



HPTLC ANALYSIS OF VARIOUS MARKET SAMPLES OF A TRADITIONAL DRUG SOURCE – KODIVELI (*PLUMBAGO ZEYLANICA* LINN)

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ABSTRACT

Kodiveli or Chitraka is a traditional drug botanically equated as *Plumbago zeylanica* Linn belonging to the family Plumbaginaceae and is used in respiratory diseases and in anemic conditions. The present study deals with Plumbagin, a 5-hydroxy-2methyl-1, 4-naphthoquinone, an active chemical constituent of the root of this plant drug. This bioactive molecule is a proven anticarcinogenic, antiatherosclerotic and antimicrobial agent. In the present paper, HPTLC analysis of market samples of the roots procured from Trichy and Chennai as well as fresh samples collected from gardens of Chennai were carried out for evaluating their Plumbagin content. The data of the results obtained revealed that the sample collected from trichy market is of better quality as it produced higher Plumbagin content. In the shade dried freshly collected samples, plumbagin peak was not detected. Hence it is inferred that only on storage the quinone content increased.

Keywords: Kodiveli, Chitraka, *Plumbago zeylanica*, Napthoquinone, HPTLC

INTRODUCTION

The plant species *Plumbago zeylanica* known vernacularly as Chitraka, Kodiveli, Chitramulam, Tellachitramulam, Agnichela, Agnimaala or by its trade or popular names as "Lead wort-white flowered" and "Ceylon Lead wort" belongs to the family Plumbaginaceae. Source taxon is distributed as a weed throughout the tropical and subtropical countries of the world¹. The roots of *Plumbago zeylanica* (Chitraka or Chitramulam) has numerous therapeutic uses^{2,3,4,5}. The root is known to be abortifacient and to have vesicant properties. It is used as an appetizer, diuretic, expectorant and in dysentery, diarrhoea, piles and peptic ulcers. The root paste is applied topically for filarial leg. It is used topically for early maturation, rupture and healing of abscess. The root powder taken orally along with honey gradually reduces hypercholesterolemia and improves blood formation (anemia). It is used to reduce obesity, vitiligo, splenomegaly, hepatomegaly and ascitis. It is also used to relieve coryza (running nose), hoarseness of voice and sore throat. It is used in the form of local applications for leucoderma, scabies, psoriasis, symptoms of leprosy and allied skin diseases. The decoction of the root is useful in checking and preventing spermatorrhoea⁶.

Plumbagin, is the main chemical constituent of this drug source and could act as the chemical marker for the *Plumbago* spp. The best sources for plumbagin are the roots of *Plumbago europea*, *P. rosea*, and *P. zeylanica*⁷. This plant derived 5-hydroxy-2methyl-1,4-naphthoquinone possesses a broad spectrum of biological activity. It has been proved to exert anticarcinogenic⁸ and antiatherosclerotic effects in animals. Plumbagin is commonly used for its antibacterial and antiviral properties besides being a good insecticidal, anti-inflammatory and antipyretic agent^{9,10,11,12}. Extracts of *P. zeylanica* and its active ingredient Plumbagin possess significant antioxidant potential¹¹. It is also considered to be an effective antimalarial¹³ and antifungal agent¹⁴. Studies have also been carried out to evaluate the antifertility activity of Plumbagin^{15,2,16,17,18,19,20}.

The present study deals with the HPTLC analysis of market samples as well as freshly collected roots of *P. zeylanica* Linn. This analysis can help in determining the Plumbagin content, the chemical marker of the drug under study. Presence of considerable amount of Plumbagin can help in deciding the genuineness of the drug source. Data obtained in the present work could be useful in the proper identification and authentication of this traditional drug which is the primary pre requisite for the international acceptance and recognition of herbal medicines.

MATERIALS AND METHODS

Collection of plant material

Various samples of kodiveli from South Indian raw drug markets of Chennai and Tiruchirappalli which were stored for 3months, 6months and 9months respectively were used for the present study. Fresh samples were collected from medicinal plant gardens of Puthanampatti, Tiruchirappalli, Tamil Nadu. Fresh samples were shade dried and subjected to HPTLC analysis along with dried market samples. Standard Plumbagin was also subjected to HPTLC finger printing.

Instrumentation and application of the sample

Commercially available pre coated plates of silica gel GF 254 of 10x10 sizes were used for the study. Application was maintained at 10 micro grams per min, using Linomate 4 applicator and automatic TLC applicator, Camag, Switzerland. A Camag TLC scanner equipped with cats V4.06 version software was used for interpretation of data. The standard is a solution containing known concentration (conc. Range 10-100µg/ml) of plumbagin in toluene. A known quantity of sample volume was applied. (For sample A, B & D - 30µl and for sample C - 15µl).

Chromatographic conditions

The experiment was performed on silica gel GF254 HPTLC plates using mobile phase comprising of toluene: formic acid (9.9:0.1). The plate was pre washed by toluene and activated in an oven at 110deg C for 1hr before use. The sample solutions were applied on the HPTLC plate as sharp bands of 20mm width with the help of Camag Linomat IV sample applicator at the distance of 10mm from the edge of the HPTLC plate. The rate of speed for Samples A, B, C & D are 20mm/s. The developed TLC plate was air dried and then scanned between 200 & 400nm using Camag TLC scanner with cat 4.06 version software. The wavelength chosen for quantification is 423nm. The overlain spectra for the samples are given in fig 2a, 2b, 2c & 2d.

The amount of plumbagin was determined from the Michaelis Menten Regression equation, of calibration graph, plotted between area and concentration.

RESULTS AND DISCUSSION

Data obtained are tabulated in Table I and presented in Fig 1 (Standard plumbagin), Fig 2a, 2b, 2c & 2d.

Table 1: Details of plant material

Sample & place of collection	Period of storage in months	Appearance	Plumbagin content
Sample A CHENNAI	3 months	Brown coloured coarse powder	0.0017%
Sample B CHENNAI	6 months	Brown coloured coarse powder	0.0035%
Sample C TRICHY	9 months	Brown coloured fine powder	0.0165%
Sample D PUTHANAMPATTI	Fresh shade dried samples	Leaves	No peak detected

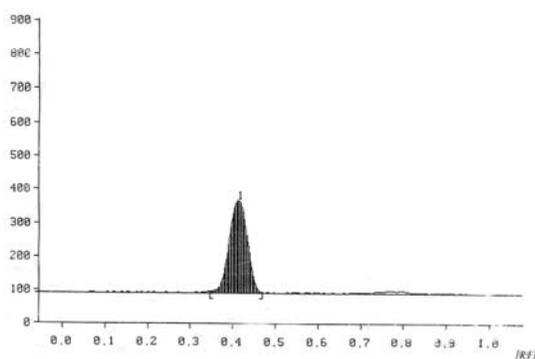


Fig. 1: It shows chromatogram of standard plumbagin

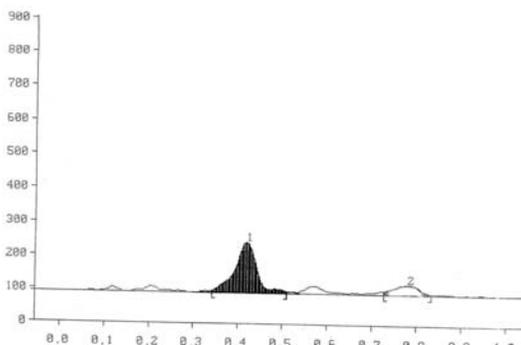


Fig. 2a: It shows chromatogram of sample A at 423nm.

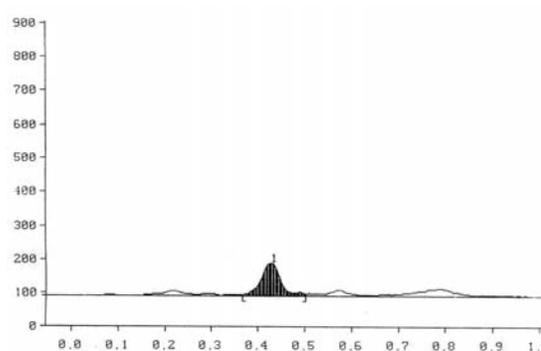


Fig. 2b: It shows chromatogram of sample B at 423nm.

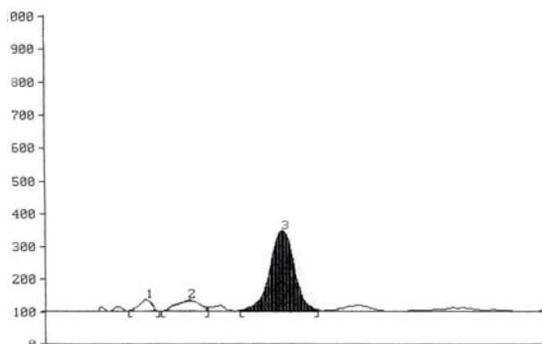


Fig. 2c: It shows chromatogram of sample C at 423nm.



Fig. 2d: It shows chromatogram of sample D at 423nm.

The results and data obtained in this study clearly revealed that plumbagin content was more (i.e. 0.0165 %) in plants from trichy compared to chennai samples (0.0035% & 0.0017%). The shade dried freshly collected puthanampatti samples revealed no peaks of Plumbagin. Hence it is inferred that only on storage there is an increase in quinone content (Plumbagin). The method adopted in

the present work is reproducible, accurate, precise and cost effective.

CONCLUSION

The present study was aimed to develop an analytical method for the estimation of plumbagin, an active chemical constituent of *P.*

zeylanica, a traditional drug source. This chemical marker has a number of proven therapeutic properties. This study stresses the importance of scientific methods of proper identification and authentication as well as impact of storage of traditional drugs. These factors can certainly contribute significantly in promoting ecofriendly herbal drugs for the health care of human society.

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