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Research Article

PHARMACOGNOSTICAL AND ANALYTICAL EVALUATION OF KARPASA (*Gossypium herbaceum* Linn.) ROOT

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Abstract

Gossypium herbaceum Linn. known as Karpasa belongs to the family Malvaceae is used in Ayurveda to treat various diseases and for processing various formulations in Rasashastra and Bhaishajyakalpana. In the present study, transverse section of fresh roots of karpasa showed, cortex with pericyclic fibres, prismatic crystals of calcium oxalate and starch grains. Vascular bundle shows the phloem above the xylem, xylem radially arranged with biseriate to multiseriate medullary rays. Presence of lysigenous cavity in the medullary ray is a special character. The physico-chemical parameters like, pH of Karpasa root (KR) was 7.2, the loss on drying was 6.47 % w/w and the alcohol soluble extractive was 8.80 % w/w. TLC profile of Karpasa root showed R_f 0.04 & 0.50 at 254 nm frequency and at 366 nm respectively.

Key Words: Karpasa root; Pharmacognosy; Phytochemical parameters.

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INTRODUCTION

Karpasa *Gossypium herbaceum* Linn. is an annual shrub. It belongs to the family Malvaceae and possesses the following synonyms viz. Karpasaaki, Samudranta, Tundikeri, Cavya, Picu. In Ayurveda the properties of roots are Katu (Pungent), Kashaya (Astringent) in rasa; Laghu (Light), Tikshna (Penetrating) guna; Ushna (Hot) virya and Katu (Pungent) vipaka. Its doshagnata is kaphapitashamaka. Karma of roots is Garbhashayasankochana (Uterus stimulant) and Artavajana (increases menses flow). It is used in Anartava (Amenorrhoea), Kashtartava (Dysmenorrhoea) and Prasutipashchatavikara (Purpueral disorders).[1][2] Roots are thermogenic, emollient, abortifacient, emmenogougue, diuretic, haematopurative[3] and root bark is anticancerous.[4] Root contains a polyphwnolix toxic compound known as Gossypol. Gossypol is a male contraceptive.[5] It also assists menstrual flow and effectively inhibits egg implantation.[6,7] Gossypol and its derivatives have been shown to have significant antimicrobial activity as well as wound healing effect.[8] It is reported to kill herpes virus.[9] Root is abortifacient and has uterus stimulating activity therefore it is mostly used in menstrual disorders.[10] In the present study an attempt has been made to establish the analytical and pharmacognostical standards especially for the roots.

MATERIAL AND METHODS

Collection:

The fresh roots of Karpasa (*Gossypium herbaceum* Linn.) were collected from Gondkhaire Dist., Nagpur. The collected sample was identified, authenticated by using various floras and texts. The verified specimen was preserved in the Pharmacognostical departmental herbarium museum of IPGT and RA, GAU, Jamnagar vide no. 6034/2012 for future reference. The sample was preserved in the solution of FAA (70% Ethyl alcohol: Glacial acetic acid: Formalin in the ratio of 90:5:5) for the histological profile.[11]

Pharmacognostic evaluation:

Macroscopic evaluation: Macroscopic characters were recorded as per visual observations and by comparing with various Floras and Texts.[12][13][14]

Organoleptic evaluation: The colour, odour and taste of the root was recorded separately.[15]

Microscopic evaluation: Free hand sections were taken cleared with chloral hydrate followed by phloroglucinol and hydrochloric acid. Microphotographs were taken by using Carl Zeiss binocular microscope.[16]

Powder microscopy: Cut pieces of the roots were dried under shade, powdered with the help of mechanical grinder and sieved through mesh no. 60. Karpasa root powder was studied under the microscope with distilled water. Microphotographs were taken by using Carl Zeiss binocular microscope.[17]

Histochemical tests:

To detect the site of location of various constituents of the drug, sections of roots was treated with various reagents like ruthenium red (for mucilage), FeCl3 for (tannin) and iodine for (starch grains). Histochemical tests for few constituents like Calcium oxalate were also carried out.[18]

Physicochemical parameters:

In physical evaluation, moisture content, ash values viz., total ash, acid insoluble ash, and extractive values viz., alcohol soluble extractive value, water soluble extractive values were determined. The determinations were performed in triplicate and results are expressed as mean ± SD. The percentage w/w values were calculated with reference to the air-dried drug. [19][20][21]

Preliminary Phytochemical:

Preliminary phytochemical investigations were carried out by following standard procedure of API. [22]

TLC:

TLC was performed as per the guidelines provided in API. Methanol extract of root powder was used for spotting. TLC was performed by using Chloroform + Methanol (8:2) v/v solvent system.[23]

RESULTS

Pharmacognostical evaluation:

Macroscopical study:

The root system consists of a long woody cylindrical tap root and a few lateral roots with their branches. The lateral roots are often as long as the tap root and fairly thick. They may attain half to one meter or more in length and about one cm. in diameter. The outer surface of the root, when fresh is light reddish yellow to yellowish brown in colour and shows the presence of lenticels, many root-lets and scars of fallen rootlets. The lenticels are many in number, prominent, protruding fairly long and tangentially elongated. Those towards the upper or basal part of the root are often arranged closer together. The surface skin is very thin. It can be easily scraped off exposing

a smooth cream white tissue. In transverse section of mature root of about one cm in diameter the entire bark appears whitish and the wood forms the bulk part of the root. It appears dull or yellowish white, minutely diffusely porous, slightly hard and with several whitish radial lines or streaks. There is no pith in the centre; the root has no particular odour or taste. (Fig.1.2)

Figure 1:Transeverse section of Karpasa root

Fig.1.1: Karpasa with flower

Fig.1.2: Root's morphology

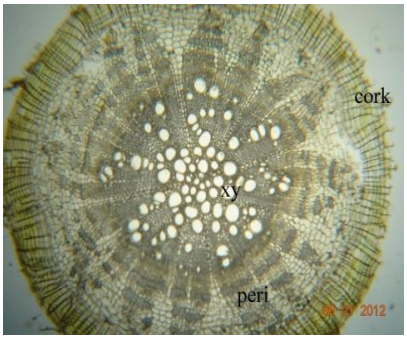

Fig.1.3: T.S of Root

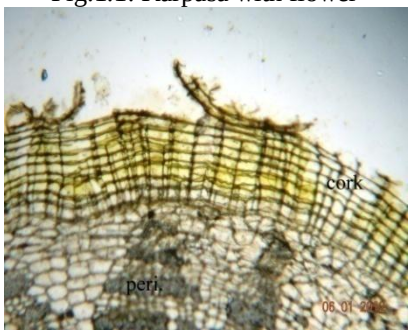

Fig.1.4: Cork cells, Pericyclic fibers.

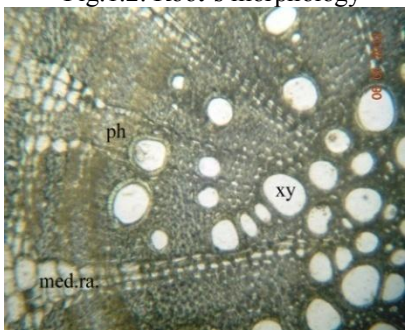

Fig.1.5: Medullary rays, xylem, phloem.

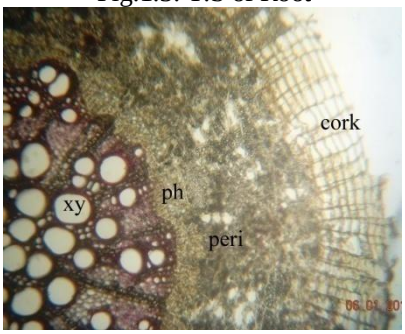

Fig.1.6: Cork, Pericyclic fiber, phloem & stained xylem & medullary rays.

Fig.1.7: Unstained phloem, stained xylem, xylem parenchyma & medullary rays.

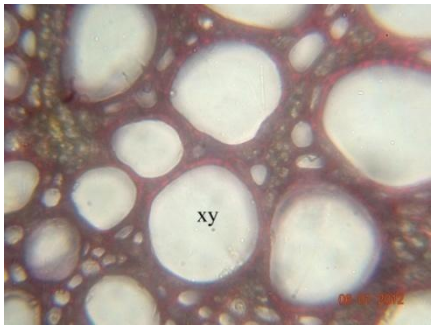

Fig.1.8: Xylem

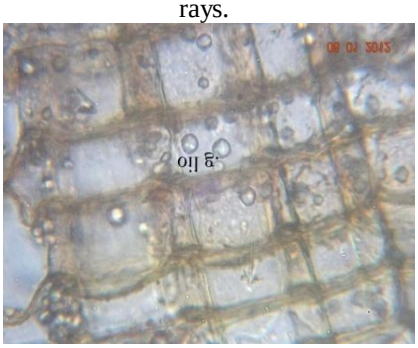

Fig.1.9: Cork cells embedded with oil globules

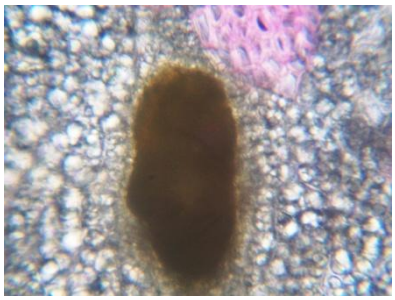

Fig.1.10: Lysigenous cavity

Microscopic study:

The outermost tissue namely the cork or phellem is composed of 12 – 18 or occasionally fewer 4-6 rows of thin walled rectangular tangentially elongated cells – the tangential length being twice the width. The peripheral rows are slightly compressed and their cell walls appear somewhat wavy and are light yellow to yellowish brown in colour. (Fig. 1.4 & Fig.1.9)

Next within is a comparatively narrow zone of phelloderm composed of more than three rows of thin walled rectangular cells, similar to those of inner rows of the cork. Many of these cells contain simple as well as compound starch grains which are generally small compared to those present in the phloem. Inner to the phelloderm is a zone of pericyclic parenchyma which extends inside as far as the phloem (Fig.1.4). The parenchyma cells are thin walled and larger being somewhat broader and more tangentially elongated.

Figure 2: Powder microscopy of Karpasa root

Fig.2.1: Powder of root

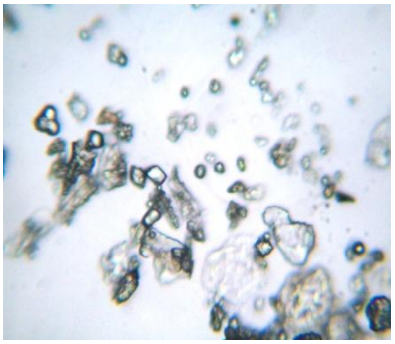

Fig.2.2: Compound starch grains & prismatic crystal

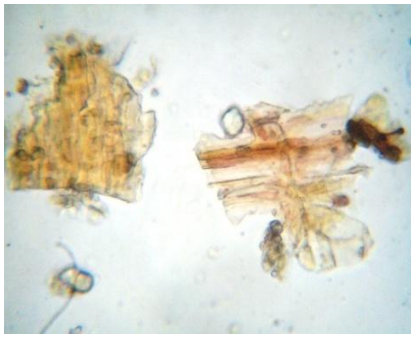

Fig.2.3:Cork cells, fiber fragment

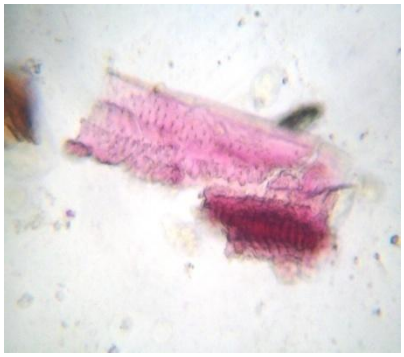

Fig.2.4: Fragment of lignified pitted vessel

Fig.2.5: Fragment of lignified annular vessel

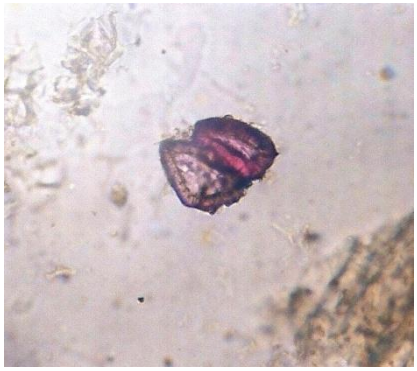

Fig.2.6: Lignified stone cells

Most of the cells are abundantly packed with starch grains both simple and compound and a considerable number of these cells contain fairly large sized prismatic crystals of calcium oxalate. The starch grains appear larger. The compound grains are composed of 2 to 4 or very rarely 6 components.

The region next inside is the phloem that occupies the major part of the bark. As seen in transverse section it occurs in the form of wedge shaped strips or patches of varying width with the apices towards the periphery and radially alternating with or separated by the widened distal ends of the vascular (phloem) rays. Each such strip of phloem shows a number of 8 to 12 or more narrow tangential bands or groups of phloem fibres separated by wider zones of thin walled phloem elements. (Fig.1.5 & Fig.1.6) The groups of fibre cells located towards the interior are larger and in transverse section the fibres are thick-walled.

In the radial strips of xylem the lignified elements such as the vessels are found distributed in a scattered manner and mostly occur in small groups of two or three or occasionally as solitary members. They are comparatively wide. The walls appear pitted. The unlignified xylem parenchyma is paratracheal. The cells are much wider than the fibres in T.S. and those present elsewhere. The centre of the root is occupied by the primary xylem surrounded by a small amount of secondary xylem. The primary xylem is tetrarch. The medullary rays are generally two to four seriate. (Fig.1.5 & Fig.1.7) Uniseriate rays are also common but are shorter than the others. Many or the rays start from very near the center of the root. All the ray cells are thin walled and radially elongated, the radial length being four or five times the breadth. They contain starch grains which may be simple or compound or occasionally aggregated. Some of the ray cells contain rhomboidal crystals of calcium oxalate.

Powder microscopy:

Organoleptical evaluation:

Organoleptic evaluation of *Gossypium herbaceum* Linn. root is Katu (Pungent) Kashaya (Astringent) taste, pale yellowish in colour, pungent odour and the texture of powdered root is smooth. (Table 1)

The diagnostic characters of powdered root showed prismatic crystals of calcium oxalate having a diagonal length of 21µ, simple and compound starch grains varies from 9 to 20 µ in diameter, fibres from cortical region, brown pigment tannin content cell from cortex zone, fragment of cork in surface view, lignified stone cells varies from 27 to 54 µ in diameter, fragment of lignified pitted and annular vessels from stelar region varies from 120 to 320 µ in length and 21 to 192 µ in diameter. (Figure 2)

Table 1: Organoleptic evaluation of Karpas root powder

Sr. No.	Character	Karpasa root
1.	Taste	Katu (Pungent) Kashaya(Astringent)
2.	Colour	Pale yellow
3.	Odour	Pungent
4.	Texture	Smooth

TLC:

In TLC profile at 254 nm frequency, in Karpasa roots one spot was observed, R_f value 0.04. At 366 nm KS root one spot was observed, R_f values 0.50. After spray it showed two spots, their R_f values are 0.06, 0.51. (Table 5)

Table 2: Histochemical tests for Karpasa root

Sr. No.	Reagents	Observation	Characteristics
1.	Phloroglucinol+Conc. Hcl	Red	Lignified cells
2.	Iodine	Blue	Starch grains
3.	Phloroglucinol+Conc. Hcl	Dissolved/ effervescence	Calcium oxalate crystals
4.	Fecl3 solution	Dark blue to black	Tannin
5.	Ruthenium red	Red	Mucilage

Phytochemical Study:

Physicochemical parameters:

The pH of Karpasa root was 7.2, the loss on drying 6.47 w/w, ash value 5.2 % w/w, the acid insoluble ash 0.02 w/w, water soluble extractive 5.6 % w/w and the alcohol soluble extractive was 8.80 % w/w. (Table 3)

Preliminary Phytochemical:

While analyzing the Karpasa roots for qualitative analysis alkaloids, glycosides, flavanoids, proteins and sterol were absent, whereas starch, phenols, tannin, saponin and carbohydrates were present. (Table 4)

Table 3: Physicochemical analysis of Karpasa root:

Parameter	Karpasa root
pH value	7.2
Loss on drying	6.47
Ash value (%w/w)	5.2
Acid insoluble ash (%w/w)	0.02
Water soluble extract (%w/w)	5.6
Alcohol soluble extract (%w/w)	8.80

Table 4: The results of qualitative test of Karpasa root for various functional groups

Tests	Karpasa root
Alkaloids	-Ve
Glycosides	-Ve
Starch	+Ve
Flavanoids	-Ve
Proteins	-Ve
Sterol/Steroid	-Ve
Tannin	+Ve
Phenols	+Ve
Saponin	+Ve
Carbohydrates	+Ve

+Ve = Present, -Ve = Absent

Table 5: R_f values of methanolic extract of Karpasa root in TLC

Sr. No.	Sample	Before spraying 254 nm (Short U.V.)		Before spraying 366 nm (Long U.V.)		After spraying 10% Ferric Chloride	
		Spots	Rf value	Spots	Rf value	Spots	Rf value
1.	Karpasa root	1	0.04	1	0.50	2	0.06, 0.51

DISSCUSSION

The microscopic studies of the transverse section showed the parts from cork to xylem. The structure of cork, pericyclic fibres, phloem, lysigenous cavities, medullary rays and xylem are the distinguishing features of the root. Calcium oxalate crystals, starch grains, lignified fibres, pitted and annular vessels, tannin content, stone cells are found in powder microscopy.

The pH of Karpasa root was 7.2, the loss on drying 6.47 w/w., ash value 5.2 % w/w, the acid insoluble ash 0.02 w/w, water soluble extractive 5.6 % w/w. and the alcohol soluble extractive 8.80 % w/w. Phenols, tannin, starch, saponin and carbohydrates were present in Karpasa root. All other components were found to be absent.TLC profile at 254 nm frequency, one spot, R_f value 0.04 & at 366 nm Karpasa root one spot, R_f values 0.50. After spray two spots, R_f values are 0.06, 0.51.

CONCLUSION

The Diagnostic morphological and microscopic characters were noted down for easy identification of plant material. Physico-chemical parameters have been established to identify the quality and degree of purity of the plant material as per pharmacopoeial requirements. Qualitative tests indicated the presence of tannin, starch, saponin, calcium, mucilage, carbohydrate, phenolic compounds and TLC studies confirmed the same. The results are being reported for the first time, could be useful in the identification and standardization of a crude drug. The data produced in the present investigation is also helpful in the preparation of the crude drug's monograph and inclusion in various pharmacopoeias.

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