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Phytochemical and Chromatographic Studies in the Latex Milk Powder of *P. Daemia. Linn*

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Abstract

India being a rich and varied flora of medicinal plants since the Vedic period. The present study deals with the scientific validation of *P. daemia.Linn* with special reference to its pharmacognostical and phytochemical investigations. This is commonly known as *P. daemia*, figure and is known to have medicinal properties, also even used as home remedies in the rural and the remotest parts of the India Qualitative & Quantitative analysis for nutritional value and amino acid detection for both sample has been estimated & detected TLC finger prints of latex of at 366 nm (before derivatization) & Rf values in TLC finger prints of latex at 254 nm (after derivatization) has been studied for both Samples. Latex is applied externally on chronic infected wounds to alleviate edema, pain and to promote the healing; therefore an attempt has been made to carried out the detailed quality control and assurance of the drug followed by HPTLC profiles, HPTLC profile of *P. Daemia* latex milk powder observed under 366 nm & 254 nm shows the variety of results A – C (Gautala forest,) B - Aurangabad (M.S.) Sample A, B & C.

Keywords: FHPTLC; Phytochemistry; Nutritional Value; *P.daemia. Linn*

Abbreviations: HPTLC: High Performance Thin Layer Chromatography; TLC: Thin Layer Chromatography.

Introduction

Medicinal plants are important component of natural resources viz. food, gum, fiber, tannin, resin, herbal medicines. The importance of herbal medicine that about 80% of the developing world's population depends on traditional medicine for their primary health care [1-4]. One such plant is *Pergularia daemia linn* whole Plant**.** It is a hispid perennial herb which grows along the road sides of India and other tropical and sub- tropical regions. It is popularly known as Veliparuthi in Tamil. This plant has been used in the traditional medicine for a wide range of aliments. The entire plant is used as anti-helmenthic, anti-pyretic, laxative, expectorant and infantile diarrhea [2]. Each part of the plant has various therapeutic values. The root of the plant is effecting in treating the disorders like asthma, mental disorder, anemia, leprosy and piles. The dried leaves are used in treating bronchitis, asthma, rheumatic fever, amenorrhea and dysmenorrhea and wounds Roots and shoots are prescribed for Whooping cough. All these medicinal properties of *Pergularia daemia* is may be due to the presence of various phytochemicals [5-7]. Hence this study aims to determine the phytochemicals so that they can be used further for the designing of the drugs in future. All natural drugs are functioning properly and whose mind, body

and spiritare cheerful. *Pergularia daemia*, the trellis-vine, is a hispid, perennial vine in the family Apocynaceae, with an extensive range in the Old World tropics and subtropics. It has been used traditionally to treat a number of ailments [6-9].

Materials and Methods

Collection of the Plant

The plant materials for the proposed study were collected in the month of August & September 2019 from Paithan road, Aurangabad District, Maharashtra India. The plant material was taxonomically identified by Prof. Arvind Dhabe, Botany Department, Dr.BAMU, Aurangabad, Maharashtra, India. The voucher herbarium specimens (Accession No. 0715) have been preserved in our laboratory for further reference.

Physico-Chemical Analysis

Air dried plant material was used for the quantitative determination of ash and extractive values (nutritional values) according to standard procedure of Indian Pharmacopoeia [10-14] and WHO/QCMMPM [15]. Fluorescence analysis of the extract(s) was carried out according to standard procedure [16]. Powdered latex was subjected to analysis under ultraviolet light after treatment with various reagents and chemicals like sulphuric acid, nitric acid, dilute hydrochloric acid and sodium hydroxide.

Preliminary Phytochemical Screening

Preliminary phytochemical evaluation was carried out by using standard procedure [17].

Fluorescence Analysis

The latex powder was treated with different chemicals and seen under the normal light and UV radiations at 254 and366 nm wavelengths as per the standard procedure. The colour development under the day light was also observed for the presence of various phytochemical compounds.

High Performance Thin Layer Chromatography (HPTLC)

For HPTLC, 5 g of coarsely powdered drug in 250 ml stoppered conical flask & extracted with 100 ml ethanol for24 hours by maceration technique with occasional shaking. The extract was extracted and volume was raised up to100 ml in a volumetric flask. 25 ml of the extract was taken from the above stock solution and concentrated on water bath to similarly, ethanol extracts were prepared for two samples of *P.daemia. Linn*, as reference Kokate CK and Lohar DR [17,18].

Results and Discussion

Physiochemical Analysis

Air dried material was used for quantitative determination of phytochemical values. Total ash, acid insoluble ash, water soluble and alcohol soluble extractive were determined for five times as per WHO recommendations. Alcohol soluble extractive value was found to be very high when compared to other extractable matter in the drug (Table 3) [18].

Preliminary Phytochemical Screening

The preliminary phytochemical test was performed on the extracts of plant of Linn. They show the presence of the alkaloids, carbohydrate, flavonoids, saponin, resin, protein and tannin (Table 1). The fluorescence analysis is a tool for the determination of constituents in the plant that gives a definite idea of the chemical nature. Thus the fluorescence analysis of the powder was carried out and data is presented in the Table 4.All amino acid test and nutritional value concentration has been determined in both samples Tables 2-8 [15,18].

S. No	Parameters	Samples A	Sample B
1	Carbohydrate	+	+
2	Alkaloid	+	+
3	Flavonoid	+	+
4	Tannin	+	+
5	Protein	+	+
6	Resin	+	+
7	Saponin	+	+

Key: + present, -absent.

Table 1: Preliminary phytochemical screening of extracts of latex milk powder of *P.daemia*: Linn.

S. No	Amino acid test	*P.Deamia* Extract	
		Sample A	Sample B
1	L-hydroxyproline	-	-
2	DL serine	+	+
3	DL- alanine	+	+
4	DL-tryptopham	-	+
5	DL Iso- leucine	+	+
6	DL valine	+	-
7	DL- nor- leucine	+	+
8	L- Cystein hydroxyl	+	+
9	L- ornithine	-	-
10	DL-2-aminobutyric acid	+	+
11	DL- aspartic acid	+	+
12	Glycine	-	+
13	3-c-3-4dihydroxy phenyl	-	-
14	L-glutamic acid	+	+
15	L-tyrosine	+	+
16	DL- threonine	+	+
17	L- proline	-	+
18	L- arginine	+	+
19	L-leucine	+	+
20	L- lysine monochloride	+	+
21	DL- methionine	+	+
22	L- histidine	-	-

Table 2: Amino acid test qualitative estimation latex milk powder of *P.daemia*: Linn.

S. No	Nutrients	Value	
		Sample A	Sample B
1	Moisture (%)	46.20 ± 0.14	48.20 ± 0.15
2	Ash (%)	3.96 ± 0.15	5.00 ± 0.14
3	Insoluble ash (%)	9.29 ± 0.10	7.11± 0.15
4	Soluble ash (%)	7.45 ± 0.10	8.36 ± 0.11
5	Crude fibre (%)	16.65 ± 0.10	15.38± 0.17
6	Crude fat (%)	4.39 ± 0.14	6.21± 0.18
7	Total nitrogen (%)	0.75 ± 0.08	1.25 ± 0.10
8	Total protein (%)	4.06 ± 0.06	3.55± 0.09
9	Carbohydrate (%)	19.78 ± 0.10	17.24± 0.11
10	Organic matter (%)	94.85 ± 0.15	90.77± 0.19
11	Na (mg/100g)	0.72 ± 0.12	0.88 ± 0.16

12	Ca (mg/100g)	1.45 ± 0.13	1.67 ± 0.15
13	pH	5.6	5.56
14	Alcohol-soluble extractive	63.45%	63.65%
15	Water-soluble extractive	22.33%	22.45%
16	K (mg/100g)	1.48 ± 0.10	2.11 ± 0.11
17	Mg (mg/100g)	0.90± 0.13	0.96± 0.14
18	P (mg/100g)	1.86 ± 0.02	1.99 ± 0.02
19	Fe (mg/100g)	0.02 ± 0.02	0.03 ± 0.04

Table 3: Nutritional value of latex powder of *P.daemia*: Linn for Both Sample.

S. No	Treatment	Samples A	Samples B
	Day light	UV light at 254 nm	UV light at 366nm
1	Powder as such	Brown Brown	Brown
2	Powder + KOH	Dark Yellow	Blue Blue
3	Powder + HCl	Brown Blues	Brown Brown
4	Powder + H2SO4	Dark Yellow	Brown Brown
5	Powder + NaOH	Dark Yellow	Brown Black
6	Powder + HNO3	Brown	Blue Blue

Table 4: Fluorescence Analysis of latex milk powder of *P.Daemia* Linn.

HPTLC fingerprint profile of alkaloid and phenolic compound fraction was developed in respective solvent system and identified in different visualizing agent as per Stahl, in both Samples [16,19].

S. No	Parameters	Samples A	Samples B
1	Rf 1 (sky blue)	0.52	0.52
2	Rf 2 (white)	0.81	0.81
3	Rf 3 (blue)	0.95	0.95

Table 5: Rf values in TLC finger prints of latex of at3 66nm (before derivatization) Rf value.

A –Daultabad forest (Aurangabad, Maharashtra), B – Gautala forest (Aurangabad, Maharashtra)

S. No	Parameters	Samples A	Samples B
1	Rf 1 (yellow)	0.63	0.63
2	Rf 2 (yellow)	0.77	0.77
3	Rf 3 (red)	0.95	0.95

Table 6: Rf values in TLC finger prints of latex of at 254 nm (after derivatization) Rf value.

S. No	Parameters	Samples A	Samples B
1	Rf 1(blue)	0.07	0.07
2	Rf 2 (sky blue)	0.38	0.38
3	Rf 3 (brown)	0.54	0.54
4	Rf 4 (white)	0.63	0.63
5	Rf 5 (yellow)	0.77	0.77
6	Rf 6 (yellow)	0.87	0.87
7	Rf 6 (red)	0.95	0.95

Table 7: Rf values in TLC finger prints of at 366nm (after derivatization) Rf value.

S. No	Parameters	Samples A	Samples B
1	Rf 1(brown)	0.63	0.63
2	Rf 2 (brown)	0.77	0.77
3	Rf 3 (black)	0.95	0.95

Table 8: Rf values in TLC finger prints of at visible light (after derivatization) Rf value.

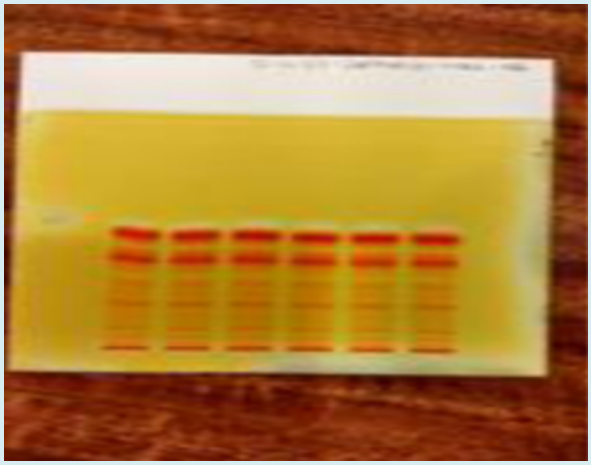

Figure 1.1: HPTLC profile of *P. Daemia* latex milk powder observed under 366 nm A – C (Gautala forest,) B - Aurangabad (M.S.) Sample A.

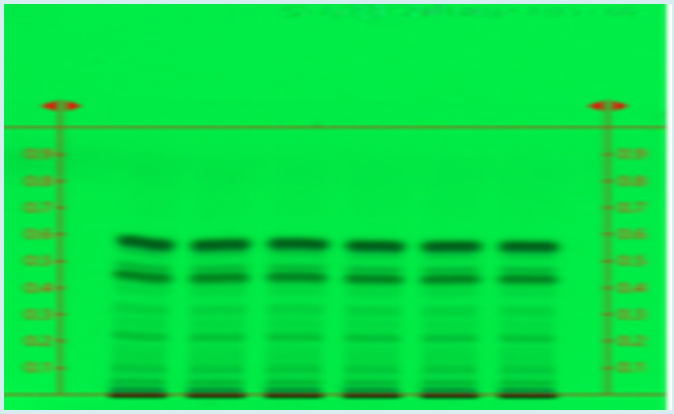

Figure 1.2: HPTLC profile of *P. Daemia* latex milk powder after spraying with 5% methanolicsulphuric acid reagent observed under 254 nm .Sample B.

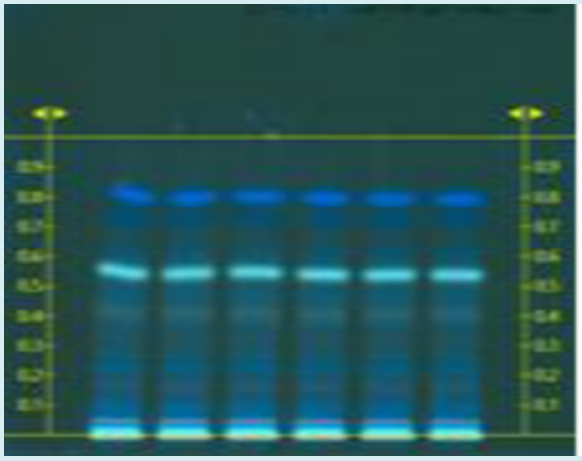

Figure 1.3: HPTLC profile of *P. Daemia* latex milk powder after spraying with 5% methanolicsulphuric acid reagent observed under 366 nm Sample A.

Figure 1.4: HPTLC profile of *P. Daemia* latex milk powder after spraying with 5% methanolicsulphuric acid reagent observed under visible light Sample B.
A B A B A B A B

Conclusion

The study of Pharmacognostical-phytochemical features of *P. Daemia* latex milk powder Linn had shown the standards which will be useful the detection of its identity and authenticity. The other study viz. Sample A & Sample B Shows the variety of results under the physiochemical analysis, preliminary phytochemical test, nutritional value determination, amino acid detection fluorescence analysis and High Performance Thin Layer Chromatography (HPTLC) add to its quality control and quality assurance for proper identification. HPLC and TLC fingerprint would be useful for the quality assessment of raw material under different wavelength frequency of and also helpful in the identification and isolation of therapeutically important phytochemicals.

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