Nyctanthes arbor-tristis* leaf

Table 1: Macroscopic study of *N. arbor-tristis* Linn. Leaf

Plant part and characters	Observation
Part	Leaves
Colour	Light to dark green
Odour	Indistinct
Taste	Bitter and astringent
Size	5-15 cm long, 2.5-5.7 cm wide
Texture	Rough
Shape	Heart
Base	Oblique
Margin	Entire
Apex	Acute
Venation	Reticulate

3.2 Microscopy Study:

During the midrib convex project resting on the inferior field, the leaf of *Nyctanthes arbor-tristis* was cut, also faintly wavy between a thin middle rise going to the superior sector. A small number of collenchymal coatings lie below each other's epidermis, with the superior coating positioned nearby on the curved xylem hole. The unicellular trichomes of various sizes produce cystolite on the bottom of the bear plain layer. Glandular trichomes were also close to a bicellular top overflowing by gloomy tanned satisfied among single-cell shadow.

The higher epidermis of the lamina cell was walled broadly quite instantly with stomata devoid. Two rows of epidermis are seen in the lamina near the midrib. By sinous stockade on spaces, cuticle striated with slanting with regular anomocytic stomata, the cell of the inferior epidermis is smaller in amount than that of the upper one. Two rows of pole cells lie in the higher epidermis, "followed by 7-9 rows of soft parenchyma transversed near vascular bundles often surrounded by parenchyma cell".



a) T.S. leaves of NAT mid-rib



(b) Vascular bundle



(c) Trichome

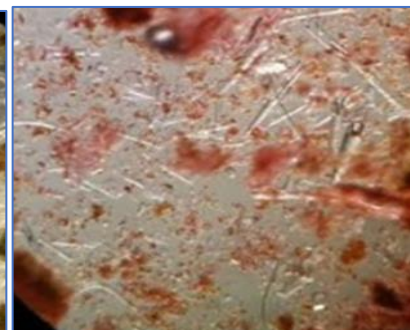
Figure 2: T.S. leaves of *Nyctanthes arbor-tristis* midrib, vascular bundle and trichome

3.3 Powder microscopy: *Nyctanthes arbor-tristis* “powder microscopy” reveals the occurrence of starch grains, palisade cells, trichomes, phloem and xylem vessels. This includes the histological study of the tissue preparation type,

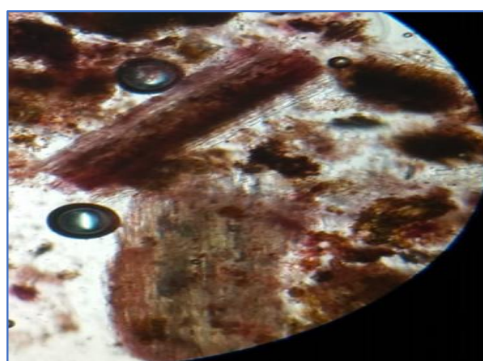
the occurrence of quality characteristics “such as trichome and starch grain using a magnification microscope”, for example.



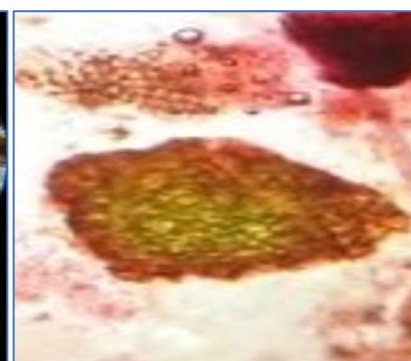
(a) Starch grains



(b) Trichomes



(c) Phloem vessels



(d) Xylem vessels

Figure 3: Powder microscopy of *Nyctanthes arbor-tristis*

3.4 Phytochemical Analysis: The crude hydroethanolic extract was screened the present “alkaloids, flavonoids, glycoside, carbohydrate, protein, starch, amino acid, steroid,

tannins and saponins using the simple chemical test as reported in a standard reference book”.

Table 2: Phytochemical screening of *Nyctanthes arbor-tristis* Linn.

Class of Drug	Chemical test	Results
Carbohydrate	Fehling	Positive
	Molisch	
	Benedict	
Alkaloids	Dragendroff test	Positive
	Hagers	
	Mayer	
	Wagners	
Flavonoid	Alkaline	Negative
	Lead acetate	
Glycoside	Legal test	Positive
	Killer Killiani	
Terpenoid and steroid	Salkowski	Positive
Protein and amino acid	Millons test	Positive
	Ninhydrane	
Saponin	Foam	Negative
Tannin	FeCl ₃	Positive

3.5 Evaluation of *Nyctanthes arbor-tristis* leaf extract:
 “The extract obtained was dark green in colour, greasy with an indistinct odour and bitter in texture”. The extracts were “soluble in an organic solvent and were considered in water to be insoluble”. The percentage yield was measured and set at 6.48% w/w.

Table 3: Observation of *Nyctanthes arbor-tristis* L. leaf extract

Extract study	Observation
Plant parts	Dried and powdered leaves
Extraction process	Simple maceration
Solvent	Ethanol (80% v/v)
Colour	Dark green
Texture	Greasy
Odour	Indistinct
Soluble	Water and alcohol
Insoluble	Acid
Practical yield	3.24 gm
Percentage yield	6.48% w/w

3.6 TLC Profile: Thin layer chromatography of purified sample of *Nyctanthes arbor-tristis* was performed”.

Table 4: TLC Finger Printing of ethanolic extract of leaf of *Nyctanthes arbor-tristis* Spots

Solvent system	Solvent run (cm)	Solute run (cm)	R _f value
Ethyl acetate: methanol: water (77: 15:8)	7.3	7.1	0.97

**Figure 4: TLC plate of ethanolic extract of *Nyctanthes arbor-tristis***

4. DISCUSSION:

As a part of standardization study, the macroscopically examination of drug was studied. *Nyctanthes arbor-tristis* leaves are plain, “5 to 14 cm long, 2.5 to 7.5 cm thick, acute to acuminate, ovate, both rough outside, whole margin or distinctly toothed base in circles to very cuneate, venation

reticulate”, 3-6 pairs of side veins, indistinguishable odour, bitter with astringent taste. The extract was dark green in colour, greasy in texture, with an indistinct smell and a bitter taste. The percentage yield was measured and set at 6.48% w/w. The microscopical evaluation of leaves powder showed that it contains characteristics features of vascular bundle, trichome, starch grains, xylem and phloem vessels. Phytochemical extract research has been conducted. In which alkaloid-containing plant extract, “glycoside, flavonoid, carbohydrate, protein and amino acid”. Phytochemical screening involves of a thin layer chromatography. The type of constituents present in the extract was obtained from the TLC. The R_f values discovered from the TLC are the constituents of Flavonoid present in the extract.

5. CONCLUSION:

The present study was focused on establishing pharmacognostic standards for the identification and authentication of the *Nyctanthes arbor-tristis*. Therefore, the outcomes of the above findings will serve as a promising source for laying down pharmacopoeial standards for the future studies and research.

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Conflict of Interest: Nil

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