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## **Research Article**

# PHARMACOGNOSTICAL EVALUATION OF DIFFERENT MARKET SAMPLES OF *VIDARIKAND* (*PUERARIA TUBEROSA* DC) IN INDIA

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**KEYWORDS:** *Vidarikand, Pueraria tuberosa* DC., Market samples Lucknow, Delhi and Mumbai, Pharmacognostical, Authenticity.

## **ABSTRACT**

The demand for natural products derived from medicinal plant is increasing in national and international market. Over 86% of raw material required for traditional medicines/herbal medicines used to be collected from wild resources. With the increase in demand of medicinal plants for the commercial herbal medicine sector led to the indiscriminate and unscientific collection without any consideration for the quality of the material collected. Therefore, it has created problem like deforestation, so some plants species have been near to endangered such as *Kutki*, *Jatamansi*, *Ativisha* etc. *Vidarikand* (*Pueraria tuberosa* DC) is a drug of high demands. Because of its increasing demand in the pharmaceutical industry, the availability of authentic drug is decreased. This decline in authenticity is due to substitutes or sub standard materials and adulterants. Objective: The main objective of present study is to evaluate the quality of the genuine/official Vidarikand and its commercial sample and to find out whether the genuine/official drug is available in the market or not. Methods: The genuine/official Vidarikand and its three different markets sample in India which are Lucknow, Delhi and Mumbai. Pharmacognostical and phytochemical analysis of official drug and three different market samples have been compared to the standards given in Ayurvedic pharmacopoeia. This research work was conducted in laboratory of State Ayurvedic College, Lucknow and CSIR-NBRI, Lucknow. Observation and Results: As per Pharmacognostical and phytochemical parameters official drug of *Vidarikand* and its market samples variations were found. Conclusion: In view of pharmacognostical and phytochemical analysis, the sample of Lucknow and Delhi market were not similar to genuine/official sample, they were totally replaced with other drug and the sample of Mumbai was mixer of two different drugs one was Vidarikand and second was other adulterated drug.

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## INTRODUCTION

Vidarikand is an important plant which is described in detail in all the classical text of Ayurveda. It is commonly known as Indian kudzu which is a perennial climber with woody tuberculated stem with large tuberous roots. In Ayurved Vidarikand used to treat many diseases such as Daha (burning sensation), Ksaya-Kasa (feeble body, cough), Sula (pain), Mutrakrcchra (dysuria), Visarpa (erysipelas) etc., many

therapeutic effect like Vatahara (vitiated Vatadosha), Pittahara (vitiated Pitta dosha), Hrdya (Heart wellness), Brihani (anabolic), Vrashya (aphrodisiacs), Mutral (diuretics), Jivniya (to give life), Rasayan (rejuvenation), Balya (strength, stamina and immunity promoter), Varnya (complexion promoters), Svarya (promoter/ beneficial for throat or voice) etc. and important formulations such as Satavaryadighrita, Nityananda

Sarasvatarista, Asvagandhad-yarista, Rasa. Mahavisaaarbha Taila etc.[1] Vidarikand (Pueraria tuberosa DC) is a drug of high demands. Because of its increasing demand in the pharmaceutical industry, the availability of authentic drug is decreased. This decline in authenticity is due to substandard substitutes or materials adulterants. Enough literature and documents are available regarding the morphology of *Vidarikand*. At present here is market dependency for the procurement of Vidarikand in crude form. In the market available official (genuine) drug is either substituted or sub-standard and adulterated. It is difficult to find out official drug among them without the help of modern scientific methods. Problem of substitutes or sub-standard materials and adulterants starts right from the collection of the drug. Human resources employed for collection of drugs, if not qualified, collect all externally similar plants. After a drug dry up, it is a tedious work to identify it. API (Ayurvedic Pharmacopoeia of India) has accepted Pueraria tuberosa DC as the official source for Vidarikand. Present research work was planned in order to carry out the pharmacognostical evaluation of different market samples of Vidarikand (Pueraria tuberosa DC.) in India. Present research work increases awareness of Vidarikand (Pueraria tuberosa DC), and its identification and authentication.

# MATERIAL AND METHODS Material

Official (genuine) drug of *Vidarikand* (*Pueraria tuberosa* DC) was taken from NBRI-CSRI, Lucknow, (U.P.) for comparative study and quality standardization. After collection of official sample, was washed with tap water, cut in slices and dried in shade. Sample packed in polybags and labelled. Three market samples of *Vidarikand* were procured. Different market samples sold in markets on the name of *Vidarikand* were collected from markets which are Lucknow, Delhi and Mumbai. 250g crude *Vidarikand* of each market sample packed in polybags and labelled. The official *Vidarikand* and its market sample were powdered with mechanical

grinder and preserved in an air-tight glass container. After preservation of each sample were symbolized as- Official- OFF, Lucknow- LKO, Mumbai- MUM and Delhi- DEL. This research work was conducted in laboratory of State Ayurvedic College, Lucknow and CSIR-NBRI, Lucknow.

#### Methods

Macroscopic study, Microscopic study<sup>[2]</sup>, Powder Microscopy<sup>[2]</sup>, Determination of Foreign matter<sup>[3]</sup>, Physicochemical Studies as Loss on drying (LOD)/Moisture content<sup>[4]</sup>, Determination of total ash value<sup>[5]</sup>, Determination of Acid insoluble ash value<sup>[6]</sup>, Determination of water soluble ash value<sup>[7]</sup>, Determination of alcohol soluble Extractive value<sup>[8]</sup>, Determination of water soluble Extractive value<sup>[9]</sup>, Qualitative Photochemical analysis<sup>[10][11]</sup> TLC<sup>[12]</sup> and HPTLC<sup>[13]</sup>were carried out by standard methods.

In chromatography genuine/official and market samples of Vidarikand and standard marker compound puerarin were applied on percolated silica gel 60F<sub>254</sub> HPTLC plate format 100.0 x 100.0 mm, the linear ascending development was carried out in the developing chamber twin trough chamber (10 X 10 cm). The saturation time of the TLC chamber in the mobile phase was optimized to 30minutes for a good resolution of the tested markers and the total run time was 30 minutes at room temperature  $(27 \pm 2^{\circ}C)$ . The mobile phase was selected using a various system where in varying ratio and polarity were tried. The mobile phase consisting of Chloroform: Methanol: Acetic acid (8:2:1) was optimized for quantitative study. TLC plate was developed up to a distance of 85mm from the point of application, scanning of the TLC plate was performed using the CAMAG TLC Scanner at single wavelength  $\lambda_{max}$  450nm in ultraviolet absorbance mode for all tracks, slit dimension was 4X0.45mm. Puerarin was quantified using Camag scanner equipped with Camag Visioncats software (slit width, 5 mmX 0.45 mm) in absorption mode. HPTLC fingerprinting of all four samples of Vidarikand (Pueraria tuberosa DC) at 20µl of applied volume. Under UV 254nm wavelength was shows presence or absence of puerarin.

### Observation and results

### Organoleptic characters of crude samples

Table 1: Organoleptic characters of crude samples of Vidarikand

Parameters	Sample - Official	Sample -Lucknow	Sample - Mumbai	Sample - Delhi
Shape (Roopa)	Multi –angled, with multiple edges	cubical	Some pieces Cubical, some are hemisphere	Cubical
Size (Roopa)	1 to 5cm large, 1 to 4cm broad	2 to 5cm large, 2 to 4cm broad	3 to 7cm large, 2 to 5cm broad	2 to 4cm large, 2 to 4cm broad
Colour	Outer surface light	Light cream	Light cream	Light cream

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(Varna)	brown, cut surface dark brown (wet form white)			
Surface (Sparsha)	Epidermis smooth with warts, cut surface rough, fibrous	Smooth, some pieces fibrous on central region	Smooth, some pieces rough and fibrous	Smooth, some fibrous on central region
odour ( <i>Gandha</i> )	No particular smell (wet form slightly sweet)	Slightly sweet	Slightly sweet	Slightly sweet
Taste (Rasa)	Slightly sweet	No taste	No taste	No taste



Fig. 1 Crude samples of Vidarikand

# Organoleptic characters of powder

Table 2: Organoleptic characters of powder of Vidarikand samples

Parameters	Sample - Official	Sample -Lucknow	Sample - Mumbai	Sample - Delhi
Texture	Coarse, fibrous	Fine to coarse	Fine to coarse	Fine to coarse
Colour	Dark brown	Light cream	Light brown	Light cream
Odour	No particular smell	Sweet	Slightly sweet	Sweet
Taste	Slightly sweet and bitter	No taste	No taste	No taste
Touch	Rough	Smooth and Rough	Smooth and Rough	Smooth and Rough





Fig. 2 Powder of Vidarikand samples

# **Microscopic features**

**Table 3: Microscopic features** 

Parameters	Sample - Official	Sample -Lucknow	Sample - Mumbai	Sample - Delhi
Cork	Found outermost position	Not seen	Not seen	Not clear
Epidermis	Well developed	Not seen	Not seen	Not seen
Cortex region	5 to7 parenchy- matous cells layers	Not seen	Not seen	Partially seen
Endodermis	Well developed	Not seen	Not seen	Not seen
Crystals	In pericycle region	Not seen	Not seen	Present
Vascular bundle	Conjoint-collateral open	Concentric hadrocentric	Concentric hadrocentric and some pieces Conjoint collateral	Concentric hadrocentric
Medullary Rays	Broad and parenchymatous	Not seen	Not seen	Not seen

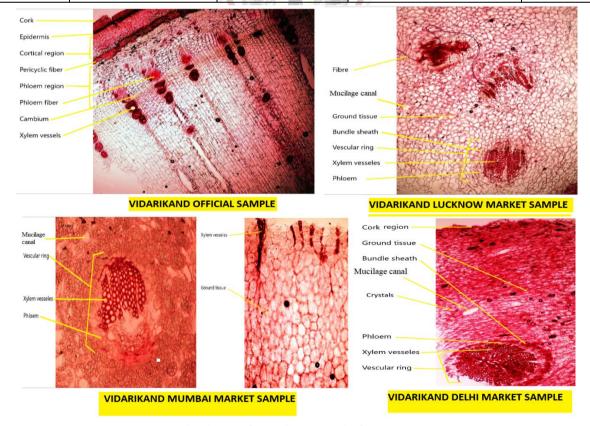
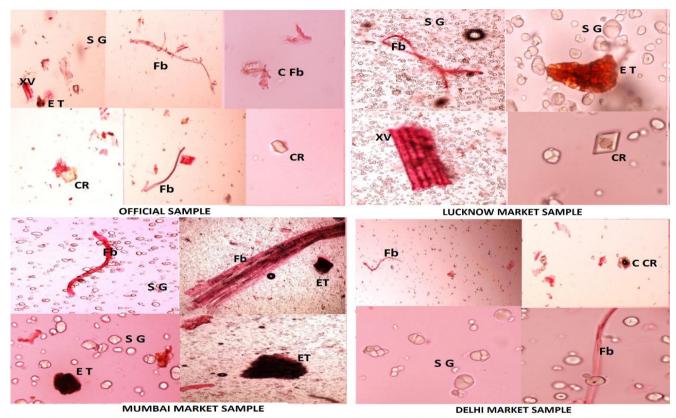


Fig. 3 Powder microscopic features

Table 4: Powder microscopic features

Characters	Sample - Official	Sample -Lucknow	Sample - Mumbai	Sample - Delhi
Fiber	present	Present	present	present
Crystal fiber	present	Not seen	Not seen	Not seen
Crystal	present	Present	Not seen	Cluster of crystal
Epidermal tissue	present	Present	present	Not seen
Xylem vessels	present	Present	Not seen	Not seen



Fb- fiber, C Fb- crystal fiber, S G- starch grain, CR- crystal, E T- epidermal tissue, C CR- cluster of crystal XV- xylem vessels

Fig. 4 Poweder microscopy

# Physicochemical analysis

Table 5: physicochemical analysis of all *Vidarikand* sample

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S.No.	Parameters	Sample - Official	Sample - Lucknow	Sample - Mumbai	Sample - Delhi	Standard as Per API
1.	Foreign Matter	Absent	1.8 %	Absent	1.4 %	Not more than 2%
2.	Total Ash Values	10.72 %	3.39 %	2.37 %	4.05 %	Not more than 11%
3.	Acid Insoluble Ash	1.11 %	0.21 %	0.25 %	0.46 %	Not less than 1%
4.	Water Soluble Ash	5.91 %	1.21 %	0.73 %	1.68 %	Not mentioned
5.	Alcohol Soluble Extractive Value	13.16 %	0.99 %	0.83 %	8.16 %	Not less than 13%
6.	WaterSoluble Extractive Value	28.16 %	13.66 %	4.49 %	19.91 %	Not less than 22%
7.	Moisture Content	7.06 %	9.88 %	11.97 %	8.47 %	Not more than 10%

# **Phytochemical screening**

### Test for water extract

Table 6: Test for water extract

Name of Tests	Sample - Official	Sample - Lucknow	Sample - Mumbai	Sample - Delhi
Flavonoids	+	+	+	+
Saponins	++	+	-	++
Tannins	-	•	-	-
Glycosides	+	+	-	+
Steroids	+	+	-	-
Terpenoids	+	+	-	-
Alkaloid (Hegar & Meyar)	+	+	+	+
Reducing Sugar	+	++	-	+
Resins	+	+	+	+

# **Test for Methanol extract**

**Table 7: Test for Methanol extract** 

Name of Tests	Sample - Official	Sample - Lucknow	Sample - Mumbai	Sample - Delhi
Flavonoids	+	+	+	+
Saponins	++	+	-	++
Tannins	++	-	-	-
Glycosides	++	++	+	+
Steroids	- 8	242	-	-
Terpenoids	- 8		-	-
Alkaloid (Hegar & Meyar)	mild+	mild +	mild +	+
Reducing Sugar	++		+	++
Resins	+		+	+

# Chromatographic study

Table 8:Chromatographic study - Presence of Purerain content in sample

rubic ordin omittogrupine study Tresence of rur crum content in sumpr				
Sample - Official	% of Purerain			
Sample - Official	55.41 microgram/ml			
Sample - Official	00			
Sample - Official	13.06 microgram/ml			
Sample - Official	00			

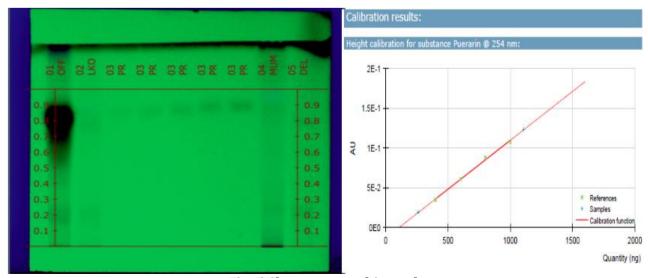


Fig. 5 Chromatographic study

### DISCUSSION

According to Table no.1 and Table no.2, we can say that organoleptically, all the market samples looked different in shape, size, colour, surface, odour and taste to official sample. Based on shape in table no.1 the Mumbai sample was look like two different drugs are mixed.

In table no.3 transverse section by compound microscope shows all the market samples looked different on the basis of type of vascular bundle, phloem and xylem structure to official sample. But in Mumbai sample was two different drugs was mix (shape, table no.1). On the basis of microscopic study some pieces were slightly match with official sample.

In table no.5, foreign matter found in samples in increasing, official and Mumbai< Delhi< Lucknow. Limit in API should not be more than 2%. Foreign matter is under limit in all samples.

The ash value is indicator of presence of inorganic and earthy material in the drug sample.

Total ash content found in samples in increasing, Mumbai<Lucknow<Delhi<Official. Limit in API- should not be more than 11%. Total ash value of all samples in limit of API.

The acid insoluble content indicates the presence of siliceous matter. Acid insoluble ash content found in different samples in increasing order, Lucknow<Mumbai<Delhi<Official. Limit in API should not be less than 1%. Acid insoluble ash value of official sample (1.11%) was within the limit of API. Acid insoluble ash value of Lucknow, Mumbai and Delhi samples were less than the limit of API.

Water soluble ash value is useful in determining authenticity and purity of sample and also these values are important qualitative standards. Limit of water soluble ash API has not mentioned. Water soluble ash content found in different samples in increasing order, Mumbai< Lucknow<Delhi<Official. Maximum percentage of water soluble ash value was found in official sample.

Extractive value is amount of active constituents in a given amount of plant material when extracted with a particular solvent whether the crude drug is exhausted or not.

Limit of Alcohol soluble extractive value in API should not be less than 13%. Alcohol soluble extractive value content found in different samples in increasing order, Mumbai<Lucknow<Delhi<Official. Alcohol soluble extractive value of official sample (13.16%) was within the limit of API. Alcohol soluble extractive value of Lucknow,

Mumbai was very less and Delhi samples were less than the limit of API.

Limit of water-soluble extractive valuein API should not be less than 22%. Water soluble extractive value content found in different samples in increasing order, Mumbai<Lucknow<Delhi< Official. Water soluble extractive value of official sample (28.16%) was within the limit of API. Water soluble extractive value of Lucknow. Mumbai was very less and a Delhi sample was less than the limit of API. In all the samples water soluble extractive values are more than alcohol extractive values which indicates the presence of more amounts of water soluble contents in the plant. Alcohol and water soluble extractive values of market samples are very less relative to genuine/official sample. Extractive values of Mumbai and Lucknow samples were very poorer.

Moisture content in drug encourages microbial growth, a presence of fungi or insects and deterioration following hydrolysis. Limit of water content should therefore be set for every given plant material. Limit in API should not be more than 10%. Moisture content found in different samples in increasing order, Official<Delhi<Lucknow<Mumbai. Moisture content of Official, Delhi and Lucknow sample were within the limit of API. Moisture content of Mumbai sample was higher the limit of API.

Identification of phytochemicals indicates pharmacological active metabolites present in the plant.

Phytochemical screening of water extract the genuine/official sample revealed the presence of flavonoids, saponins, glycosides, steroids, terpenoids, alkaloid, reducing sugar and resins. Lucknow sample showed the presence of all those phytochemicals which were present genuine/official sample. Mumbai sample was different with Official sample, because it showed the absence of saponins, glycosides, terpenoids and reducing sugar. While flavonoids, alkaloid and resins were present in Mumbai sample which was just common of genuine/official sample. Delhi sample was different with Official sample; because it showed the absence of steroids and terpenoids. While present of flavonoids, saponins, glycosides, alkaloid, reducing sugar and resins which was just common of genuine/official sample. Phytochemical screening of water extract found in different samples in increasing order, Mumbai< Delhi<Lucknow and Official (Table no.6).

Phytochemical screening of methanolic extract the genuine/official sample revealed the of flavonoids, saponins, glycosides, alkaloid, reducing sugar and resins. Lucknow and Delhi sample showed the presence of all those phytochemicals which were present in genuine/official sample, except tannins. Mumbai sample showed the presence of all those phytochemicals which present were genuine/official sample, except saponins and tannins. Phytochemical screening of methanolic extract found in different samples in increasing order, Mumbai<Delhi and Lucknow<Official (Table no.7).

HPTLC finger printing of all samples of *Vidarikand* at 20µl of applied volume under UV 254nm wavelength. The reference marker Puerarin was found only two samples Official (Rf 0.823 to 0.935) and Mumbai (Rf 0.827 to 0.937). Delhi and Lucknow sample were not found Puerarin. Quantification of Puerarin by HPTLC in *Vidarikand* was carried out on the basis of calibration curve of standard. The amount of Puerarin was found to be  $55.41\mu g/ml$  and  $13.06\mu g/ml$ , in Official and Mumbai sample respectively. Amount of Puerarin maximum  $55.41\mu g/ml$  in genuine/official sample (Table no.8).

#### CONCLUSION

On the basis of above discussion and result it has been concluded that the presence of marker puerarin in the genuine/official sample of *Vidarikand* and this sample fulfil the parameters of API, that means genuine/official sample of Vidarikand was pure and genuine. The sample of Lucknow and Delhi market were not similar to genuine/official sample, they were totally replaced other drug, because of organoleptic, with microscopic, physico-chemical observation and absence of marker Puerarin in HPTLC, they were totally different to genuine/official sample. On the basis of organoleptically and microscope character it has been concluded that the sample of Mumbai market was mixer of two different drugs. Microscopic character (Some pieces) and the basis of HPTLC (presence of marker Puerarin) Mumbai market sample slightly similar with genuine/official sample. That means Mumbai sample was mixer of two different drugs one was Vidarikand and second was another adulterated drug.

Hence, standardization and quality specifications of herbal drug market samples are a matter of concern. Condition of crude herbal drug market is very poor.

Success of Ayurvedic therapy relies on effectiveness of plant raw materials which directly

would depend on their authenticity, quality and safety. Classical methods of identification help usually morphology of plant but not for raw drug. Thus, identification of raw drug should with the modern scientific methods of pharmacognosy. Present time needs attention and promotion of medicinal plant cultivation, for authentic and quality raw material. That is necessary for its long term stability and prevention of adulteration and substitution.

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