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A comparative pharmacognostical evaluation of *Dhataki pushpa* (*Woodfordia fruticosa* (L.) Kurz.) as a substitute for *Yashtimadhu moola* (*Glycyrrhiza glabra* Linn.)

***Dr. Ibameaimon Pohtam[1], Dr Anuradha KN[2]**
** [1]PG Scholar, [2]Assistant Professor*
Department of Dravyaguana,
Sri Dharmasthala Manjunatheswara College of Ayurveda & Hospital, Hassan, Karnataka

Abstract

: *Glycyrrhiza glabra* Linn. is one of the most widely used herb due to its high value index in food grade, pharmaceutical grade , cosmetic grade, feed grade and other fibres materials. The part used is root which is a destructive form of harvesting. To meet the high demand the drug is over exploited and also adulterated with roots of *Abrus precatorius* Linn and roots of *Glycyrrhiza uralensis* Fish (Manchurian liquorice). Stem pieces of *Glycyrrhiza glabra* Linn. are also sold in place of root. *Ayurvedic* literature mentions *Dhatakipushpa* as the substitute of *Yashtimadhumoola. Dhataki* (*Woodfordia fruticosa* (L.) Kurz.) is abundantly found throughout India. Thus to prevent adulteration as well to maintain the quality of the drug the present study is taken up to evaluate Dhataki (Woodfordia fruticosa (L.) Kurz.) as abhava pratinidhi dravya for Yashtimadhu (Glycyrrhiza glabra Linn.). This study aims at comparative pharmagonostic evaluation of root of Yashtimadhu (Glycyrrhiza glabra Linn.) and flower of Dhataki (Woodfordia fruticosa (L.) Kurz.).

Materials and Method : Flowers of *Woodfordia fruticosa* (L.) Kurz. was collected from Botanical garden of *Shri Dharmasthala Manjunatheshwara (SDM) college of Ayurveda and Hospital, Hassan* . The roots of *Glycyrrhiza glabra* Linn. was collected from market sample of Kajrekhar Pharmacy, Belgaum, Karnataka. Both roots of Yashtimadhu (Glycyrrhiza glabra Linn.) and flower of Dhataki (Woodfordia fruticosa (L.) Kurz.) were subjected to macroscopic, microscopic, physicochemical, preliminary phytochemical and HPTLC evaluation as per standard method.

Results: Carbohydrate, Tannin, Flavanoids, Saponins, Phenols, Carboxylic acid, Quinone are common chemical constituents in both Glycyrrhiza glabra Linn and Woodfordia fruticosa (L.) Kurz. Alkaloids , Steroids, Triterpenoid, Resins are present only in Glycyrrhiza glabra. Coumarins, Amino acids are absent in both Glycyrrhiza glabra Linn and Woodfordia fruticosa (L.) Kurz. Total 17 numbers of active components were detected in Glycyrrhiza glabra Linn. Total 26 number of active components were detected in Woodfordia fruticosa (L.) Kurz.

Conclusion: The pharmacognostic evaluation reveals various chemical constituents that are common in both the drugs. The results from HPTLC finger prints, scanned at various wavelengths revealed that there are some similarities in both drugs i.e. *Glycyrrhiza glabra* Linn & *Woodfordia fruticosa* (L.) Kurz. which detected the common active component.

Keywords: Glycyrrhiza glabra Linn.; Woodfordia fruticosa (L.) Kurz.; Pharmacognosy, Preliminary phytochemical , HPTLC

INTRODUCTION

Yashtimadhu (*Glycyrrhiza glabra* Linn.) belonging to Papilionaceae family is one of the most widely used herb due to its high value index in food grade, pharmaceutical grade, cosmetic grade, feed grade and other fibres material.[1] Its active constituents Glycyrrhizine cannot be produced synrthetically.[2] The drug is over exploited and adulterated to meet its high demand.[3] R.N Chopra's list listed the drugs as threatened/ depleted.[4] In red data book *Glycyrrhiza glabra* Linn has been placed as an endangered medicinal plant.[5] *Ayurvedic* literature mentions *Dhataki pushpa* (*Woodfordia fruticosa* (L.) Kurz.) as the substitute of *Yashtimadhu moola* (*Glycyrrhiza glabra* Linn.).[6-8] *Yashtimadhu* (*Glycyrrhiza glabra* Linn) is a hard, semi-perennial, erect herb or under shrub growing to a height of upto 1.7m. It has a characteristic pleasant taste.[9-11] It is distributed in the Sub- tropical and warm temperate regions of the world, chiefly in the Mediterranean countries, South Europe, Asia Minor, Egypt, Turkistan, Iran, Siberia, Persia, Arab countries and Afghanistan.[12] Medicinal uses of licorice includes cough suppression,[13] gastric ulcer treatment,[14] treatment of early Addison disease,[15,16] treatment of liver disease and as a laxative.[17,18]

Dhataki (*Woodfordia fruticosa* (L.) Kurz) belonging to Lythraceae family is abundantly present throughout India and also in a majority of the countries of South East and Far East Asia like Malaysia, Indonesia, Sri Lanka, China, Japan and Pakistan as well as Tropical Africa.[19,20,21] It is extensively used in the preparation of "Asavas and Arishtas" containing self generated alcohol. The flowers of *Woodfordia fruticosa* (L.) Kurz. are brilliant red in colour are commonly used for the treatment of several ailments which includes rheumatism, leucorrhea, menorrhagia, asthma, liver disorder, and inflammatory conditions.[22]

MATERIALS AND METHODS

Plant Materials

Flowers of *Woodfordia fruticosa* (L.) Kurz. was collected from Botanical garden of *Shri Dharmasthala Manjunatheshwara (SDM) College of Ayurveda and Hospital, Hassan* . The roots of *Glycyrrhiza glabra* Linn. was collected from market sample of Kajrekhar Pharmacy, Belgaum, Karnataka. The authentication was done at Department of *DravyaGuna, Shri Dharmasthala Manjunatheshwara (SDM) college of Ayurveda and Hospital, Hassan, Karnataka.*

Macroscopy evaluation

The external features of the test samples were documented using Canon IXUS digital camera. The macroscopic features were compared to local flora for authentication.

Microscopy evaluation

Sample was preserved in fixative solution. The fixative used was FAA (Formalin-5ml + Acetic acid-5ml + 70% Ethyl alcohol-90ml). The materials were left in FAA for more than 48 hours. The preserved specimens were cut into thin transverse section using a sharp blade and the sections were stained with safranin. Transverse sections were photographed using Zeiss AXIO trinocular microscope attached with Zeiss AxioCam camera under bright field light. Magnifications of the figures are indicated by the scale-bars.

Physico- chemical parameters

Physico- chemical constants of both test drug Yashtimadhu (*Glycyrrhiza glabra* Linn.) and Dhataki (*Woodfordia fruticosa* (L.) Kurz.) like moisture content, total ash, acid insoluble ash, water soluble ash, aqueous and ethanol soluble extractives were calculated following the procedures recommended by standard text books.[23]

Preliminary phytochemical tests

The phytochemical screening for the detection of organic and inorganic constituents like test for carbohydrates, alkaloids, steroids, saponins, tannins, flavonoids, phenol, coumarins, triterpenoids, amino acids, carboxylic acid, resin, quinone were following the procedures recommended by standard textbooks.[24]

HPTLC

1g of each of *Yastimadhu* (*Glycyrrhiza glabra* Linn.) root coarse powder and flower of *Dhataki* (*Woodfordia fruticosa* (L.) Kurz.) were extracted with 10 ml of alcohol. 4µl, 8µl and 12µl of the above extract was applied on a pre-coated silica gel F_{254} on aluminum plates to a band width of 7 mm using Linomat 5 TLC applicator. The plate was developed in n-Butanol: Water: Glacial acetic acid (7.0: 2.0: 1.0) for *Yastimadhu* (*Glycyrrhiza glabra* Linn.) and *Dhataki* (*Woodfordia fruticosa* (L.) Kurz.) Toluene: Chloroform: Ethyl Acetate: Formic acid (2.0: 6.0: 6.0: 2.0); Toluene: Ethyl Acetate: Methanol: Formic acid (3.0: 3.0: 0.8: 0.2). The developed plates were visualized in short UV, long UV and then derivatised with vanillin sulphuric acid (post derivatisation under white light). Subsequently scanned under UV 254nm, 366nm and 620nm (after derivatisation). R_f, colour of the spots and densitometric scan were recorded.

RESULTS AND OBSERVATIONS

Macroscopic evaluation

Figure 1: Macroscopy of Yashtimadhu (Glycyrrhiza glabra Linn) root

Table No 1: Showing macroscopic evaluation of root of *Yashtimadhu* (*Glycyrrhiza glabra* Linn.)

Root of *Yashtimadhu* (*Glycyrrhiza glabra* Linn.)	
Appearance	Cylindrical, longitudinally wrinkled pieces, transversely cut and smoothened surface
Texture	Rough
Colour	Yellowish brown
Taste	Sweet
Odour	Characteristic odour

Microscopic evaluation

Root of *Glycyrrhiza glabra* Linn.

Transverse section

Cork: 10-20 or more layers of tabular cells, outer layers with reddish brown amorphous contents, inner 3 or 4 rows having thicker, colourless walls

Secondary Cortex: usually of 1-3 layers of radially arranged parenchymatous cells

Phloem: a broad band, parenchymatous cells of inner part cellulosic and outer lignified;

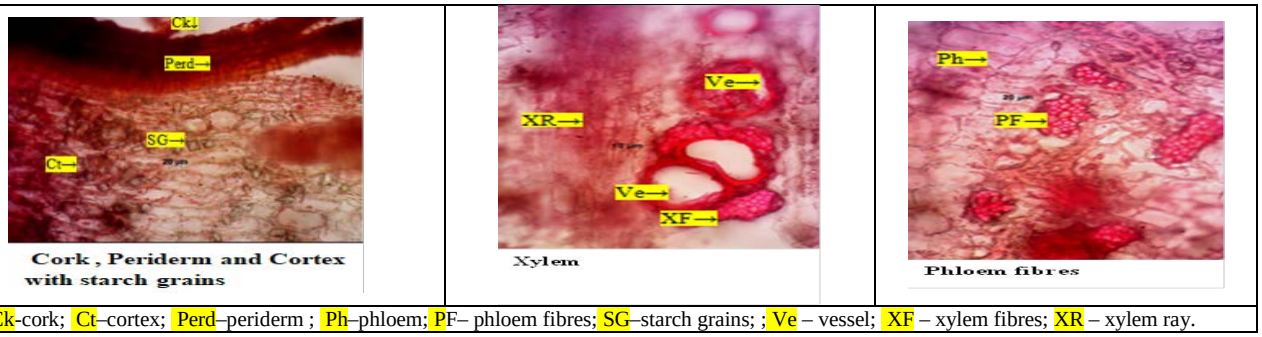
radially arranged groups of about 10 or 50 fibre.

Cambium: form a tissue of 3 or more layers of cells

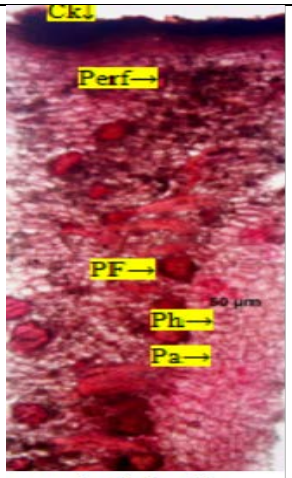
Secondary xylem: distinctly radiate with medullary rays, 3 or 5 cells wide, yellow, pitted, reticulately thickened walls, groups of lignified fibres with crystal sheaths similar to those of phloem

Pith: parenchymatous cells in longitudinal rows, with inter- cellular spaces

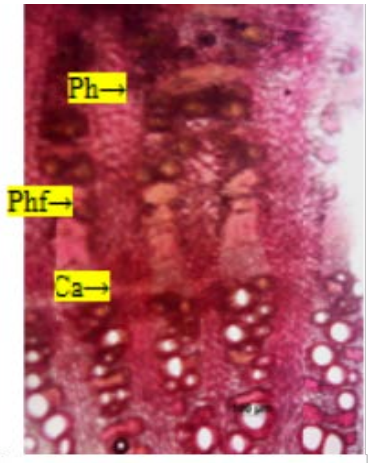
Figure 2 : Microscopy of Yashtimadhu (*Glycyrrhiza glabra* Linn) root



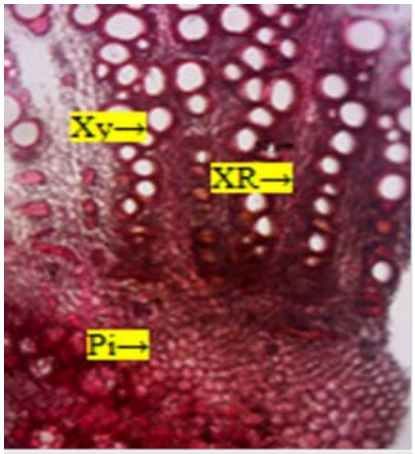
Ck-cork; Ct–cortex; Perd–periderm ; Ph–phloem; PF– phloem fibres; SG–starch grains; ; Ve – vessel; XF – xylem fibres; XR – xylem ray.



T.S of Yashtimadhu root



Phloem, Cambium, Xylem



Xylem and Pith

Ca – cambium; Ck – cork; Pa – parenchyma; PF – phloem fibres; Ph – phloem; Perf – pericyclic fibre; Xy–xylem; XR – xylem ray; Pi - Pith

Physico- chemical parameters

Table No – 2: Showing Physico- chemical parameter of *Yashtimadhu* (*Glycyrrhiza glabra* Linn)

	Standard value	Obtained value
Foreign matter	Not more than 2%	absent
Loss on drying	Not more than 12%	11.8%
Total Ash	Not more than 10%	7.5%
Acid Insoluble Ash	Not more than 2.5%	1%
Alcohol soluble extractive value	Not less than 10%	10%
Water soluble extractive value	Not less than 20%	31%

Preliminary Phytochemicals:

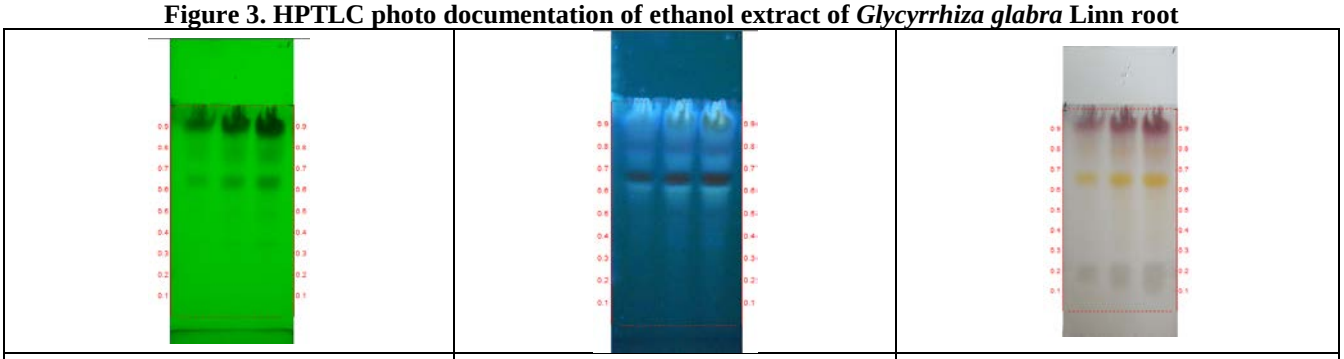
Table No – 3: Showing results of preliminary phytochemical screening of aqueous extract

Test	**Inference**
	***Glycyrrhiza glabra* Linn. (Aqueous extract)**
Alkaloid	+
Steroid	+
Carbohydrate	+
Tannin	+
Flavanoids	+
Saponins	+
Tri terpenoid	+
Coumarins	-
Phenols	+
Carboxylic acid	+
Amino acids	-
Resin	+
Quinone	+

(+) - present; (-) – negative

HPTLC

Figure 3. HPTLC photo documentation of ethanol extract of *Glycyrrhiza glabra* Linn root



Short UV	**Long UV**	**After derivatisation**

Solvent system – n-butanol: Water: Glacial acetic acid (7.0: 2.0: 1.0)

Track 1 – Yashtimadhu root – 4µl ; Track 2 – Yashtimadhu root – 8µl; Track 2 – Yashimadhu root – 12µl

Table No - 4: R_f values of sample of *Glycyrrhiza glabra* Linn root

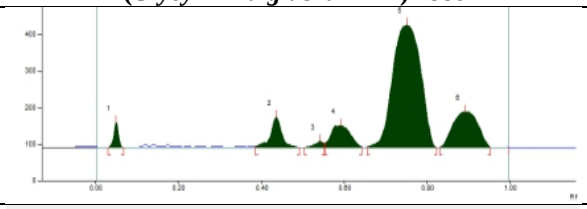
Solvent system – n-butanol: Water: Glacial acetic acid (7.0: 2.0: 1.0)

Short UV	Long UV	After derivatisation
		0.20 (Purple)
0.35 (L. green)	-	-
-	0.37 (F. blue)	-
0.49 (L. green)	-	-
-	0.61 (F. blue)	-
0.65 (D. green)	0.65 (F. blue)	0.65 (Yellow)
-	0.74 (F. blue)	-
0.78 (D. green)	0.78 (FD. blue)	0.78 (Yellow)

***D – dark; L – light; F – fluorescent**

In *Glycyrrhiza glabra* Linn. {Solvent system – n-butanol: Water: Glacial acetic acid (7.0: 2.0: 1.0)] - 8 spots were detected in different Rf value.

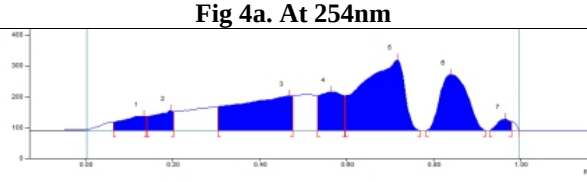
Figure 4. Densitometric scan of Yashtimadhu (*Glycyrrhiza glabra* Linn) root



Track 3, ID: Yastimadhu root extract

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.03 Rf	0.0 AU	0.05 Rf	67.9 AU	10.31 %	0.07 Rf	0.1 AU	543.0 AU	2.17 %
2	0.39 Rf	4.3 AU	0.44 Rf	82.0 AU	12.46 %	0.49 Rf	0.1 AU	1627.9 AU	6.51 %
3	0.50 Rf	0.2 AU	0.54 Rf	16.5 AU	2.51 %	0.55 Rf	11.5 AU	254.2 AU	1.02 %
4	0.55 Rf	11.9 AU	0.59 Rf	60.0 AU	9.12 %	0.64 Rf	2.0 AU	2025.6 AU	8.10 %
5	0.66 Rf	2.0 AU	0.75 Rf	333.8 AU	50.72 %	0.82 Rf	0.2 AU	16238.6 AU	64.97 %
6	0.83 Rf	0.5 AU	0.89 Rf	98.0 AU	14.88 %	0.95 Rf	0.7 AU	4305.3 AU	17.22 %

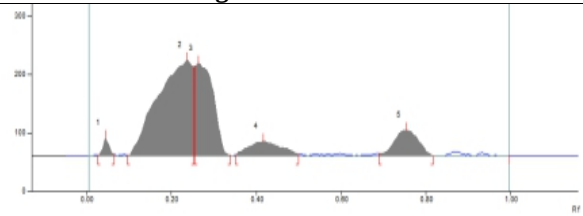
Fig 4a. At 254nm



Track 3, ID: Yastimadhu root extract

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.06 Rf	27.2 AU	0.13 Rf	46.3 AU	5.83 %	0.14 Rf	45.1 AU	1832.5 AU	4.25 %
2	0.14 Rf	45.2 AU	0.20 Rf	64.8 AU	8.17 %	0.20 Rf	60.9 AU	2103.5 AU	4.87 %
3	0.30 Rf	77.4 AU	0.47 Rf	112.6 AU	14.18 %	0.48 Rf	11.7 AU	10126.4 AU	23.47 %
4	0.53 Rf	112.5 AU	0.56 Rf	124.7 AU	15.71 %	0.60 Rf	12.3 AU	4764.1 AU	11.04 %
5	0.60 Rf	112.5 AU	0.72 Rf	228.6 AU	28.79 %	0.77 Rf	0.0 AU	15467.8 AU	35.85 %
6	0.78 Rf	0.0 AU	0.84 Rf	180.6 AU	22.75 %	0.92 Rf	0.1 AU	8075.3 AU	18.71 %
7	0.93 Rf	0.3 AU	0.97 Rf	36.3 AU	4.58 %	0.98 Rf	28.2 AU	780.1 AU	1.81 %

Fig 4b. At 366nm



Track 3, ID: Yastimadhu root extract

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.03 Rf	1.7 AU	0.05 Rf	30.3 AU	7.22 %	0.07 Rf	2.6 AU	327.4 AU	1.80 %
2	0.10 Rf	0.8 AU	0.24 Rf	163.4 AU	38.96 %	0.25 Rf	52.5 AU	9619.2 AU	53.01 %
3	0.25 Rf	152.7 AU	0.26 Rf	157.7 AU	37.60 %	0.34 Rf	0.8 AU	4983.3 AU	27.46 %
4	0.35 Rf	2.0 AU	0.42 Rf	24.5 AU	5.85 %	0.50 Rf	3.1 AU	1393.2 AU	7.68 %
5	0.69 Rf	3.8 AU	0.75 Rf	43.5 AU	10.37 %	0.82 Rf	0.1 AU	1821.7 AU	10.04 %

Fig 4c. At 620nm

HPTLC Densitometric Scan

Total 17 numbers of active components were detected in *Glycyrrhiza glabra* Linn. having Rf value (0.05, 0.44, 0.54, 0.59, 0.75, 0.89, 0.13, 0.20, 0.47, 0.56, 0.72, 0.84, 0.97, 0.24, 0.26, 0.42, 0.75)

Under short UV (254nm) , 6 peaks were detected . Among them maximum percentage of area were occupied by Rf 0.75 (50.72%), 0.89 (14.88%), 0.44 (12.46%).

Under long UV (366nm), 7 peaks were detected . Among them maximum percentage of area were occupied by Rf 0.72 (28.79%), 0.84 (15. 71%).

After derivatization (620nm), 5 peaks were detected. Among them maximum percentage of area were occupied by Rf 0.24 (38.96%) , 0.26 (37.60%)

***Dhataki* (*Woodfordia fruticosa* (L.) Kurz)**

Macroscopic evaluation

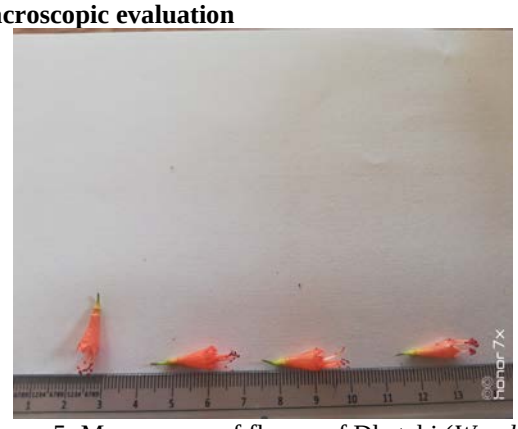


Figure 5: Macroscopy of flower of Dhataki (*Woodfordia forticosa* (L.) Kurz

Table No - 5. Macroscopic evaluation of Flower of *Dhataki* (*Woodfordia fruticosa* (L.) Kurz)

Appearance	Flower is about 1.2 cm long, occurs as singles or in bunches; with campanulate base and oblique apex; very minute accessory sepals attached outside at the juncture of calyx tooth
Texture	Smooth, Glabrous
Colour	Scarlet red in colour
Taste	Astringent
Odour	Characteristic odour

Microscopic evaluation

Flowers of *Woodfordia fruticosa* (L.) Kurz.

Transverse section of stalk shows an epidermis formed by thick wall cells with cuticle; 2 - 3 layers of collenchyma; cortex formed by 4 - 5 layers of reticulate parenchyma; inner to cortex continuous ring of phloem followed by xylem. The canter is occupied by pith formed by pitted parenchyma

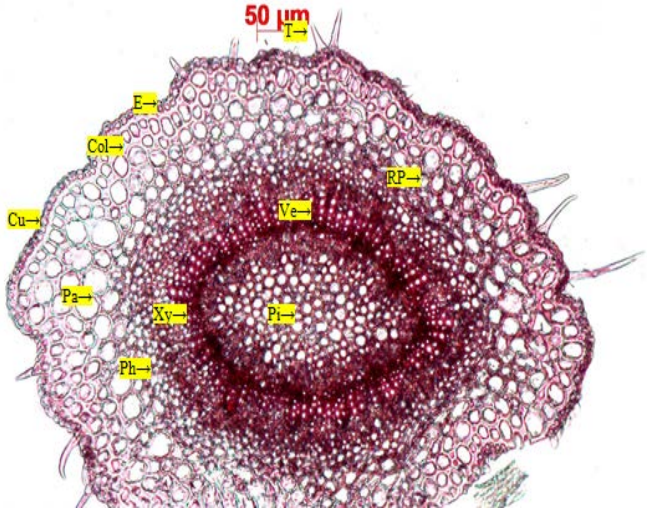
Transverse section of Calyx shows upper and lower epidermis with few covering trichomes, present of vascular bundle. Beneath the upper epidermis shows layer of palisade cells. The lower epidermis having spongy parenchyma.

Transverse section of Anther shows epidermis. Underneath the epidermis shows presence of pollen grains

Transverse section of Ovary shows an epidermis with covering trichome; cortex shows group of thin walled

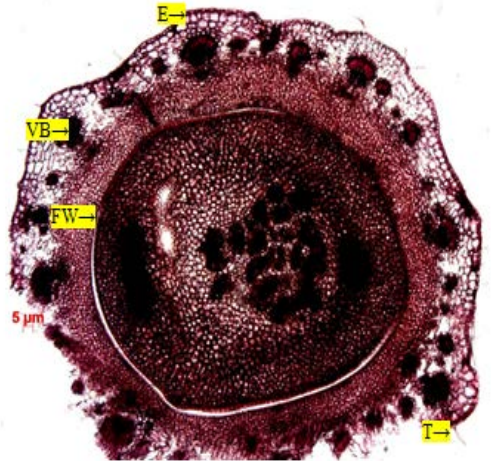
fibers; rudimentary cells which would form fruit wall and seed wall are seen. The central portion is formed by thin walled parenchyma which shows pith like tissue of the axis of the ovary.

Figure 6: Microscopy of flower of Dhataki (*Woodfordia fruticosa* (L.) Kurz



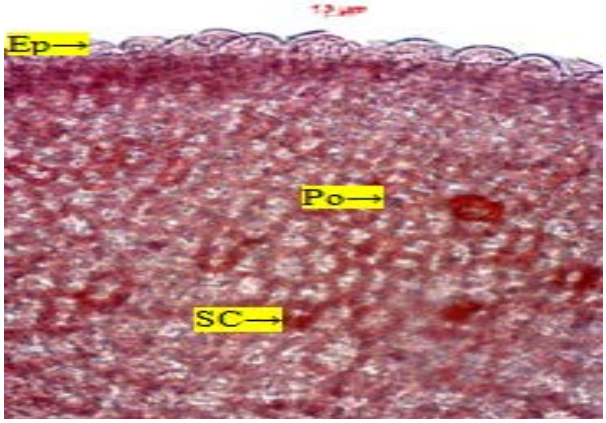
Transverse section of stalk

Col – collenchymas; Ct – cortex; Cu – cuticle; Ep – epidermis; Pa – parenchyma; Ph – phloem; Pi – pith; RP – reticulated parenchyma; T – trichome; Ve – vessels; Xy – xylem.



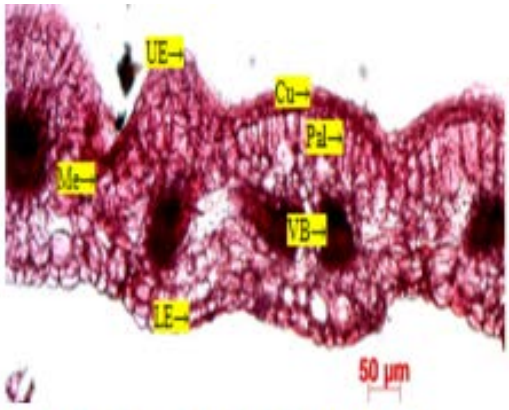
TS of flower through ovary

E–epidermis; FW–fruit wall; T–trichomes; VB–vascular bundle

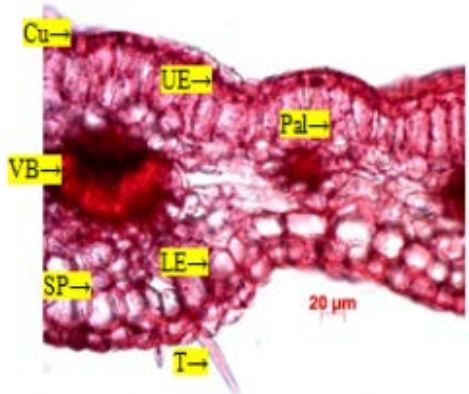


TS of Anther

Ep – epidermis; Po – pollen sac with pollen grains; SC – sclerified layer.



Transverse section of Calyx



A portion enlarged

Cu – cuticle; LE – lower epidermis; Me – mesophyll; Pal – palisade cells; SP – spongy parenchyma; T –trichome; UE – upper epidermis; VB – vascular bundle.

Physico-chemical parameters

Table No – 6: Showing Physiochemical parameter of *Woodfordia fruticosa* (L.) Kurz

	Standard value	Obtained value
Foreign matter	Not more than 2%	0.17%
Loss on drying	-	13.1 %
Total Ash	Not more than 10%	6%
Acid Insoluble Ash	Not more than 0.5%	8%
Alcohol soluble extractive value	Not less than 8 %	18%
Water soluble extractive value	Not less than 20%	41%

Preliminary Phytochemicals:

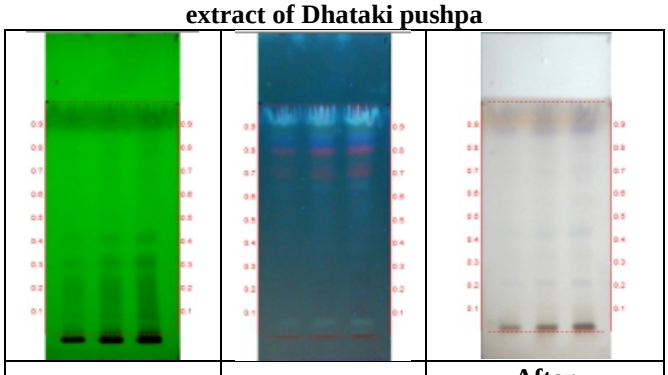
Table No – 7: Showing Results of preliminary phytochemical screening of aqueous extract

Test	Inference
	***Woodfordia fruticosa* (L.) Kurz (Aqueous extract)**
Alkaloid	-
Steroid	-
Carbohydrate	+
Tannin	+
Flavanoids	+
Saponins	+
Tri terpenoid	-
Coumarins	-
Phenols	+
Carboxylic acid	+
Amino acids	-
Resin	-
Quinone	+

(+) - present; (-) – negative

HPTLC

Figure 7. HPTLC photo documentation of ethanol extract of Dhataki pushpa



Short UV	Long UV	After derivatisation

Solvent system - Toluene: Chloroform: Ethyl Acetate: Formic acid (2.0: 6.0: 6.0: 2.0)

Track 1 – **Dhataki pushpa**– 4µl

Track 2 – **Dhataki pushpa**– 8µl

Track 2 – **Dhataki pushpa**– 12µl

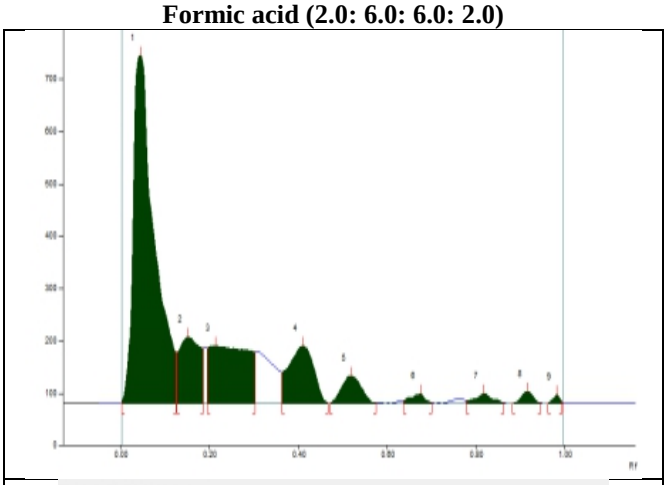
In *Woodfordia fruticosa* (L.) Kurz [Solvent system - Toluene: Chloroform: Ethyl Acetate: Formic acid (2.0: 6.0: 6.0: 2.0)] - 13 spots were detected in different Rf value.

Table No – 8: R_f values of sample of Dhataki pushpa (*Woodfordia fruticosa* (L.) Kurz) Solvent system - Toluene: Chloroform: Ethyl Acetate: Formic acid (2.0: 6.0: 6.0: 2.0)

Short UV	Long UV	After derivatisation
0.08 (Green)	-	-
-	0.06 (F. blue)	-
-	-	0.21 (Purple)
0.32 (Green)	-	-
0.42(Green)	-	-
-	-	0.44 (Purple)
-	-	0.57 (Purple)
-	0.65 (F. blue)	-
0.70 (Green)	0.70 (F. red)	0.71 (Purple)
0.78 (Green)	-	-
-	0.81 (F. red)	-
0.84 (Green)	0.84 (F. blue)	-
-	0.87 (F. blue)	-

***D – dark; L – light; F – fluoresce**

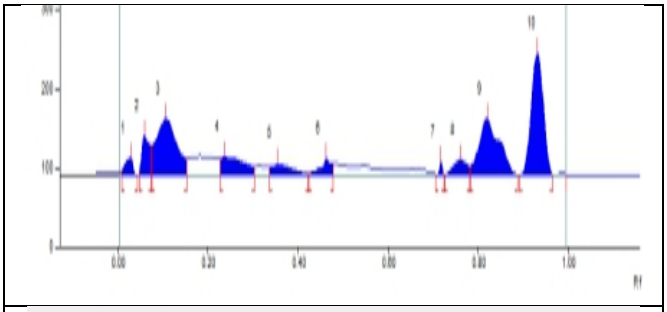
Figure 8. Densitometric scan of Dhataki pushpa (*Woodfordia fruticosa* (L.) Kurz) Solvent system - Toluene: Chloroform: Ethyl Acetate: Formic acid (2.0: 6.0: 6.0: 2.0)



Track 3, ID: Dhataki pushpa

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.00 Rf	3.3 AU	0.05 Rf	664.7 AU	58.24 %	0.13 Rf	98.5 AU	22746.7 AU	54.08 %
2	0.13 Rf	98.5 AU	0.15 Rf	127.3 AU	11.15 %	0.19 Rf	06.4 AU	4360.1 AU	10.37 %
3	0.20 Rf	106.8 AU	0.21 Rf	109.9 AU	9.63 %	0.30 Rf	99.1 AU	7141.5 AU	16.98 %
4	0.36 Rf	60.2 AU	0.41 Rf	109.5 AU	9.59 %	0.47 Rf	0.4 AU	4529.3 AU	10.77 %
5	0.47 Rf	0.2 AU	0.52 Rf	52.7 AU	4.62 %	0.58 Rf	1.6 AU	1702.8 AU	4.05 %
6	0.64 Rf	4.7 AU	0.68 Rf	18.6 AU	1.63 %	0.70 Rf	2.1 AU	431.3 AU	1.03 %
7	0.78 Rf	5.0 AU	0.82 Rf	18.9 AU	1.66 %	0.86 Rf	1.4 AU	522.3 AU	1.24 %
8	0.88 Rf	0.3 AU	0.92 Rf	23.2 AU	2.03 %	0.95 Rf	1.0 AU	446.5 AU	1.06 %
9	0.96 Rf	0.3 AU	0.98 Rf	16.5 AU	1.44 %	1.00 Rf	4.3 AU	183.0 AU	0.43 %

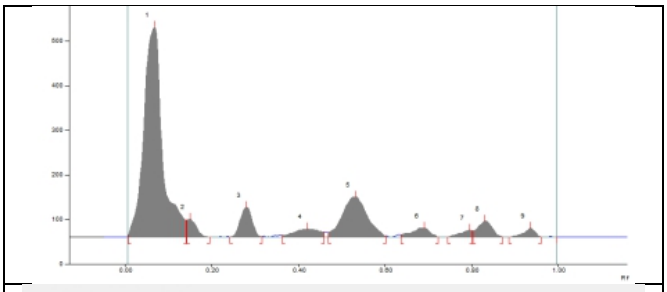
Fig 8a. At 254nm



Track 3, ID: Dhataki pushpa

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.01 Rf	5.4 AU	0.03 Rf	21.2 AU	4.63 %	0.04 Rf	0.5 AU	269.6 AU	2.57 %
2	0.05 Rf	3.1 AU	0.06 Rf	49.5 AU	10.79 %	0.07 Rf	36.7 AU	623.3 AU	5.94 %
3	0.08 Rf	36.9 AU	0.11 Rf	70.5 AU	15.37 %	0.15 Rf	20.7 AU	2296.3 AU	21.87 %
4	0.23 Rf	20.7 AU	0.24 Rf	23.6 AU	5.15 %	0.30 Rf	10.5 AU	846.3 AU	8.06 %
5	0.34 Rf	10.4 AU	0.35 Rf	13.5 AU	2.94 %	0.42 Rf	2.1 AU	436.6 AU	4.16 %
6	0.42 Rf	2.2 AU	0.46 Rf	21.0 AU	4.59 %	0.48 Rf	13.8 AU	342.0 AU	3.26 %
7	0.71 Rf	1.0 AU	0.72 Rf	17.0 AU	3.71 %	0.73 Rf	0.4 AU	75.0 AU	0.71 %
8	0.73 Rf	0.6 AU	0.76 Rf	18.5 AU	4.02 %	0.78 Rf	10.9 AU	407.6 AU	3.86 %
9	0.78 Rf	11.1 AU	0.82 Rf	70.3 AU	15.32 %	0.89 Rf	0.1 AU	2250.0 AU	21.43 %
10	0.89 Rf	0.1 AU	0.93 Rf	153.5 AU	33.48 %	0.97 Rf	0.7 AU	2951.3 AU	28.11 %

Fig 8b. At 366nm



Track 3, ID: Dhataki pushpa

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.01 Rf	3.7 AU	0.07 Rf	469.5 AU	60.28 %	0.14 Rf	38.1 AU	14714.7 AU	62.87 %
2	0.14 Rf	38.3 AU	0.15 Rf	40.6 AU	5.22 %	0.20 Rf	0.0 AU	611.2 AU	2.61 %
3	0.24 Rf	0.3 AU	0.28 Rf	67.1 AU	8.61 %	0.32 Rf	1.1 AU	1319.5 AU	5.64 %
4	0.36 Rf	3.6 AU	0.42 Rf	18.2 AU	2.34 %	0.46 Rf	11.7 AU	785.3 AU	3.36 %
5	0.47 Rf	10.8 AU	0.53 Rf	90.1 AU	11.57 %	0.60 Rf	2.8 AU	3752.7 AU	16.03 %
6	0.64 Rf	5.3 AU	0.69 Rf	20.5 AU	2.63 %	0.72 Rf	0.0 AU	646.5 AU	2.76 %
7	0.75 Rf	0.1 AU	0.80 Rf	16.0 AU	2.05 %	0.80 Rf	14.1 AU	320.3 AU	1.37 %
8	0.80 Rf	14.1 AU	0.83 Rf	36.6 AU	4.70 %	0.87 Rf	0.6 AU	868.0 AU	3.71 %
9	0.89 Rf	0.0 AU	0.94 Rf	20.2 AU	2.59 %	0.96 Rf	0.4 AU	386.3 AU	1.65 %

Fig 8c. At 620nm

HPTLC Densitometric scan

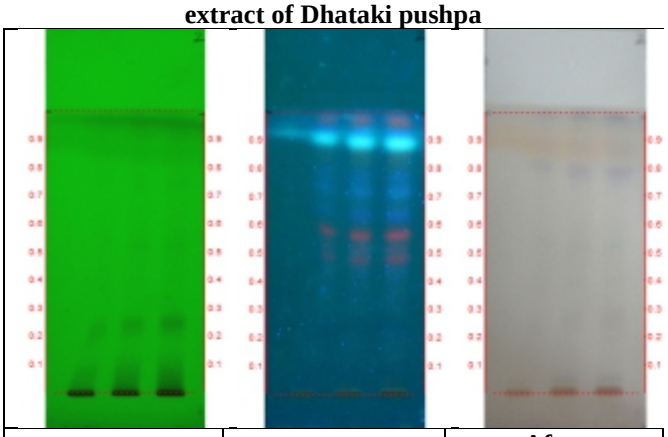
Total 26 number of active components were detected in *Woodfordia fruticosa* (L.) Kurz having Rf value (0.05, 0.15, 0.21, 0.41, 0.52, 0.68, 0.82, 0.92, 0.98, 0.03, 0.06, 0.11, 0.24, 0.35, 0.46, 0.72, 0.76, 0.93, 0.07, 0.28, 0.42, 0.53, 0.69, 0.80, 0.83, 0.94)

Under short UV (254 nm) , 9 peaks were detected. Among them maximum percentage of area were occupied by Rf 0.05 (58.24%), 0.15 (11.15%)

Under long UV (366 nm) , 10 peaks were detected. Among them maximum percentage of area were occupied by Rf 0.93 (33.38%) , 0.11 (15.37%), 0.82 (15.32%)

After derivatization (620 nm), 9 peaks were detected. Among them maximum percentage of area were occupied by Rf 0.07 (60.28%), 0.53 (11.57%)

Figure 9. HPTLC photo documentation of ethanol extract of Dhataki pushpa



Short UV	Long UV	After derivatisation

Solvent system - Toluene: Ethyl Acetate: Methanol: Formic acid (3.0: 3.0: 0.8: 0.2)

Track 1 – **Dhataki pushpa**– 4µl
Track 2 – **Dhataki pushpa**– 8µl
Track 2 – **Dhataki pushpa**– 12µl

Table No - 9: R_f values of sample of Dhataki pushpa (Woodfordia fruticosa (L.) Kurz

Short UV	Long UV	After derivatisation
0.25 (Green)	-	-
-	-	0.27 (L. purple)
-	-	0.41 (L. purple)
-	0.43 (F. blue)	-
-	0.48 (F. red)	-
0.53 (Green)	-	0.53 (L. purple)
-	0.56 (F. red)	-
-	0.64 (F. blue)	-
-	0.72 (F. blue)	-
0.75 (Green)	-	-
-	-	0.79 (D. purple)
-	0.89 (F. blue)	-

***D – dark; L – light; F – fluorescent**

HPTLC Densitometric Scan

Total 25 number of active components were detected in *Woodfordia fruticosa* (L.) Kurz having Rf value (0.05, 0.32, 0.43, 0.58, 0.63, 0.66, 0.87, 0.04, 0.06, 0.20, 0.23, 0.27, 0.39, 0.50, 0.53, 0.55, 0.62, 0.67, 0.76, 0.85, 0.92, 0.10, 0.34, 0.49, 0.93)

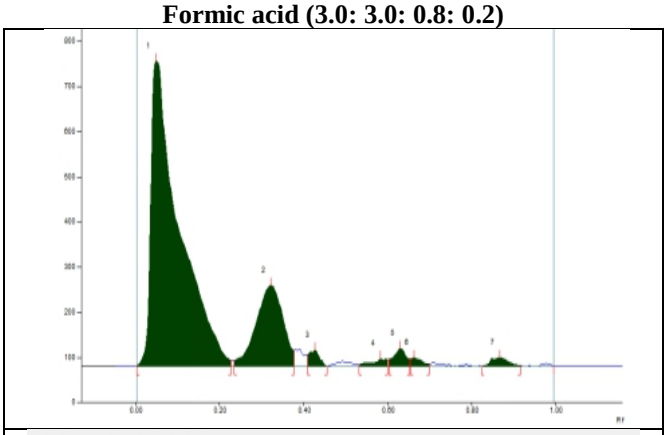
Under short UV (254nm), 7 peaks were detected. Among them maximum percentage of area were occupied by Rf 0.05 (68.70%), 0.32 (18.27%)

Under long UV (366nm), 15 peaks were detected. Among them maximum percentage of area were occupied by Rf 0.67 (17.14%), 0.58 (10.35%), 0.55 (8.82%)

After derivatization (620nm), 7 peaks were detected. Among them maximum percentage of area were occupied by Rf 0.06 (38.40%), 0.05 (38.32%)

In *Woodfordia fruticosa* (L.) Kurz **[**Solvent system - Toluene: Ethyl Acetate: Methanol: Formic acid (3.0: 3.0: 0.8: 0.2)] - 12 spots were detected in different Rf value.

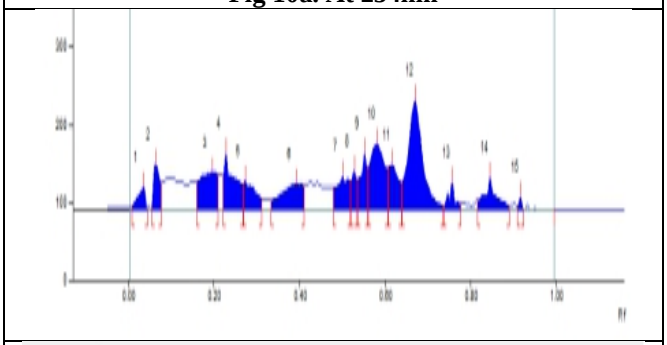
Figure 10. Densitometric scan of Dhataki pushpa (*Woodfordia fruticosa* (L.) Kurz)
Solvent system - Toluene: Ethyl Acetate: Methanol: Formic acid (3.0: 3.0: 0.8: 0.2)



Track 3, ID: Dhataki pushpa

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.00 Rf	0.8 AU	0.05 Rf	674.3 AU	68.70 %	0.23 Rf	13.2 AU	32468.8 AU	74.57 %
2	0.24 Rf	13.0 AU	0.32 Rf	179.3 AU	18.27 %	0.38 Rf	35.5 AU	8235.5 AU	18.91 %
3	0.41 Rf	25.2 AU	0.43 Rf	35.7 AU	3.64 %	0.46 Rf	0.5 AU	647.3 AU	1.49 %
4	0.53 Rf	3.5 AU	0.58 Rf	16.1 AU	1.64 %	0.60 Rf	14.6 AU	438.3 AU	1.01 %
5	0.60 Rf	15.6 AU	0.63 Rf	39.1 AU	3.99 %	0.65 Rf	16.0 AU	815.5 AU	1.87 %
6	0.65 Rf	16.1 AU	0.66 Rf	18.2 AU	1.85 %	0.70 Rf	5.7 AU	368.3 AU	0.85 %
7	0.82 Rf	0.6 AU	0.87 Rf	18.8 AU	1.91 %	0.92 Rf	1.4 AU	566.8 AU	1.30 %

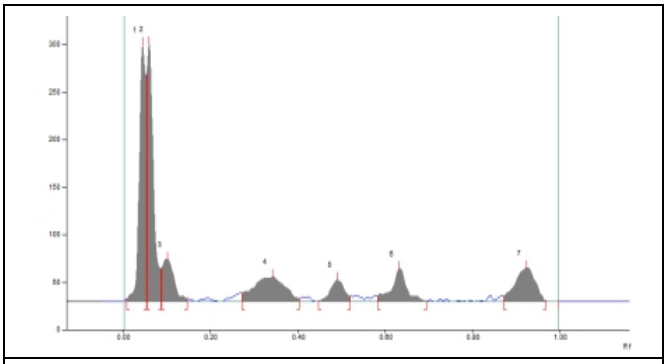
Fig 10a. At 254nm



Track 3, ID: Dhataki pushpa

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.01 Rf	4.3 AU	0.04 Rf	27.9 AU	3.42 %	0.05 Rf	0.6 AU	365.1 AU	2.37 %
2	0.05 Rf	4.0 AU	0.06 Rf	58.5 AU	7.17 %	0.08 Rf	35.2 AU	614.0 AU	3.99 %
3	0.16 Rf	36.1 AU	0.20 Rf	48.2 AU	5.91 %	0.21 Rf	46.3 AU	1403.5 AU	9.13 %
4	0.22 Rf	44.2 AU	0.23 Rf	71.3 AU	8.74 %	0.27 Rf	32.9 AU	1329.2 AU	8.65 %
5	0.27 Rf	33.4 AU	0.27 Rf	37.0 AU	4.53 %	0.31 Rf	15.1 AU	740.5 AU	4.82 %
6	0.33 Rf	12.8 AU	0.39 Rf	33.8 AU	4.15 %	0.41 Rf	31.1 AU	1221.0 AU	7.94 %
7	0.48 Rf	28.4 AU	0.50 Rf	42.8 AU	5.25 %	0.52 Rf	36.2 AU	873.9 AU	5.68 %
8	0.52 Rf	36.3 AU	0.53 Rf	48.7 AU	5.97 %	0.54 Rf	39.1 AU	422.8 AU	2.75 %
9	0.54 Rf	39.6 AU	0.55 Rf	71.9 AU	8.82 %	0.56 Rf	52.9 AU	777.9 AU	5.06 %
10	0.56 Rf	53.9 AU	0.58 Rf	84.4 AU	10.35 %	0.61 Rf	53.6 AU	2007.1 AU	13.06 %
11	0.61 Rf	54.1 AU	0.62 Rf	57.3 AU	7.03 %	0.64 Rf	35.8 AU	968.8 AU	6.30 %
12	0.64 Rf	36.0 AU	0.67 Rf	139.7 AU	17.14 %	0.74 Rf	5.1 AU	3418.8 AU	22.24 %
13	0.74 Rf	5.0 AU	0.76 Rf	35.3 AU	4.32 %	0.78 Rf	8.8 AU	364.1 AU	2.37 %
14	0.82 Rf	11.7 AU	0.85 Rf	41.4 AU	5.08 %	0.89 Rf	4.7 AU	782.6 AU	5.09 %
15	0.91 Rf	3.3 AU	0.92 Rf	17.1 AU	2.10 %	0.93 Rf	0.5 AU	83.6 AU	0.54 %

Fig 10b. At 366nm



Track 3, ID: Dhataki pushpa

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.01 Rf	3.0 AU	0.05 Rf	270.6 AU	38.32 %	0.05 Rf	35.6 AU	2895.5 AU	27.42 %
2	0.05 Rf	241.7 AU	0.06 Rf	271.2 AU	38.40 %	0.09 Rf	34.3 AU	2872.2 AU	27.20 %
3	0.09 Rf	34.9 AU	0.10 Rf	44.9 AU	6.36 %	0.15 Rf	3.6 AU	868.6 AU	8.23 %
4	0.27 Rf	8.7 AU	0.34 Rf	26.2 AU	3.71 %	0.40 Rf	3.4 AU	1358.5 AU	12.87 %
5	0.45 Rf	0.6 AU	0.49 Rf	22.8 AU	3.23 %	0.52 Rf	5.7 AU	522.0 AU	4.94 %
6	0.58 Rf	5.5 AU	0.63 Rf	35.1 AU	4.97 %	0.70 Rf	0.3 AU	857.9 AU	8.13 %
7	0.87 Rf	5.9 AU	0.93 Rf	35.4 AU	5.02 %	0.97 Rf	0.8 AU	1183.3 AU	11.21 %

Fig 10c. At 620nm

DISCUSSION`

The macroscopic and microscopic evaluation have confirmed the authenticity and purity of both Yashtimadhu (*Glycyrrhiza glabra* Linn) and Dhataki (Woodfordia fruticosa (L.) Kurz). Physico- chemical evaluation of the drug indicates their quality, purity and presence of other admixture. Loss on drying determine the moisture content of a drug. It aids to prevent the decomposition of the drugs either due to chemical change or microbial contamination. Ash value is the residue remaining after incineration is the ash content of the drug. These could be inorganic salts such as carbonates, sulphates, phosphates, silicates etc. naturally occurring in the drug or adhered to it or deliberately added to it in order to adulterate the drug. Total ash is to measure the total amount of plant material remaining after ignition of the drug. Acid insoluble ash is the residue obtained after boiling the total ash either with dilute hydrochloric acid or water which measures the amount of sand and silica matter present in the drug. Extractive value measures the nature of the chemical constituents present in a crude drug. It is essential for the estimation of specific chemical constituents soluble in that particular solvent used for extraction. In preliminary phytochemical analysis showed Carbohydrate, Tannin, Flavanoids, Saponins, Phenols, Carboxylic acid, Quinone are common chemical constituents in both *Glycyrrhiza glabra* Linn. and *Woodfordia fruticosa* (L.) Kurz. Alkaloids , Steroids, Triterpenoid, Resins are present only in *Glycyrrhiza glabra* Linn. Coumarins, Amino acids are absent in both *Glycyrrhiza glabra* Linn. and *Woodfordia fruticosa* (L.) Kurz. According to previous studies- Tannins, Flavonoids, Saponins, Phenols, Carbohydrates, Carboxylic acids, Quinone showed antimicrobial properties.[25- 33] Saponins have been reported to possess a

wide range of biological activities. The mode of action of antibacterial effects of saponins seems to involve membranolytic properties, rather than simply altering the surface tension of the extracellular medium, thus being influenced by microbial population density (Killeen *et al*., 1998).[34] The action mechanisms of saponins may lie in damage to the membrane and leakage of cellular materials, ultimately leading to cell death (Mshvildadze *et al*., 2000).[35] Flavonoids are phenolic structures containing one carbonyl group. Their activity is probably due to their ability to complex with extracellular and soluble proteins and to complex with bacterial cell walls (Cowan, 1999).[36] More lipophilic flavonoids may also disrupt microbial membranes (Tsuchiya *et al*., 1996).[37] Alkaloids isolated from plant are commonly found to have antimicrobial properties (Omulokoli *et al*., 1997).[38] Berberine and harmane are important representatives of the alkaloid group. Their mechanism of action is attributed to their ability to intercalate with DNA of bacteria (Phillipson and O′Neill, 1987).[39] The astringent character of tannins may induce complexation with enzymes or substrates. Many microbial enzymes were found to be inhibited when raw culture filtrates or purified enzymes were mixed with tannins.[40] Quinones are known to complex irreversibly with nucleophilic amino acids in proteins , often leading to inactivation of the protein and loss of function. For that reason, the potential range of quinone antimicrobial effects is great. Probable targets in the microbial cell are surface-exposed adhesins, cell wall polypeptides, and membrane-bound enzymes. Quinones may also render substrates unavailable to the microorganism.[36]

HPTLC

The bands run through TLC with numerous Rf values represents the individual phytoconstituents. In the result common Rf value of the both plants is detected.

In *Dhataki* (*Woodfordia fruticosa* (L.)kurz) using Solvent system – Toluene: Chloroform: Ethyl Acetate: Formic acid (2: 6: 6 : 2) and *Yashtimadhu* (*Glycyrrhiza glabra* Linn) using Solvent system – n- butanol: Water: Glacial acetic acid (7: 2: 1) , the common active component was detected in **Rf value 0. 05** which occupied 2.17% (254 nm) and 1.80% (620 nm) area in *Glycyrrhiza glabra* Linn. and 54.08% (254 nm) area in *Woodfordia fruticosa* (L.) kurz. **Rf value 0.24** which occupied 53.01% (620 nm) area in Glycyrrhiza glabra Linn. and 8.06% (366 nm) area in *Woodfordia fruticosa* (L.) Kurz. **Rf value 0.42** which occupied 7.68% (620 nm) in *Glycyrrhiza glabra* Linn. and 3.36% (620 nm) area in *Woodfordia fruticosa* (L.) Kurz. **Rf value 0.72** which occupied 35.85% (366 nm) in *Glycyrrhiza glabra* Linn. and 0.71% (366 nm) area in *Woodfordia fruticosa* (L.) Kurz .

In *Dhataki* (*Woodfordia fruticosa* (L.) Kurz) using Solvent system – Toluene: Ethyl Acetate: Methanol: Formic acid (3: 3: 8: 2) and *Yashtimadhu* (*Glycyrrhiza glabra* Linn.) using Solvent system – n- butanol: Water: Glacial acetic acid (7: 2: 1) , the common active component was detected in **Rf value 0.05** which occupied 2.17% (254 nm) area and 1.80% (620 nm) area in *Glycyrrhiza glabra* Linn. and 74.57% (254 nm) area and 27.42% (620 nm) area in *Woodfordia fruticosa* (L.) Kurz. **Rf value 0.20** which occupied 4.87% (366 nm) area in *Glycyrrhiza glabra* Linn. and 9.13% (366 nm) area in *Woodfordia fruticosa* (L.) Kurz.

The common and nearer value is required to bring same or nearer therapeutic effect. The results from HPTLC finger prints, scanned at various wavelengths revealed that there are many compounds in both drugs i.e. *Glycyrrhiza glabra* Linn. & *Woodfordia fruticosa* (L.) Kurz. From those chromatograms, it has been found that alcoholic extracts of either of the drugs contain multiple spots and peaks representing multiple components responsible for specific therapeutic effect in the body. Major therapeutic effect is brought by the Rf value which are in Higher concentration.[178]

These results provide a very important clue for further isolation of pure compounds which are shown in higher concentration in chromatography to elicit the exact bio chemical compound involve in performing the respective activities.

CONCLUSION

The pharmacognostical studies have confirmed the authenticity and purity of both the *Yashtimadhu* (*Glycyrrhiza glabra* Linn.) and *Dhataki* (*Woodfordia fruticosa* L. Kurz.). Both the drugs have few commom phytochemicals like carbohydrates, tannins, flavanoids, saponins, phenol, carboxylic acid, quinone**.** According to previous studies- Tannins, Flavonoids, Saponins, Phenols, Carbohydrates, Carboxylic acids, Quinone showed antimicrobial properties**.** Chromatographic fingerprinting showed some similarities in both Yashtimadu (Glycyrrhiza glabra Linn.) and Dhataki (Woodfordia fruticosa (L.) Kurz.) which detected the common active component. Thus one can used *Dhataki* (*Woodfordia fruticosa* L. Kurz.) as a substitute for *Yashtimadhu* (*Glycyrrhiza glabra* Linn.) in its absence.

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REFERENCES

1. BadkhaneYogesh,Yadav A.S, Bajaj. A, Sharma. AjitK,Raghuwanshi D.K. Glycyrrhizaglabra L. A miracle medicinal herb . Indo American Journal of Pharmaceutical Research. 2014; 4(12): p. 5808- 16
2. KR. Akshay, N. Sudharanal, et.al. Biodiversity and strategies for conservation of rare, endangered and threatened medicinal plants. Journal of Pharmacognosy and Phytochemistry. 2014; Vol 2(3) : p. 12- 20
3. SharmaP.C, Yelne M.B, DennisT.J.Database on Medicinal Plants Used in Ayurveda Vol3. 2nd . New Delhi: Central Counsil for Research in Ayurveda & Siddha; 2005: p.561- 6
4. Chaudhuri. A.B. Endangered Medicinal Plants. New Delhi: Daya Publishing House; 2014: p. 111

5. Srivastava. M , et.al. In vitro conservation of Glycyrrhiza glabra by slow growth culture. International Journal of Bio- Technology & Research. (IJBTR) 2013; Vol 3(1): p. 49- 58
6. Misra Siddhi Nandan, editor. Bhaisajya Ratnavali of Kaviraj Govind Sen; Abhavaprakarana: Chapter 4, Verse 29. Varanasi: Chaukhambha Surbharati Prakashan, 2016; p. 72.
7. Shastri Brahmashankar Bhisagratna, editor. Yogaratnakara by Vaidya Laksmipati Shastri, Purvaardhagata; Abhavavarga, Verse 13. Varanasi: Chaukhambha Prakashan, 2015; p. 171
8. Misra Bhavamasankara, Vaisya Rupalalaji, editor, 9th. Bhavaprakasha of Sri Bhavamisra Prathamabhaga; Verse 148. Varanasi: Chaukhambha Sanskrit Sansthan, 1999; p. 182
9. Sastry. J.L.N. Dravyaguna Vijnana, 1st ed. Varanasi: Chaukhambha Orientalia; 2014. p. 152
10. Hegde Prakash, A Harini. A text bok of dravyaguna vijnana, Vol 2. 1st ed. New Delhi: Chaukhambha publication; 2014. p. 903
11. The Ayurvedic Pharmacopoeia of India; Ministry of Health and Family Welfare, Department of Health, Govt of India, Vol1; 1st ed. p. 127- 8
12. SharmaP.C, Yelne M.B, DennisT.J.Database on Medicinal Plants Used in Ayurveda Vol3.2nd . New Delhi: Central Counsil for Research in Ayurveda & Siddha; 2005: p.561- 6
13. Anderson DM, SmithWG. The antitussive activity of of glycyrrhetinic acid and its derivatives. *J.Pharm. Pharmacol*. Jul 1961;13:396-404
14. Krausse R, Bielenberg J, Blaschek W, Ullmann U. In vitro anti-Helicobacter pylori activity of Extractum liquiritiae, glycyrrhizin and its metabolites. *J Antimicrob Chemother* Jul 2004;54(1): 243-6
15. Cooper H, Bhattacharya B, Verma V, McCulloch AJ, Smellie WS, Heald AH. Liquorice and soy sauce, a life-saving concoction in a patient with Addison's disease. *Ann Clin Biochem.* Jul 2007;44(4):397-9.
16. Ross EJ. Liquorice and Addison's disease. *Br Med J. Jun* 20 1970;2(5711):733
17. Dhiman RK, Chawla YK. Herbal medicines for liver diseases. *Dig Dis Sci*. Oct 2005;50(10): 1807-12
18. Kim YW, Kang HE, Lee MG, Hwang SJ, Kim SC, Lee CH, et al. Liquiritigenin, a flavonoid aglycone from licorice, has a choleretic effect and the ability to induce hepatic transporters and phase-II enzymes. *Am J Physiol Gastrointest Liver Physiol*. Feb 2009;296(2):372-81
19. Kirtikar K R, Basu BD. Indian Medicinal Plants, 2nd ed. Dehradun: International Book Distributors; 2008. Vol 2. p. 1074- 5
20. Dey Dipali, Das M N, Sharma A K, O P Gupta editor , Pharmacognosy of Indigenous Drugs, New Delhi: Central Council for Research In Ayurveda and Siddha; 2005. Vol 3. p. 1163
21. SharmaP.C, Yelne M.B, DennisT.J.Database on Medicinal Plants Used in Ayurveda Vol3. 2nd . New Delhi: Central Counsil for Research in Ayurveda & Siddha; 2005: p. 206
22. Baravalia Y, Chanda S; Protective effect of Woodfordia fruticosa flowers against acetaminophen-induced hepatic toxicity in rats. Pharm Biol, 2011;49(8):826-832
23. Gupta A.K, Tandon Neeraj, Sharma Madhu. Quality standards of Indian Medicinal Plants. New Delhi: Indian Council of Medical Research ; 2006
24. Khandelwal KR. Practical Pharmacognosy; 29th edition. Pune: Nirali Prakashan; 2018
25. Hisanori Akiyama,et.al. Antibacterial action of several tannins against Staphylococcus aureus, Journal of Antimicrobial Chematherapy, 2001
26. Augustin Scalbert. Antimicrobial properties of tannins, Phytochemistry, 1991
27. Nouioua wafa, et.al. The antioxidant and antimicrobial activities of flavonoids and tannins extracted from Phlomis bovei De Noe, European Journal of Experimental Biology, 2016
28. Michal Arabski, et.al. Effect of Saponins against clinical E.coli strains and Eukaryotic cell line, Journal of Biomedicine and Biotechnology, 2012
29. Joshita Sabbineni. Phenol- An efffective antibacterial agent, Journal of Medical & Organic Chemistry, 2016
30. WD Armstrong. Antibacterial effects of Quinones, SAGE journals, 1943
31. Paul M Borick. Antimicrobial activity of some higher Amine Salts of Carboxylic acids, Applied Microbiology, 1961
32. Escoula L. Effeect of carbohydrates on the antbacterial activity of patulin on Steptococcus bovis, Pubmed , 1982
33. Sugars in human mother's milk are non toxic antibacterial agents, Vanderbilt institute, 2017
34. Killeen, G., Madigan, C., Connolly, C., Walsh, G., Clark, C., Hynes, M., Timmins, B., James, P., Headon, D., Power, R., 1998. Antimicrobial saponins of Yucca schidigera and the implications of their in vitro properties for their in vivo impact. J. Agric. Food Chem., 46(8):3178- 86. [doi:10.1021/jf970928j]
35. Mshvildadze, V., Favel, A., Delmas, F., Elias, R., Faure, R., Decanosidze, Q., Kemertelidze, E., Balansard, G., 2000. Antifungal and antiprotozoal activities of saponins from Hedera colchica. Pharmazie, 55(4):325- 6.
36. Cowan M., 1999. Plant products as antimicrobial agents. Clinical Microbiology Review, 12(4):564- 82.
37. Tsuchiya, H., Sato, M., Miyazaki, T., Fujiwara, S., Tanigaki, S., Ohyama, M., Tanaka, T., Iinuma, M., 1996. Comparative study on the antibacterial activity of phytochemical flavanones against methicillin-resistant Staphylococcus aureus. J. Ethnopharmacol., 50(1):27-34.
38. Omulokoli, E., Khan, B., Chhabra, S., 1997. Antiplasmodial activity of four Kenyan medicinal plants. J. Ethnopharmacol., 56(2):133- 7. [doi:10.1016/S0378-8741(97)01521-3]
39. Phillipson, J., O'Neill, M., 1987. New leads to the treatment of protozoal infections based on natural product molecules. Acta Pharm. Nord., **1**:131- 44.
40. Scalbert Augustin. Antimicrobial properties of Tannins. Phytochemistry, Vol. 30, No. 12. pp. 3875- 83, 1991.