ORIGINAL ARTICLE

Pharmacognostic and phytochemical investigation of root of *Solanum nigrum* Linn.: An ethnomedicinally important herb

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Background: Roots of *Solanum nigrum* Linn. (Solanaceae), commonly known as black night shade (*Kakamachi*), is traditionally used in the treatment of worms and abdominal pain. Until date no scientific evaluation has been reported on its roots. **Aim:** The present study deals with the microscopical, histochemical, physicochemical, fluorescence analysis, preliminary phytochemical and chromatographic study of roots of *S. nigrum* L. **Materials and Methods:** Thin sections of fresh root were used for the microscopical and histochemical evaluation. Root powder was used for physicochemical and fluorescence analysis. For preliminary phytochemical study water, methanol and ether soluble extractives were used. Chromatographic study was carried out on methanol soluble extractive. **Results:** Diagnostic features of the roots are presence of prismatic crystals, border pitted vessels, and tannin content. Physico-chemical analysis of root powder shows ash value 3.99% w/w, 13.04% w/w and 11.85% w/w of water and alcohol soluble extractive values respectively. Phytochemical investigations of root shows presence of alkaloids, tannins and saponins along with other phytoconstituents in different extracts. In chromatographic study, alcoholic extract of root shows 10, 7 and 8 number of spots at 256 nm, 366 nm wavelengths and after spray of vanillin sulphuric acid, respectively. **Conclusion:** The information generated in this study will be helpful for the proper identification and authentication of roots of this herb.

Key words: *Kakamachi*, pharmacognosy, phytochemistry, *Solanun nigrum* Linn

INTRODUCTION

Since times immemorial, medicinal plants are playing a vital role in alleviating human sufferings in the form of source of food and medicine. The global demand of herbal medicine, especially, Ayurvedic herbs, is not only large but growing.[1] Safety, efficacy and inexpensiveness have led to rapid expansion of Ayurvedic herbal pharmaceutical industry.[2,3] Therefore, The World Health Assembly has emphasized the need to ensure the quality of medicinal plant products by using modern control techniques and applying suitable standards.[4] Modern physico-chemical parameters and chromatographic studies provide the unique direction and a scientific basis towards this approach.

Kakamachi (*Solanum nigrum* Linn.) is herb of Solanaceae family used mostly in Traditional Medicinal prescriptions for liver and skin disorders.[5] Its leaves and tender shoots are widely used as vegetables throughout the world and have provided a food source since early times. In India, an infusion of the plant is used as an enema for infants with abdominal upsets. The Houmas Indians, in North America, use an infusion made from boiled roots of this plant to administer to babies with worms. Raw roots are also found to be eaten for abdominal pain in Tanzania.[6] No systematic pharmacognostic and phytochemical studies of its roots have been reported until date and therefore, a detailed investigation of the powdered roots of *S. nigrum* L. has been carried out using various pharmacognostical and physico-phytochemical parameters.

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MATERIALS AND METHODS

Collection of the Sample

The whole plant was collected, by uprooting the plant without damaging the root system, from the campus of Gujarat Ayurved University (GAU), Jamnagar (Gujarat, India) in the month of January 2012 [Figure 1a]. The plant specimen was authenticated by the Pharmacognosist, GAU, Jamnagar. Roots were separated and washed under running tap water; shade dried; pulverized; sieved through 80 meshes and preserved in an airtight glass bottle.

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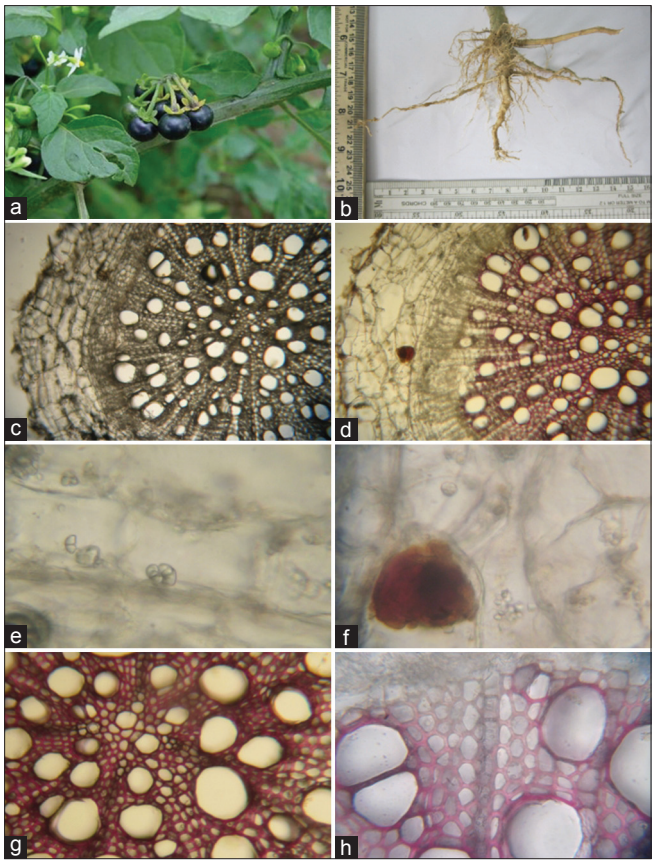

Figure 1: (a) Plant of *Solanum nigrum* Linn.; (b) Root system of *Solanum nigrum* Linn.; (c) Cortex; (d) Phloem; (e) Starch grains; (f) Tannin content; (g) Xylem; (h) Medullary rays

Macroscopic and Microscopic Evaluation

Thin sections of fresh root were taken by maceration method[7] and were treated with phloroglucinol, hydrochloric acid, and iodine for identification of various contents.[8] Photomicrographs were taken by using canon digital camera attached to Carl Zeiss binocular microscope. Powder microscopy was also carried out with stain and without stain and photomicrographs were taken.

Histochemical Tests

To detect the site of location of various constituents of the drug, sections of root were treated with various reagents such as ruthenium red (for mucilage), $FeCl_3$ to (tannin), and iodine for (starch grains) etc.[9]

Physico and Phytochemical Evaluation

The dried sample was used for physicochemical and the preliminary phytochemical investigations by standard procedure adopted by Ayurvedic pharmacopoeia of India.[10]

Fluorescence Analysis

It was carried out as per the method of Chase and Pratt (1949).[11] Two grams of dried root powder was soaked under solvents and the colours of the extracts were observed under ordinary light and Ultra Violet (UV) light for the fluorescent colour.

High Profile Thin Layer Chromatography Study

A CAMAG (Switzerland) HPTLC system equipped with a sample applicator Linomat V was used for application of samples. CAMAG TLC Scanner 3, Reprostar and Wincats 4.02 were used for scanning the plates. CAMAG twin through glass chamber was used for developing the plates. Pre-coated silica gel plate was used as stationary phase and Toulene: Ethyl acetate: Formic acid (7:2:1 v/v/v) was used as mobile phase. After development, the plate was visualized under short UV (254 nm) and long UV (366 nm) and thereafter sprayed with Vaniline sulphuric acid for color reaction.

RESULTS AND DISCUSSION

Macroscopic Features

Tap root with few branches and numerous small lateral roots [Figure 1b], externally smooth, pale brown in colour; bark is thin, easily peeled off exposing pale yellow wood.

Microscopic Study

Transverse section of root

Root shows cork consisting of 2-4 rows of tangentially elongated cells; cortex of large, slightly elongated, thin walled cells having patches of the cortical cells contain oval to round starch grains [Figure 1c], measuring 2.5-11 μ in diameter [Figure 1e], single or with two or rarely three components; phloem consists of thin walled polygonal cells, phloem rays uniseriate, and filled with starch grains [Figure 1d]; xylem composed of vessels and parenchyma; vessels arranged in groups of 2-4 in radial rows [Figure 1g]; parenchyma thick walled containing prismatic crystals of calcium oxalate and tannin content cells [Figure 1f]; medullary rays composed of thin walled, radially elongated cells [Figure 1h].

Organoleptic characters of powder

Root powder is creamish white in colour; tasteless and with characteristic odour [Figure 2a].

Powder microscopy

The diagnostic characters of powder microscopy show fragments of border pitted vessels [Figure 2b], scleroids [Figure 2c], oil globules [Figure 2d], prismatic crystals of calcium oxalate [Figure 2e], and simple starch grains [Figure 2f].

Histochemical test

Various histochemical tests were conducted on the

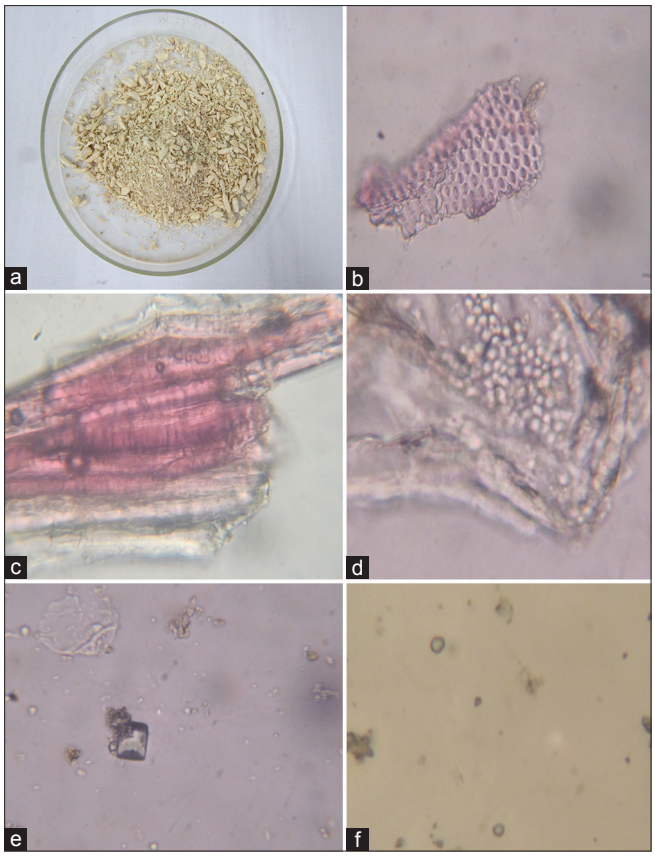

Figure 2: (a) Root powder; (b) border pitted vessels; (c) scleroids; (d) Oil globules; (e) Prismatic crystal of calcium oxalate; (f) Simple starch grains

root powder of *S. nigrum* L. The results are depicted in Table 1.

Physicochemical tests, preliminary phytochemical screening, and test of fluorescence of *S. nigrum* L. root extract: The results are shown in Tables 2-4 respectively.

Chromatographic study

HPTLC fingerprint profile of alcohol extract revealed different number of phytoconstituents at 254 nm and 366 nm wavelengths [Table 5 and Figure 3].

CONCLUSION

For correct identification and standardization of *S. nigrum* L. roots; these studies provide the referential information. The pharmacognostical studies reveal that the presence of prismatic crystals of calcium oxalate, border pitted vessels, scleroids, oil globules, simple and compound starch grains are the striking characters of identification of this plant material. The preliminary phytochemical screening of three extracts of the root powder shows the presence of alkaloids, tannins, and saponins along with other phytoconstituents. In HPTLC studies, the alcoholic extract shows presence of ten and seven phytoconstituents at 254 nm and 366 nm wavelengths respectively. The information in this study is helpful to differentiate *S. nigrum* L. from the other closely related species of Solanaceae family.

Table 1: Histochemical tests for *Solanum nigrum* Linn. roots

Reagents	Observation	Characteristics
Phloroglucinol+Conc. HCL	Red	Lignified cells
Iodine	Blue	Starch grains
Phloroglucinol+Conc. HCl	Dissolved	Calcium oxalate crystals
$FeCl_3$ solution	Dark blue to black	Tannin cells
Ruthenium red	Red	Mucilage

HCl – Hydrochloric Acid, $FeCl_3$ – Ferric Chloride,

Table 2: Physicochemical analysis of *Solanum nigrum* Linn. roots

Parameters	Root
Foreign matter	Nil
Loss on drying (% w/w)	5.50
Ash value (% w/w)	3.99
Acid insoluble ash (% w/w)	0.19
Water soluble extractive value (% w/w)	13.04
Alcohol soluble extractive value (% w/w)	11.85
pH	6

Table 3: Preliminary phytochemical screening of *Solanum nigrum* Linn. roots

Tests for phytoconstituents	WE	ME	CE
Carbohydrates	+	+	–
Proteins	+	–	–
Amino acids	+	–	–
Steroids	–	–	–
Alkaloids	+	+	+
Glycosides	+	+	–
Cardiac glycosides	+	–	–
Phenols	–	–	–
Flavonoids	+	+	+
Terpenoids	–	–	–
Tannins	+	+	+
Saponins	+	+	+
Anthraquinone	–	–	–
Volatile oil	+	–	–

WE – Water extract; ME – Methanol extract; CE – Chloroform extract; + – Present; – – Absent

Table 4: Fluorescence analysis of *Solanum nigrum* Linn. root

Treatment	Observations under		
	Ordinary light	UV light	
		254 nm	366 nm
1 g powder+methanol	Light yellow	Colorless	Blue
1 g powder+water	Light yellow	Colorless	Green
1 g powder+chloroform	Light yellow	Colorless	Purple
1 g powder+hexane	Colorless	Colorless	Green
1 g powder+6 N HCL	Yellow	Colorless	Green
1 g powder+4% NaOH	Yellow	Yellow	Light green

UV – Ultra violet; HCl – Hydrochloric Acid; NaOH – Sodium Hydroxide

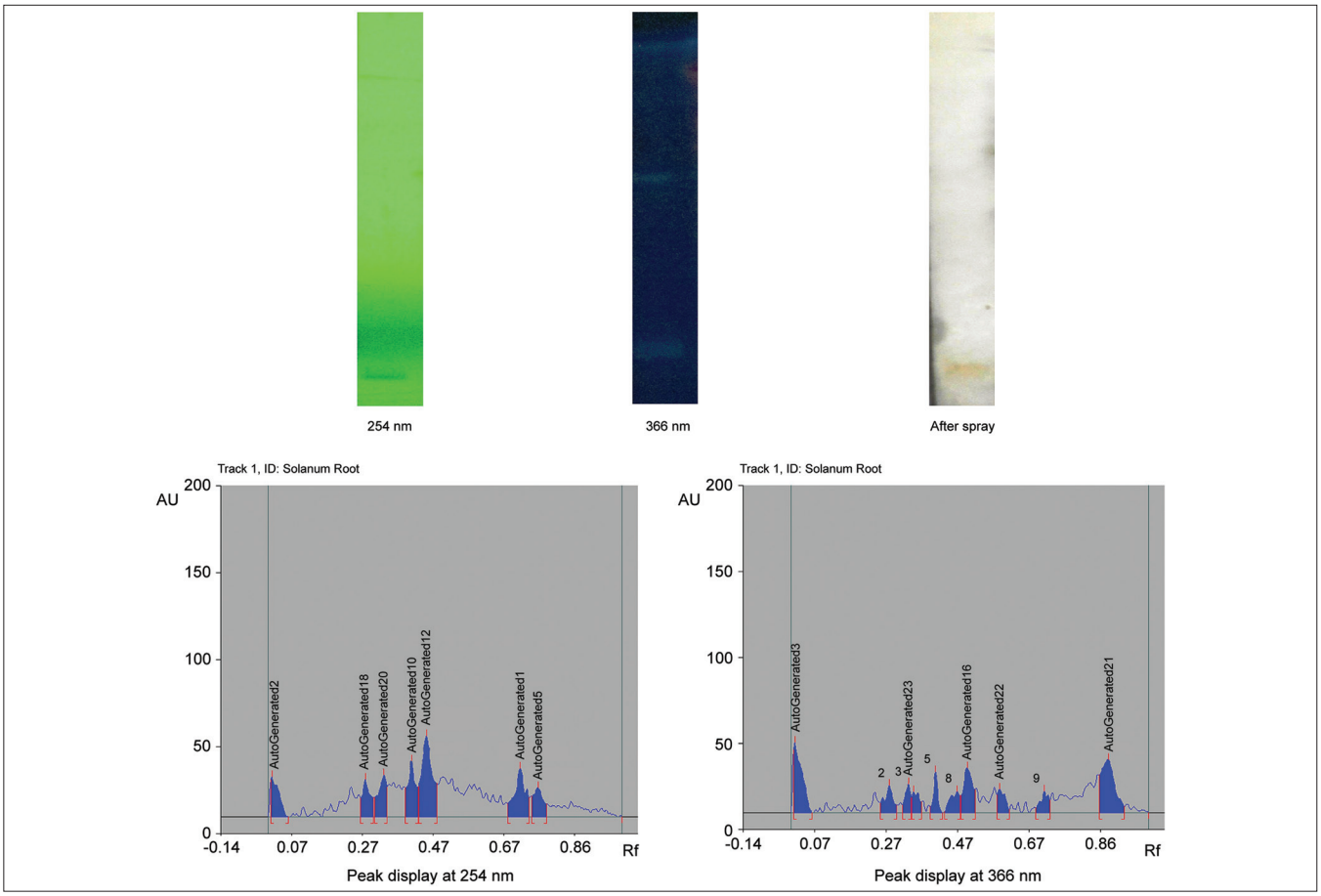

Figure 3: High profile thin layer chromatography study of alcoholic extract of *Solanum nigrum* Linn. root

Table 5: Chromatographic results of *Solanum nigrum* Linn. root extract

Conditions	Number of spots	Rf value
Short ultra violet (254 nm)	10	0.01, 0.27, 0.33, 0.34, 0.40, 0.46, 0.49, 0.58, 0.71, 0.89
Long ultra violet (366 nm)	7	0.01, 0.27, 0.33, 0.41, 0.45, 0.71, 0.76
After spraying (Vaniline sulphuric acid)	8	0.07, 0.13, 0.15, 0.37, 0.48, 0.55, 0.68, 0.94

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